e-ISSN: 2458-8377 http://sjafs.selcuk.edu.tr

Selcuk Journal of Agriculture and Food Sciences

Number: 36 Volume: 3 Year: 2022
DECEMBER

Selcuk Journal of Agriculture and Food Sciences

Selcuk Journal of Agriculture and Food Sciences

Product Information

Publisher	Selçuk University Faculty of Agriculture
Owner (On Behalf of SUAF)	Prof. Dr. Zeki BAYRAMOĞLU Dean
Editor in Chief	Prof. Dr. Ercan CEYHAN, Selcuk University, Türkiye
Printing House	Selçuk University
Date of Publication	25.12.2022
Language	English
Frequency	Published three times a year
Type of Publication	Double-blind peer-reviewed, widely distributed periodical
Indexed and Abstracted in	TR DİZİN GOOGLE SCHOLAR SCIENTIFIC INDEXING SERVICES (SIS) ARAŞTIRMAX CAB ABSTRACTS CROSSREF CAB DİRECT MIAR SCILIT ESJİ Dimensions OAJI.net
Aims and Scope	Selcuk Journal of Agriculture and Food Sciences is unique journal covering mostly theoretical and applied all disciplines of agriculture, food and energy sciences such as agronomy, crop sciences, animal and feed sciences, poultry sciences, field crops, horticulture, agricultural microbiology, soil science, plant nutrition, agricultural engineering and technology, irrigation, land scape, agricultural economics, plant pathology, entomology, herbology, energy, biofuels and biomass, food chemistry, aroma, microbiology, food science and technology, biotechnology, food biotechnology, agricultural production, nutrition and related subjects.
Web Address	http://sjafs.selcuk.edu.tr/
Address	Selçuk University, Faculty of Agriculture, 42075, Konya, Türkiye Telephone : +90 (332) 223 28 56 Fax : +90 (332) 241 01 08 E-mail: eceyhan@selcuk.edu.tr

Selcuk Journal of Agriculture and Food Sciences

CONTENTS

Research Article

Christelle JOU-NTEUFA Ercan CEYHAN	Determination of Combining Ability and Heredity Through Diallel Analysis Method in F2 Populations of Cowpea	299-311
Ayşen AKAY Hasan Hüseyin ÖZAYTEKIN	Using Contamination Indices for Assessments of Heavy Metals Status of Soils around Mercury Mine, in Kurşunlu, (Konya) Province	312-319
Nur Banu TEKIN Ercan CEYHAN	Genetic Variability Studies in F2 Generations of Determinate High Yield Dry Bean Lines for Seed Yield and Yield Components	320-330
Şemsi TAMÜKSEK Ercan CEYHAN	Genetic Variability Studies in F2 Generations of Determinate High Yield Fresh Bean Lines for Seed Yield and Yield Components	331-341
Nuraniye ERUYGUR Mehmet TEKIN Erol DÖNMEZ	Pharmacognostic, Physicochemical, Phytochemical Screening and In-Vitro Antioxidant Activity of Chrysochamela Noeana, Endemic in Türkiye	342-348
Mehtap GÜRSOY	Importance of Priming Application Times on Growth, Relative Water Content and Photosynthetic Pigments of Rapeseed (Brassica napus ssp. oleifera L.) Cultivars Under Salinity Stress	349-355
Muhsin YEŞILYOL Mustafa TERIN İbrahim YILDIRIM	Factors Affective on The Consumers' Purchasing Behaviours in Shopping Malls: A Case Study in Eastern Part of Turkey	356-361
Metin TURAN Mostafa Fraidoon Faaek ALI Lütfi PIRLAK	Biochemical Effects of Drought Stress in Some Strawberry Cultivars	362-367
Osaihiiomwan OJOGHO Osagie Stephen IMADE	Food Expenditure Pattern of Household in Delta State, Nigeria: Economic Rationality Essentials	368-374
Khalil BOUNAAMA Abdelkader El-Amine DAHOU Zakaria MESKINI Abderrahmen ABBAD Hafida TAHLAITI Houari YEROU Abdelkader HOMRANI	Identification and Technological Characterisation of Lactic Acid Bacteria Isolated From Traditional Algerian Cheese "J'ben"	375-379
Nazik Meziyet DİLEK Mustafa KARAKAYA	Natural Alternative Curing Agent in Fermented Sucuk Production: Sugar beet (Beta vulgaris var. saccharifera L.) Molasses	380-386
Muzaffer İPEK Ahmet EŞİTKEN	Effects of Rhizobacteria on Plant Growth and Fruit Quality of Blackberry in Alkaline Soil	387-392
Jale BİLGİN Rafet ASLANTAŞ	The Effects of Some Post-Harvest Organic Acid Treatments on the Storage Quality of Brussels Sprouts	393-398
Ali Samet BABAOĞLU	Effects of Replacing Breadcrumbs with Buckwheat, Chickpea, Corn and Mil-let Flour in Gluten-Free Meatball Formulation	399-404
Seyfi TANER Şah İsmail CERİT Enes YAKIŞIR Musa TÜRKÖZ	Relationships between Some Spectral Traits and Grain Yield in Bread Wheat under Rainfed Conditions	405-409
Emine Burcu TURGAY Sibel BÜLBÜL Merve Nur ERTAŞ ÖZ Öztekin URLA	The Incidence of Barley (Hordeum vulgare L.) Leaf Diseases in Central Anatolia Region of Turkey in 2020	410-413
Fatma Rana BAYRAM Nuh POYRAZ Kemal KESENCİ	Determination of Disease Severity and Anastomosis Groups of Rhizoctonia solani Isolates from Chickpea Plant in Konya Province	414-420
Talha DEMİRCİ	Enumeration Of Bifidobacterium Spp., Lactobacillus Acidophilus and Starter Cultures from Commercial Probiotic Yogurts and Freeze-Dried Yogurt Starter Mixes	421-427

Ahmet TAMKOÇ Cafer TÜRKER	Effect of Temperature and Storage Time on Germination in Forage Peas	428-437
Resul MUT Hamza NEGİŞ Cevdet ŞEKER	Effectiveness of Conventional and Minimized Tillage Practices on Soil Quality Properties and Maize Yield Attributes	438-446
Esra YILMAZ Özden ÖZTÜRK	The Effect of Different Nitrogen Doses on Yield and Quality of Some Winter Canola (Brassica napus L.) Cultivars	447-457
Osagie Stephen IMADE Joseph AHMADU	Profitability Analysis of Catfish (Clarias gariepinus) Production in Edo State, Nigeria	458-465
Gürhan ÖZAYDIN Mithat DİREK	Evaluation of Agro-Industries According To Management Functions: Case of Konya Province	466-475
Ozcan CATALTAS Kemal TUTUNCU	Determination of the ADF and IVOMD Content of Sugarcane Using Near Infrared Spectroscopy Coupled with Chemometrics	476-481
Zeki KARA Burcu Nur UĞUR Osman DOĞAN	The effects of Ortho Silicon (Optysil) and Ascophyllum nodosum Based Sea-weed Extract (KelpGreen) Applications on the Quality of Table Grape cvs. Gök Üzüm and Müşküle	482-492
Nilgün ARIKAN İzzet KADIOĞLU	Determination of Densities and Frequencies of Problematic Weed Species in Onion Planting Areas of Ankara and Çorum Provinces	493-500
Muhammet KÜÇÜK Ercan CEYHAN	Determination of Chemical Fertilizer and Various Organic Fertilizers on Some Agricultural Characteristics of Green Bean (Phaseolus vulgaris L.)	501-506
Sevil KARAASLAN Barbaros Salih KUMBUL Kamil EKİNCİ	Drying of Melons in a Solar Tunnel Dryer: The Effect of Ascorbic Acid Solution on Drying Kinetics and Color Parameters	507-514
Yavuz Selim TAŞPINAR	Classification and Analysis of Tomato Species with Convolutional Neural Networks	515-520
Günel MİKAİLSOY	A Research on Food Safety in Turkey after the Covid-19 Pandemic	521-526

Research Article

Nursel ÇÖL KESKIN Gülsüm KAYIŞOĞLU	Erratum to: The Effects of Iron Application Foliarly at Different Times and Amounts on Agricultural Characteristics in Some Peanut Varieties	527

Selcuk Journal of Agriculture and Food Sciences

Selcuk Journal of Agriculture and Food Sciences SJAFS

http://sjafs.selcuk.edu.tr/sjafs/index

(2022) 36 (3), 299-311

e-ISSN: 2458-8377

DOI:10.15316/SJAFS.2022.039

Research Article

Determination of Combining Ability and Heredity Through Diallel Analysis Method in F_2 Populations of Cowpea

Christelle JOU-NTEUFA[1], Ercan CEYHAN[2,*]

[1] Selçuk University, The Graduate School of Natural and Applied Science, Department of Field Crops, Konya, Türkiye
[2] Selçuk University, Faculty of Agriculture, Department of Field Crops, Konya, Türkiye

ARTICLE INFO

Article history:
Received date: 09.08.2022
Accepted date: 12.08.2022

Keywords:
Cowpea
Diallel analysis
GCA
Heredity
SCA

ABSTRACT

Cowpea is an essential economic crop in underdeveloped and developing countries mainly grown by small farmers. The parents and F_2 populations obtained for the F_1 generation were planted, in 3 rows of 2 m in length, with row spacing of 45 cm, row spacing of 10 cm and in the ecological conditions of Konya in 2021. The 20 F_2 populations and 5 parents were evaluated in the 2021 grown season. General combining ability (GCA) effects for various parents differed significantly. "Sırma" gave significant, positive GCA effects for the number of pod per plant, number of seeds per plant, seed yield per plant and protein yield in plant. Significant, positive specific combining ability (SCA) effects were smaller and less significant than GCA effects. The non-additive dominant gene effect was determined for all the traits. Heterosis of the F_2 over the high-parent was observed in five F_2 populations. In the terms of combining ability for yield, the best parent was "Sırma". The heterosis for a yield of F_2 hybrids resulted mainly from an increased number of seeds per plant, hundred seed weight. These results suggest that high yielding F_2 cowpea populations can be developed that may contain acceptable levels of superior agronomic and technological characteristics.

1. Introduction

Cowpea [*Vigla unguiculata (L.) Wap*.] (2n=22) is native and domesticated in Africa; it is widespread and cultivated in tropical and subtropical regions. Cultivated for the collection of immature pods, ripe seeds, leaves, richness in proteins, vitamins (*thiamine, riboflavin*), photo chemicals (*phenolic acids, anthocyanins*) and minerals (potassium, phosphorus iron, calcium (Sert and Ceyhan, 2012; Abdulazeez et al., 2019). The very regular consumption of pulses including cowpeas is associated with poverty (Sert and Ceyhan, 2012; Harmankaya et al., 2016). It is an important component of human and animal nutrition, especially on the African continent, where it is mainly cultivated. Cowpea plays a major role as a concentrated source of cheaper protein like fish, meat, poultry or dairy products. Thus, it combines well with cereals in the human diet hence its name "food of the poor men" (Porch and Hall, 2013). It is one of the favorite crops grown in India for green pods, ripe seeds, and richness in nutrients. The consumption of meat or dairy products in India is sometimes questioned for customary and religious reasons. However, cowpea is consumed for the rational and nutritional balance of these populations (Penchalaraju and Don Bosco, 2022). Cowpea seeds contain substantial amounts of protein (about 25%), carbohydrates (about 64%), vitamins and fiber. When a part of cowpea is combined with three cereal seeds, it provides an almost complete food (Hal, 2012).

The most important function of legumes in nutrition is that they are a source of protein. Legumes are important nutritious sources of proteins, vitamins and above all rich in minerals like potassium, phosphorus, calcium and iron. Legumes are more widely used as a source of protein, especially in less developed and developing third world countries (Sing, 2020). Pulses are important sources that potentially provide the 15 minerals needed by humans, the amount of which varies depending on genetic and environmental factors (Horn and Shimelis, 2020).

Cowpeas are better than other legumes in heat and drought tolerance in semi-arid and tropical regions of the world (Boukar et al., 2019). It is undeniable that crossbreeding studies are necessary to increase the genetic variation of cowpea, which has an important place in the world. However, its importance has not been demonstrated in Turkey and only selection studies have focused on selection by selection. The development of

* Corresponding author email: *eceyhan@selcuk.edu.tr*

cowpea cultivars in these variations can be formed by hybridization, improved productivity and quality in the unit area. Thus, by improving the breeding varieties resistant to the diseases observed in Turkey; the creation of genetic resources would show how much more important the study is in terms of temperature and dry weather (Veeranagappa et al., 2022). However, Turkey's total number of local varieties, especially in extreme and remote areas, is declining Since Turkey is a micro gene for many plants such as cowpea, the demand for protein-rich foods like cowpea is increasing daily, especially in rural areas where malnutrition becomes a problem. These protein-rich varieties are of profitable economic importance, of an undeniable role in the diet of populations. Consequently, their insufficiency could contribute to exacerbate this problem of malnutrition and to lower the economy.

Genotypes with significantly positive general and specific combining ability (GCA and SCA) were reported for plant height, pod lenght, pod width, number of pods per plant, number of seed per pod, seed yield per plant, hundred weight, protein ratio and protein yield of cowpea hybrids (Pejic et al., 2013; Tchiagam et al., 2011; Magar et al., 2016; Kumari and Chauhan, 2018; Kalambe et al., 2019; Walle et al., 2019). But for the number of seeds per plant, GCA was negative and SCA was positive. The genotypes with a high GCA variance can be used as a basic breeding source in such breeding studies (Ayo-Vaughan et al., 2013).

It was found that non-additive genes were effective on pod length (Pallavi and Chaudhary, 2020; Yadji Haman, 2020). Non-additive genes were also effective on number of pod per plant, number of seed per plant, plant height and protein yield per plant (Magar et al., 2016; Kumari and Chauhan, 2018), then effective on the number of seed per pod, protein ratio (Ayo-Vaughan et al., 2013), on hundred-seed weight, seed yield per plant of cowpea (Singh, 2020). Whereas, additive genes were effective in the inheritance of pod width (Umaharan et al., 1997; Tchiagam et al., 2011; Verma et al., 2020).

This study aimed to study the genetic structure of the F_2 hybrids, identify the appropriate parents and combinations, determine the heritability, heterosis, and heterobeltiosis of the characters studied, and finally identify the hybrids with superior agronomic and technological characteristics and available for mechanical agriculture.

2. Materials and Methods

Field experiments were carried out on an apple (In this study, 5 high-yield cowpea varieties of seed collection have been used: Sırma (shows stunted and upright development; Seed shape is rhombic; hilum ring color is yellowish brown; the number of fruit seeds is 8-12), Amazon (It shows stunted and upright development; The shape of the seed is that of a pastry; the color of the hilum ring is black; The number of fruit seeds is 8-12), Karagöz (They are black-eyed peas with cylindrical seeds that are off-white. Fruit color is dark green. The circumference of the hilum is black. Plant height is 60-90 cm. Fruit size is about 16-21 cm long, 10-12 mm in diameter. The number of seeds in the fruit is 10-14), Ülkem (There is no pubescence on the stem and leaves of the plant, which produces abundant vegetative parts, develops spreading, growth type is pole (unlimited), growth type is spreading (creeper); Seeds are reddish brown; flower color is purple violet) and Pekşen (There is a climbing type, which most of lower branches touched the ground).

The soil of research area was clay loam, with pH 8.05 and phosphorous, potassium, iron, zinc, organic matter and $CaCO_3$ contents of 55.9, 17.9 kg ha^{-1}, 14.74, 0.32 ppm and 37.6, 2.25%, respectively. 20 year annual precipitation is 109.6 mm per year, annual mean temperature is 19.5 ^oC and average relative humidity is 48.0%. Total annual precipitation was 134.3 mm, which was more than 20 years on average (109.6 mm) of the site. During the experimental period, average temperature was 19.8 ^0C and average relative humidity was 44.4%.

The parents and F_2 populations obtained for the F_1 generation were planted on 04.29.2021, in 3 rows of 2 m of length, row spacing of 45 cm and row spacing of 10 cm. Mother plants were planted first, then father plants were planted in each row of the hybrid group. In the trial field of Selcuk University Faculty of Agriculture and according to the “Random Blocks Trial Design”, the trials were set up with 3 replicates (Yurtsever, 1984). At the sowing’s time, 15 kg of 46 % DAP (Diammonium phosphate) fertilizer was added per decare. After planting, the plots were irrigated by the sprinkler irrigation method to ensure germination and emergence. Using a hoe, weeding was done manually and mechanically. When the plants reached harvest maturity, they were harvested separately and measurement, weighing, analysis, and evaluation processes were carried out.

Measurements and counts of the properties examined in the research have been carried out in all plants obtained in each parcel in F_2 populations. Plant height, pod length, number of pods per plant, number of seed per pod, number of seed per plant, seed yield per plant, hundred-seed weight, protein ratio, and protein yield were investigated in this study.

The breeding value of the plant material was evaluated by analyzing the data on heterosis or combining ability for all the traits in the F_2. The studied data were analyzed with the program TARPOPGEN PC Program (Ozcan and Acikgoz, 1999).

3. Results and Discussion

In Table 1 are given the results of the mean squares of the initial analysis of variance and the combination of the analysis of variance of the capacities for the characters studied in the complete diallel hybrid set.

The mean squares of the crosses were determined to be statistically significant for all traits in the full diallel analysis of trait variance. Genotypes varied at the 5%

and 1% significance levels for all traits studied (Table 1).

In the F_2 generation, in the full diallel hybrid set, significant differences were determined for all the investigated traits between GCA and SCA for the traits whose combinatory ability variances were examined. Among the variances of the reciprocal effect, it was found to be statistically significant in all properties except pod width and pod length (Table 1).

Table 1
Mean squares of initial variance analysis and combining ability variance analysis for investigated traits in a full-diallel hybrid set

Source of Variation	SD	Plant Height	Pod Length	Pod Width	Number of Pods per Plant	Number of Seeds per Pod
Blocks	2	131,745	3,715	0,005	0,549	0,2250
Genotypes	24	1494,545**	10,731**	0,021**	19,248**	3,497**
Error	48	395,064	3,34	0,002	6,448	0,729
GCA	4	935,949**	935,949**	11,927**	0,008**	10,509**
SCA	10	248,160	248,160	2,195	0,004**	5,884**
Reciprocal Effect	10	573,096**	573,096**	1,619	0,009**	5,310*
Error	48	131,688	131,688	1,114	0,001	2,149
Source of Variation	SD	Number of Seed per Plant	Seed Yield	Hundred Seed Weight	Protein Ratio	Protein Yield in Plant
Blocks	2	131,513	6,180	6,737	0,052	0,438
Genotypes	24	3288,605**	244,852**	22,251**	2,234**	19,264**
Error	48	853,570	46,405	3,181	0,074	3,601
GCA	4	2229,474**	181,394**	7,158**	1,177**	12,842**
SCA	10	865,068**	93,441**	10,843**	0,422**	7,391**
Reciprocal Effect	10	874,026**	29,883	4,094**	0,894**	2,882*
Error	48	284,523	15,468	1,060	0,025	1,200

* : significant at 5% level , ** : significant at 1% level

3.1. Plant height:

In the F_2 generation, the plant height ranged between 80.52 cm (Karagöz x Amazon) and 153.53 cm (Sırma x Karagöz) (Table 2). In previous studies, some researchers have determined similar results (Peksen et al., 2004; Pekşen, 2012; Sert and Ceyhan, 2012; Kadam et al., 2013; Pekşen, 2013; Walle et al., 2019; Joshi et al., 2022).

The fact that σ^2SCA was greather than σ^2GCA and $H/D^{1/2}$ ratio was also greater than 1 (Table 2), showed us that, the non-additive gene effect and superior dominance are important in the inheritance on plant height. Owusu et al. (2020) determined in their research that the additive gene effect was more dominant in the inheritance of plant height. But contrary to (Magar et al., 2016; Kumari and Chauhan, 2018; Verma et al., 2020) non-additive gene effect was predominant in this trait and these results were in agreement with ours.

In the F_2 generation plant height, while examining the parental GCA, Pekşen, Sırma and Ülkem genotypes had positive and significant GCA values, while Amazon and Karagöz genotypes showed negative and significant GCA values (Table 3). Pekşen, Sırma and Ülkem genotypes with positive and significant GCA effect value can be used to increase plant height, and Amazon and Karagöz genotypes, which are negative and important, can be easily used in the development of Cowpea varieties with less grading. Magar et al. (2016), Kumari and Chauhan (2018), Ravelombola et al. (2018), Owusu et al. (2020), Pallavi et al. (2020), Verma et al. (2020), who studied the plant height feature found the GCA and SCA values of different numbers of parents and crosses to be significant.

While examining the SCA effects of hybrids in the F_2 generation, "Sırma x Karagöz" hybrid combinations are positive and significant, "Pekşen x Sırma", "Ülkem x Pekşen", "Karagöz x Pekşen", "Ülkem x Sırma", "Amazonx Pekşen", "Karagöz x Sırma", hybrid combination showed a negative and significant effect (Table 3). When Table 4.5 sub-diagonal reciprocal effects are examined, "Sırma x Karagöz" was positively significant. This shows us that cytoplasm or cytoplasm x nucleus interactions cause significant changes in this feature.

The average heterosis value determined in the F_2 generation is 4.45%, the heterobeltiosis value is -10.61%.Heterosis values in the F_2 generation ranged between -27.05% (Ülkem x Karagöz) and 54.32% (Pekşen x Ülkem), while heterobeltiosis values ranged between -33.90% (Karagöz x Pekşen) and 22.35% (Sırma x Karagöz) (Table 4). In terms of plant height, the majority of hybrids have negative heterosis and heterobeltiosis values. 16,31. Analyzing the heterosis and heterobeltiosis values for the same trait, Sarath and Reshma (2017), and Joshi et al. (2022) reported that they detected high or low heterosis and heterobeltiosis values for this feature and hybrids that both were desirable and significant.

In the F_2 generation, broad and narrow sense heritability was calculated as 0.74 and 0.21, respectively (Table 2). The high heritability in F_2 generation reveals that genetic factors, as well as the environment, as well as the environment, significantly impact the emergence of this trait. Thereby, it can be said that a selection to be-

Table 2
Mean values for investigated traits in full-diallel hybrid set

Parents	Plant Height		Pod Length		Pod Width		Number of Pods per Plant		Number of Seeds per Pod	
Pekşen	121,89	a-d	21,48	a	0,81	c-f	12,91	f	9,41	ab
Sırma	125,49	abc	19,77	abc	0,84	b-e	18,88	a-e	9,60	ab
Ülkem	116,07	a-d	16,51	be	0,73	fgh	16,31	a-f	10,26	ab
Amazon	96,74	b-e	16,58	be	0,85	b-e	13,76	ef	9,96	ab
Karagöz	58,00	e	15,67	de	0,99	a	13,67	ef	5,67	d
F_2 Populations										
Pekşen X Sırma	96,42	b-e	17,26	be	0,87	cd	17,66	a-f	10,07	ab
Pekşen X Ülkem	133,61	ab	19,94	ab	0,81	c-f	17,89	a-f	9,92	ab
Pekşen X Amazon	133,73	ab	21,65	a	0,84	b-e	18,85	a-e	9,67	ab
Pekşen X Karagöz	135,92	ab	18,25	a-e	0,92	ab	18,09	a-f	11,27	a
Sırma X Pekşen	109,63	bcd	18,25	a-e	0,78	d-g	21,10	ab	9,40	b
Sırma X Ülkem	137,06	ab	17,96	a-e	0,86	bcd	17,92	a-f	10,42	ab
Sırma X Amazon	112,84	a-d	16,92	b-e	0,90	abc	18,40	a-f	9,54	ab
Sırma X Karagöz	153,53	a	17,05	b-e	0,69	gh	16,06	a-f	7,43	cd
Ülkem X Pekşen	115,43	a-d	16,84	b-e	0,76	efg	20,40	abc	9,03	bc
Ülkem X Sırma	100,82	b-e	15,82	cde	0,72	fgh	16,76	a-f	10,44	ab
Ülkem X Amazon	103,10	bcd	15,92	cde	0,71	gh	20,06	a-d	10,11	ab
Ülkem X Karagöz	94,44	b-e	16,27	b-e	0,85	b-e	13,13	f	9,93	ab
Amazon X Pekşen	103,67	bcd	19,32	a-d	0,87	abc	14,54	def	9,88	ab
Amazon X Sırma	97,88	b-e	15,01	e	0,86	bcd	21,56	a	9,70	ab
Amazon X Ülkem	88,96	cde	15,75	de	0,84	b-e	13,98	ef	9,13	bc
Amazon X Karagöz	95,17	b-e	15,98	b-e	0,83	b-e	18,15	a-f	10,20	ab
Karagöz X Pekşen	80,57	de	15,81	cde	0,66	h	14,46	ef	9,03	bc
Karagöz X Sırma	83,34	cde	16,22	b-e	0,84	b-e	15,95	b-f	9,22	bc
KaragözX Ülkem	83,91	cde	15,91	cde	0,65	h	14,99	c-f	9,80	ab
KaragözX Amazon	80,52	de	14,85	e	0,85	b-e	16,04	a-f	8,59	bc
GCA	160,85		2,16		0,00		1,67		0,42	
SCA	349,42		3,24		0,01		11,21		2,90	
Reciprocal	441,41		0,51		0,01		3,16		0,41	
σ^2GCA/σ^2SCA	0,46		0,67		0,13		0,15		0,14	
$H/D^{1/2}$	1112,53		8,07		0,02		17,71		4,15	
H^2	0,74		0,71		0,93		0,74		0,85	
h^2	0,21		0,38		0,12		0,14		0,17	

GCA: General Combining Ability; SCA: Specific Combining Ability; $H/D^{1/2}$: Mean Degree of Dominance; H^2: Broad Sense Heritability; h^2: Narrow Sense

made in the direction of plant height should be considered together with seed yield. For this reason, it may be more appropriate to start the selection in future generations. Owusu ve ark. (2020); Verma ve ark. (2020) found the similar results.

3.2. Pod Length:

The parental values in the F_2 generation ranged from 15.67 mm (Karagöz) to 21.48 mm (Pekşen), and the pod length per plant in the F_2 generation was 14.85 mm (Karagöz x Amazon) to 21.65 mm (Pekşen x Amazon) (Table 2). Peksen et al. (2004), Pekşen (2012), Kadam et al. (2013), Pekşen (2013), da Costa et al. (2017), Walle et al. (2019), and Joshi et al. (2022) obtained similar results.

The fact that in F_2 generation σ^2SCA was greather than σ^2GCA in the inheritance of pod length, then σ^2GCA/σ^2SCA ratio was lower than 1 and $H/D^{1/2}$ ratio was greater than 1 showed us the effectivity of non-additive gene effect and superior dominance in the inheritance of this trait. It reveals that selection in future generations would be appropriate because the genes that affect the pod length in the plant are not additive in the examined hybrid generations (Pallavi et al., 2020; Verma et al., 2020), found different results. But Magar et al. (2016), Kumari and Chauhan (2018), and Olunloyo et al. (2019) found similar results.

Considering the SCA effects of hybrids in the F_2 generation, "Pekşen x Amazon" hybrid combinations are positive and significant, "Ülkem x Pekşen", "Amazon x Pekşen", "Karagöz x Pekşen", "Pekşen x Sırma", "Amazon x Sırma" ", "Ülkem x Sırma" hybrids combinations showed a negative and significant effect (Table 2). When the reciprocal effects are examined, in the F_2 generation, "Pekşen x Amazon" was found to be positive and significant. This shows us that cytoplasm or cytoplasm x nucleus interactions cause significant changes in this feature(Table 2). Working on the pod length, Dias et al. (2016), Magar et al. (2016), Kumari and Chauhan (2018), Olunloyo et al. (2019), Owusu et al. (2020), Pallavi et al. (2020), and Verma et al. (2020) found the GCA and SCA values of different numbers of parents and crosses to be important for this trait.

In the F2 generation, the determined mean heterosis value was -5.16%, while the heterobeltiosis value was -12.08%. From this feature, most hybrids have negative values of heterosis and heterobeltiosis. Heterosis values are between -17.42% (Amazon x Sırma) and 13.79% (Pekşen x Amazon), heterobeltiosis values are between -26.39% (Karagöz x Pekşen) and 0.83% (Pekşen x Amazon) (Tables 4 and 8). These findings were supported by Ceyhan (2004), Kadam et al. (2013), Ceyhan et al. (2014), Sarath and Reshma (2017), and Joshi et al. (2022). Moreover, Kadam et al. (2013), and Sarath and

Table 3
Genetic components for investigated traits in full-diallel hybrid set

Parents	Plant Height	Pod Length	Pod Width	Number of Pods per Plant	Number of Seeds per Pod
Pekşen	8,926	1,789**	0,000	0,020	0,201
Sırma	7,899	0,163	0,006	1,456*	0,036
Ülkem	2,598	-0,497	-0,047**	-0,086	0,423*
Amazon	-5,414	-0,383	0,027*	0,050	0,167
Karagöz	-14,009*	-1,073*	0,015	-1,440*	-0,827**
F_2 Populations					
Pekşen X Sırma	-20,152*	-1,436*	0,007	1,045	-0,006
Pekşen X Ülkem	6,645	-0,140	0,019	2,348*	-0,658*
Pekşen X Amazon	8,836	1,842*	0,015	-0,237	-0,100
Pekşen X Karagöz	6,982	-0,925	-0,037*	0,837	1,265**
Sırma X Pekşen	6,605	0,493	-0,045**	1,723*	-0,336
Sırma X Ülkem	2,095	-0,017	0,016	-0,892	0,464
Sırma X Amazon	-3,475	-1,055	0,034*	1,612	-0,092
Sırma X Karagöz	18,193*	0,302	-0,069**	-0,872	-0,389
Ülkem X Pekşen	-9,092*	-1,550**	-0,022*	1,257*	-0,443*
Ülkem X Sırma	-18,120**	-1,073*	-0,074**	-0,582	0,008
Ülkem X Amazon	-7,501	-0,527	-0,018	0,196	-0,479
Ülkem X Karagöz	-5,764	0,419	-0,029	-1,276	0,762*
Amazon X Pekşen	-15,030**	-1,166*	0,015	-2,157**	0,108
Amazon X Sırma	-7,484	-0,955*	-0,022*	1,584*	0,078
Amazon X Ülkem	-7,072	-0,084	0,061**	-3,037**	-0,492*
Amazon X Karagöz	0,920	-0,368	-0,014	1,626	0,549
Karagöz X Pekşen	-27,675**	-1,223*	-0,132**	-1,816*	-1,120**
Karagöz X Sırma	-35,096**	-0,415	0,076**	-0,051	0,897**
KaragözX Ülkem	-5,263	-0,180	-0,099**-	0,932	-0,067
KaragözX Amazon	-7,323	-0,568	0,009	-1,054	-0,801**
G_i	10,535	0,089	0,000	0,172	0,019
S_{ij}	44,774	0,379	0,000	0,731	0,083
R_{ij}	65,844	0,557	0,000	1,075	0,122

G_i : GCA, S_{ij}: SCA; R_{ij}: Reciprocal effect, ** : significant at 1% level; * : significant at 5% level

Reshma (2017), who examined the heterosis and heterobeltiosis values pod length, reported that heterosis was expressed in hybrids and heterobeltiosis was recorded in crosses. In comparison, Joshi et al. (2022) found that heterosis values were positive and significant.

In the F_2 generation, broad and narrow sense heritability rates were 0.71 and 0.38, respectively (Table 2). The low heritability in both generations indicates that genetic factors are more dominant in the emergence of this trait. With these results, it can be said that it would be more appropriate to start the selection in terms of the pod length after the F_2 generation. Ceyhan (2004), and Owusu et al. (2020) were in harmony.

Table 4
Heterosis (%) values for investigated traits in full-diallel hybrid set

F_2 Populations	Plant Height	Pod Length	Pod Width	Number of Pods	Number of Seed per Pod
Pekşen X Sırma	-15,64	-16,29**	5,87**	11,10	5,98**
Pekşen X Ülkem	54,32**	5,00	4,80**	22,41	0,82
Pekşen X Amazon	9,36	13,79*	1,16**	41,35*	-0,20
Pekşen X Karagöz	-18,91	-1,71	2,35**	36,18*	49,46**
Sırma X Pekşen	5,95	-11,51*	-4,98**	32,78*	-1,08
Sırma X Ülkem	-8,12	-0,99	9,93**	1,83	4,94**
Sırma X Amazon	6,37	-6,91	6,99**	12,70	-2,47*
Sırma X Karagöz	47,68*	-3,79	-24,66**	-1,34	-2,66*
Ülkem X Pekşen	1,14	-11,32*	-0,82**	39,62*	-8,19**
Ülkem X Sırma	-5,43	-12,82*	-8,91**	-4,78	5,09**
Ülkem X Amazon	-13,68	-3,80	-9,77**	33,38*	-0,03
Ülkem X Karagöz	-27,05	1,12	-1,37**	-12,43	24,70**
Amazon X Pekşen	-2,77	1,54	4,82**	9,01	2,03*
Amazon X Sırma	22,82	-17,42**	1,74**	32,10*	-0,88
Amazon X Ülkem	-22,00	-4,82	5,64**	-7,01	-9,77**
Amazon X Karagöz	-8,69	-0,87	-9,74**	32,34*	30,48**
Karagöz X Pekşen	-8,68	-14,88*	-26,84**	8,84	19,74**
Karagöz X Sırma	37,45	-8,48	-8,17**	-1,97	20,83**
KaragözX Ülkem	-14,07	-1,11	-24,24**	0,00	23,01**
KaragözX Amazon	-0,44	-7,91	-7,83**	16,96	9,97**
Mean	4,45	-5.16	-4.20	15.15	8.59

** : significant at 1% level; * : significant at 5% level

3.4. Number of pods:

According to the average number of pods in the F_2 generation, the parental values were between 12.91 per plant (Pekşen) and 18.88 per plant (Sırma), in the F_2 generation the number of pods per plant was 13.13 per plant (Ülkem x Karagöz) and 21.56 per plant (Ülkem x Sırma) (Table 2). These findings were supported by Peksen et al. (2004), Pekşen (2012), Kadam et al. (2013), da Costa et al. (2017), Walle et al. (2019), and Joshi et al. (2022).

For the number of pods per plant in the F_2 generation, the GCA variance was less than SCA, then σ^2GCA/σ^2SCA ratio ratio was also less than 1 and the $H/D^{1/2}$ ratio was greather than 1 The fact that the σ^2GCA/σ^2SCA ratios examined for the number of pods in the plant were found to be less than 1 reveals that the non-additive gene effect is effective in the inheritance of this trait. Likewise, the fact that the $(H/D)^{1/2}$ ratio is greater than 1 in the F_2 generation indicates superior dominance and supports this result. Magar et al. (2016), Gupta et al. (2017), Kumari and Chauhan (2018), and Purnamasari et al. (2019) agreed with these results. Hall et al. (2003), and Verma et al. (2020) stated that additive genes were effective on the number of pods in the plant.

In the F_2 generation, examining the effects of parents and hybrids on GCA and SCA in terms of the number of pods per plant; Magar et al., (2016), Olunloyo et al. (2019), and Pallavi and Chaudhary (2020) obtained similar results to our findings. (Table 2). Considering the reciprocal effect, the hybrid is in F_2 generation, "Pekşen x Ülkem" hybrid has Pekşen cytoplasm. This result shows that cytoplasm and cytoplasm x nucleus interactions cause significant changes in this feature. Increasing the number of pods in the plant is also theoretically increasing the seed yield (Pekşen et al., 2004), examining the effects of parents and hybrids on GCA and SCA in terms of the number of pods per plant. Dias et al. (2016), Magar (2016), Gupta et al. (2017), Kumari and Chauhan (2018), Purnamasari et al. (2019), Owusu et al. (2020), Pallavi et al. (2020), and Verma et al. (2020) obtained similar results to our findings.

While the average heterosis value determined in the F_2 generation was 15.15%, the heterobeltiosis value was 5.57%. Heterosis values varied between -12.43% (Ülkem x Karagöz) and 39.62% (Ülkem x Pekşen), while heterobeltiosis values varied between -19.53% (Ülkem x Karagöz) and 36.96% (Pekşen x Amazon). Likewise, heterobeltiosis values were mostly found to be significant and positive (Tables 4 and 8). These findings were supported by Kadam et al. (2013), Sarath and Reshma (2017), and Joshi et al. (2022), who found significant heterosis and heterobeltiosis values for this feature.

Table 5
Heterobeltiosis (%) values for investigated traits in full-diallel hybrid set

F_2 Populations	Plant Height	Pod Length	Pod Width	Number of Pods	Number of Seed per Pod
Pekşen X Sırma	- 22,05	47,48*	41,28**	92,68**	- 18,29
Pekşen X Ülkem	12,30	20,90	21,29	- 34,35*	40,68**
Pekşen X Amazon	22,33	14,65	37,70**	43,80*	51,83**
Pekşen X Karagöz	51,12	- 18,32	15,26	- 52,07**	84,65**
Sırma X Pekşen	- 11,37	19,87	- 19,15	61,29**	5,06
Sırma X Ülkem	13,48	26,58	- 3,40	- 7,03	3,77
Sırma X Amazon	1,56	-5,76	0,00	31,11	- 18,29
Sırma X Karagöz	67,34	-33,89	- 42,21**	- 49,58**	- 42,22**
Ülkem X Pekşen	- 2,99	39,78	33,11*	15,41	5,32
Ülkem X Sırma	- 16,52	20,22	2,55	- 7,07	- 4,86
Ülkem X Amazon	- 3,10	-14,66	0,26	- 1,24	- 37,60*
Ülkem X Karagöz	8,50	1,70	- 25,97*	17,12	- 20,00
Amazon X Pekşen	- 5,17	6,76	- 5,76	8,57	4,39
Amazon X Sırma	-11,92	1,24	- 6,17	57,07**	17,51
Amazon X Ülkem	-16,40	-18,34	- 24,61*	- 17,90	1,32
Amazon X Karagöz	23,00	-26,60	- 50,97**	31,96*	4,60
Karagöz X Pekşen	-10,42	-21,18	- 2,92	1,46	33,87
Karagöz X Sırma	-9,16	- 8,67	- 52,92**	11,94	8,09
KaragözX Ülkem	-3,59	-32,75	- 64,29**	- 18,52	30,60*
KaragözX Amazon	4,07	- 31,35	-60,71**	2,70	-17,76
Mean	-10.61	-12.08	-9.78	5.75	-3.13

** : significant at 1% level; * : significant at 5% level

Heritability in the broad and narrow sense was 0.7 and 0.14, respectively, in the F_2 generation (Table 2). In the study, it was revealed that the heritability in the broad sense was large and the heritability in the narrow sense was low, and the number of pods was also affected by the environment. Working on similar issues, Purnamasari et al. (2019), and Owusu et al. (2020) found high heritability broad and narrow sense. It can be argued that it would be better to start breeding after 3-4 generations, since non-additive genetic effects are important in the generation examined.

3.5. Number of seeds per pod:

According to the number of seeds per pod in the F_2 generation, the parental values were between 5.67 units/pod (Karagöz) and 10.26 units/pod (Ülkem), and in the F_2 generation, the number of seeds per pod was 7.43 units/pod (Sırma x Karagöz) to 11.27 units/pod

(Pekşen x Karagöz) (Table 2). Which are in harmony with our research findings (Peksen and Artık, 2004; Kadam et al., 2013; Kumari and Chauhan, 2018; Walle et al., 2019; Joshi et al., 2022).

When Table 2 is examined, the σ^2GCA was less than The σ^2SCA, σ^2GCA/σ^2SCA ratio was less than 1 and $(H/D)^{1/2}$ ratio was greather than 1. The fact that the σ^2GCA/σ^2SCA ratios are less than 1 and the $H/D^{1/2}$ ratio is greater than 1 for the number of seeds per pod shows us that the non-additive dominant gene effect is effective in the inheritance of this trait (Magar ve ark., 2016; Gupta ve ark., 2017; Kumari ve Chauhan, 2018; Purnamasari ve ark., 2019).

When GCA was examined regarding the number of seeds in the pod, Pekşen, Sırma, Ülkem and Amazon genotypes were found to have significant and positive values in the F_2 generation. In contrast, Karagöz genotype had a significant negative effect (Table 2). As a result, Pekşen, Sırma, Ülkem and Amazon genotypes, which are positively important in increasing the number of seeds per pod, were determined as suitable parents to be used in breeding studies for this purpose.

Looking at the SCA effects of crosses in the F_2 generation, "Ülkem x Karagöz" and "Pekşen x Karagöz", "hybrid have a positive significant SCA effect. Pekşen x Ülkem" and "Ülkem x Pekşen", "x "Karagöz x Pekşen", "Amazon x Ülkem"and "Karagöz x Amazon" hybrids have negative significant SCA effect (Table 2). Considering the reciprocal effect values, "hybrids are positive and have high SCA. They can be considered as suitable combinations that can be used to increase the number of seeds in the pod, as they have the effect of SCA. In many studies, the effects of GCA and SCA for the number of seeds per pod were determined (Dias et al., 2016; Magar et al., 2016; Gupta et al., 2017; Kumari and Chauhan, 2018; Olunloyo et al., 2019; Purnamasari et al., 2019; Owusu et al., 2020; Pallavi et al., 2020; Verma et al., 2020).

In the F_2 generation, the heterosis average value determined for the number of seeds per pod was 8.59%, and the heterobeltiosis value was calculated as -3.13%. Heterosis value was found to be significant and positive, and heterobeltiosis value was found to be significant and negative. Heterosis values varied between -9.77% (Amazon x Ülkem) and 49.46% (Pekşen x Karagöz), while heterobeltiosis values ranged between -22.61% (Sırma x Karagöz) and 19.75% (Pekşen x Karagöz) (Tables 4 and 8). Examining the heterosis and heterobeltiosis values for the seed number feature per pod, Kadam et al. (2013), Sarath and Reshma (2017), and Joshi et al. (2022) reported that in hybrids they detected significate positive and negative performance in heterosis. According to Kadam et al. (2013), Sarath and Reshma (2017), and Joshi et al. (2022), their results were similar to ours.

In the F_2 generation, broad and narrow sense heritability was 0.85 and 0.17, respectively (Table 2). The fact that the heritability in the narrow sense is at medium levels indicates that genetics and the environment influence the number of seeds per pod to a certain extent. Analyzing seed number inheritance in pods Purnamasari et al. (2019), and Owusu et al. (2020) found that broad sense heritabilities were generally largest but varied among crosses. It would be more appropriate to start the selection in advanced generations, since non-additive gene effects are important and hybrid strength is low.

3.6. Number of seeds per plant:

It was determined that the parent values in terms of the number of seeds per plant in the F_2 generation ranged between 76.67 units/plant (Karagöz) and 181.11 units/plant (Sırma), and the number of seeds per pod in the F_2 generation ranged between 119.92 units/plant (Sırma x Karagöz) and 209.13 (Amazon x Sırma) (Table 5). With our research findings, Peksen et al. (2004), Kumari and Chauhan (2018), Walle et al. (2019) are in harmony.

In the F_2 generation, it is seen that the σ^2GCA was less than the σ^2SCA, the σ^2GCA/σ^2SCA ratio was less than 1 and the $H/D^{1/2}$ ratio was greather than 1. These results supports the non-additive gene effect. The gene effect and the superiority of dominance reduce the success of selection for this trait in early generations. Kumari and Chauhan (2018) found the similar results.

When GCA was examined for the number of seeds per plant in the F_2 generation, Pekşen, Sırma, Amazon and Ülkem had significant positive values, while negative and significant GCA values were determined for Karagöz genotype (Table 5). Pekşen, Sırma, Amazon and Ülkem, which were found to be positively significant, were determined as the parents that could be used to increase the number of seeds per plant in crossing studies. Kumari and Chauhan (2018) were in agreement.

Looking at the SCA effects of hybrids in the F_2 generation, "Pekşen x Karagöz", "Karagöz x Sırma", hybrids have positive significant SCA effect and these combinations are genotypes with breeding potential for high seed number (Table 5). When the subdiagonal reciprocal effects are examined in Table 5, in the F_2 generation, Pekşen cytoplasm of "Pekşen x Karagöz" was found to increase the number of seeds per plant. This shows us that cytoplasm or cytoplasm x nucleus interactions cause significant changes in this feature (Table 5). "Pekşen x Karagöz", "Karagöz x Sırma", hybrids have emerged as suitable combinations that can be used to increase the number of seeds per plant, since they have a positive significant SCA effect. Kumari and Chauhan (2018) found the similar results in previous studies.

In the F_2 generation, the average heterosis value was determined as 25.00%, while the heterobeltiosis value was 5.02%. Heterosis values varied between -16.73% (Amazon x Ülkem) and 106.48% (Pekşen x Karagöz), while heterobeltiosis values varied between -33.79% (Sırma x Karagöz) and 68.70% (Pekşen x Karagöz) (Tables 7 and 9).While analysing their results Kumari and Chauhan (2018) found that heterosis and heterobeltiosis were significant in this feature.

Broad and narrow sense heritability was 0.79 and 0.00, respectively in the F_2 generation (Table 2). The high heritability in the broad sense and the low heritability in the narrow sense indicates that the effect of genotype variance on this trait is low. Therefore, it would be more appropriate to start selection in future generations.

3.7. Seed yield per plant

According to the average seed yields in the F_2 generation, the parental values varies between 17.04 g/plant (Karagöz) and 52.06 g/plant (Sırma), and in the F_2 generation, the single-plant seed yields were 24.45 g/plant (Sırma x Karagöz) and 51.41 g/plant (Pekşen x Karagöz) (Table 6). With the findings of this study, Ceyhan (2004), Peksen and Artık (2004), Ceyhan et al. (2008), Borivoj et al. (2013), Kadam et al. (2013), Ceyhan et al. (2014), Rodrigues et al. (2018), Walle et al. (2019), and Joshi et al. (2022) are in harmony.

In the F_2 generation, it was determined that the σ^2GCA was less than the σ^2SCA, the σ^2GCA/σ^2SCA ratio was less than 1 and the $H/D^{1/2}$ ratio was greather than 1 (Table 6). Thus, Dias et al. (2016), Magar et al. (2016), Kumari and Chauhan (2018), Rodrigues et al. (2018), Purnamasari et al. (2019), Owusu et al. (2020), and Verma et al. (2020) reported that additive gene effects are important in seed yield. Olunloyo et al. (2019) stated that cowpea seed yield was influenced by both added and non-added genes.

When we look at the effect value of GCA, it is seen that Pekşen, Sırma, Ülkem and Amazon genotypes among the parent genotypes show significant and positive values in the F_2 generation. Karagöz genotype had a significant negative effect (Table 6). Pekşen, Sırma, Ülkem and Amazon genotypes with positive and significant GCA effect values were determined as promising parents that could be used in terms of plant yield in crossing studies.

In the F_2 generation, "Pekşen x Karagöz", and "Amazon x Karagöz" hybrids were positive and has a significant SCA effect. "Amazon x Pekşen", "Karagöz x Pekşen", "Amazon x Ülkem", "Sırma x Karagöz", hybrids showed high negative significant SCA effect (Table 6). When Table 4.29 is examined, in the F_2 generation "Pekşen x Karagöz" showed a significant negative and positive reciprocal effect. This shows us that cytoplasm or cytoplasm x nucleus interactions cause significant changes in this feature. These results are in agreement with the previous studies of Magar al. (2016), Gupta et al. (2017), Kumari and Chauhan (2018), Rodrigues et al. (2018), Olunloyo et al. (2019), Purnamasari et al. (2019), Owusu et al. (2020), Pallavi et al. (2020), and Verma et al. (2020).

Table 6
Mean values for investigated traits in full-diallel hybrid set

Parents	Number of Seeds per Plant		Seed Yield per Plant		Hundred Seed Weight		Protein Ratio		Protein Yield in Plant	
Pekşen	120,91	1fg	27,37	hıj	17,86	fgh	28,49	cde	7,80	hıj
Sırma	181,11	a-f	52,06	a	26,27	a	26,49	k	13,80	abc
Ülkem	168,34	a-f	43,58	a-g	23,65	abc	28,09	efg	12,24	a-g
Amazon	135,83	d-g	30,89	f-j	17,68	fgh	28,37	def	8,76	e-j
Karagöz	76,67	g	17,04	j	17,09	gh	28,12	efg	4,79	j
F_2 Populations										
Pekşen X Sırma	178,15	a-f	46,93		22,29	bcd	26,29	kl	12,33	a-g
Pekşen X Ülkem	177,30	a-f	44,17	a-d	21,31	c-f	28,88	a-d	12,75	a-f
Pekşen X Amazon	182,78	a-f	45,14	a-g	21,48	c-f	29,01	abc	13,10	a-d
Pekşen X Karagöz	203,97	ab	51,41	a-g	22,77	a-d	29,25	a	15,04	a
Sırma X Pekşen	199,41	a-d	41,02	a-h	18,21	e-h	28,37	def	11,64	a-h
Sırma X Ülkem	185,65	a-e	50,80	ab	23,36	abc	28,37	def	14,43	ab
Sırma X Amazon	176,50	a-f	45,88	a-e	23,46	abc	25,87	l	11,87	a-h
Sırma X Karagöz	119,92	fg	24,45	ı j	15,15	h	28,52	cde	6,96	ıj
Ülkem X Pekşen	185,11	a-e	44,14	a-g	20,50	c-g	29,12	ab	12,85	a-e
Ülkem X Sırma	174,99	a-f	46,72	a-d	23,44	abc	27,99	e-h	13,07	a-d
Ülkem X Amazon	202,53	abc	45,70	a-f	19,86	c-g	27,37	ıj	12,51	a-g
Ülkem X Karagöz	130,42	efg	31,71	e-j	21,36	c-f	27,13	j	8,60	f-j
Amazon X Pekşen	143,85	b-f	37,20	a-ı	22,36	bcd	28,53	b-e	10,62	b-ı
Amazon X Sırma	209,13	a	48,57	abc	20,90	c-g	28,48	cde	13,82	abc
Amazon X Ülkem	126,64	efg	33,31	d-ı	23,13	a-d	27,71	g-j	9,22	d-ı
Amazon X Karagöz	185,82	a-e	42,65	a-g	19,30	d-g	28,01	efg	11,95	a-h
Karagöz X Pekşen	131,01	efg	35,69	bc-ı	22,47	a-d	27,40	hıj	9,78	c-ı
Karagöz X Sırma	147,43	a-f	30,36	g-j	17,98	e-h	27,61	g-j	8,38	g-j
KaragözX Ülkem	146,68	a-f	37,13	b-ı	21,79	b-e	28,51	cde	10,59	b-ı
KaragözX Amazon	139,24	c-g	39,39	a-h	25,48	ab	27,82	f-ı	10,96	a-ı
GCA	388,99		33,19		1,22		0,23		2,33	
SCA	1741,63		233,92		29,35		1,19		18,57	
Reciprocal	589,50		14,41		3,03		0,87		1,68	
σ^2GCA/σ^2SCA	0,22		0,14		0,04		0,19		0,13	
$H/D^{1/2}$	3109,12		314,70		34,82		2,52		24,91	
H^2	0,79		0,88		0,91		0,97		0,88	
h^2	0,20		0,18		0,06		0,18		0,16	

GCA: General Combining Ability; SCA: Specific Combining Ability; $H/D^{1/2}$: Mean Degree of Dominance; H^2: Broad Sense Heritability; h^2: Narrow Sense

In the F_2 generation, the average heterosis value determined in this study for seed yield was 25.87%, while the heterobeltiosis value was 1.76%. Heterosis values varied between -29.23% (Sırma x Karagöz) and 131.55% (Pekşen x Karagöz), and heterobeltiosis values ranged between -53.03% (Sırma x Karagöz) and 87.83% (Pekşen x Karagöz) (Tables 7 and 9). In their previous studies, Kadam et al. (2013), Magar et al. (2016), Gupta et al. (2017), Rodrigues et al. (2018), Owusu et al. (2020), and Joshi et al. (2022) reported that heterosis was more significant than herobeltiosis in seed yield.

Broad and narrow sense heritability was 0.88 and 0.18 in the F_2 generation, respectively (Table 5). In the F_2 generation, the low heritability in the narrow sense in the seed yield means that the effect of the environmental variance of this feature may be high. Purnamasari et al. (2019), and Owusu et al. (2020) found high heritability in the broad sense and low heritability in the narrow sense for this trait.

Table 7
Genetic components for investigated traits in full-diallel hybrid set

Parents	Number of Seeds per Plant	Seed Yield per Plant	Hundred Seed Weight	Protein Ratio	Protein Yield in Plant
Pekşen	3,165	0,311	-0,456	0,390**	0,256
Sırma	14,164*	4,153*	0,569	-0,544**	0,894*
lkem	5,424	2,352	1,039*	0,135*	0,737
Amazon	2,640	0,229	-0,034	-0,038	0,043
Karagöz	-25,392**	-7,045**	-1,118*	0,058	-1,930**
F_2 Populations					
Pekşen X Sırma	10,273	-0,223	-1,028	-0,509**	-0,280
Pekşen X Ülkem	11,444	1,759	-0,845	0,483**	0,693
Pekşen X Amazon	-3,664	0,896	1,244	0,426**	0,446
Pekşen X Karagöz	28,544*	10,551**	3,026**	-0,115	2,969**
Sırma X Pekşen	10,630	-2,956	-2,043**	1,042**	-0,344
Sırma X Ülkem	-0,443	2,521	0,630	0,601**	1,006
Sırma X Amazon	14,838	3,108	0,478	-0,238*	0,791
Sırma X Karagöz	-16,273	-9,432**	-4,050**	0,562**	-2,410**
Ülkem X Pekşen	3,903	-0,015	-0,407	0,117*	0,052
Ülkem X Sırma	-5,332	-2,040	0,040	-0,191*	-0,679
Ülkem X Amazon	-4,657	-2,807	-0,678	-0,549**	-1,029
Ülkem X Karagöz	-2,659	-0,616	0,487	-0,365**	-0,324
Amazon X Pekşen	-19,463*	-3,972*	0,439	-0,243**	-1,244*
Amazon X Sırma	16,313*	1,346	-1,282*	1,305**	0,975*
Amazon X Ülkem	-37,947**	-6,196**	1,637**	0,167*	-1,641**
Amazon X Karagöz	24,111	8,103**	2,377**	-0,094	2,230**
Karagöz X Pekşen	-36,480**	-7,862**	-0,150	-0,922**	-2,628**
Karagöz X Sırma	13,757*	2,956	1,413**	-0,455**	0,706
KaragözX Ülkem	8,129	2,709	0,213	0,693**	0,995*
KaragözX Amazon	-23,290**	-1,627	3,090**	-0,095	-0,495
G_i	22,762	1,237	0,085	0,002	0,096
S_{ij}	96,738	5,259	0,361	0,008	0,408
R_{ij}	142,262	7,734	0,530	0,012	0,600

G_i : GCA, S_{ij}: SCA; R_{ij}: Reciprocal effect, ** : significant at 1% level; * : significant at 5% level

3.8. Hundred seed weight:

The parent values ranged from 17.09 g (Karagöz) to 26.27 g (Sırma), while the hundred-seed weight of the F_2 generation ranged from 15.15 g (Sırma x Karagöz) to 25.48 g (Karagöz x Amazon) (Table 5). Some researchers have obtained similar results (Peksen and Artık, 2004; Sert and Ceyhan, 2012; Kadam et al., 2013; Sarath and Reshma, 2017; Rodrigues et al., 2018; Walle et al., 2019).

The σ^2GCA in the F_2 generation was less than the σ^2SCA, the ratio of σ^2GCA/σ^2SCA was also less than 1, and $H/D^{1/2}$ ratio was greather than 1. The fact that σ^2GCA/σ^2SCA ratios are less than 1 and $H/D^{1/2}$ ratio is greater than 1 shows that the non-additive gene effect is effective and the superior dominance of these genes. Looking at these results selection could be started in the future generations. Contrary to us, Magar et al. (2016), Gupta et al. (2017), Kumari and Chauhan (2018), Rodrigues et al. (2018), Olunloyo et al.(2019), Purnamasari et al.(2019), Owusu et al.(2020), and Verma et al. (2020) found that for the hundred seed weight the additive gene effect was important.

In F_2 generation, Sırma, and Ülkem cultivars have significant and positive values while Pekşen, Karagöz and Amazon genotypes have significant and negative values. (Table 6). Sırma and Ülkem cultivars, which were found to be positively important in increasing the hundred-seed weight in this generation, emerged as suitable parents for breeding studies for this trait. In their previous studies, Dias et al. (2016), Magar et al. (2016), Gupta et al. (2017), Kumari and Chauhan (2018), Rodrigues et al. (2018), Olunloyo et al. (2019), Purnamasari et al. (2019), Verma et al. (2020) were in harmony.

In the F_2 generation, "Karagöz x Sırma", "Amazon x Ülkem", "Karagöz x Amazon", "Pekşen x Karagöz", "Amazon x Karagöz" hybrids were determined to have positive and significant effects and emerge as a promising genotype that can be used in breeding studies for this purpose (Table 6). "Sırma x Pekşen", "Amazon x Sırma", "Sırma x Karagöz" are negative had a significant effect. Dias et al. (2016), Magar et al. (2016), Gupta et al. (2017), Kumari and Chauhan (2018), Rodrigues et al. (2018), Olunloyo et al. (2019), Purnamasari et al. (2019), Owusu et al. (2020), Pallavi et al. (2020), and Verma et al. (2020) found genotypes with significant positive SCA for a hundred seed weight in their studies.

Table 8
Heterosis (%) values for investigated traits in full-diallel hybrid set

F_2 Populations	Number of Seeds per Plant	Seed Yield per Plant	Hundred Seed Weight	Protein Ratio	Protein Yield in Plant
Pekşen X Sırma	17,97	18,16	1,03	-4,37**	14,20
Pekşen X Ülkem	22,60	24,51	2,68	2,10**	27,23*
Pekşen X Amazon	42,39	54,96	20,90**	2,06**	58,27**
Pekşen X Karagöz	106,48	131,55	30,30**	3,33**	138,97**
Sırma X Pekşen	32,05	3,27	-17,49*	3,21**	7,82
Sırma X Ülkem	6,25	6,23	-6,40*	3,97**	10,84
Sırma X Amazon	11,38	10,61	6,76*	-5,70**	5,23
Sırma X Karagöz	-6,96	-29,23	-30,11**	4,47**	-25,06*
Ülkem X Pekşen	28,00	24,42	-1,24	2,92**	28,27*
Ülkem X Sırma	0,15	-2,30	-6,08*	2,58**	0,41
Ülkem X Amazon	33,17	22,75	-3,91	-3,04**	19,08*
Ülkem X Karagöz	6,46	4,64	4,86	-3,49**	1,02
Amazon X Pekşen	12,06	27,69	25,84**	0,35**	28,22*
Amazon X Sırma	31,97	17,10	-4,91	3,82**	22,51*
Amazon X Ülkem	-16,73	-10,54	11,94*	-1,86**	-12,16
Amazon X Karagöz	74,90	77,97	11,03*	-0,82**	76,43**
Karagöz X Pekşen	32,62	60,73	28,58**	-3,18**	55,44**
Karagöz X Sırma	14,39	-12,11	-17,07**	1,13**	-9,87
KaragözX Ülkem	19,73	22,52	6,96*	1,45**	24,38*
KaragözX Amazon	31,06	64,39	46,58**	-1,49**	61,80**
Mean	20.00	25.87	5.51	0.37	26.65

** : significant at 1% level; * : significant at 5% level

3.8. Protein ratio:

According to the average of the protein ratio in the F_2 generation, it was determined that the parental values ranged from 26.69 % (Sırma) to 28.82% (Pekşen), and in the F_2 generation the protein ratio ranged between 25.64% (Sırma x Amazon) and 29.51% (Pekşen x Karagöz) (Table 5). Researching this subject, Hall et al. (2003), Sert and Ceyhan (2012), Kadam et al. (2013), Harmankaya et al. (2016), Joshi et al. (2022) found similar results.

It has been determined that in the F_1 generation, The σ^2GCA was less than The σ^2SCA, the ratio of σ^2GCA/σ^2SCA was also less than 1 and the $H/D^{1/2}$ ratio was greater than 1. The fact that the σ^2GCA/σ^2SCA ratios of the protein ratio were less than 1 shows us that the non-additive gene effect is effective in the inheritance of this trait. Likewise, a ratio of $H/D^{1/2}$ greater than 1 indicates superior dominance. Researching this subject, Sharma and Mehta (2014), Magar et al. (2016), and Purnamasari et al. (2019) determined that the additive gene effect is preponderant for the protein feature in cowpea.

When we examine the effect value of GCA in the F_1 generation, we find that among the genotypes, Pekşen and Ülkem show significant and positive values. It was determined that the Sırma, Amazon and Karagöz genotypes had a negative significant GCA effect (Table 6). Pekşen and Ülkem genotypes, which were found to have a positive and significant GCA effect value, were determined as promising parents that could be used in terms of protein ratio in crossing studies.

Looking at the SCA effects of hybrids in the F_2 generation, "Sırma x Pekşen", "Ülkem x Pekşen", "Amazon x Sırma", "Pekşen x Ülkem", "Sırma x Ülkem", "Pekşen x Amazon", "Sırma x Karagöz" hybrids have positive and significant SCA effect. Hybrids which showed high positive SCA effect with high reproductive potential for protein ratio, which could be used as genotype in future generations. In this study, parents and hybrids with significant positive SCA effect and high positive SCA effect for protein ratio are highlighted as suitable materials that can be used in protein ratio based selection studies Tchiagam et al. (2011), Sharma and Mehta (2014), Magar et al. (2016), Kumari ve Chauhan (2018), Purnamasari et al. (2019), found that the GCA and SCA values of different numbers of parents and crosses were important for the protein ratio in cowpea.

In the F_2 generation, the average heterosis value determined in this study for the protein ratio was -0.46%, while the heterobeltiosis value was -2.33%. Heterosis values varied between -8.17% (Karagöz x Pekşen) and 4.81% (Sırma x Karagöz), while heterobeltiosis values ranged between -9.10% (Karagöz x Pekşen) and 2.78% (Ülkem x Amazon) (Tables 7 and Table 9). More than half of the hybrids showed positive heterosis for this trait and were statistically significant, indicating that suitable

material for high protein is available. Analyzing the heterosis and heterobeltiosis values for the protein ratio, Sharma and Mehta (2014), Sarath and Reshma (2017), Joshi et al. (2022) found low or high mean heterosis and heterobeltiosis values for this feature.

Table 9
Heterobeltiosis (%) values for investigated traits in full-diallel hybrid set

F_2 Populations	Number of Seeds perPlant	Seed Yield per Plant	Hundred Seed Weight	Protein Ratio	Protein Yield in Plant
Pekşen X Sırma	-1,63	-9,86	-15,15**	-7,72**	-10,62
Pekşen X Ülkem	5,33	1,36	-9,89	1,39*	4,14
Pekşen X Amazon	34,57*	46,13**	20,29*	1,85*	49,56**
Pekşen X Karagöz	68,70**	87,83**	27,50**	2,67**	92,85**
Sırma X Pekşen	10,10	-21,22*	-30,70**	-0,41	-15,62
Sırma X Ülkem	2,51	-2,43	-11,08*	1,00	4,60
Sırma X Amazon	-2,54	-11,88	-10,71*	-8,82**	-13,97
Sırma X Karagöz	-33,79*	-53,03**	-42,32**	1,43*	-49,53**
Ülkem X Pekşen	9,96	1,29	-13,33*	2,21**	4,99
Ülkem X Sırma	-3,38	-10,26	-10,77*	-0,35	-5,24
Ülkem X Amazon	20,31	4,88	-16,04**	-3,51**	2,16
Ülkem X Karagöz	-22,53	-27,22*	-9,68	-3,53**	-29,73*
Amazon X Pekşen	5,91	20,42	25,21**	0,14	21,17*
Amazon X Sırma	15,47	-6,71	-20,46*	0,38	0,16
Amazon X Ülkem	-24,77*	-23,56*	-2,20	-2,34**	-24,65*
Amazon X Karagöz	36,81*	38,06*	9,18	-1,26*	36,42*
Karagöz X Pekşen	8,36	30,38*	25,82**	-3,80**	25,44*
Karagöz X Sırma	-18,60	-41,68**	-31,57**	-1,80*	-39,29**
KaragözX Ülkem	-12,87	-14,79	-7,88	1,40*	-13,49
KaragözX Amazon	2,52	27,52*	44,14**	-1,93**	25,11*
Mean	5.02	1.76	-3.98	-1.15	3.22

** : significant at 1% level; * : significant at 5% level

The heritability in the broad sense calculated for the protein ratio in the F_2 generation was 0.97, and the heritability in the narrow sense was determined as 0.18 (Table 5). The high heritability in the broad sense and the low heritability in the narrow sense in the protein ratio means that the effect of the environmental variance of this feature may be high. Analyzing the inheritance of the protein ratio, Sharma and Mehta (2014), Purnamasari et al. (2019) found high heritability in narrow and broad in this trait. Since the non-additive gene effects are important in the heritability of protein ratio and the heritability in the narrow sense is low, it is more appropriate to start the selection after a few generations.

3.9. Protein yield in plant:

According to the single plant proteins yields in the F_2 generations, the parental values ranged from 4.79 g/plant (Karagöz) to 13.80 g/plant (Sırma), and the plant proteins yields in the F_2 generation varies between 6.96 g/plant (Sırma x Karagöz) and 15.04 g/plant (Pekşen x Karagöz) (Table 5). The findings of this study are in harmony with the findings of (Kumari and Chauhan, 2018).

For the protein yields in the F_2 generation, it is seen that σ^2GCA was less than σ^2SCA, the ratio of σ^2GCA/σ^2SCA was less than 1 and the $H/D^{1/2}$ ratio was greather than 1 (Table 5). The fact that σ^2GCA/σ^2SCA ratios were less than 1 and $H/D^{1/2}$ ratio was greater than 1 indicates that the non-additive gene effect and superior dominance are effective in the inheritance of this trait. After their analyses Magar et al. (2016), Kumari and Chauhan, (2018) found the similar results.

When GCA is examined, while Pekşen, Sırma, Amazon and Ülkem genotypes have significant and positive values in the F_2 generation, Karagöz genotype has significant and negative values (Table 6). Considering the SCA effects of the hybrids, "Amazon x Sırma", "Karagöz x Ülkem", "Pekşen x Karagöz" and "Amazon x Karagöz" crosses which were positive emerge as a promising genotype that can be used in breeding studies for this purpose(Table 6). Magar et al. (2016), Kumari and Chauhan (2018), in their previous studies were in harmony

Average heterosis value determined for biological yield in F_2 generation was 26.65%, heterobeltiosis value was calculated as 3.22%. Heterosis values varied between -25.06% (Sırma x Karagöz) and 138.97% (Pekşen x Karagöz), while heterobeltiosis values ranged between -49.53% (Sırma x Karagöz) and 92.85% (Pekşen x Karagöz) (Table 4.42), ın conformity with those obtained by Kadam et al. (2013), Joshi et al. (2022).

Broad and narrow sense heritability was 0.88 and 0.16, respectively in the F_2 generation (Table 5). The determination of the non-additive gene effect in terms of this feature and the presence of positive heterosis in the examined generations revealed that selection should be made in advanced generations. Purnamasari et al. (2019) showed high heritability in the broad and narrow senses.

4. Conclusions

Considering the agronomic characteristics of the studied population, we can affirm that this population presented a sufficient variation. This study reveals that the non-additive and dominant genes were more effective in

the traits studied. Thus, in future generations, some selections could be considered.

5. Acknowledgements

This research was supported by the Selcuk University BAP project, number 21211007. This paper was summarized from Ph. D. Thesis of the first author (Christelle JOU-NTEUFA). The authors would like to acknowledge the financial support of Selcuk University.

6. References

Abdulazeez R, Akhadelor MO, Danazumi IB, Umar AA, Auta J (2019). Effect potash and onion on the nutritional and anti-nutritional composition of cooked brown-eyed cowpea, *Fudma Journal of Sciences*, 3: 381-386.

Ayo-Vaughan M, Ariyo O, Olusola A (2013). Combining ability and genetic components for pod and seed traits in cowpea lines. *Italian Journal of Agronomy*, 8(2):73-78.

Borivoj P, Mackic K, Mikic A, Cupina B, Pekşen E, Krstic D, Antanasovic S (2013). Effect of water stress on the yield of cowpea (*Vigna unguiculata* (L.) Walp.) in temperate climatic conditions. *Contemporary Agriculture, Novi Sad*, 62: 160-168.

Boukar O, Belko N, Chamarthi S, Togola A, Batieno J, Owusu E, Haruna M, Diallo S, Umar ML, Olufajo O, Fatokun C (2019). Cowpea (*Vigna unguiculata*): Genetics, genomics and breeding. *Plant Breeding*, 138 (4): 415-424.

Ceyhan E (2004). Bezelye Ebeveyn ve melezlerinde bazı tarımsal özelliklerin ve kalıtımlarının çoklu dizi analiz metoduyla belirlenmesi. Doktora, *Selçuk Üniversites*, Konya, 103.

Ceyhan E, Avcı MA, Karadaş S (2008). Line x tester analysis in pea (*Pisum sativum* L.): Identification of superior parents for seed yield and iıts components, *African Journal of Biotechnology*, 7(16): 2810-2817.

Ceyhan E, Kahraman A, Avcı MA, Dalgıç H (2014). Combining ability of bean genotypes estimated by line x tester analysis under highly-calcareous soils. *The Journal of Animal and Plant Sciences*, 24 (29): 579-584.

da Costa AF, do Vale LS, de Oliveira AB, de Brito Neto JF, Ribeiro, WS, Cardoso GD (2017). Evaluation of yield performance in cowpea genotypes (*Vigna unguiculata* (L.) Walp.). *Australian Journal of Crop Science*, 11(3): 308-312.

Dias FTC, Bertini CHCD, Filho FRF (2016). Genetic effects and potential parents in cowpea. *Crop Breeding and Applied Biotechnology*, 16(4): 315-320.

Gupta RP, Patel SR, Modha K, Wadekar PD (2017). Generation Mean analysis for yield and yield components in cowpea [*Vigna unguiculata* (L.) Walp.]. *International Journal of Current Microbiology and Applied Sciences*, 6(7): 2231-2240.

Hall AE (2004). Breeding for adaptation to drought and heat in cowpea. *European Journal of Agronomy*, 21: 447–454.

Hall AE (2012). Phenotyping cowpeas for adaptation to drought. *Frontiers in Physiology*, 3(155): 1-8.

Hall AE, Cisse N, Thiaw S, Elawad HOA, Ehlers JD, Ismail MA, Fery RL, Roberts PA, Kitch LW, Murdock LL, Boukar O, Phillips RD, McWattersj KH (2003). Development of cowpea cultivars and germplasm by the bean/cowpea CRSP. *Field Crops Research*, 82(2): 103-134

Harmankaya M, Ceyhan E, Çelik AS, Sert H, Kahraman A, Özcan MM (2016). Some chemical properties, mineral content and amino acid composition of cowpeas [*Vigna sinensis* (L.) Savi]. *Quality Assurance and Safety of Crops & Foods*, 8 (1): 111-116.

Horn LN, Shimelis H (2020). Production constraints and breeding approaches for cowpea ımprovement for drought prone agro-ecologies in Sub-Saharan Africa *Annals of Agricultural Sciences*, 65 (2): 83-91.

Joshi SN, Desai SS, Sawardekar SV, Sanap PB, Mane AV, Kunkerkar RL (2022). Heterosis for yield and yield-related traits in red cowpea (*Vigna unguiculata* L. Walp). *Pharma Innovation*, 11 (4): 1125-1130.

Kadam YR, Patel AI, Patel JM, Chaudhari PP, More SJ (2013). Heterosis study in vegetable cowpea [*Vigna unguiculata* (L.) Walp.]: *Crop Research*, 45 (1,2,3): 202-205.

Kumari J, Chauhan DA (2018). Genetic analysis for yield and contributing characters in cowpea (*Vigna unguiculata* (L.) Walp). *Journal of Pharmacognosy and Phytochemistry*, 7(4): 2453-2458.

Magar NM, Badhe PL, Raut DM, Borole DN, Pawar VY (2016). Diallel analysis in cowpea (*Vigna unguiculata* (L.) Walp.), *Electronic Journal of Plant Breeding*, 7 (2): 291-295.

Olunloyo AA, Adedokun S, Aderemi F, Alarape A, Aderemi A, Sulaiman O (2019). Diallel analysis of cowpea cultivar ıfe brown and its mutants. *International Journal of Forest, Animal and Fisheries Research*, 3(5): 182-187

Owusu EY, Mohammed H, Manigben KA, Adjebeng-Danquah J, Kusi F, Karikari B, Sie EK (2020). Diallel analysis and heritability of grain yield, yield components, and maturity traits in cowpea (*Vigna unguiculata* (L.) Walp.). *The Scientific World Journal*, 2020 (9390287): 1-9.

Özcan K, Açıkgöz N (1999). Populasyon genetiği için bir istatistik paket program geliştirmesi. 3. Tarımda Bilgisayar Uygulamaları Sempozyumu, Çukurova Üniversitesi, 3-6 Ekim, Adana.

Pallavi, Singh A, AC (2020). Diallel analysis for combining ability in cowpea (*Vigna unguiculata* (l.) Walp). *The Pharma Innovation*, 9(9): 500-502.

Peksen E, Artık C (2004). Comparison of some cowpea (*Vigna unguiculata* L. Walp) genotypes from Turkey for seed yield and yield related characters. *Journal of Agronomy*, 3(2): 137-140.

Pekşen A, Pekşen E (2013). Agronomic and morphological characters of newly registered peksen and reyhan vegetable cowpea cultivars in Turkey. *International Journal of Current Microbiology and Applied Sciences*, 2(9): 133-140.

Pekşen E, Pekşen A (2012). Evaluation of vegetable cowpea (*Vigna unguiculata* (L.) Walp.) breeding lines for cultivar development. *Igdır University Journal of Institute of Science and Technology*, 2(4): 9-18.

Penchalaraju M, Don Bosco SJ (2022). Legume protein concentrates from green gram, cowpea, and horse gram. *Journal of Food Processing and Preservation*, 46 (4):1-11.

Porch TG, Hall AE (2013). Heat tolerance, In: Genomics and breeding for climate-resilient crops, Eds: Kole, C., *Berlin*: Springer, p. 167-202.

Purnamasari I, Sobir, Syukur M (2019). Diversity and inheritance in cowpea (*Vigna unguiculata*) on protein and yield components characters. *Biodiversitas*, 20(5): 1294-1298.

Ravelombola W, Shi A, Weng Y, Motes D, Clark J, Chen P, Srivastava V, Qin J, Dong L, Yang W, Bhattarai G, Sugihara Y (2018). Association Analysis of salt tolerance in cowpea (*Vigna unguiculata* (L.) Walp) at germination and seedling stages. *Theoretical and Applied Genetics*, 131(1): 71-91.

Rodrigues EV, Damasceno-Silva KJ, Rocha MDM, Bastos EA, Santos AD (2018). Diallel analysis of tolerance to drought in cowpea genotypes. *Revista Caatinga*, 31(140-47): 40.

Sarath PS, Reshma T (2017). Heterosis in cowpea (*Vigna unguiculata* L. Walp) for selected traits. *International Journal of Current Microbiology and Applied Sciences*, 6(7): 522-526.

Sert H, Ceyhan E (2012). Hatay ili ekolojik şartlarında börülce (*Vigna sinensis* (L.) Savi) çeşitlerinin tane verimi ve bazı tarımsal özellikleri üzerine farklı bitki sıklıklarının etkileri. *Selçuk Tarım ve Gıda Bilimleri Dergisi*, 26(1): 34 -43.

Sharma D, Mehta N (2014). Combining ability analysis for protein content in relation to heterosis and green pod yield in vegetable cowpea. *Indian Journal of Horticulture*, 71(4): 577-580.

Sing BB (2020). Cowpea: The food legume of the 21st century, *USA*, Crop Science Society of America, p. 1-192.

Tchiagam JBN, Bell JM, Nassourou AM, Njintang NY, Youmbi E (2011). Genetic analysis of seed proteins contents in cowpea (*Vigna unguiculata* L. Walp.). *African Journal of Biotechnology*, 10(16): 3077-3086.

Veeranagappa P, Manu B, Prasad G, Blair MW, Hickok D, Naveena NL, Manjunath L, Tripathi, K (2022). advanced breeding strategies for abiotic stress tolerance in cowpea, ın: genomic designing for abiotic stress resistant pulse crops. Eds: Kole, C., *Cham*: Springer, p. 115-144.

Verma AK, Mehta AK, Sharma D and Singh RP (2020). Genetic Analysis of Pod Yield and Its Contributing Traits in Cowpea (*Vigna unguiculata* L. Walp), *International Journal of Chemical Studies*, 8 (1), 965-970

Walle T, Mekbib F, Amsalu B, Gedi M (2019). Genetic diversity of Ethiopian cowpea [*Vigna unguiculata* (L) Walp] genotypes using multivariate analyses. *Ethiopian Journal of Agricultural Sciences*, 29(3): 89-104.

Yurtsever N (1984). Deneysel istatistik metotlar, *Toprak ve Gübre Araştırma Enstitüsü*, Ankara.

Selcuk Journal of Agriculture and Food Sciences SJAFS

http://sjafs.selcuk.edu.tr/sjafs/index

(2022) 36 (3), 312-319

e-ISSN: 2458-8377

DOI:10.15316/SJAFS.2022.040

Research Article

Using Contamination Indices for Assessments of Heavy Metals Status of Soils around Mercury Mine, in Kurşunlu, (Konya) Province

Ayşen AKAY[1], Hasan Hüseyin ÖZAYTEKIN[2,*]

[1]Selçuk University, Faculty of Agriculture, Department of Soil Science and Plant Nutrition, Konya, Türkiye

[2] Karamanoğlu MehmetBey University, Vocational School of Technical Sciences, Department of Crop and Animal Production, Karaman, Türkiye

ARTICLE INFO

Article history:
Received date: 23.02.2022
Accepted date: 16.08.2022

Keywords:
Mercury mine
Soil pollution
Contamination index.

ABSTRACT

The objective of this study was to establish contamination level for, Pb, Zn, Cu, Ni, Cr, As, Hg, Sb, Co, and Cd in soils around the old Hg mine area central part of Turkey. 34surface samples from mine area were collected. Samples were digested and the metals were determined by ICP-OES and assess metal contamination in soils by using Enrichment factor (EF), contamination factor (Cf), Geoaccumulation index (Igeo), hazard quotient (HQ) (individual metal), contamination index (Cd), modified contamination index (mCd), pollution load index (PLI), and hazard load index (HLI) (multi metal).
Enrichment factor (EF), contamination factor (Cf), Geoaccumulation index (Igeo), hazard quotient (HQ)indicate that the soil in the most of studied points were unpolluted, slightly polluted or moderate degree of pollution with respect to Pb, Cr, Cd, Zn, Cu, Co and Ni, while the soils in all studied stations were extremely contaminated with respect to Hg, Sb and As. When the obtained result to by evaluated the cumulative effect of metal pollution load index (PLI), the contamination degree (Cd) and modified Contamination Degree (mCd), and hazard load index were found to be very high high pollution, ultra-high degree pollution and medium pollution respectively indicating that the studied soils in were polluted by total of studied heavy metals.

1. Introduction

C Soil is a dynamic natural resource for the survival of human life and due to its complex matrix is the principal receiver of the persistent contaminants such as heavy metals (Luo et al., 2006). Every soil comprises some natural quantities of heavy metals, at concentrations called backgrounds. The magnitude of a metal's background depends upon the composition of the parent rock material from which the soil was derived (Scazzola et al., 2003).Heavy metals pollution assessment nowadays is becoming an important task due to the increase in anthropogenic actions related to industrialization central sources of heavy metals in terms of petrochemicals, compost, pesticides, animal manures, sewage sludge, leaded paints as well as the indiscriminate dumping of wastes inland fills (Sajn et. all 1998;Davidson et al, 2006;Stafilov et al., 2010; Dumitrescu et al. 2012). Pollution of the natural environment by heavy metals is a universal problem because these metals are indestructible and most of them have toxic effects on living organisms, when permissible concentration levels are exceeded.

Heavy metal contamination of soils concern several scientists because of the potential toxicity of metals (Homa et al., 2003). Heavy metals accumulation has shown to be detrimental to both plants and animals in human body, for example, it is capable of causing neurological disorders, damage to the internal organs of the body and even death while in plants it shows negative effects on photosynthesis and absorption /exchange of gases (Dumitrescuet al, 2012; Edward et al, 2013; Senila et al, 2013; Alia et al, 2015).

Heavy metals contamination assessment in soils has been carried out successfully all over the world using quality index method. These quality index methods have proved to be a significant tool for effectively gathering composite influence of indicators to the overall contamination (Bhuiyan et al. 2015). Several evaluation methods have been utilized by various authors in heavy metal pollution assessment in soils and sediments: Single in-

* Corresponding author email: *hhozaytekin@gmail.com*

dex factor (Pi), Nemerow's pollution index (PN), potential ecological risk index (RI), enrichment factor (EF), contamination factor (CF), Geoaccumulation index (Igeo), contamination index (Cd), pollution load index (PLI) as well as hazard quotient. Pi and CF are computed as basis for obtaining PN and PLI respectively. Igeo can be used to distinguish the effects that human activities have on the environment (Bello et al. 2016). RI considers the toxicity of the pollutant as a means of evaluating the ecological risk, the value does so by comparing the concentration of the pollutant with the back ground value. EF represents the values that assess anthropogenic influences on heavy metals in sediments; the measurement uses aluminum (Al) as a conservative element (Zhang et al. 2015).

Studies on heavy metal levels in soils and waters of different fresh water systems in Turkey. There are many studies on the determination of pollution status due to anthropogenic (urban, agriculture, industrialization, road and railway environment) activities and the determination of the effects of various applications on heavy metal contents using pollution quality indexes. (Uğulu, 2015¸ Uğulu et al.2019; Coşkun et al. 2006; Yılmaz et al. 2003; Çelebi and Kara 2007; Demir et al. 2016; Göçmez 2006; Haktanır et al., 1995; Kara et al., 2004; Tokalıoğlu and Kartal 2005; Sezgin et al., 2003; Horasan and Arık 2019; Sungur et al., 2014). In addition, the mobility and bioavailability of heavy metals were determined by sequential extraction method in soils and greenhouses in various regions (Sungur et al., 2015; Sungur et al., 2021).

Understanding the contamination characteristics of heavy metals in soils and identifying their environmental exposure risks not only are the basic preconditions for soil pollution prevention, but also provide important information for making decisions for remediation of contaminated soils. The objectives of this study are to identify the heavy metal pollution level of soils around Ladik-Sızma mercury mine (about 30 km north - northwest of Konya province) using enrichment factor (EF), contamination factor (Cf), Geoaccumulation index (Igeo), hazard quotient (HQ) (individual metal), contamination index (Cd), modified contamination index (mCd), pollution load index (PLI), and hazard load index (HLI) (multi metal).

2. Materials and Methods

Site description

Kurşunlu Hg mine (33° 04' N; 6° 38' W), which is placed north - northwest of the Konya Province, Turkey. Ladik-Kurşunlu Hg bed located in an environment where the metamorphic rocks located like as a lot of Hg bed in Turkey. Phyllite, schist and carbonated rocks are the main lithological units observed around the beds. The carbonate rocks are composed of gray-dark gray colored limestone, marble and dolomitic marbles which are recrystallized locally. This unit (Bozdağ formation; is a Silurian-Carboniferous age (Wesner, 1968) and it is generally composed of phyllite (sericite-quartzphyllite, chlorite-quartz phyllite), schist (sericite-biotite-(Bagnkurt formation), which consists of quartzite, quartz schist, sericite-quartz schist), quartzite, metaconglomera and transported methacarbonate blocks (Aydın, 1996). Both carbonates and phyllites and schists were cut by Karatepe metamorphic rocks after the Carboniferous age (Yıldız, 1978). These magmatites, which are called metaporfir by Bayiç (1968), contain mainly feldspar, and less frequently, muscovite, quartz and sfen. The study area is under the influence of semi-arid continental climate in Central Anatolia. The average annual precipitation in the region is 323,6 mm, and the annual evaporation is 978,2 mm. The annual average temperature is 11,3ºC, the average soil temperature at 50 cm is 14,2. A significant part of the precipitation falls in the winter months, and drought is observed in June, July and August. (MGM, 2016). In the light of these data, the climate type of the region is "BSk" Semi-Arid Steppe Climate (Cold) according to the Köppen climate classification. When the De Martonne-Bottman drought index formula is applied to the climate data of the study region, it is determined that the semi-arid-less humid Mediterranean climate is dominant (Akman 1990). According to the diagrams (precipitation-evaporation-temperature) prepared in the light of the information obtained, the temperature regime of the study area is mesic and the humidity regime is xeric (Nachtergaele 2001).

Soil samples collection and analysis

Surface soil samples (0 to 20 cm) were collected in the Kurşunlu Hg mine area. Soil samples were taken from randomly chosen spots at certain distances from the land around the mine. Each sample was composed of five subsamples collected around the point. In total, 34 composed samples were analyzed. Partical size distribution, pH, electrical conductivity (EC) and heavy metal contents were determined after drying the samples at 50°C in a hot air oven (USDA 2004). Element concentrations include Cu, Ni, Co, Zn, Pb, As, Cr, Sb, Hg and Cd were determined in Aqua regia acid mixture (HNO3: HCl: 1: 3), and the extracts obtained using Atomic Absorption Spectrometers (AAS) with standard solutions in a similar manner to described by procedure(Sparks et al.,1996).

Pollution Indices

Enrichment factor (EF) (Lacatusu, R. 1998), contamination factor (Cf) (Tomlinson ve ark 1980), geoaccumulation index (Igeo) (Muller 1979), hazard quotient (HQ) (Epa, 1992) (individual metal), contamination index (Cd) (Hakanson (1980), modified contamination index (mCd) (Abrahim and Parker 2008), pollution load index (PLI) (Tomlinson et al. 1980), and hazard load index (HLI) (multi metal) were employed to assess the pollution of metals in the soil situated the mine. Calculating the degree of contamination by a specified heavy metal obliges that the contaminant metal concentration be compared with a reference material (geochemical background). Such reference material should be an uncontaminated substance that is comparable with the studied samples, as reported with Maanan et al. (2014),

Nouri and Haddıouı (2016). To calculating the hazard quotient (HQ) for Sb, Canadian Soil Quality Guidelines for the Protection of Environmental and Human Health (Turekian and Wedepohl 1961).For other elements, Ministry of Environment and Forestry Regulation on Control of Soil Pollution were used. By considering Fe as a reference element the enrichment factor for heavy metals were calculated.

-Enrichment factor is calculated with the following formula.

EF = (CX/CFe) soil (CX/CFe) reference

Here, (CX/CFe) Soil: The ratio of the metal concentration studied in the soil sample to the Fe concentration

(CX/CFe)Reference: The ratio of the metal concentration studied in the reference sample to the Fe concentration The metal concentration of the reference sample is the abundance of elements in the earth's crust.

The contamination factor (Cf) was calculated by the ratio of the metal content at each sampling point to the abundance of the element in the earth's crust of that metal. Cf is found by the equation given below (Hakanson, 1980).

Cf=Cmetal/Co

Cmetal: The metal concentration in the soil sample,

Co: The abundance value of the metal in the earth's crust

Contamination index (Cd)" is the sum of all pollution factors of a given basin. Cd was calculated as the sum of Cf for each sample.

Cd=∑_(i=1)^(i=n)▒Cf

Similarly, it is possible to approximate the exact degree of impurity using the modified contamination index (mCd), method, and for this purpose, the modified contamination index (mCd) is calculated by the following equation.

MCd=(∑_(i=1)^(i=n)▒Cf)/n

The geoaccumulation index (Igeo) of any metal is found by calculating the base 2 logarithm of the measured total metal concentration over the background concentration using the following mathematical relationship

Igeo=log2(Cn/(1.5xBn))

Here, Cn is the measured concentration of "n" heavy metal in the soil sample, Bn is the geochemical background (reference) value of the element "n" in the earth's crust, and 1.5 is the background matrix correction factor resulting from lithogenic effects.

Pollution load index (PLI) is frequently used to compare the pollution status of different places, since all the metals examined are handled in a single index. The index is obtained by calculating the pollution factors (Cf) of each metal. Pollution load index (PLI) is calculated with the following formula al.,

PLI=(Cf1 * Cf2 * Cf3 * ….. * Cfn)1/n

Cf= Cmetal / Co

Here Cmetal : Metal concentration in the sediment sample

Co : The basic (background) value of the metal

Cf : Pollution factor

n : Number of metals

-The soil Hazard Quotient (HQ) is the ratio of the heavy metal concentration of surveyed soil samples to reference permissible limit and is computed using the relation,

HQ=Cc/Cp (3)

Where, Cp and Cc=reference maximum permissible limit of heavy metal concentration and the concentration obtained in the sampled area, respectively

The hazard load index: Similar to the pollution load index that takes contamination factors as inputs, an analogous relation termed hazard load index was developed by adoption and amendment of the pollution load equation and was presented in equation

3. Results and Discussion

Soil Properties

Some soil properties were presented in table1. According to Table 1 soil pH was slightly alkaline and changed between 6.76 and 7.55, EC values ranged from 59.1 to 270 (µS/cm) and lime contents were ranged 1.03% to 21.06%. The organic matter content was highly variable and varied between 1.43% and 7.16%. Clay content is very low in most of samples and changedbetween4.2% and 28.8%. Sand contents were very high and ranged from 48% to 89.2%. High coefficients of variation were observed in some of the soil properties, and this was due to the fact that the sampling points were located on different physiographic units and were accordingly affected by the transport and accumulation processes. The high coefficient of variation observed especially in organic matter, EC, clay and silt values supports this situation.

The metals concentration

Heavy metal statuses of soils were shown in Table 2. According to table highest content of Cu concentration is 109.70 ppm while mean value is 52.86 ppm. The Pb content was changed from 26.9 to 154.0 ppm. Zn concentrations were changed between 8.0 and 152 ppm and all Zn values were under reference value. Ni content ranged between 30.2 and 165.7 ppm. Co contents ranged 10.3 and 44.2 ppm. Mean Co value is upper then reference value. As contents are changed between 21.0 and 382 ppm and all As content bigger than background value. Cd concentrations were changed between 0.25 and 3.30 ppm and mean Cd value were upper reference value. Sb content ranged between 11.0 and 2443.5 ppm. All Sb values very high according to reference value. Similar results were observed for Hg. Hg content ranged between 0.6 and 100 ppm and all Hg values were upper reference value. Cr contents ranged 50.0 and 333.0 ppm. Mean Cr value is upper then reference value.

Table 1
Some soil properties of soil samples

Soil properties							
Samplenumber	pH	Organic matter (%)	EC (µS/cm)	Lime (%)	Particle size distribution		
					Sand (%)	Silt (%)	Clay (%)
1	7,13	3,61	123,30	2,85	54,0	35,2	10,8
2	7,17	4,40	120,05	2,28	69,2	22,0	8,8
3	7,33	2,02	139,75	7,06	68,0	22,6	9,4
4	7,53	3,49	113,00	2,62	68,0	25,2	6,8
5	6,96	3,90	94,85	2,51	73,2	16,0	10,8
6	7,10	3,08	75,25	1,82	89,2	4,0	6,8
7	7,37	4,60	106,80	2,39	85,2	4,0	10,8
8	6,82	4,80	79,35	2,62	57,2	14,0	28,8
9	7,15	7,16	79,45	3,98	79,2	16,0	4,8
10	6,84	4,03	216,60	2,05	65,2	8,0	26,8
11	6,89	4,95	108,75	1,72	67,2	18,0	14,8
12	6,76	1,99	65,80	2,17	65,2	22,0	12,8
13	7,13	3,28	165,75	2,73	48,0	38,2	13,8
14	7,55	4,68	154,30	5,12	55,2	20,0	24,8
15	7,15	3,93	96,45	2,73	68,0	27,8	4,2
16	7,13	5,34	59,10	1,03	50,0	39,8	10,2
17	7,10	1,88	139,30	1,72	61,2	10,0	28,8
18	7,14	3,44	162,35	1,59	53,4	33,8	12,8
19	7,08	2,88	67,35	1,26	68,0	25,2	6,8
20	7,10	3,70	102,95	1,59	71,2	16,0	12,8
21	7,14	5,90	97,95	1,26	73,2	18,0	8,8
22	7,21	4,91	269,00	21,06	49,0	37,5	13,50
23	7,26	1,43	134,45	4,32	52,0	39,2	8,8
24	7,24	2,01	88,65	3,30	67,4	23,8	8,8
25	7,04	2,13	86,15	2,05	54,0	33,2	12,8
26	7,35	6,74	124,55	3,87	54,0	41,2	4,8
27	7,50	3,14	66,60	1,94	68,0	25,9	6,1
28	6,99	3,39	91,90	1,71	75,2	14,0	10,8
29	7,13	1,99	270,00	1,71	61,2	30,0	8,8
30	7,13	3,52	59,90	1,59	50,0	39,8	10,2
31	7,36	6,29	114,15	5,35	65,4	29,2	5,4
32	7,13	6,83	219,50	6,72	58,0	35,2	6,8
33	7,29	2,19	80,00	2,62	68,0	25,9	6,1
34	7,43	4,79	259,50	20,58	57,4	36,8	5,8
Min	6,76	1,43	59,1	1,03	48.0	4.0	4,2
max	7,55	7,16	270	21,06	89,2	41,2	28,8
mean	7,17	3,89	124,49	3,82	63,78	24,93	11,29
SD	0,19	1,54	59,50	1,47	11,73	10,06	7,68
CV (%)	2,6	39,5	47,7	2,3	18,4	40,3	68,0

Table2
Means and ranges of heavy metals of soil samples

	Metals									
	Cu (ppm)	Pb (ppm)	Zn (ppm)	Ni (ppm)	Co (ppm)	As (ppm)	Cd (ppm)	Sb (ppm)	Cr (ppm)	Hg (ppm)
Min	19,60	26,90	8,00	30,20	10,30	21,00	0,25	11,00	50,00	0,60
Max	109,70	154,00	152,00	165,70	44,20	382,00	3,30	2443,50	333,00	100,00
Mean	52,86	58,96	105,15	98,47	25,43	74,16	0,94	492,56	160,78	22,23
SD	19,63	32,86	24,03	38,01	9,87	73,70	0,78	595,31	77,75	23,24
CV(%)	37,14	55,73	22,85	38,60	38,83	99,38	82,81	120,86	48,36	104,58

Pollution assessment of soil metals using geochemical indicators

Popular soil contamination assessment methods can be classified into two categories: quantitative and qualitative. The qualitative methods, such as principal component analysis (PCA), factor analysis, and cluster analysis, are inferential and indicative. These multivariate analyses require that each variable shows a normal distribution and that the whole dataset shows a multivariate normal distribution Some of the most commonly used quantitative methods are the contamination factor (CF), enrichment factor (EF), and Geoaccumulation index(Igeo).

Table 3 presents the contamination status of metals in the topsoil of the research site. As shown in Table 3 the results showed that the average EF values of Pb, Hg, Sb, As, Cr, Cd, Zn, Cu, Co, Ni, were 3.89, 53.22, 468.2, 8.10, 1.78, 4.19, 1.23, 1.26, 1.36 and 2.22 respectively. The mean EF values of Cr, Zn, Co were < 2, suggesting relatively minimal enrichment. EF is upper 2 Pb, Cd, Zn and Pb shown medium enrichment and for Hg, Sb and As very high enrichment.

The average Contamination factor (Cf) of Pb, Cr, Cd, Zn, Cu, Co, Ni, were, respectively which approves that the soils have a low or medium degree of pollution. Contamination factor of Hg, Sb and As with a mean of 59.46, 355.59, 8.10 respectively which approves that the soils have a very high degree of pollution

The mean Igeo values of vary the most but most of values have negative value for Pb, Cr, Cd, Zn, Cu, Co and Ni. But for Hg, Sb, and As the mean Igeo values very high. The result can be drawn from Table 3 that the Igeo values for Hg, Sb, and As are extremely contaminated level.

When HQ values are >1.0, the soils are considered to be contaminated by anthropogenic inputs. The degree of contamination of soils is low or medium (1 < PLI) for Pb, Cr, Cd, Zn, Cu, Co, and Ni but only for Hg, Sb and As which have very high (PI>5) contamination

Table 3

Assessed level of contamination effect founded on the enrichment factor (EF)[A]; Contamination factor (Cf)[B]; geo-accumulation index (Igeo)[C]; Hazard quation index (HQ)[D]; pollution load indices (PLI)[E]; degree of contamination (Cd)[F], modified degree of contamination (mCd)[G]and Hazard Load Index(HLI)[H]. Metals are reported in mg g-1 (*n=34*).

Individual Metal												
	Pb				Hg				Sb			
	EF	Cf	Igeo	HQ	EF	Cf	Igeo	HQ	EF	Cf	Igeo	HQ
Min	0,97	1,35	-0,16	0,09	1,01	1,50	0,00	1,01	8,0	7,33	2,29	0,55
Max	15,94	7,70	2,36	0,51	345,03	250,00	7,38	345,03	2258,1	1629,0	10,08	122,18
Mean	3,89	3,04	0,82	0,20	53,22	59,46	4,20	53,22	468,2	355,59	6,49	26,67
SD	3,76	1,78	0,73	0,12	80,27	64,89	2,12	80,27	679,3	439,88	2,37	32,99
	As				Cr				Cd			
	EF	Cf	Igeo	HQ	EF	Cf	Igeo	HQ	EF	Cf	Igeo	HQ
Min	1,51	1,62	0,11	1,05	0,63	0,56	-1,43	0,50	0,65	0,83	-0,85	0,08
Max	46,46	29,38	4,29	19,10	2,79	3,70	1,30	3,33	16,34	11,0	2,87	1,10
Mean	8,10	6,25	1,55	4,06	1,78	1,81	0,09	1,62	4,19	3,29	0,69	0,33
SD	11,03	6,72	1,11	4,37	0,56	0,91	0,73	0,82	4,74	2,83	1,10	0,28
	Zn				Cu				Co			
	EF	Cf	Igeo	HQ	EF	Cf	Igeo	HQ	EF	Cf	Igeo	HQ
Min	0,10	0,08	-4,15	0.03	0,49	0,44	-1,78	0,14	0,62	0,54	-1,47	0,52
Max	2,57	1,60	0,09	0.51	2,70	2,44	0,70	0,78	1,96	2,33	0,63	2,21
Mean	1,23	1,09	-0,61	0.35	1,26	1,19	-0,45	0,38	1,36	1,34	-0,27	1,28
SD	0,53	0,31	0,89	0.10	0,51	0,48	0,60	0,16	0,32	0,54	0,58	0,51
	Ni				Multi-Metal							
	EF	Cf	Igeo	HQ	PLI		Cd		mCd		HLI	
Min	0,76	0,44	-1,76	0,40	1,31		20,36		2,04		0,40	
Max	3,27	2,44	0,70	2,21	7,21		1700,23		170,02		2,19	
Mean	2,22	1,45	-0,19	1,31	3,99		428,01		42,80		1,22	
SD	0,60	0,58	0,66	0,53	1,88		474,77		47,48		0,57	

A Minimal enrichment (EF < 2), moderate enrichment (2 ≤ EF < 5), significant enrichment (5 ≤ EF < 20), very high enrichment (20 ≤ EF < 40) or extremely high enrichment (EF ≥ 40).

B Contamination factor No pollution (PI ≤ 1), slight pollution (1 < PI ≤ 3), moderate pollution (3 < PI ≤ 6) sever pollution (PI > 6).

C Uncontaminated (*I*geo ≤ 0), uncontaminated to moderately contaminated (0 <*I*geo ≤ 1), moderately contaminated (1 <*I*geo ≤ 2), moderately to heavily contaminated (2 <*I*geo≤3), heavily contaminated (3 <*I*geo≤4), heavily to extremely contaminated (4 <*I*geo ≤ 5), or extremely contaminated (*I*geo> 5).

D No pollution (HQ=PI ≤ 1), slight pollution (1 < PI ≤ 3), moderate pollution (3 < PI ≤ 5) and sever pollution (PI > 5).d *Cd* < 6 indicates a low degree of pollution; 6 <*Cd* < 12 is a moderate degree of pollution; 12 <*Cd* < 24 is a considerable degree of pollution; and *Cd* > 24 is a high degree of pollution indicating serious anthropogenic pollution.

E Background concentration (PLI = 0), uncontaminated (0 < PLI ≤ 1), uncontaminated to moderately contaminated (1 < PLI ≤ 2), moderately contaminated (2 < PLI ≤ 3), moderately to highly contaminated (3 < PLI ≤ 4), highly contaminated (4 < PLI ≤ 5), or very highly contaminated (PLI > 5).

F *Cd*< 6 indicates a low degree of pollution; 6 <*Cd* < 12 is a moderate degree of pollution; 12 <*Cd* < 24 is a considerable degree of pollution; and *Cd* > 24 is a high degree of pollution indicating serious anthropogenic pollution

G m*Cd*< 1.5 is nil to a very low degree of pollution; 1.5 ≤ m*Cd*< 2 is a low degree of pollution; 2 ≤ m*Cd*< 4 is a moderate degree of pollution; 4 ≤ m*Cd*< 8 is a high degree of pollution; 8 ≤ m*Cd*< 16 is a very high degree of pollution; 16 ≤ m*Cd*< 32 is an extremely high degree of pollution; m*Cd*≤ 32 is an ultra-high degree of pollution.

H Low contamination HLI ≤1, medium contamination 1< HLI ≤3, 3< HLI ≤6 Considerable, PI>6 very high contamination

In addition to give a comprehensive situation of heavy metals, the integrated pollution load index (PLI) for each sample was evaluated. The results showed that the PLI values of heavy metals in soils ranged from 1.31 to 7.21 with an average of 3.99 also indicating highly contaminated. The degree of contamination varied from 20.36 to 7.01 with a mean of 1700.23, which approves that the soils have high degree of pollution. The revised Hakanson formula was utilized to determine the mCd for all the studied elements. The results are shown in Table 3. The values vary from 2.04 to 170.02 with an average 42.80, representing that the investigate site presents an ultra-high degree of pollution. Hazard load index varied from 0.50 to 2.19 with a mean of 171.22, which approves that the soils have medium degree of pollution.

Heavy metals in uncontaminated soils and sediments are found in their silicate and primary mineral forms and are essentially inert. However, heavy metals in contaminated soils are more mobile and bind to other phases (Rauret, 1998). For this reason, the mentioned elements are considered to be of parent material origin, that is, of lithosphere origin, since there is no source of pollutants in the vicinity of the region.

There was a very high variability in the general analysis results of the studied soil samples. This situation is also reflected in the standard deviation and coefficient of variation of the results. However, the variability of the topography in the sampling area and the variability of the erosion and sedimentation timers accordingly caused the high coefficient of variation at the sample points.

Similarly, the coefficient of variation was found to be high in heavy metal contents. Especially in Hg, As and Sb, these values are quite high. This is due to the sampling strategy and the factors described below that affect the metal contents. While sampling, the mine area and waste dumping areas were chosen as the center, and sampling was made from different distances from the waste dumping area. Accordingly, the heavy metal contents of the soil samples showed significant changes. Especially the high coefficients of variation (CV) seen in Hg, Sb, As explain this situation. Much lower CV rates in other metals compared to these three metals also support the findings.

It is known that the ecological effects of heavy metals in the soil are closely related to the composition of the solid and liquid phase, the presence and behavior of heavy metals, the mobility and transformation and accumulation processes and forms in the ecosystem, the type of bedrock from which the soil is formed, the degree of weathering, pH, redox conditions, oxidation conditions, temperature. It depends on factors such as the presence of organic matter, conversion of heavy metals during the biocycle, and microbiological activity. Movement of heavy metals depends on temperature, speed and direction of movement of surface waters, circulation of air masses and wind speed. Apart from these, there are other factors such as polarity, pressure and molecular stability that affect the distribution and movement of these pollutants (Briffa et al., 2020). The geological and geomorphological differences in the region, the distance to the waste area, and the changes in the above-mentioned factors caused the heavy metal contents to show a high coefficient of variation between the sample points.

When the average values of metals are compared with the values allowed in the Turkish soil pollution regulation, it is seen that the values of Cd, Cu, Pb, Co and Zn are below or slightly above the allowable values, while the values of Cr, Ni, and especially As, Sb and Hg are found to be very high. It has been determined that these high-value metals are in close contact with plant roots and thus can potentially affect soil fertility, and further trace metal entry into soils in these areas should be avoided by agricultural management or other means in order to avoid long-term threats to productivity and food security. As, Sb and Hg are a toxic substance for living things and this state of As, Sb and Hg in the study area causes concern for the environment

In addition to this, the mobility of heavy metals in the region should be determined as well as the total content of the metals in different fractions of the soils by sequential extraction methods. Because the high amount of heavy metals associated with the non-residual phases is in a condition that can be easily transferred to the food chain through water sources, uptake by soil-grown plants or any other mechanism..

For example, although the total Sb content of the soils is below the maximum permissible pollutant concentration, its high concentrations in the mobile fractions have been observed to require caution in this metal. Because, in general, the total Sb content in the soil does not significantly correlate with the Sb in the plant, but it can be positively correlated with the exchangeable Sb content in the soil, because plants tend to readily absorb soluble or exchangeable Sb from the soil (Baroni et al., 2000). Although the proportion of bioavailable Sb is only 0.15–2.45% of total Sb in rhizospheric soil, high Sb concentrations in contaminated soil resulted in high bioavailable Sb, which could mean high uptake and accumulation potential by plants.

4. Conclusion

Different useful methods and indices were employed to evaluate soil pollution and contamination status of soils around Kurşunlu mine. According to the results of Enrichment factor (EF), contamination factor (Cf), Geoaccumulation index (Igeo), hazard quotient (HQ) (individual metal), contamination index (Cd), modified contamination index (mCd), pollution load index (PLI), and hazard load index(HLI) (multi metal) based on the averages, heavy metal pollution in soils of study area was observed considerable level for the studied metals for both individual metals and multi-metal. These results indicate that the soils around Kurşunlu mine are not contaminated by Cr, Zn, Ni, Cd, Cu, Pb, Co but contamination is maximum for Hg, Sb As. Considering the geological structure of the region rock formations it was explain sources of Hg, Sb, As. Therefore, the source of the very limited pollution seen in the region is not anthropogenic but natural source. According to results very high contamination for Hg, Sb,As is caused by natural geological factors related to rock formation of studied soils.

5. References

Abrahim GMS and Parker RJ (2008). Assessment of Heavy Metal Enrichment Factors and the Degree of Contamination in Marine Sediments from Tamaki Estuary, Auckland, New Zealand. Environmental Monitoring and Assessment, 136, 227-238.http://dx.doi.org/10.1007/s10661-007-9678-2

Alia N, Sardar K, Said M, Salma K, Sadia A, (2015). Toxicity and bioaccumulation of heavy metals in spinach (Spinaciaoleracea) grown in a controlled environment. Int J Environ Res Public Health 12: 7400-7416

Aydın Y (1996). Karadağ-Sızma (Konya) yöresindeki mavi şist metamorfizmasının kökeni: S. Korkmaz ve M. Akçay (ed.), Jeoloji Müh. Böl. 30. Yıl Sempozyumu Bildirileri, KTÜ-Trabzon, 1,186-194.

Baroni F, Boscagli A, Protano G, Riccobono F, (2000). Antimony accumulation in Achillea ageratum, Plantago lanceolata and Silene vulgaris growing in an old Sb-mining area. Environ Pollut 109(2), 347–352.

Bayiç A (1968).Sizma-Konya metaporfiritleri hakkında: MTA Dergisi, 70, 214-229.

Bello S, Zakari YI, Ibeanu IGE, Muhammad BG (2016).Characterization and assessment of heavy metal pollution levels in soils of Dana steel limited dumpsite, Katsina state, Nigeria using geo-accumulation, ecological risk and hazard indices American Journal of Engineering Research (AJER) e-ISSN: 2320-0847 p-ISSN : 2320-0936 Volume-5, Issue-1, pp-49-61

Bhuiyan MAH, Dampare SB, Islam MA, Suzuki S(2015). Source apportionment and pollution evaluation of heavy metals in water and sediments of Buriganga River, Bangladesh, using multivariate analysis and pollution evaluation indices. Environ. Monit. Assess. 187, 4075

Briffa J, Sinagra E, Blundell R. (2020). Heavy metal pollution in the environment and their toxicological effects on humans. Heliyon, 6(9), e04691.

Coşkun M, Steinnes E, Frontasyeva VM, Sjobakk TE, Demkına S (2006). Environmental Monitoring and Assessment (2006) 119: 545–556 DOI: 10.1007/s10661-005-9042-3

Çelebi H, Kara EE (2007) Niğde ili Bor ilçesi Tabakhanelerinden Çıkan Atıksuların Tarım Topraklarının Kirliliğine Olan Etkisinin Araştırılması. Uluslararası Küresel İklim Değişikliği ve Çevresel Etkileri Kongresi. 18-20 Ekim Konya-TÜRKİYE

Davidson CM, Urquhart GJ, Ajmone-Marsan F, Biasioli M, Costa Duarte A, Díaz E, Barrientos H, Grčman I, Hossack AS, Hursthouse L, Madrid S, Rodrigues, MZ (2006). Fractionation of potentially toxic elements in urban soils from five European cities by means of a harmonised sequential extraction procedure Anal. Chim. Acta, 565 (2006), pp. 63-72

Demir G, Özcan HK, Özdemir H, Pektaş AO, Oruc I, Büyükyıldız M (2016) Heavy Metal Concentrations of Selected Public Parks of İstanbul City. 2nd International Conference on Chemical Materials and Process (ICCMP 2016).May 11-13 Copenhagen, DenmarkBook Series:

Dumitrescu DV, Gurzău ES,Gurzău AE, Marchean D, Chera I (2012).CopşaMică: Area and heavy metals pollution. Proc of PRE9: Protection and restoration of the environment IX, Greece.

Edward M, Muntean N, Duda M (2013). Heavy metal contamination of soil in CopşaMică Area. ProEnvironment 6: 469-473.

EPAUS (1992). Framework for Ecological Risk Assessment. EPA-630/R92/001. U.S. Environmental Protection Agency, Washington, DC. 366 pp.

Göçmez S (2006) Menemen Ovası Topraklarında İz Su Kentsel Arıtma Çamuru Uygulamalarının Mikrobiyal Aktivite ve Biyomas ile Bazı Fiziksel ve KimyasalToprak Özellikleri Üzerine Etkisi. Ege Üniversitesi, Fen Bilimleri Enstitüsü, Toprak Anabilim Dalı, Doktora Tezi (Yayımlanmamış)., Bornova, İZMİR

Hakanson LL (1980). An ecological risk index aquatic pollution control, a sedimentological approach .Water Res., 14 (8), pp. 975-1001

Haktanır K, Arcak S, Erpul G, Tan A (1995). Yol Kenarlarındaki Topraklarda Trafikten Kaynaklanan Ağır Metal Birikimi. Tr., J., of Engineering and Environmental Sciences. 19, 423-431.

Homa J, Niklinska M, Plytycz B (2003). Effect of heavy metals on coelomocytes of the earthworm Allolobophorachlorotica. Pedobiologia, 47, 640−645. doi: 10.1078/0031-4056-00239.

Horasan BY, Arık F (2019). Assessıng Heavy Metal Pollutıon in The Surface Soils of Central Anatolia Region of Turkey. Carpathian Journal of Earth and Environmental Sciences, February 2019, Vol. 14, No. 1, p. 107 - 118; DOI:10.26471/cjees/2019/014/063

Kara EE, Pırlak U, Özdilek HG (2004). Evaluation of Heavy Metals'(Cd, Cu, Ni, Pb and Zn) Distribution in Sowing Regions of Potatofields in the Province of Niğde, Turkey. Water, Air and Soil Pollution;153, 173-186

Lacatusu R (1998.) 'Appraising level of soil contamination and pollution with heavy metals. In: Land Information Systems: Developments for planning the sustainable use of land resources', H. J. Heineke, W. Eckelmann, A. J. Thomasson, R. J. A. Jones, L. Montanarella and B. Buckley (eds.). European Soil Bureau Research Report No.4, EUR 17729 EN, 393–402.

Luo XS, Zhou DM, Liu XH, Wang YJ (2006). Solid/solution partitioning andspeciation of heavy metals in the contaminated agricultural soils arounda copper mine in eastern Nanjing city, China. Journal of Hazardous Materials 131, 19-27.

Maanan M, Saddik M, Maanan M, Chaibi M, Assobhei O, Zourarah B (2014). Environmental and ecological risk assessment of heavy metals in sediments of Nador lagoon, Morocco. Ecological Indicators, 48, 616−626. doi: 0.1016/j.ecolind.2014.09.034

Müller G (1979). Schwermetalle in den sedimenten des RheinseVeranderungenseitt 1971. Umschau; 778– 783

Nouri M, Haddioui A (2016). Assessment Of Metals Contamination And Ecological Rısk In Aıt Ammar Abondoned Iron Mine Soil Morocco Ekológia (Bratislava) Vol. 35, No. 1, P. 32–49

Rauret G (1998). Extraction procedures for the determination of heavy metals in contaminated soil and sediment. Talanta. 46(3), 449–455

Sajn R, Bidovec M. Andjelov S. Pirc M. G (1998). Geochemical Atlas of Ljubljanaand Environs, Institute of Geology, Geotechnique and Geophysics, Ljubljana,

Scazzola R, Avezzu S, Biancotto, R, Chiamenti E, Chiozzotto E, Gerotto M, Palonta M,Roiter S (2003). Assessment of heavy metal background values in the soils of inland coastal areas of Venice, Italy. Ann. Chim., 93, 465−470

Senila M, Tanaselia C, Rimba E (2013). Investigations on arsenic mobility changes in rizosphere of two ferns species using DGT technique. Carpathian J Earth Environ Sci 8: 145-154.

Sezgin N, Ozcan HK, Demir G, Nemlioglu S, Bayat C (2003) Determination of Heavy Metal Concentrations in Street Dusts in Istanbul E-5 Highway. Environment International Vol: 29, 979– 985.

Sparks DL, Page AI, Helmke DA, Loeppert RH, Soltanpour PN, Abatabai PN, Johnston CT, Sumuer ME (1996). Method of soil analysis Part. 3. Chemical methods. Inc. Madison Wisconsin, U.S.A.

Stafilov T, ŠajnR, Pančevski Z, Boev B, Frontasyeva MV, Strelkova LP (2010). Heavy metal contamination of topsoils around a lead and zinc smelter in the Republic of Macedonia. J. Hazard. Mater., 175, 896−914. doi: 10.1016/j.jhazmat.2009.10.094

Sungur A, Soylak M, Yilmaz E, Yilmaz S, Ozcan H (2014) Soil and Sediment Contamination, 24:1–15, Copyright © Taylor & Francis Group, LLC ISSN: 1532-0383 print / 1549-7887 online DOI: 10.1080/15320383.2014.907238

Sungur A, Soylak M, Yilmaz E, Yilmaz S, Ozcan H (2015). Characterization of Heavy Metal Fractions in Agricultural Soils by Sequential Extraction Procedure: The Relationship Between Soil Properties and Heavy Metal Fractions. Soil Sediment Contam 24(1), 1–15

Sungur A, Soylak M, Ozcan H (2016). Chemical fractionation, mobility and environmental impacts of heavy metals in greenhouse soils from Çanakkale, Turkey. Environ Earth Sci 75:334 DOI 10.1007/s12665-016-5268-3

Sungur A, Kavdir Y, Ozcan H, İlay R, Soylak M (2021). Geochemical fractions of trace metals in surface and core sections of aggregates in agricultural soils Catena 197:104995. https://doi.org/ 10.1016/j.catena.2020.104995

Tokalıoğlu S, Kartal S (2005). Comparison of Metal Fractionation Results Obtained from Single and BCR Sequential

Extractions. Bull. Environ. Contam. Toxicol. (2005) 75:180–188 © 2005 Springer Science+Business Media, Inc. DOI: 10.1007/s00128-005-0736-6

Tomlinson DL, WilsonJG, Harris CR, Jeffery DW (1980). Problems in the assessment of heavy metals levels in estuaries and the formation of a pollution index. HelgolWiss. Meeresunters, 33 (1-4), 566-575.

Turekıan KK, Wedepohl KH (1961). Distribution of the Elements in Some Major Units of the Earth's Crust Geological Society of America Bulletin, v. 72, p. 175-192,

Uğulu İ (2015). Determination of Heavy Metal Accumulation in Plant Samples by Spectrometric Techniques in TurkeyApplied Spectroscopy Reviews, Volume 50, 2015 - Issue 2

Ugulu İ, Khan ZI, Rehman S, Ahmad K, Munir M, Bashir Hİ, Nawaz K (2019). Trace Metal Accumulation in Trigonella foenum graecum Irrigated with Wastewater and Human Health Risk of Metal Access Through the Consumption. Bulletin of Environmental Contamination and Toxicology 103,468–475

USDA (2004). Soil Survey Laboratory Methods Manual. United States Department of Agriculture- Natural Resources Conservation Service, Soil Survey Investigations Report No. 42, Washington DC, USA

Wiesner K (1968). Konya civa yatakları ve bunlar üzerindeki etüdler: MTA Dergisi, 70, 178-214.

Yıldız M (1978). Türkiye'de bazı civa madenlerinin oluşum ve mukayesesi: MTA Yayınlan, Ankara.

Yılmaz F, Yılmaz ZY, Ergin M, Erkol AY, Muftuoglu AE, Karakelle B (2003). 'Heavy metal concentrations in surface soils of Izmit gulf region Turkey', J. Trace Microprobe Techn. 21, 523–531

Zhang Z, Abuduwaili J, Jiang F (2015). Heavy metal contamination, sources and pollution assessment of surface water in the Tianshan Mountains of China. Environ Monit Assess 187: 33.

Selcuk Journal of Agriculture and Food Sciences SJAFS

http://sjafs.selcuk.edu.tr/sjafs/index

(2022) 36 (3), 320-330

e-ISSN: 2458-8377

DOI:10.15316/SJAFS.2022.041

Research Article

Genetic Variability Studies in F_2 Generations of Determinate High Yield Dry Bean Lines for Seed Yield and Yield Components

Nur Banu TEKİN [1], Ercan CEYHAN[2,*]

[1] Selçuk University, The Graduate School of Natural and Applied Science, Department of Field Crops, Konya, Türkiye
[2] Selçuk University, Faculty of Agriculture, Department of Field Crops, Konya, Türkiye

ARTICLE INFO

Article history:
Received date: 09.08.2022
Accepted date: 18.08.2022

Keywords:
Diallel analysis
Dry bean
GCA
Heredity
SCA

ABSTRACT

Dry bean is the third most important pulse crop which is widely adapted and can improve soil fertility in Türkiye. The genetic variability and association studies help in selection which would increase the yield potential of dry bean. The F_2 generation and parents (Kınalı, Alberto, Great, Göynük, Özmen) obtained by diallel hybrid method (5 x 5 and 20 combinations were obtained) were sown in the experimental field of Selçuk University Faculty of Agriculture in 2021.In the research, measurement, counting, weighing and analysis of plant height, pod height, number of seeds per plant, number of seeds per pod, seed yield per plant, hundred-seed weight, protein ratio, protein yield in the plant were made. In the F_2 generation, the seed yield values in plant of the hybrids are between 23.63 g/plant (Kınalı x Özmen) and 97.45 g/plant (Alberto x Kinalı). Heterosis values of seed yield in plant range from -50.72% (Kınalı x Özmen) to 93.01% (Kınalı x Göynük), heterobeltiosis values varies from -58.92% (Kınalı x Özmen) to 77.26% (Göynuk x Özmen). The protein ratio of the crosses in the F_2 generation ranged from 23.05% (Alberto x G Northern 59) to 29.12% (Özmen x Alberto). Heterosis values of protein ratio are between -18.33% (Alberto x G Northern 59) and 7.23% (Özmen x Alberto), heterobeltiosis values are between -22.27% (Alberto x G Northern 59) and 5.82% (Özmen x Alberto). As a result of this research, a sufficient level of genetic variation was determined in the population, considering the agronomic characteristics examined. Determination of suitable parents and hybrids for green bean breeding in terms of sustainability of calcareous soils, agronomic characteristics and inheritance of these parents and hybrids were determined.

1. Introduction

The chemical inputs applied to increase production have brought about the problem of agricultural environmental pollution. One of the alternative agriculture systems that will reduce agricultural environmental pollution is sustainable agriculture (Cukur and Isın 2008; Disbudak 2008). Sustainable agriculture is a system in which agricultural technologies are used in a controlled manner from tillage to the last stage of production without harming the environment (Tan and Koksal 2004; Turhan 2005; Menalled et al. 2008; Eryılmaz and Kılıç 2018). It has been revealed how essential bean farming is for sustainable agricultural practices and a clean environment (Isık 2001). Since the sulfur-containing amino acid content of the bean is higher than that of other legumes, the biological value of its protein is high (Broughton et al. 2003).

It is close to meat protein due to the high protein content in its seeds and the amino acid composition of its proteins. It is abruption from other legumes due to its richness in carbohydrates, calcium, iron, phosphorus, vitamin B_1, prebiotics, various micronutrients and minerals. It has been determined that the protein, fiber, phosphorus, potassium, calcium, sulfur, iron, zinc and magnesium ratios in the bean seed vary depending on the genotype (Sprent et al. 1990; Onder and Ozkaynak 1994b; Broughton et al. 2003; Ceyhan 2007; Câmara et al. 2013; Duc et al. 2015).

In self-pollinating plants such as beans, seed yield is a quantitative trait and is governed by polygenes (Arunga et al. 2010). Selections made by considering only seed yield are not effective. In plant breeding programs, if selection is made with seed yield and other yield components, it is reliable and is thought to increase yield

* Corresponding author email: *eceyhan@selcuk.edu.tr*

(Muhammad et al. 1994; Shimelis 2006; Kwaye et al. 2008).

In the study conducted on hybrids for dry beans, they determined that (SCA) specific combination ability and (GCA) general combination ability were positive and important for plant height, pod height, number of seeds per pod, number of seeds per plant, seed yield per plant, hundred-seed weight, protein ratio, and protein yield in the plant (Zimmermann et al. 1985; Singh and Urrea 1994; Oliveira et al. 1997; Rodrigues et al. 1998; Barelli et al. 2000; Bozoglu and Sozen 2007; Ceyhan et al. 2014a;b). They determined the effect of additive and non-additive genes on plant height, seed yield and hundred-seed weight of bean plants (Ceyhan et al. 2014b). They determined that additive genes were effective on seed yield and harvest index (Zimmermann et al. 1985; Singh and Urrea 1994; Oliveira et al. 1997; Rodrigues et al. 1998; Barelli et al. 2000). They found that a single gene allele was effective on the number of ovaries in a pod (Al-Mukhtar and Coyne 1981), and non-additive genes were effective on the inheritance of pod traits and plant height (Rodrigues et al. 1998; Ceyhan et al. 2014b).

In this study aimed to select high yield and quality genotypes by determining the agronomic characteristics, heterosis and heterobeltiosis values of the F_2 generation, and heritability of the examined traits within the scope of sustainable agriculture.

2. Materials and Methods

The hybridization process was carried out according to Ceyhan (2003). Hybridization was done according to the diallel analysis method as 5 x 5 (20 combinations). Hybrid combinations was obtained by applying diallel hybridization method in Selcuk University plant breeding greenhouse. The obtained F_1 combinations were planted in the Selcuk University experimental field in 2020.

It was determined that the organic matter content of the trial field, which has a clayey soil structure, is at a medium level of 2.25% at a soil depth of 0-30 cm, and at a low level of 1.23% at a soil depth of 30-60 cm. The trial field, which has a high level of lime content (37.6%, 34.4%), has an alkaline structure (pH = 8.05 - 8.00), and there is no problem in terms of salinity. The amount of usable phosphorus (1.79 kg/da – 1.34 kg/da) and zinc (0.32 ppm – 0.34 ppm) in the trial field is quite low. Considering the analysis results of the trial field, it is sufficient in terms of iron (14.74 ppm – 8.74 ppm), copper (1.70 ppm – 1.74 ppm), and manganese (7.50 ppm – 5.76 ppm).

In the province of Konya, which corresponds to the research area, the average temperature was determined as 19.4 ^{0}C by the 21-year meteorological data, and 19.8 ^{0}C in 2021 when the parent and F_2 plants were grown. In the research, the total precipitation during the 21-year growing period was determined as 109.1 mm, and the total precipitation during the cultivation period in 2021 was determined as 134.3 mm. In the research, the average relative humidity was determined as 47.8% in the 21-year growing period and the average relative humidity in the growing period in 2021 was 44.4%.

F_2 generations were sowed on April 29, 2021. It was made on rows of 2 m in length, with a distance of 10 cm between rows of 40 cm. 20 seeds in each row were sown by hand at a depth of 5 cm. Parents and F_2 generations were sowed in the middle 3 rows of hybrids with 2 rows of parent. The experiment was established in the experimental field of the Selcuk University Faculty of Agriculture with 3 replications according to the randomized blocks trial design. To examine the soil characteristics, 15 kg of urea 46% N fertilizer was given at planting times. The trace element was applied to the leaves of the plants in which trace element deficiency was determined. Weed control was done manually and mechanically with a hoe. Harvesting of plants that reached harvest maturity was done in August-September. Measurement and counting of harvested plants were made separately for each plant. In the study, the data on plant characteristics were made according to Ceyhan (2003) and Akcin (1974).

In the research, the measurements made in the F_2 hybrids were first calculated analysis of variance according to the "Random Blocks Trial Design". Calculations were made using the Diallel Analysis Method for traits with significant variance values of 1% and 5% between hybrids. In the research, analysis and calculations were determined in TARPOPGEN PC Program (Ozcan and Acikgoz 1999).

3. Results and Discussion

The squares mean of diallel variance analysis of some agricultural characteristics examined in the F_2 population are given in the Table 1. General and specific combination variances and their genetic parameters are given in the Table 2. The results of full dialell variance analysis and the mean of squares of all the analyzed features were statistically significant in the study. SCA was important for the combination ability variance analysis in all traits except pod length, plant protein yield, and seed yield per plant. GCA was significant in all properties except protein yield in the plant. In the F_2 generation, υ^2GCA/υ^2SCA negative for plant height indicates an additive gene effect. For other traits, the variances of υ^2GCA/υ^2SCA were less than 1 and $H/D^{1/2}$ variances were more significant than 1, indicating that non-additive gene effect and superior dominance were effective. Our study agrees with other studies (Oliveira Junior et al. 1997; Barelli et al. 2000; Ceyhan et al. 2014b). Rodrigues et al. (1998), Barelli et al. (2000), Ceyhan et al. (2014b). They found is an additive gene effect for the number of pods per plant in beans. Zimmermann et al. (1985), Singh and Urrea (1994), Oliveira Junior et al. (1997), Rodrigues et al. (1998), Barelli et al. (2000), determined the additive gene effect and the dominant gene effect for seed yield in bean plant.

Table 1
Mean squares of initial variance analysis and combining ability variance analysis for investigated traits in a full-diallel hybrid set

Source of Variation	SD	Plant Height	Number of Pods	Number of Seeds per Pod	Number Seeds in Plant
Blocks	2	24,751	41,912	1,207	3527,575
Genotypes	24	827,032**	526,358*	1,855**	7527,468**
Error	48	307,666	258,311	0,421	2544,817
GCA	4	595,971**	146,928	0,941**	2669,416*
SCA	10	70,880	127,167	0,437**	928,961
Reciprocal Effect	10	352,357**	235,149**	0,671**	4025,247**
Error	48	102,555	86,104	0,140	848,272
Source of Variation	SD	Seed Yield per Plant	Hundred Seed Weight	Protein Ratio	Protein Yield in Plant
Blocks	2	1464,523	100,397	0,091	97,494
Genotypes	24	988,453**	107,657**	10,211**	64,37**
Error	48	373,774	16,982	0,104	26,635
GCA	4	339,252*	70,796**	3,797**	19,127
SCA	10	181,879	20,115**	2,449**	10,931
Reciprocal Effect	10	473,18**	37,693**	4,200**	32,918**
Error	48	124,592	5,661	0,035	8,878

* : significant at 5% level , ** : significant at 1% level

3.1. Plant height:

According to the plant height averages of the F_2 generation, parental values are between 49.64 (Göynük) and 93.41 (Kınalı), hybrids are between 35.00 cm (Göynük x Kınalı) and 102.10 cm (Kınalı x Alberto) (Table 2). Other studies show parallelism with our results (Ceyhan 2004; Peksen 2005; Ulker and Ceyhan 2008b; Ceyhan et al. 2009; Babagil et al. 2011; Ceyhan and Kahraman 2013; Ceyhan et al. 2014b; Elkoca and Cınar 2015; Girgel et al. 2018; Yolci 2020).

The plant height trait in the F2 generation of the parents GCA was negative and significant in the Göynük cultivar (Table 3). Kınalı, Alberto and G Northern 59 parents are recommended for use in growing tall plants as they have a positive effect. The Göynük genotype, which has a negative and important GCA effect value, is recommended for the development of short bean varieties in order to transfer resistance for lodging to the next generations. When the SCA effects of crosses in the F_2 generation were examined, it was found that G Northern 59 x Göynük, G Northern 59 x Alberto were positive and significant, Alberto x Kınalı, Göynük x Kınalı, Göynük x Alberto, G Northern 59 x Kınalı, Özmen x Alberto, Özmen x G Northern 59 were determined as negative and significant (Table 3) Among the hybrids, those with positive and significant SCA effects are tall plants. Those with negative and significant effects are combinations that can be used to develop short or medium height varieties. Barelli et al. (2000), Rodrigues et al. (1998), Arunga et al. (2010), Ceyhan et al. (2014b), Ceyhan and Simsek (2021), in their study on plant height, they found the GCA and SCA values of different numbers of parents and crosses to be significant.

The low average of heterosis and heterobeltiosis values in the F_2 generation indicates the presence of additive gene effect. F_2 generation heterosis values ranged from -51.06% (Göynük x Kınalı) to 38.54% (G Northern 59 x Göynük) (Table 4). Rodrigues et al. (1998); Arunga et al. (2010); Ceyhan et al. (2014a) researchers working on heterosis and heterobeltiosis values for plant height stated that they obtained high or low heterosis and heterobeltiosis values in this feature.

Table 2
Mean values for investigated traits in full-diallel hybrid set

Parents	Plant Height		Number of Pods		Number of Seeds per Pod		Number Seeds in Plant	
Kınalı	93,41	ab	50,11	a-f	4,98	b-f	57,52	a-e
Alberto	82,41	a-d	43,28	b-g	4,22	c-g	45,27	cde
Göynük	49,64	de	38,44	c-g	3,78	fg	43,11	cde
G Northern 59	84,70	0a-d	57,07	a-d	5,20	b-f	64,05	a-e
Özmen	65,25	b-e	38,86	c-g	4,33	b-g	38,40	cde
F_2 Hybrids								
Kınalı x Alberto	102,10	a	53,50	a-f	4,98	b-f	195,47	a-e
Kınalı x Göynük	75,17	a-d	56,33	a-e	4,22	c-g	251,83	a
Kınalı x G Northern 59	89,33	abc	51,89	a-f	3,78	fg	174,39	a-f
Kınalı x Özmen	78,00	a-d	22,00	g	5,20	b-f	74,00	fg
Alberto x Kınalı	70,17	a-e	55,00	a-e	7,00	a	219,00	abc
Alberto x Göynük	90,50	ab	28,00	fg	4,00	d-g	86,00	efg
Alberto x G Northern 59	67,46	a-e	31,17	d-g	4,58	b-g	123,08	b-g
Alberto x Özmen	77,67	a-d	39,50	c-g	4,00	d-g	119,83	c-g
Göynük x Kınalı	35,00	e	28,00	fg	3,33	g	62,33	g
Göynük x Alberto	56,67	b-e	42,17	c-g	3,92	efg	116,08	c-g
Göynük x G Northern 59	52,00	cde	37,00	d-g	4,00	d-g	113,00	c-g
Göynük x Özmen	49,10	de	70,67	a	5,00	b-f	180,33	a-f
G Northern 59 x Kınalı	72,53	a-e	48,55	a-f	4,78	b-f	183,11	a-f
G Northern 59 x Alberto	87,17	a-d	63,44	abc	4,89	b-f	232,55	ab
G Northern 59 x Göynük	93,06	ab	69,39	ab	4,39	b-g	196,00	a-e
G Northern 59 x Özmen	81,28	8a-d	45,50	a-g	5,39	bcd	151,16	a-g
Özmen x Kınalı	71,55	a-e	32,50	d-g	5,50	bc	210,00	a-d

Table 2 (Continue)
Mean values for investigated traits in full-diallel hybrid set

Özmen x Alberto	57,25	b-e	30,67	efg	5,33	b-e	135,00	b-g
Özmen x Göynük	61,13	b-e	35,80	d-g	4,13	c-g	113,80	c-g
Özmen x G Northern 59	63,67	b-e	31,67	d-g	4,67	b-g	119,00	c-g
GCA	29,28		12,16		0,16		364,23	
SCA	39,23		123,19		0,89		242,07	
Reciprocal	18,59		149,05		0,53		3176,97	
υ^2GCA / υ^2SCA	0,75		0,10		0,18		1,50	
$H/D^{1/2}$	116,37		296,56		1,74		4147,50	
H^2	0,46		0,54		0,79		0,62	
h^2	0,23		0,04		0,15		0,11	

GCA: General Combining Ability; SCA: Specific Combining Ability; $H/D^{1/2}$: Mean Degree of Dominance; H^2: Broad Sense Heritability; h^2: Narrow Sense

3.2. Number of Pods:

Regarding the F_2 generation, the pod length values of the parents vary between 38.44 units/plant (Göynük) and 57.07 units/plant (G Northern 59), while those of the hybrids vary between 22.00 units/plant (Kınalı x Özmen) and 70.67 units/plant (Göynük x Özmen) (Table 2). Bozoğlu and Gülümser (2000); Ülker and Ceyhan (2008b); Varankaya and Ceyhan (2012); Elkoca and Çınar (2015); Girgel et al. (2018); Konuk and Uzun (2021) found similar results. The G Northern 59 genotype can be developed and used in breeding studies. It was determined that the recessive genes were effective in the Alberto and Özmen parents (Table 3). Conserning the SCA, hybrids G Nord 59 x Alberto, G Nord 59 x Göynük, Göynük x Özmen were positive and significant. While Göynük x Kınalı, Özmen x Göynük, Kınalı x Özmen hybrids were negative and significant (Table 3). It was determined that the hybrids with positive and significant SCA effect could be used to increase the number of pods (Table 3). In the F_2 generation, heterosis values are between -50.55% (Kınalı x Özmen) and 82.83% (Göynük x Özmen), and heterobeltiosis values are between -56.10% (Kınalı x Özmen) and 81.85% (Göynük x Özmen) (Table 4.5). The fact that the ratio of υ^2GCA / υ^2SCA was less than 1 in the F_2 generation indicates that the non-additive gene effect is effective in the inheritance of this trait. The heritability in the broad sense is high in the F_2 generation. The heritability in the narrow sense is low (Table 2). The results of Heterosis and heterobeltiosis obtained in hybrids show that this characteristic is affected by environmental conditions (Ceyhan 2003). In a study conducted for the number of pod characteristics, the positive and negative values of heterosis and heterobeltiosis were obtained and given (Barelli et al. 2000; Arunga et al. 2010; Ceyhan et al. 2014b). This result shows that the number of pod is affected by the environmentals conditions. Since non-additive gene effects are important in this study, it is thought that it would be appropriate to start selection after 3-4 generations.

Table 3
Genetic components for investigated traits in full-diallel hybrid set

Parents	Plant Height	Number of Pods	Number of Seeds per Pod	Number Seeds in Plant
Kınalı	5,818	0,780	0,350*	19,418
Alberto	5,132	-1,020	0,071	-1,975
Göynük	-11,058*	0,404	-0,490**	-18,123
G Northern 59	5,341	5,254	0,091	13,721
Özmen	-5,234	-5,418	-0,021	-13,042
F_2 Populations				
Kınalı x Alberto	2,933	10,471	1,112**	39,104*
Kınalı x Göynük	-11,925	-3,037	-0,049	5,099
Kınalı x G Northern 59	-2,477	0,167	-0,383	-5,080
Kınalı x Özmen	1,943	-12,132*	-0,268	-15,065
Alberto x Kınalı	-15,964**	0,750	0,778**	11,765
Alberto x Göynük	7,261	-8,321	-0,312	-29,549
Alberto x G Northern 59	-5,408	-0,949	-0,117	15,384
Alberto x Özmen	-4,686	-2,498	-0,072	-8,255
Göynük x Kınalı	-20,083**	-14,167**	-1,167**	-94,750**
Göynük x Alberto	-16,917**	7,083	-0,042	15,042
Göynük x G Northern 59	5,997	3,516	-0,096	8,213
Göynük x Özmen	-0,839	14,227*	0,389	27,543
G Northern 59 x Kınalı	-8,403*	-1,666	0,028	4,361
G Northern 59 x Alberto	9,854*	16,138**	0,152	54,735**
G Northern 59 x Göynük	20,528**	16,194**	0,194	41,500**
G Northern 59 x Özmen	0,117	-5,273	0,268	-16,286
Özmen x Kınalı	-3,225	5,250	0,750**	68,000**
Özmen x Alberto	-10,207*	-4,417	0,667**	7,583
Özmen x Göynük	6,017	-17,433**	-0,433*	-33,267*
Özmen x G Northern 59	-8,805*	-6,917	-0,361*	-16,082
G_i	8,204	6,888	0,011	67,862
S_{ij}	34,869	29,275	0,048	288,413
R_{ij}	51,278	43,052	0,070	424,136

G_i : GCA, S_{ij}: SCA; R_{ij}: Reciprocal effect, ** : significant at 1% level; * : significant at 5% level

3.3.Number of seeds per pod:

Parental values for the number of seeds per pod in the F_2 generation ranged from 3.78 units/pod (Göynuk) to 5.20 units/pod (G Northern 59), while the values of crosses were between 3.33 units/pod (Göynük x Kınalı) and 7.00 units. Pieces/pod (Alberto x Kınalı) (Table 2). Many studies conducted have similar results with the result we have obtained (Pekşen 2005; Ülker and Ceyhan 2008a; Varankaya and Ceyhan 2012; Elkoca and Çınar 2015; Girgel et al. 2018; Bildirici and Demir 2019; Gülnur 2019; Aydoğan et al. 2020a; Sirat 2020). In the F_2 generation, the GCA of the parents was positive and significant for Kınalı, and the Göynük genotype was found to be negative and significant. The Kınalı genotype, whose GCA is positive and significant, is considered the parent to be used to increase the number of seeds in the pod (Table 3). Crosses with positive and significant SCA have been determined as combinations that could be used to increase pod length (Table 3). In the studies carried out for the number of seeds in the broad bean, they determined that the GCA and SCA are important (Al Mukhtar and Coyne 1981; Rodrigues et al. 1998; Barelli et al. 2000; Arunga et al. 2010; Ceyhan et al. 2014b). In addition, they determined that SCA and GCA effects are important in determining additive and non-additive gene effects (Griffing 1956; Arunga et al. 2010). Heterosis values in the F_2 generation range from -23.84% (Göynük x Kınalı) to 52.23% (Alberto x Kınalı), and heterobeltiosis values are between -33.02% (Göynük x Kınalı) and 40.66% (Alberto x Kınalı) (Table 4,5). Ceyhan et al., (2014b) observed that heterosis and heterobeltiosis values were negative and positive for the number of grains in the pod. The heritability in the broad sense is high in the F_2 generation The heritability in the narrow sense is low (Table 2). This result shows that the environment affects the number of grains per pod. Since non-additive gene effects are important in this study, it is thought that it would be appropriate to start selection after 3-4 generations.

Table 4
Heterosis (%) values for investigated traits in full-diallel hybrid set

F_2 Populations	Plant Height	Number of Pods	Number of Seeds per Pod	Number seeds in plant
Kınalı x Alberto	16,13	14,58	18,38**	32,29
Kınalı x Göynük	5,09	27,23	29,47**	87,51
Kınalı x G Northern 59	0,31	-3,18	-7,24**	2,17
Kınalı x Özmen	-1,67	-50,55	-14,04**	-51,02
Alberto x Kınalı	-20,19	17,79	52,23**	48,21
Alberto x Göynük	37,07	-31,47	0,04	-26,24
Alberto x G Northern 59	-19,27	-37,88	-2,69*	-19,54
Alberto x Özmen	5,20	-3,82	-6,43**	-10,14
Göynük x Kınalı	-51,06	-36,76	-23,84**	-53,59
Göynük x Alberto	-14,17	3,20	-2,04*	-0,44
Göynük x G Northern 59	-22,58	-22,52	-10,88	-19,01
Göynük x Özmen	-14,52	82,83	23,36**	50,4
G Northern 59 x Kınalı	-18,56	-9,39	-6,15**	7,28
G Northern 59 x Alberto	4,32	26,45	3,75**	52,02
G Northern 59 x Göynük	38,54	45,30	-2,22*	40,48
G Northern 59 x Özmen	8,41	-5,14	13,07**	-3,28
Özmen x Kınalı	-9,81	-26,94	18,19**	39,01
Özmen x Alberto	-22,45	-25,33	24,76**	1,23
Özmen x Göynük	6,43	-7,38	1,97*	-5,09
Özmen x G Northern 59	-15,08	-33,98	-2,06*	-23,86
Mean	-4,39	-3,85	5,38	7,42

** : significant at 1% level; * : significant at 5% level

3.4. Number of Seeds per Plant:

Parental values for the number of seeds in plant in the F_2 generation ranged from 103.14 number/ plant (Göynük) to 175.90 number/ plant (G Northern 59), hybrid data from 62.33 number/plant (Göynük x Kınalı) to 251.83 number/plant (Kınalı x Göynük) (Table 2). Many researchers have obtained results similar to our studies (Ülker and Ceyhan, 2008a; Varankaya and Ceyhan 2012; Bildirici and Demir 2019; Ceyhan and Şimşek 2021). When the GCA of the parents in the F_2 generation is examined, Kınalı and G Northern 59 genotypes are recommended as parents to be used in breeding studies to increase the number of seeds in the plant (Table 3). When the SCA of the crosses were examined for the number of seeds per plant in the F_2 generation, the combinations of Özmen x Kınalı, G Northern 59 x Alberto, G Northern 59 x Göynük, Kınalı x Alberto were found positive and significant, while the combinations of Göynük x Kınalı, Özmen x Göynük were found to be negative and significant (Table 3). Positive and significant hybrids in the F_2 generation can be suggested as combinations to be used to increase the number of seeds in the plant. Heterosis values in the F_2 generation range from -53.59% (Göynük x Kınalı) to 87.51% (Kınalı x Göynük) and heterobeltiosis values range from -62.33% (Göynük x Kınalı) to 52.20% (Kınalı x Göynük) is changing (Table 4,5). The low values of mean heterosis in the F_2 generation indicates that there is an additive gene effect. It shows that complex genes manage this trait. It has been determined that the heritability of the broad sense

is high and the heritability is low in the narrow sense, and that the number of seeds per pod is affected by the environment (Table 2). In addition, it was determined that the effect of genotype variance on this trait was low. The high and low heritability in the broad and narrow sense indicates that the non-additive gene effect effectively inherits this trait. Therefore, it would be appropriate to start selection in advanced generations.

3.5. Seed Yield per Plant:

Seed yield values per plant of the parents in the F_2 generation are between 38.40 g/plant (Özmen) and 64.05 g/plant (G Northern 59), hybrids values are between 23.63 g/plant (Kınalı x Özmen) and 97.45 g/plant (Alberto x Kınalı) (Table 6). Many studies on seed yield in plants are similar to our results (Yeken et. al. 2018; Bildirici and Demir 2019; Taşkesen 2019; Ceyhan and Şimşek 2021). When the GCA of the parents for the seed yield per plant in the F_2 generation was examined, the Özmen cultivar was found to be negative and significant. Kınalı and G Northern 59, which were determined as positive, can be recommended for use in breeding studies for seed yield in the plant (Table 7). When the SCA of the hybrids in the F_2 generation was examined, Alberto x Kınalı, Özmen x Kınalı, G Northern 59 x Alberto, Kınalı x Alberto, G Northern 59 x Göynük were positive and significant, Göynük x Kınalı, Özmen x Göynük were negative and significant (Table 7). Positive and important like Alberto x Kınalı, Özmen x Kınalı, G Northern 59 x Alberto, Kınalı x Alberto, G Northern 59 x Göynük can be used as hybrids that will increase the seed yield of the plant in advanced generations. In a study they carried out for GCA and SCA for seed yield, they determined that there were in different numbers and significant effects of SCA and GCA in the generations they obtained (Zimmermann et al. 1985; Singh and Urrea 1994; Oliveira Junior et al. 1997; Rodrigues et al. 1998; Barelli et al. 2000; Arunga et al. 2010; Ceyhan et al. 2014b). Heterosis values in the F_2 generation range from -50.72% (Kınalı x Özmen) to 93.01% (Kınalı x Göynük), heterobeltiosis values range from -58.92% (Kınalı x Özmen) to 77.26% (Göynük x Özmen) (Table 8,9). Generally, seed yield is quantitative feature and is governed by polygenes (Arunga et al. 2010). The low heritability in the narrow sense for the seed yield trait shows that the non-additive gene effect is effective in the inheritance of this trait (Table 6). This reduces the success of selection to be made in early generations for seed yield. For this reason, it would be more appropriate to select for traits with high heritability and easily evident rather than seed yield in early generations.

Table 5
Heterobeltiosis (%) values for investigated traits in full-diallel hybrid set

F_2 Populations	Plant Height	Number of Pods	Number of Seeds per Pod	Number Seeds in Plant
Kınalı x Alberto	9,30	6,77	9,38	18,13
Kınalı x Göynük	-19,53	12,42	13,86	52,20*
Kınalı x G Northern 59	-4,36	-9,08	-9,23	-0,86
Kınalı x Özmen	-16,50	-56,10*	-19,62*	-55,28*
Alberto x Kınalı	-24,88	9,76	40,66**	32,35
Alberto x Göynük	9,81	-35,30	-5,21	-33,87
Alberto x G Northern 59	-20,36	-45,39*	-11,86	-30,03
Alberto x Özmen	-5,76	-8,73	-7,62	-12,32
Göynük x Kınalı	-62,53**	-44,12*	-33,02**	-62,33**
Göynük x Alberto	-31,24*	-2,56	-7,19	-10,74
Göynük x G Northern 59	-38,61*	-35,16	-23,08*	-35,76
Göynük x Özmen	-24,75	81,85*	15,47	31,95
G Northern 59 x Kınalı	-22,36	-14,92	-8,16	4,10
G Northern 59 x Alberto	2,91	11,17	-6,03	32,21
G Northern 59 x Göynük	9,86	21,59	-15,60	11,43
G Northern 59 x Özmen	-4,04	-20,27	3,61	-14,06
Özmen x Kınalı	-23,40	-35,14	10,52	26,91
Özmen x Alberto	-30,53*	-29,14	23,17*	-1,22
Özmen x Göynük	-6,30	-7,87	-4,54	-16,73
Özmen x G Northern 59	-24,83	-44,51*	-10,26	-32,35
Mean	-16,40	-12,24	-2,24	-4,81

** : significant at 1% level; * : significant at 5% level

3.6. Hundred-seed weight:

In the F_2 generation, the values for the 100 seed weight of the parents are between 25.11 g (Özmen) and 36.57 g (Göynük), the values for the hundred seed weight of the crosses are between 24.50 g (Özmen x Kınalı) and 43.33 g (Özmen x G Northern) (Table 6). The work done by many researchers for hundred seed weight is consistent with our results (Bıyıklı et al. 2015; Elkoca and Çınar 2015; Yeken et al. 2018b; Bildirici and Demir 2019; Soydaş et al. 2019; Ceyhan and Şimşek 2021). In the F_2 generation, when the GCA of the parents was examined, the Göynük genotype was determined as positive and significant, Özmen as negative and significant. Göynük cultivar, which was found to be positive and important, was determined as the parent to be used to increase the hundred seed weight (Table 7). When the SCA of one hundred seed weight of the crosses in the F_2 generation were examined, the combinations Alberto x Kınalı, Göynük x Kınalı, Alberto x Göynük, Özmen x G

Northern 59, G Northern 59 x Özmen were positive and significant, G Northern 59 x Kınalı, Özmen x Alberto, G Northern 59 x Göynük, Özmen x Göynük, Kınalı x Özmen were negative and significant (Table 7). Hybrids with positive and significant SCA effect were determined as suitable combinations that can be used to increase hundred seed weight. In a study conducted for hundred seed weight, they determined that the GCA and SCA values of the crosses and parents examined were positive and significant (Ceyhan et al. 2014b; Ceyhan and Şimşek 2021; Kepildek and Ceyhan 2021). It has been observed that the heritability extent in the broad sense is high, and the heritability in the narrow sense go out low, there is an environmental effect in the emergence of hundred seed weight (Table 6). The fact that non-additive gene effects are important in the inheritance of hundred seed weight indicates that it would be appropriate to start selection in late generations.

Table 6
Mean values for investigated traits in full-diallel hybrid set

Parents	Seed Yield per Plant		Hundred Seed Weight		Protein Ratio		Protein Yield in Plant	
Kınalı	57,52	a-e	31,47	c-f	25,83	j-m	14,86	a-d
Alberto	45,27	cde	33,70	b-e	26,80	gh	12,13	bcd
Göynük	43,11	cde	36,57	abc	26,39	hıj	11,38	bcd
G Northern 59	64,05	a-e	31,33	3c-f	29,66	a	19,00	abc
Özmen	38,40	cde	25,11	ef	27,52	ef	10,58	bcd
F_2 Populations								
Kınalı x Alberto	65,38	a-e	30,05	c-f	24,32	n	15,98	a-d
Kınalı x Göynük	97,12	ab	35,17	a-d	25,17	m	24,44	a
Kınalı x G Northern 59	67,79	a-d	35,55	a-d	24,42	n	16,55	a-d
Kınalı x Özmen	23,63	e	25,00ef	ef	23,19	op	5,48	d
Alberto x Kınalı	97,45	a	42,00ab	ab	25,38	klm	24,74	a
Alberto x Göynük	44,11	cde	43,00	a	25,95	ı-l	11,45	bcd
Alberto x G Northern 59	48,55	cde	35,25	a-d	23,05	p	11,20	bcd
Alberto x Özmen	52,60	cde	38,83	abc	23,89	no	12,57	bcd
Göynük x Kınalı	31,67	de	41,00	ab	24,40	n	7,72	cd
Göynük x Alberto	55,11	b-e	42,83	a	26,08	ıjk	14,37	a-d
Göynük x G Northern 59	47,38	cde	37,00	abc	27,17	fg	12,87	bcd
Göynük x Özmen	76,41	abc	38,33	abc	26,11	hıj	19,99	ab
G Northern 59 x Kınalı	61,87	a-e	30,22	c-f	27,88	de	17,25	abc
G Northern 59 x Alberto	78,74	abc	31,89	c-f	24,31	n	19,11	ab
G Northern 59 x Göynük	65,14	a-e	30,33	c-f	26,59	ghı	17,28	abc
G Northern 59 x Özmen	48,10	cde	26,89	def	25,32	lm	12,15	bcd
Özmen x Kınalı	57,58	a-e	24,50	f	28,11	cde	16,19	a-d
Özmen x Alberto	38,25	cde	25,55	ef	29,12	ab	11,15	bcd
Özmen x Göynük	39,94	cde	31,57	c-f	28,25	cd	11,29	bcd
Özmen x G Northern 59	57,00	a-e	43,33	a	28,80	bc	16,45	a-d
GCA	42,93		13,03		0,75		2,05	
SCA	171,86		43,36		7,24		6,16	
Reciprocal	348,59		32,03		4,17		24,04	
σ^2GCA/σ^2SCA	0,25		0,30		0,10		0,33	
$H/D^{1/2}$	606,32		101,45		12,91		34,30	
H^2	0,59		0,83		0,99		0,54	
h^2	0,08		0,21		0,12		0,06	

GCA: General Combining Ability; SCA: Specific Combining Ability; $H/D^{1/2}$: Mean Degree of Dominance; H^2: Broad Sense Heritability; h^2: Narrow Sense

3.7. Protein content:

In the F_2 generation, the protein ratio of the parents is between 25.83% (Kınalı) and 29.66% (G Northern 59), and the protein ratio of the crosses is between 23.05% (Alberto x G Northern 59) and 29.12% (Özmen x Alberto) (Table 6). The results of our study are consistent with the results of other researchers (Varankaya and Ceyhan 2012; Gülnur 2019; Aydoğan et al. 2020; Sirat 2020; Yolci 2020; Kepildek and Ceyhan 2021). When the GCA of the parents for the protein ratio in the F_2 generation was examined, G Northern 59, Özmen cultivars were found to be positive and significant, while Kınalı and Alberto cultivars were found to be negative and significant (Table 7). GCA positive and significant genotypes in F_2 generations can be used to increase the protein ratio in breeding studies. When examining the SCA for protein ratio in the F_2 generation, Alberto x Kınalı, G Northern 59 x Kınalı, Özmen x Kınalı, G Northern 59 x Alberto, Özmen x Alberto, Alberto x Göynük, Özmen x Göynük, Özmen x G Northern 59, Alberto x Özmen, Göynük x Özmen were positive and significant, Göynük x Kınalı, Kınalı x Göynük, G Northern 59 x Göynük, Alberto x G Northern 59, Kınalı x Özmen, G Northern 59 x Özmen were negative and significant (Table 7). It has been determined that hybrids that are positive and important in the F_2 generation are combinations that can

be used to increase the protein ratio. Heterosis values in the F_2 generation ranged from -18.33% (Alberto x G Northern 59) to 7.23% (Özmen x Alberto), heterobeltiosis values were between -22.27% (Alberto x G Northern 59) and 5.82% (Özmen x Alberto) (Table 8,9). All values calculated for heterosis and heterobeltiosis were found to be significant. The negative mean heterosis and heterobeltiosis values determined for the protein ratio generally affect the protein ratio in a decreasing way. The heritability in the broad sense was high and the heritability in the narrow sense is low (Table 6). As a result, it is thought that genetic variance and environment are effective in the inheritance of protein ratio. Considering the non-additive gene effects in the inheritance of protein ratio, selection should be started in advanced generations.

Table 7
Genetic components for investigated traits in full-diallel hybrid set

Parents	Seed Yield per Plant	Hundred Seed Weight	Protein Ratio	Protein Yield in Plant
Kınalı	5,667	-1,217	-0,694**	1,160
Alberto	0,986	1,822	-0,579**	-0,163
Göynük	-1,777	3,378**	0,103	-0,430
G Northern 59	4,179	-0,546	0,537**	1,439
Özmen	-9,055*	-3,437**	0,634**	-2,006
F_2 Populations				
Kınalı x Alberto	18,673*	1,562	-0,024	4,716*
Kınalı x Göynük	4,416	2,063	-0,770**	0,703
Kınalı x G Northern 59	-1,104	0,791	0,160	-0,348
Kınalı x Özmen	-12,089	-4,456**	-0,436**	-2,967
Alberto x Kınalı	16,034**	5,973**	0,534**	4,377**
Alberto x Göynük	-5,682	3,858*	0,346**	-1,143
Alberto x G Northern 59	2,395	-1,566	-2,427**	-0,766
Alberto x Özmen	-2,594	-0,051	0,300*	-0,621
Göynük x Kınalı	-32,725**	2,917*	-0,387**	-8,363**
Göynük x Alberto	5,500	-0,083	0,064	1,462
Göynük x G Northern 59	-2,232	-3,024	0,093	-0,579
Göynük x Özmen	12,921	1,149	0,296*	3,427
G Northern 59 x Kınalı	-2,958	-2,666*	1,730**	0,349
G Northern 59 x Alberto	15,099**	-1,682	0,626**	3,958**
G Northern 59 x Göynük	8,880*	-3,333**	-0,289**	2,203
G Northern 59 x Özmen	1,339	5,234**	-0,261*	0,217
Özmen x Kınalı	16,975**	-0,250	2,462**	5,354**
Özmen x Alberto	-7,172	-6,640**	2,617**	-0,709
Özmen x Göynük	-18,238**	-3,383**	1,069**	-4,350**
Özmen x G Northern 59	4,452	8,223**	1,744**	2,149
G_i	9,967	0,453	0,003	0,710
S_{ij}	42,361	1,925	0,012	3,019
R_{ij}	62,296	2,830	0,017	4,439

G_i : GCA, S_{ij}: SCA; R_{ij}: Reciprocal effect, ** : significant at 1% level; * : significant at 5% level

3.8. Protein yield:

The parents values of protein yield in the F_2 generation are between 11.38 g/plant (Göynuk) and 19.00 g/plant (G Northern 59), hybrid values are between 5.48 g/plant (Kınalı x Özmen) and 24.74 g/plant (Alberto x Kınalı) (Table 6). Our results are similar to those of other researchers (Ülker and Ceyhan 2008a; Varankaya and Ceyhan 2012; Kepildek and Ceyhan 2021). When we examine the data of the parents' GCA's on protein yield in the plant in the F_2 generation, G Northern 59 and Henna parents are suggested as varieties that can be used in breeding studies (Table 7). When we examine the SCA of hybrids in the F_2 generation, Alberto x Kınalı, Özmen x Kınalı, Kınalı x Alberto, G Northern 59 x Alberto have positive and significant value, and Göynük x Kınalı and Özmen x Göynük combinations have negative and significant value (Table 7). Crosses with positive and significant SCA in the F_2 generation can be recommended for use in breeding studies. Heterosis values in the F_2 generation range from -56.92% (Kınalı x Özmen) to 86.29% (Kınalı x Göynük) heterobeltiosis values range from -48.09% (Göynük x Kınalı) to 75.66% (Alberto x Özmen) (Table 8,9). For protein yield in the plant, heritability in the broad sense was high and heritability in the narrow sense have low (Table 6). This shows that in addition to the genotype variance effect, the environment also has an effect on protein yield. Considering the non-additive gene effects in the inheritance of protein yield, starting selection in late generations is recommended.

Table 8
Heterosis (%) values for investigated traits in full-diallel hybrid set

F_2 Populations	Seed Yield per Plant	Hundred Seed Weight	Protein Ratio	Protein Yield in Plant
Kınalı x Alberto	27,21	-7,76	-7,59**	18,40
Kınalı x Göynük	93,01	3,38	-3,59**	86,29
Kınalı x G Northern 59	11,52	13,23	-11,98**	-2,27
Kınalı x Özmen	-50,72	-11,62	-13,06**	-56,92
Alberto x Kınalı	89,60	28,90	-3,53**	83,25
Alberto x Göynük	-0,17	22,39	-2,40**	-2,62
Alberto x G Northern 59	-11,18	8,41	-18,33**	-28,07
Alberto x Özmen	25,73	32,06	-12,04**	10,65
Göynük x Kınalı	-37,06	20,52	-6,55**	-41,19
Göynük x Alberto	24,73	21,91	-1,92**	22,25
Göynük x G Northern 59	-11,57	8,98	-3,04**	-15,26
Göynük x Özmen	87,50	24,30	-3,12**	82,05
G Northern 59 x Kınalı	1,78	-3,75	0,50**	1,85
G Northern 59 x Alberto	44,07	-1,94	-13,89**	22,78
G Northern 59 x Göynük	21,57	-10,66	-5,10**	13,76
G Northern 59 x Özmen	-6,10	-4,73	-11,45**	-17,87
Özmen x Kınalı	20,06	-13,39	5,40**	27,25
Özmen x Alberto	-8,56	-13,10	7,23**	-1,83
Özmen x Göynük	-2,01	2,36	4,81**	2,81
Özmen x G Northern 59	11,28	53,55	0,75**	11,19
Mean	16,53	8,65	-4,95	10,83

** : significant at 1% level; * : significant at 5% level

Table 9
Heterobeltiosis (%) values for investigated traits in full-diallel hybrid set

F_2 Populations	Seed Yield per Plant	Hundred Seed Weight	Protein Ratio	Protein Yield in Plant
Kınalı x Alberto	13,66	-10,82	-9,25**	7,53
Kınalı x Göynük	68,83**	-3,84	-4,61**	64,44*
Kınalı x G Northern 59	5,84	12,99	-17,66**	-12,91
Kınalı x Özmen	-58,92*	-20,55*	-15,73**	-63,13*
Alberto x Kınalı	69,40**	24,63**	-5,26**	66,43*
Alberto x Göynük	-2,55	17,58*	-3,14**	-5,65
Alberto x G Northern 59	-24,20	4,60	-22,27**	-41,07
Alberto x Özmen	16,19	15,23	-13,20**	3,56
Göynük x Kınalı	-44,95	12,11	-7,54**	-48,09*
Göynük x Alberto	21,75	17,13*	-2,66**	18,44
Göynük x G Northern 59	-26,03	1,18	-8,38**	-32,26
Göynük x Özmen	77,26**	4,82	-5,11**	75,66*
G Northern 59 x Kınalı	-3,40	-3,96	-5,99**	-9,24
G Northern 59 x Alberto	22,95	-5,38	-18,05**	0,59
G Northern 59 x Göynük	1,70	-17,05*	-10,33**	-9,06
G Northern 59 x Özmen	-24,90	-14,19	-14,64**	-36,07
Özmen x Kınalı	0,10	-22,14*	2,17*	8,91
Özmen x Alberto	-15,50	-24,17*	5,82**	-8,12
Özmen x Göynük	-7,36	-13,68	2,66**	-0,79
Özmen x G Northern 59	-11,00	38,30**	-2,88**	-13,45
Mean	3,94	0,64	-7,80	-1,71

** : significant at 1% level; * : significant at 5% level

4. Conclusions

It has been determined that the plant height trait has an additive gene effect. Non-additive gene effects were identified for other traits. The majority of the hybrids showed positive heterosis and heterobeltiosis values for seed yield, indicating that they are suitable for seed yield. The generation we obtained determined that gifted genotypes could be selected and used in advanced generations to develop new bean varieties and increase yield and quality.

5. Acknowledgements

This research was supported by the Selcuk University BAP project, number 21211001. This paper was summarized from Ph. D. Thesis of the first author (Nur Banu TEKİN). The authors would like to acknowledge the financial support of Selcuk University.

6. References

Akçin A (1974). Erzurum Şartlarında Yetiştirilen Kuru Fasulye Çeşitlerinde Gübreleme, Ekim Zamanı ve Sıra Aralığının Tane Verimine Etkisi ile Bu Çeşitlerin Bazı Fenolojik, Morfolojik ve Teknolojik Karekterleri Üzerinde Bir Araştırma, Atatürk Üniversitesi Ziraat Fakültesi Yayın No: 157, S:1-112, Erzurum, Türkiye.

Al Mukhtar FA and Coyne D (1981). Inheritance and Association of Flower, Ovule, Seed, Pod, and Maturity Characters in Dry Edible Beans *(Phaseolus vulgaris L.), Journal American Society for Horticultural Science*.

Arunga EE, Van Rheenen, HA, Owuoche JO (2010). Diallel Analysis of Snap Bean *(Phaseolus vulgaris L.)* Varieties for Important Traits, *African Journal of Agricultural Research*, 5 (15), 1951-1957.

Aydoğan C, Elkoca E, Haliloğlu K, Aydın M (2020). Bazı İspir Kuru Fasulye *(Phaseolus vulgaris L.)* Hatlarının Seleksiyonu Üzerine Bir Ön Çalışma, *Anadolu Ege Tarımsal Araştırma Enstitüsü Dergisi*, 30 (2), 251-265.

Babagil GE, Tozlu E ve Dizikısa T (2011). Erzincan ve Hınıs Ekolojik Koşullarında Yetiştirilen Bazı Kuru Fasulye *(Phaseolus vulgaris L.)* Genotiplerinin Verim Ve Verim Unsurlarının Belirlenmesi, *Atatürk Üniversitesi Ziraat Fakültesi Dergisi*, 42 (1), 11-17.

Barelli MAA, Goncalves Vidigal MC, Amaral Junior ATD, Vidigal Filho PS, Scapim CA, Sagrilo E (2000). Diallel analysis for seed yield and yield components in *Phaseolus vulgaris* L.. *Acta Scien-tiarum* 22(4), 883-887.

Bıyıklı B, Elkoca E, Aydin M (2015). İspir Kuru Fasulye *(Phaseolus vulgaris L.)* Popülasyonunun Karakterizasyonu ve Seleksiyon Yoluyla Islahı, *Anadolu Tarım Bilimleri Dergisi*, 36 (1), 20-33.

Bildirici N, Demir S (2019). Hakkâri Ekolojik Koşullarında Bazı Kuru Fasulye *(Phaseolus vulgaris L.)* Çeşitlerinin Verim ve Verim Özelliklerinin Belirlenmesi, *Bitlis Eren Üniversitesi Fen Bilimleri Dergisi*, 8 (4), 1250-1257.

Bozoğlu H ve Gülümser A (2000). Kuru Fasulyede *(Phaseolus vulgaris L.)* Bazı Tarımsal Özelliklerin Genotip Çevre Interaksiyonları ve Stabilitelerinin Belirlenmesi Üzerine Bir Araştırma, *Turkish Journal Agriculture and Forestry*, 24, 211-220.

Bozoğlu H, Sözen Ö (2007). Some Agronomic Properties of The Local Population of Common Bean *(Phaseolus vulgaris L.)* of Artvin Province, *Turkish Journal of Agriculture and Forestry*, 31 (5), 327-334.

Broughton WJ, Hernandez G, Blair M, Beebe S, Gepts P, Vanderleyden J. J. P, (2003). Beans (Phaseolus spp.)–model food legumes, 252 (1), 55-128.

Çukur T, Işın F (2008). İzmir İli Torbalı İlçesinde Sanayi Domatesi Üreticilerinin Sürdürülebilir Tarım Uygulamaları, *Ege Üniversitesi Ziraat Fakültesi Dergisi*, 45 (1), 27-36.

Câmara CR, Urrea CA, Schlegel V (2013). Pinto beans *(Phaseolus vulgaris L.)* as a Functional Food, *Agriculture Implications on Human Health*, 3 (1), 90-111.

Ceyhan E (2003). Bezelye Ebeveyn ve Melezlerinde Bazı Tarımsal Özelliklerin ve Kalıtımlarının Çoklu Dizi Analiz Metoduyla Belirlenmesi, *Selçuk Üniversitesi Fen Bilimleri Enstitüsü*. Selçuk Üniversitesi, Konya, 103.

Ceyhan E (2004). Effect of Sowing Dates on Some Yield Components and Yield of Dry Bean *(Phaseolus vulgaris L.)* Cultivars, *Turkish Journal of Field Crops*, 9 (2), 87-95.

Ceyhan E (2007). Yemeklik Tane Baklagiller Ders Notları, *Selçuk Üniversitesi Ziraat Fakültesi Tarla Bitkileri Bölümü*. 33-141.

Ceyhan E, Önder M, Kahraman A (2009). Fasulye Genotiplerinin Bazı Tarımsal Özelliklerinin Belirlenmesi, *Selcuk Journal of Agriculture and Food Sciences*, 23 (49), 67-73.

Ceyhan E, Kahraman A (2013). Genetic analysis of yield and some characters in peas, *Legume Research-An International Journal*, 36 (4), 273-279.

Ceyhan E, Harmankaya M, Kahraman A (2014a). Combining ability and heterosis for concentration of mineral elements and protein in common bean (*Phaseolus vulgaris* L.), *Turkish Journal of Agri-culture* and Forestry 38(5): 581-590.

Ceyhan E, Kahraman A, Avcı MA, Dalgıç H (2014b). Combining ability of bean genotypes estimated by line x tester analysis under highly-calcareous soils. *The Journal of Animal and Plant Sciences* 24(29): 579-584.

Ceyhan E, Şimşek D (2021). Fasulyede Tarımsal Özelliklerin Kalıtımlarının Çoklu Dizi Analiz Metoduyla Belirlenmesi, *Türk Tarım ve Doğa Bilimleri Dergisi*, 8 (1), 215-225.

Dişbudak K (2008). Avrupa Birliği'nde Tarım Çevre İlişkisi ve Türkiye'nin Uyumu.

Duc G, Agrama H, Bao S, Berger J, Bourion V, De Ron AM, Gowda CL, Mikic A, Millot D, Singh KBJC-RiPS (2015). Breeding annual seed legumes for sustainable agriculture: new methods to approach complex traits and target new cultivar ideotypes, 34 (1-3), 381-411.

Elkoca E, Çınar T (2015). Bazı Kuru Fasulye *(Phaseolus vulgaris L.)* Çeşit ve Hatlarının Erzurum Ekolojik Koşullarına Adaptasyonu, Tarımsal ve Kalite Özellikleri, *Anadolu Tarım Bilimleri Dergisi*, 30 (2), 141-153.

Eryılmaz GA, Kılıç O (2018). Türkiye'de Sürdürülebilir Tarım ve İyi Tarım Uygulamaları, *Tarım ve Doğa Dergisi*, 21 (4), 624.

Falconer DS (1980). Introduction to Quantitative Genetics. London. *Oliver and Boyd Ltd.*, 365.

Fonseca S, Patterson FL (1968). Hybrid vigor in a seven parent diallel cross in common winter wheat. *Crop Science* 8: 85-88

Genchev D (1995). Assessment of Tolerance to Stress Factors in Breeding Material of Kidney Beans *(Phaseolus vulgaris L.), Bulgarian Journal of Agricultural Science*, 1 (4), 415-422.

Gülnur Ç (2019). Bazı Kuru Fasulye *(Phaseolus vulgaris L.)* Genotiplerinin Eskişehir Ekolojik Koşullarına Adaptasyonu ile Tarımsal ve Kalite Özelliklerinin Belirlenmesi, *ESOGÜ, Fen Bilimleri Enstitüsü*.

Girgel Ü, Çokkizgin A, Çölkesen M (2018). Bayburt Koşullarında Organik Olarak Yetiştirilen Bazı Yerel Fasulye *(Phaseolus vulgaris L.)* Genotiplerinin Bazı Morfolojik ve Agronomik Özelliklerinin Belirlenmesi Üzerine Bir Araştırma, *Türk Tarım-Gıda Bilim ve Teknoloji dergisi*, 6 (5), 530-535.

Griffing B (1956). Concept of general and specific combining ability in relation to diallel crossing systems, *Australian journal of biological sciences*, 9 (4), 463-493.

Işık M (2001). Yemeklik Dane Bitkiler Yetiştirme Tekniği, *Anadolu Tarımsal Araştırma Enstitüsü, Eskişehir*.

Kepildek R, Ceyhan E (2021). Determination of Some Agronomic Traits of Fresh Bean Parents and Hybrids and Their Heritability with Diallel Analysis Method, *Selcuk Journal of Agriculture and Food Sciences*, 35 (2), 71-82.

Konuk A ve Uzun T (2021). Kuru Fasulye Genotiplerinde Bazı Tarımsal Özelliklerinin Belirlenmesi, *Bahri Dağdaş Bitkisel Araştırma Dergisi*, 10 (2), 161-168.

Kwaye GR, Shimelis H, William PM (2008). Combining ability analysis and association of yield and yield components among selected cowpea lines. Euphytica, 162: 205–210.

Menalled F, Bass T, Buschena D, Cash D, Malone M, Maxwell B, McVay K, Miller P, Soto R, Weaver DJMSU (2008). An introduction to the principles and practices of sustainable farming.

Muhammad G, Ramazan CM, Aslam M, Chaudhry GA (1994). Performance of cowpea cultivars under rain fed conditions. Journal of Agricultural Research (Pakistan), 32:55–61

Oliveira Junior A, Miranda G, Cruz C (1997). Evaluation of the combining ability of dry bean cultivars based on unbalanced circulating and partial diallel crossing systems, *Revista Ceres (Brazil)*.

Önder M, Özkaynak İ (1994). Bakteri Aşılaması ve Azot Uygulamasının Bodur Kuru Fasulye Çeşitlerinin Tane Verimi ve Bazı Özellikleri Üzerine Etkileri, *Tr. J. of Agricultural and Forestry*, 18, 463-471.

Özcan K ve Açıkgöz N (1999). Populasyon genetiği için bir istatistik paket program geliştirmesi. 3. Tarımda bilgisayar uygulamaları sempozyumu, Çukurova üniversitesi, 3(6).

Pekşen E (2005). Samsun Koşullarında Bazı Fasulye (*Phaseolus vulgaris L.*) Genotiplerinin Tane Verimi ve Verimle İlgili Özellikler Bakımından Karşılaştırılması, *Anadolu Tarım Bilimleri Dergisi*, 20 (3), 88-95.

Rodrigues R, Leal NR, Pereira MG (1998). Diallel analysis of six agronomic traits in *Phaseolus vul-garis* L. *Bragantia* 57(2): 241-250.

Singh SP, Urrea CA (1994). Selection for Seed Yield and Other Traits Among Early Generations of Intra and Interracial Populations of the Common Bean, *Revısta Brasıleıra De Genetıca*, 17, 299-299.

Sirat A (2020). Yerel Kuru Fasulye *(Phaseolus vulgaris L.)* Genotiplerinin Tane Verimi, Verim Unsurları ve Bazı Kalite Özelliklerinin Belirlenmesi, *Adnan Menderes Üniversitesi Ziraat Fakültesi Dergisi*, 17 (2), 245-254.

Soydaş V, Aydın M, Elkoca E, İlhan E (2019). Gümüşhane İli Yerel Fasulye Genotiplerinin Morfolojik ve Tarımsal Özellikler Yönünden Karakterizasyonu Üzerine Bir Ön Çalışma, *Anadolu Ege Tarımsal Araştırma Enstitüsü Dergisi*, 31 (2), 143-160.

Shimelis HA (2006). Associations of yield and yield components among selected durum wheats (*Triticum turgidum L.*). South African Journal of Plant and Soil, 23(4), 305–310.

Sprent JI, Sprent PJNFOP (1990). Nitrogen fixing organisms: pure and applied aspects.

Tan S, Köksal H (2004). Sürdürülebilir Tarım. Tarımsal Ekonomi ve Araştırma Enstitüsü. TEAE-BAKIŞ Sayı 5 Nüsha:2, 4 s.

Taşkesen S (2019). Bazı Kuru Fasulye *(Phaseolus vulgaris L.)* Genotiplerinin Erzincan Koşullarındaki Verim ve Verim Özelliklerinin Belirlenmesi, *Fen Bilimleri Enstitüsü*.

Turhan Ş (2005). Tarımda Sürdürülebilirlik ve Organik Tarım, *Tarım Ekonomisi Dergisi*, 11 (1 ve 2), 13-24.

Ülker M, Ceyhan E (2008a). Orta Anadolu Ekolojik Şartlarında Yetiştirilen Fasulye *(Phaseolus vulgaris L.)* Genotiplerinin Protein ve Bazı Mineral Oranlarının Belirlenmesi, *Selcuk Journal of Agriculture and Food Sciences*, 22 (46), 90-97.

Ülker M, Ceyhan E (2008b). Orta Anadolu Ekolojik Şartlarında Yetiştirilen Fasulye *(Phaseolus vulgaris L.)* Genotiplerinin Bazı Tarımsal Özelliklerinin Belirlenmesi, *Selcuk Journal of Agriculture and Food Sciences*, 22 (46), 77-89.

Varankaya S, Ceyhan E (2012). Yozgat Ekolojik Şartlarında Yetiştirilen Fasulye *(Phaseolus vulgaris L.)* Genotiplerinin Bazı Tarımsal Özelliklerinin Belirlenmesi, *Selcuk Journal of Agriculture and Food Sciences*, 26 (1), 27-33.

Yeken MZ, Kantar F, Çancı H, Özer G, Çiftçi V (2018). Türkiye'deki Yerel Phaseolus vulgaris Populasyonlarını Kullanarak Kuru Fasulye Çeşitlerinin Islahı, *Uluslararası Tarım ve Yaban Hayatı Bilimleri Dergisi*, 4 (1), 45-54.

Yolci MS (2020). Erciş (Van) ekolojik koşullarında bazı fasulye (Phaseolus Vulgaris L.) çeşitlerinin verim ve verim unsurlarının belirlenmesi, *Avrupa Bilim ve Teknoloji Dergisi* (18), 562-567.

Zimmerman MJO, Rosielle AA, Foster KW, Waines JG (1985). Gene action for seed yield and harvest index of common bean grown as sole crop and in intercrop with maize. *Field Crops Research* 12: 319-329.

Selcuk Journal of Agriculture and Food Sciences SJAFS

http://sjafs.selcuk.edu.tr/sjafs/index

(2022) 36 (3), 331-341

e-ISSN: 2458-8377

DOI:10.15316/SJAFS.2022.042

Research Article

Genetic Variability Studies in F_2 Generations of Determinate High Yield Fresh Bean Lines for Seed Yield and Yield Components

Şemsi TAMÜKSEK[1], Ercan CEYHAN[2,*]

[1] Selçuk University, The Graduate School of Natural and Applied Science, Department of Field Crops, Konya, Türkiye
[2] Selçuk University, Faculty of Agriculture, Department of Field Crops, Konya, Türkiye

ARTICLE INFO

Article history:
Received date: 09.08.2022
Accepted date: 23.08.2022

Keywords:
Bean
Diallel analysis
GCA
Heterosis
SCA

ABSTRACT

Fresh bean is the most important pulse crop which are sold fresh, canned, and frozen. The genetic variability and association studies help in selection which would increase the yield potential of fresh bean. The experiment was conducted using randomized block design in 2021 at Konya with 25 genotypes (5 parents and 20 F_2 Population). General and special combination abilities, heterosis and heterobeltiosis values, broad and narrow sense heritability, as well as the relationships between the investigated traits, were determined in the parent and F_2 hybrids suitable for the diallel analysis method. It was found that there is an additive gene effect in the inheritance of the seed yield trait, and non-additive gene effect in the inheritance of the protein ratio trait. It was determined that the heritability in the narrow sense was low for the seed yield and protein ratio traits. It was also determined that non-additive gene effects were effective in the inheritance of seed yield and protein ratio traits. In F_2 generation, the mean heterosis value determined in terms of seed yield was positive and the mean heterobeltiosis value was negative. As a result of this research, a sufficient level of genetic variation was determined in the population, considering the agronomic characteristics examined. Determination of suitable parents and hybrids for green bean breeding in terms of sustainability of calcareous soils, agronomic characteristics and inheritance of these parents and hybrids were determined.

1. Introduction

Sustainable agriculture is formed due to the combination of an agricultural structure in which agricultural inputs and technologies are used, both by ensuring the protection of natural resources in the long term and by not harming the environment. Sustainable agriculture is a system that protects natural resources such as water and soil and includes many other agricultural practices (integrated pharma management, etc.) (Turhan, 2005). As the calcareous rate in the soil increases, it combines with phosphorus, iron and calcium, which can be taken in the soil to form insoluble compounds. Necrosis, also called “lime chlorosis”, occurs due to iron deficiency in plants growing under a high amount of calcareous soil. Due to the high amount of lime in the soil, it is difficult to feed the plant, causing a decrease in yield and quality (Griffing, 1956; Falconer, 1989; Kwaye et al., 2008; Ceyhan et al., 2014).

Bean (*Phaseolus vulgaris* L.), an important cultural plant of the Leguminosae family, which is widely consumed worldwide, has an important place in terms of both high nutritional value and human nutrition. The bean plant has an important place in human nutrition with its very different usage areas as a fresh vegetable or canned form, and dry seed and seed canning (Bozoğlu, 1995). In the world, 23,276 million tons of green beans were produced in 15,796 million hectares of land in 2020. While China is in the first place with 18 million tons, Turkey is in the fourth place with approximately 547 thousand tons (FAO, 2022).

In their research, Arunga et al. (2010), determined the effects of the additive gene in the inheritance of plant height, pod number, and pod length traits, and Ceyhan et al. (2014b), Kepildek and Ceyhan (2021), determined the effects of non-additive genes in the inheritance of plant height, pod number, and pod length traits in their studies. Iqbal et al. (2010) determined the additive gene effect on the inheritance of pod length and hundred-seed weight traits, while they determined the non-additive

* Corresponding author email: *eceyhan@selcuk.edu.tr*

gene effect on the inheritance of pod number, seed number per pod, seed yield, and protein ratio traits.

The genetic structure of the plant and environmental conditions are important factors affecting the yield and quality of the plants obtained by breeding programs. To develop new varieties with breeding studies, it is necessary to find productive and high-quality genotypes suitable for environmental conditions or to develop the insufficient aspects of existing varieties. For this reason, green bean breeding is very important for the sustainability of calcareous soils. At this point, the aim of the study has led to the development of bean breeding programs depending on the selection of lines based on the desired genetic characteristics of the variable yield of beans under high calcareous soils. For this purpose, crosses were made between 5 green bean genotypes with very superior agricultural and technological properties. The research was carried out to investigate the genetic structure of the parent and the F_2 hybrid, to determine the appropriate parents and genotypes, heritability, heterosis, and heterobeltiosis values of the examined traits. At the same time, it was also aimed to determine the yield and quality of green bean parents and genotypes in terms of the sustainability of calcareous soils.

2. Materials and Methods

In the research, two registered cultivars (Beyzade and Lida) developed by Prof. Dr. Ercan CEYHAN and 3 commercial cultivars (Ribera, Albeni, and Garrafol) were used. These cultivars are suitable for Central Anatolian conditions, show superiority in terms of various yield components and quality characteristics, and have morphologically different characteristics. In the research, green bean seeds were planted in the Plant Breeding Greenhouse in 2019 using the diallel analysis method at four different sowing times. Thus, simultaneous flowering of green bean genotypes used in hybridization was ensured. The hybridization process was carried out according to Ceyhan (2004). In the research by diallel hybridization, 20 hybrid combinations were obtained as a result of 5 x 5 equation.

In the research, parents and F_1 hybrids were carried out in 2020 in the Experimention Field of the Selcuk University's Faculty of Agriculture. On 29.04.2021 Parents and F_2 hybrids were planted in the Experimention Field of the Selcuk University's Faculty of Agriculture. The experiments were set up on blocks of 2 m length with 3 replications according to the "Random Blocks Trial Design". To meet the nutritional needs of the parent and F_2 hybrids, 15 kg of urea was given to the trial area uniformly. To ensure germination and emergence, after planting, the plots were irrigated five times with the sprinkler irrigation method. Weed control was done mechanically or manually.

It was determined that the organic matter content of the trial field, which had a clayey soil structure, was at a moderate level of 2.25% at a soil depth of 0-30 cm, and at a low level of 1.23% at a soil depth of 30-60 cm (Table1). The trial field, which has a high level of lime content (37.6%, 34.4%), has an alkaline structure (pH = 8.05 - 8.00), and there is no problem in terms of salinity (Table1). The amount of usable phosphorus (1.79 kg/da - 1.34 kg/da) and zinc (0.32 ppm - 0.34 ppm) in the trial field is quite low (Table1). Considering the analysis results of the trial field, it is sufficient in terms of iron (14.74 ppm - 8.74 ppm), copper (1.70 ppm - 1.74 ppm), and manganese (7.50 ppm - 5.76 ppm)

In the province of Konya which corresponds to the area of research', the average temperature was determined as 19.4 ^{0}C by the 21-year meteorological data, and 19.8 ^{0}C in 2021 when the parent and F_2 plants were grown. In the research, the total precipitation during the 21-year growing period was determined as 109.1 mm, and the total precipitation during the cultivation period in 2021 was determined as 134.3 mm. In the research, the average relative humidity was determined as 47.8% in the 21-year growing period and the average relative humidity in the growing period in 2021 was 44.4%.

In the study, measurements, weighing and analyzes were made in terms of the examined characteristics in the parent and F_2 hybrids obtained from each plot In the research, the applications made in terms of plant height, pod length, pod width, number of pods per plant, number of seeds per pod, number of seeds per plant, seed yield per plant, hundred-seed weight, protein ratio, and protein yield were performed according to the method proposed by Ceyhan et al. (2014a) and Ceyhan et al. (2014b).

In the research, the measurements made in the F_2 hybrids were first calculated analysis of variance according to the "Random Blocks Trial Design". Calculations were made using the Diallel Analysis Method for traits with significant variance values of 1% and 5% between hybrids. In the research, analysis and calculations were determined in TARPOPGEN PC Program (Özcan and Açıkgöz, 1999).

3. Results and Discussion

In terms of the characteristics examined in the study, mean squares of preliminary variance analysis and combining ability variance analysis in full diallel hybrid set are given in Table 1.

In the study, as a result of full diallel variance analysis, it was determined that the mean squares of the crosses were statistically significant in terms of the examined characteristics. In the F_2 generation, when the genotypes were examined, there was significant variation at the level of 5% in pod width and number of seeds per pod, while there was significant variation at the level of 1% when all other characteristics are considered (Table 1).

In the F_2 generation, when the combining ability variances were examined in the full diallel hybrid group, significant changes were noted in the GCA values for all the examined traits except for the plant height trait. When the SCA values were examined, there were significant differences in all the investigated properties except the pod width and the number of seeds per pod.

When the reciprocal effect values were examined, it was found to be statistically significant in terms of all the investigated properties except pod width, seed yield per plant, hundred-seed weight, and protein yield per plant (Table 1).

Table 1
Mean squares of initial variance analysis and combining ability variance analysis for investigated traits in a full-diallel hybrid set

Source of Variation	SD	Plant Height	Number of Pods per Plant	Number of Seeds per Pod	Number of Seeds per Plant
Blocks	2	255,110	97,469	1,433	3788,182
Genotypes	24	72,110**	71,807**	0,959*	883,355**
Error	48	11,497	21,052	0,457	273,713
GCA	4	78,525	56,708**	0,533*	644,030**
SCA	10	17,715**	15,245*	0,108	222,624*
Reciprocal Effect	10	8,563*	19,518**	0,446**	226,448*
Error	48	3,832	7,017	0,152	91,238
Source of Variation	SD	Seed Yield	Hundred Seed Weight	Protein Ration	Protein Yield
Blocks	2	34,781	639,091	0,321	50,140
Genotypes	24	105,782**	128,655**	3,475**	10,417**
Error	48	32,181	36,143	0,456	2,866
GCA	4	91,272**	134,467**	1,836**	9,803**
SCA	10	27,892*	28,832*	1,374**	2,577*
Reciprocal Effect	10	20,225	20,305	0,672**	1,836
Error	48	10,727	12,048	0,152	0,956

* : significant at 5% level , ** : significant at 1% level

3.1. Plant height

In terms of plant height, it was determined that the parental values varied between 27.44 cm (Ribera) and 40.22 cm (Albeni), and in the F_2 generation, it varied between 28.77 cm (Ribera x Lida) and 50.00 cm (Beyzade x Garrafol) (Table 2). Researchers working on this subject have found similar results (Genchev, 1995; Ülker and Ceyhan, 2008a; Varankaya and Ceyhan, 2012; Kepildek and Ceyhan, 2021).

If the $\upsilon^2GCA/\upsilon^2SCA$ ratio of plant height was less than 1 in the F_2 generation, it showed that the non-additive gene effect was effective in the inheritance of this trait. Likewise, if the ratio of $(H/D)^{1/2}$ was more significant than 1, it was found that this feature had superior dominance (Table 2).They found that the non-additive gene effect and the dominant gene effect were effective in the inheritance of plant height of the bean plant (Rodrigues et al., 1998; Ceyhan et al., 2014b; Kepildek and Ceyhan, 2021).

When the parental GCA was examined for plant height, the Garrafol cultivar showed significant and positive effects at the level of 1%, while Lida (5%) and Riberia (1%) cultivars showed significant and negative effects (Table 3). Garrafol variety, whose parental GCA value was positive and significant, can be preferred as parents in breeding studies to increase plant height. Lida and Ribera varieties, which were negative and important, can be preferred in breeding studies to obtain short or medium-sized varieties suitable for machine harvesting. Kepildek and Ceyhan (2021) found significant GCA values in terms of plant height in their study.

Considering the SCA values of the hybrids, in the F_2 generation, the "Beyzade x Garrafol" cross was found to be significant and positive at the level of 5%, and the "Beyzade x Garrafol" cross was significant and positive at the level of 1%, while the "Albeni x Garrafol" cross was significant and negative at the level of 5% (Table 3). These combinations were genotypes with breeding potential for plant height. While hybrid combinations with positive and significant SCA values were preferred to increase plant height, hybrid combinations with negative and significant SCA values can be preferred in breeding studies to obtain short or medium-sized plants suitable for machine harvesting. In a study, significant, positive, and negative SCA values in terms of plant height (Kepildek and Ceyhan, 2021) were determined.

In terms of plant height in the F_2 generation, the average heterosis value was 7.74% and the heterobeltiosis value was -2.49%. In terms of plant height in the F_2 generation, heterosis values ranged between -9.60% (Lida x Albeni) and 45.19% (Beyzade x Garrafol), while heterobeltiosis values ranged between -21.00% (Albeni x Ribera) and 25.57% (Beyzade x Garrafol) (Table 4 and 5) . Investigating the heterosis and heterobeltiosis values in terms of plant height, Arunga et al. (2010), Ceyhan et al. (2014b), Kepildek and Ceyhan (2021) stated that they found high or low rates of heterosis and heterobeltiosis.

For plant height, heritability in the broad and narrow sense detected in the F_2 generation were 0.78 and 0.31 respectively (Table 2). In terms of plant height, the high heritability in the broad sense and the lower heritability in the narrow sense indicate that the effect of environmental variance on this trait was high. These results revealed that selection in terms of plant height could be made from the next generation.

Table 2
Mean values for investigated traits in full-diallel hybrid set

Parents	Plant Height		Number of Pods per Plant		Number of Seeds per Pod		Number of Seeds per Plant	
Lida	33,77	c-h	16,22	cde	6,11	ab	66,00	cde
Beyzade	29,05	gh	16,66	cde	5,33	a-e	61,44	cde
Ribera	27,44	h	12,99	e	4,33	g	40,44	e
Albeni	40,22	bc	24,89	abc	5,78	a-e	80,22	a-d
Garrafol	39,82	bcd	13,55	de	5,77	a-e	44,89	cde
F_2 H Populations								
Lida x Beyzade	32,66	d-h	24,11	abc	5,44	a-f	68,66	b-e
Lida x Ribera	32,00	e-h	16,55	cde	5,78	a-e	65,33	cde
Lida x Albeni	33,44	c-h	16,33	cde	4,61	fg	56,05	cde
Lida x Garrafol	39,03	b-f	22,08	b-e	4,92	d-g	63,19	cde
Beyzade x Lida	33,22	c-h	18,78	b-e	5,75	a-g	60,33	cde
Beyzade x Ribera	34,22	c-h	16,89	cde	5,00	c-g	51,88	cde
Beyzade x Albeni	42,11	b	23,33	a-d	6,00	a-d	81,11	abc
Beyzade x Garrafol	50,00	a	33,22	a	5,11	b-g	115,78	a
Ribera x Lida	28,77	gh	13,11	e	4,55	fg	44,78	de
Ribera x Beyzade	36,11	b-g	15,66	cde	5,44	a-f	56,66	cde
Ribera x Albeni	33,33	c-h	14,88	cde	5,11	b-g	51,22	cde
Ribera x Garrafol	37,66	b-f	17,89	b-e	4,89	efg	55,00	cde
Albeni x Lida	34,66	c-h	21,83	b-e	6,33	a	66,67	cde
Albeni x Beyzade	40,11	bc	24,66	abc	5,55	a-f	78,11	bcd
Albeni x Ribera	31,77	fgh	17,44	b-e	5,55	a-f	50,11	cde
Albeni x Garrafol	37,44	b-f	22,33	b-e	6,11	ab	67,89	b-e
Garrafol x Lida	39,77	bcd	16,89	cde	6,00	a-d	67,88	b-e
Garrafol x Beyzade	37,97	b-f	17,11	b-e	6,05	abc	66,55	cde
Garrafol x Ribera	37,11	b-f	19,22	b-e	6,22	a	67,89	b-e
Garrafol x Albeni	39,33	b-e	27,00	ab	6,00	a-d	103,66	ab
GCA	14,94		9,94		0,08		110,56	
SCA	41,65		24,68		-0,13		394,16	
Reciprocal	4,73		12,50		0,29		135,21	
υ^2GCA / υ^2SCA	0,36		0,40		---		0,28	
$H/D^{1/2}$	76,26		57,06		----		750,49	
H^2	0,78		0,70		0,39		0,64	
h^2	0,31		0,24		0,19		0,19	

GCA: General Combining Ability; SCA: Specific Combining Ability; $H/D^{1/2}$: Mean Degree of Dominance; H^2: Broad Sense Heritability; h^2: Narrow Sense

3.2. Number of Pods

In terms of the number of pods, it was determined that the parent values varied between 13.55 numbers/plant (Garrafol) and 24.89 numbers/plant (Albeni), and the F_2 generation values varied between 13.11numbers/plant (Ribera x Lida) and 33.20 numbers/plant (Beyzade x Garrafol) (Table 2). Some researchers also reached similar results (Ceyhan, 2004; Ülker and Ceyhan, 2008a; Varankaya and Ceyhan, 2012; Ceyhan et al., 2014b; Kepildek and Ceyhan, 2021; Mutari et al., 2022).

In terms of the number of pods, it was determined that the non-additive gene effect was effective if the υ^2GCA / υ^2SCA ratio was less than 1, and the superior dominance was effective if the $(H/D)^{1/2}$ ratio was more than 1 (Table 2). In some studies, the number of pods was found to be augmented by non-additive genes (Iqbal et al., 2010; Ceyhan et al., 2014b; Kepildek and Ceyhan, 2021; Mutari et al., 2022) and additive genes (Barelli et al., 2000; Arunga et al., 2010) reported that it was effective.

When the parental GCA was examined, the Albeni cultivar, which had a significant and positive GCA value of 5%, can be preferred as a parent that can be used in breeding studies to increase the number of pods. When the SCA values of the hybrids were examined, the "Beyzade x Garrafol" combination had a significant and positive SCA effect at the level of 5% in the F_2 generation. Since the "Beyzade x Garrafol" combination had a significant and positive SCA value at the 5% level, it had been determined as the appropriate hybrid combination in breeding studies to increase the number of pods (Table 3). Arunga et al. (2010), Iqbal et al. (2010), Ceyhan et al. (2014b), Kepildek and Ceyhan (2021), Mutari et al. (2022), examined the GCA and SCA values of the parents and hybrids in terms of pod number traits, obtained similar results.

In the F_2 generation, the mean heterosis value was 20.09% and the heterobeltiosis value was 6.30%. Heterosis values were found to vary between -21.42% (Ribera x Albeni) and 119.88% (Beyzade x Garrafol), and

heterobeltiosis values between -40.20% (Ribera x Albeni) and 99.36% (Beyzade x Garrafol) (Table 4 and 5). The large differences between the heterosis and heterobeltiosis values of hybrid combinations revealed that the pod number feature was affected by environmental conditions. Investigating the heterosis and heterobeltiosis values in terms of the number of pods, Ceyhan et al. (2014b), Kepildek and Ceyhan (2021) determined both negative and positive heterosis and heterobeltiosis values.

Heritability in the broad and narrow sense was 0.70 and 0.24 respectively in the F_2 generation (Table 2). High heritability in the broad sense and low heritability in the narrow sense revealed that the number of pods was under the influence of environmental conditions. Therefore it would be more appropriate to start the selection process in the next generations.

Table 3
Genetic components for investigated traits in full-diallel hybrid set

Parents	Plant Height	Number of Pods per Plant	Number of Seeds per Pod	Number of Seeds per Plant
Lida	-1,930*	-1,134	0,009	-2,781
Beyzade	0,409	1,364	0,033	4,928
Ribera	-3,455**	-3,582**	-0,390*	-12,895**
Albeni	1,222	2,413*	0,172	6,256
Garrafol	3,753**	0,939	0,175	4,492
F_2 Populations				
Lida x Beyzade	-1,578	1,867	-0,164	-2,920
Lida x Ribera	-0,271	0,202	0,035	5,460
Lida x Albeni	-1,281	-1,544	-0,220	-7,386
Lida x Garrafol	1,536	0,334	-0,234	-1,442
Beyzade x Lida	0,278	-2,665*	-0,057	-4,167
Beyzade x Ribera	2,170	-0,851	0,068	-3,029
Beyzade x Albeni	3,434*	0,875	0,061	3,157
Beyzade x Garrafol	3,780**	3,518*	-0,137	16,474*
Ribera x Lida	-1,612*	-1,720	-0,613**	-10,277*
Ribera x Beyzade	0,945	-0,612	0,223	2,390
Ribera x Albeni	-1,257	-2,014	0,037	-7,967
Ribera x Garrafol	1,046	1,850	0,259	4,576
Albeni x Lida	0,608	2,750*	0,860**	5,306
Albeni x Beyzade	-1,000	0,667	-0,222	-1,500
Albeni x Ribera	-0,778	1,278	0,222	-0,557
Albeni x Garrafol	-2,631*	1,967	0,198	9,758
Garrafol x Lida	0,373	-2,597*	0,542**	2,345
Garrafol x Beyzade	-6,013**	-8,055**	0,473*	-24,613**
Garrafol x Ribera	-0,278	0,665	0,667**	6,445
Garrafol x Albeni	0,945	2,333*	-0,057	17,888**
Gi	0,307	0,561	0,012	7,299
Sij	1,303	2,386	0,052	31,021
Rij	1,916	3,509	0,076	45,619

G_i : GCA, S_{ij}: SCA; R_{ij}: Reciprocal effect, ** : significant at 1% level; * : significant at 5% level

3.3.Number of seeds per pod

In terms of the number of seeds per pod, it was determined that the parent values varied between 4.33 numbers/pod (Ribera) and 6.11 numbers/pod (Lida), and the F_2 generation values varied between 4.55 numbers/pod (Ribera x Lida) and 6.33 numbers/pod (Albeni x Lida) (Table 2). Ülker and Ceyhan (2008a), Iqbal et al. (2010), Varankaya and Ceyhan (2012), Ceyhan et al. (2014b), Kepildek and Ceyhan (2021), and Mutari et al. (2022) showed similar results to these research findings.

In terms of the number of seeds per pod, it had been determined that the non-additive gene effect was effective in the case of the υ^2GCA/υ^2SCA ratio of less than 1, and the superior dominance was effective in the case of $(H/D)^{1/2}$ ratio more than 1 in the F_2 generation (Table 2). Ceyhan et al. (2014b), Kepildek and Ceyhan (2021), and Mutari et al. (2022) showed similar results with this research findings.

When GCA was examined regarding the number of seeds in the pod, Albebi genotypes were found to have significant and positive values in the F_2 generation. (Table 3). As a result, Albeni genotype, which was positively important in increasing the number of seeds per pod, was determined as suitable parents for breeding studies for this purpose.

Looking at the SCA effects of crosses in the F_2 generation, "Beyzade x Garrafol", "Albeni x Lida" and "Garrafol x Albeni" hybrid had a positive significant SCA effect (Table 3). They can be considered suitable combinations thatcan be used to increase the number of seeds in the pod, as they had the effect of SCA. In many

studies, the effects of GCA and SCA for the number of seeds per pod were determined (Arunga ve ark. 2010; Iqbal ve ark. 2010; Ceyhan ve ark. 2014b; Kepildek ve Ceyhan 2021; Mutari ve ark. 2020).

When the F_2 generation was examined, the mean heterosis value was determined as -0.54% and the mean heterobeltiosis value was determined as -6.78%. Heterosis values were determined between -22.43% (Lida x Albeni) and 23.13% (Garrafol x Ribera), heterobeltiosis were determined between -25.33% (Ribera x Lida) and 7.74% (Garrafol x Ribera) (Table 4 and 5). Determined the heterosis and heterobeltiosis values in terms of the number of pods in beans, Ceyhan et al. (2014b), and Kepildek and Ceyhan (2021) found both negative and positive heterosis and heterobeltiosis values.

The heritability in the broad and narrow sense determined in terms of the number of seeds in the pod was 0.39 and 0.19, respectively (Table 2). It had been revealed that the heritability in the broad sense was high and the heritability was low in the narrow sense, the number of seeds per pod was affected by the environmental factor.

Table 4
Heterosis (%) values for investigated traits in full-diallel hybrid set

F_2 Populations	Plant Height	Number of Pods per Plant	Number of Seeds per Pod	Number of Seeds per Plant
Lida x Beyzade	3,98	46,63	-8,18**	7,76
Lida x Ribera	4,54	13,32	10,66**	22,76
Lida x Albeni	-9,60	-20,54	-22,43**	-23,33
Lida x Garrafol	6,07	48,34	-17,25**	13,98
Beyzade x Lida	5,75	14,21	-10,09**	-5,32
Beyzade x Ribera	21,14	13,88	-0,83	1,85
Beyzade x Albeni	21,57	12,30	4,08**	14,51
Beyzade x Garrafol	45,19*	119,88	-11,34**	117,77
Ribera x Lida	-6,00	-10,24	-12,84**	-15,86
Ribera x Beyzade	27,83	5,63	8,04**	11,23
Ribera x Albeni	-1,48	-21,42	1,06	-15,10
Ribera x Garrafol	11,99	34,76	-3,27*	28,91
Albeni x Lida	-6,32	6,22	6,51**	-8,81
Albeni x Beyzade	15,79	18,72	-3,62*	10,28
Albeni x Ribera	-6,08	-7,92	9,83**	-16,95
Albeni x Garrafol	-6,44	16,18	5,80**	8,53
Garrafol x Lida	8,09	13,45	0,98	22,44
Garrafol x Beyzade	10,27	13,25	5,09**	25,18
Garrafol x Ribera	10,34	44,78	23,13**	59,12
Garrafol x Albeni	-1,72	40,46	3,84*	65,72
Mean	7,74	20,09	-0,54	16,23

** : significant at 1% level; * : significant at 5% level

3.4. Number of Seeds per Plant

In terms of the number of seeds per plant, were found to vary the parent values between 40.44 numbers/plant (Ribera) and 80.22 numbers/plant (Albeni), and the F_2 generation values were between 44.78 numbers/plant (Ribera x Lida) and 115.78 numbers/plant (Beyzade x Garrafol) (Table 2). Our study results agree with some of the results of Ülker and Ceyhan (2008a), Ceyhan et al. (2014b), Kepildek ve Ceyhan, (2021).

In terms of the number of seeds in the plant, it had been determined that the non-additive gene effect was effective in the case of the υ^2GCA / υ^2SCA ratio of less than 1, and the superior dominance was effective in the case of $(H/D)^{1/2}$ ratio more than 1 in the F_2 generation (Table 2). However, Barelli et al. (2000) reported that additive and non-additive genes had equal effects on the inheritance of the seed number in pods in beans, while Ceyhan et al. (2014b), Kepildek and Ceyhan (2021), on the other hand, reported that the effect of non-additive genes was important in the inheritance of seed number in pods in beans.

Considering the SCA values of the hybrids, the "Beyzade x Garrafol" hybrid with a significant and positive SCA value of 5% in the F_2 generation was determined as the appropriate hybrid combination in breeding studies aimed at increasing the number of seeds in the plant (Table 3). In the study by Kepildek and Ceyhan (2021), GCA and SCA values were significant.

It was determined in terms of the number of seeds in the plant, the average heterosis value was 16.23% and the average heterobeltiosis value was -0.02%. Heterosis values varied between -23.33% (Lida x Allure) and 117.77% (Beyzade x Garrafol), while heterobeltiosis values ranged between -37.54% (Albeni x Ribera) and 88.40% (Beyzade x Garrafol) (Table 4 and 5).

Heritability in the broad and narrow sense was 0.64 and 0.19, respectively (Table 2). High heritability in the broad sense and low heritability in the narrow sense explains that environmental conditions had an effect on the heritability of the number of seeds per pod. Any selection process to be made in terms of the number of seeds in the plant should be done in later generations.

Table 5
Heterobeltiosis (%) values for investigated traits in full-diallel hybrid set

F_2 Populations	Plant Height	Number of Pods per Plant	Number of Seeds per Pod	Number of Seeds per Plant
Lida x Beyzade	-3,29	44,67*	-10,91	4,04
Lida x Ribera	-5,26	2,06	-5,46	-1,01
Lida x Albeni	-16,85**	-34,38*	-24,55**	-30,13*
Lida x Garrafol	-1,98	36,16*	-19,53*	-4,25
Beyzade x Lida	-1,64	12,68	-12,77	-8,59
Beyzade x Ribera	17,78**	1,34	-13,05	-15,56
Beyzade x Albeni	4,69	-6,26	3,81	1,11
Beyzade x Garrafol	25,57**	99,36**	-11,55	88,43**
Ribera x Lida	-14,80*	-19,16	-25,53**	-32,15*
Ribera x Beyzade	24,29**	-6,00	-5,28	-7,78
Ribera x Albeni	-17,13**	-40,20**	-11,60	-36,15*
Ribera x Garrafol	-5,41	31,97*	-15,36	22,52
Albeni x Lida	-13,82*	-12,28	3,60	-16,90
Albeni x Beyzade	-0,28	-0,90	-3,87	-2,63
Albeni x Ribera	-21,00	-29,92	-3,92	-37,54*
Albeni x Garrafol	-6,91	-10,27	5,77	-15,37
Garrafol x Lida	-0,11	4,13	-1,80	2,86
Garrafol x Beyzade	-4,64	2,68	4,85	8,31
Garrafol x Ribera	-6,81	41,79*	7,74	51,24*
Garrafol x Albeni	-2,21	8,48	3,81	29,22*
Mean	-2,49	6,30	-6,78	-0,02

** : significant at 1% level; * : significant at 5% level

3.5. Seed yield

In terms of seed yield per plant, the parent values differed between 29.68 g/plant (Lida) and 51.61 g/plant (Garrafol), and the values of the F_2 generation differed between 27.48 g/plant (Lida x Ribera) and 51.03 g/plant (Garrafol x Lida) (Table 6). Ülker and Ceyhan (2008a), Varankaya and Ceyhan (2012), Ceyhan et al. (2014b), and Kepildek and Ceyhan (2021), reported similar results with the present study.

In terms of seed yield in the plant, it was determined that the non-additive gene effect was effective when the υ^2GCA/υ^2SCA ratio was less than 1. In addition, if the ratio $(H/D)^{1/2}$ was more than 1, it showed that there was superior dominance (Table 6). Our study revealed that there was no simple inheritance of the seed yield of a plant in the bean. Some researchers (Zimmermann et al., 1985; Singh and Urrea, 1994; Oliveira Junior et al., 1997) who researched this subject stated that additive genes were effective in the inheritance of grain yield in beans, while others (Barelli et al., 2000; Iqbal et al., 2010; Ceyhan et al., 2014b; Kepildek and Ceyhan, 2021) reported that non-additive genes were effective. In the study of Chung and Stevenson (1973), it was found that the dominant gene effect was more than the additive gene effect in the inheritance of seed yield in the plant.

When the GCA values were examined, it was determined that the Garrafol variety had an important and positive effect at the level of 1% and the Albeni variety had an important and positive effect at the level of 5%. Garrafol and Albeni varieties, which had positive and important GCA values, emerge as parents that can be recommended to be used in breeding studies in terms of seed yield in the plant. When the SCA values of the hybrids were examined, the "Lida x Garrafol" hybrid, which showed a significant and positive SCA value at the level of 5%, can be used as a breeding material in terms of seed yield (Table 7). Many studies had been carried out in terms of GCA and SCA values in beans, and as a result of these studies, it had been determined that there were a parent and hybrid combinations with significant GCA and SCA values in different numbers in terms of seed yield (Arunga et al., 2010; Iqbal et al., 2010; Ceyhan et al., 2014b; Kepildek and Ceyhan, 2021).

The mean heterosis value was determined as 0.32% and the mean heterobeltiosis value was determined as -10.06%. Heterosis values varied between -22.08% (Lida x Riberia) and 25.54% (Garrafol x Lida), while heterobeltiosis values ranged between -32.72% (Lida x Ribera) and 11.64% (Beyzade x Ribera) (Table 8 and 9).

Heritability in the broad sense was 0.74, and heritability in the narrow sense was 0.26 (Table 6). In terms of seed yield feature, the fact that the heritability in the broad sense was higher and the heritability degree in the narrow sense was less indicates that it was due to the effect of the environmental factor. For this reason, it was necessary not to carry out the selection processes in terms of seed yield in the early period. To have a high success rate, it was necessary to perform the selection process for the next generations.

Table 6
Mean values for investigated traits in full-diallel hybrid set

Parents	Seed Yield		Hundred Seed Weight		Protein Ration		Protein Yield	
Lida	29,68	ef	20,13	d-g	28,12	abc	5,68	ef
Beyzade	37,67	b-f	24,00	c-g	25,00	ef	6,05	def
Ribera	40,85	a-e	16,86	g	27,85	a-d	4,68	f
Albeni	37,15	b-f	30,17	a-f	24,31	f	7,37	b-f
Garrafol	51,61	a	23,31	c-g	26,77	cd	6,24	def
F_2 Populations								
Lida x Beyzade	41,60	a-e	26,57	b-g	27,01	bcd	7,20	b-f
Lida x Ribera	27,48	f	18,59	efg	29,12	a	5,44	ef
Lida x Albeni	37,20	b-f	20,14	d-g	28,09	abc	5,63	ef
Lida x Garrafol	42,50	a-d	27,43	b-g	26,85	bcd	7,37	b-f
Beyzade x Lida	41,61	a-e	25,39	b-g	27,22	bcd	6,95	b-f
Beyzade x Ribera	45,60	abc	22,99	c-g	27,69	a-d	6,34	def
Beyzade x Albeni	33,70	c-f	28,10	b-g	27,01	bcd	7,59	b-f
Beyzade x Garrafol	36,82	b-f	42,62	a	27,85	a-d	11,87	a
Ribera x Lida	37,06	b-f	17,14	fg	26,89	bcd	4,61	f
Ribera x Beyzade	40,57	a-e	23,31	c-g	27,88	abc	6,49	def
Ribera x Albeni	35,20	c-f	18,51	efg	27,03	bcd	4,99	ef
Ribera x Garrafol	40,14	a-e	22,63	c-g	26,39	de	5,98	def
Albeni x Lida	33,02	def	22,63	c-g	27,73	a-d	6,27	def
Albeni x Beyzade	35,84	c-f	28,49	b-g	26,69	cd	7,61	b-f
Albeni x Ribera	40,02	a-e	20,56	d-g	27,66	a-d	5,70	ef
Albeni x Garrafol	35,27	c-f	24,68	c-g	28,02	abc	6,90	c-f
Garrafol x Lida	51,03	a	34,96	abc	29,11	a	10,20	abc
Garrafol x Beyzade	49,14	ab	33,21	a-d	28,28	ab	9,41	a-d
Garrafol x Ribera	43,79	a-d	30,79	a-e	27,95	abc	8,62	a-e
Garrafol x Albeni	37,37	b-f	38,10	ab	27,94	abc	10,65	ab
GCA	16,11		24,48		0,34		1,77	
SCA	51,50		50,35		3,67		4,86	
Reciprocal	9,50		8,26		0,52		0,88	
υ^2GCA / υ^2SCA	0,31		0,49		0,09		0,36	
$H/D^{1/2}$	93,21		107,58		4,86		9,28	
H^2	0,74		0,64		0,92		0,66	
h^2	0,26		0,29		0,13		0,25	

GCA: General Combining Ability; SCA: Specific Combining Ability; $H/D^{1/2}$: Mean Degree of Dominance; H^2: Broad Sense Heritability; h^2: Narrow Sense

3.6. Hundred-seed weight

It was determined that the parent values in terms of hundred-seed weight ranged between 16.86 g (Ribera) and 30.17 g (Albeni), and the F_2 generation values ranged between 17.14 g (Ribera x Lida) and 42.62 g (Beyzade x Garrafol) (Table 6). Researchers, who conducted similar studies, found close results (Iqbal et al., 2010; Ceyhan et al., 2014b; Senbetay et al., 2015; Kepildek and Ceyhan, 2021; Mutari et al., 2022).

In the F_2 generation, the non-additive gene effect of having υ^2GCA / υ^2SCA ratios less than 1 is effective in the inheritance of the hundred-seed weight trait, while the $(H/D)^{1/2}$ ratio being more than 1 revealed that superior dominance was effective (Table 6). Chung and Stevenson (1973), and Iqbal et al. (2010) stated that there was an additive gene effect in the inheritance of hundred-seed weight in beans, while Ceyhan et al. (2014b), and Kepildek and Ceyhan (2021) determined that non-additive genes were effective in the inheritance of hundred-seed weight in beans.

When the GCA was examined, the Garrafol variety, which had an important and positive value at the level of 5%, can be preferred as the appropriate parent in breeding studies to increase the hundred-seed weight. When the SCA values of the hybrids were examined, it was determined that the "Beyzade x Garrafol" cross in the F_2 generation was the genotype that could be preferred in breeding studies to increase the hundred-seed weight, since it had an important and positive value at the 5% level (Table 7). Iqbal et al. (2010), Ceyhan et al. (2014b), Senbetay et al. (2015), Kepildek and Ceyhan (2021), Mutari et al. (2022) determined the genotypes with significant and positive GCA and SCA values for hundred-seed weight in their studies.

The mean heterosis value was determined as 15.46% and the mean heterobeltiosis value was determined as 3.33%. Heterosis values varied between -21.28% (Riberia x Albeni) and 80.20% (Beyzade x Garrafol), while heterobeltiosis values ranged between -38.64% (Riberia x Albeni) and 77.60% (Beyzade x Garrafol) (Table 8 and 9).

Table 7
Genetic components for investigated traits in full-diallel hybrid set

Parents	Seed Yield	Hundred Seed Weight	Protein Ration	Protein Yield
Lida	-2,191	-2,342*	0,449*	-0,532
Beyzade	0,746	2,216*	-0,415*	0,524
Ribera	-0,120	-4,829**	0,252	-1,280**
Albeni	-3,085*	0,504	-0,501*	-0,026
Garrafol	4,650**	4,452*	0,215	1,314**
F_2 Populations				
Lida x Beyzade	3,772	0,456	-0,296	0,049
Lida x Ribera	-4,695*	-0,619	-0,074	-0,198
Lida x Albeni	1,108	-2,429	0,584	-0,529
Lida x Garrafol	5,026*	3,433	-0,060	0,965
Beyzade x Lida	0,007	-0,592	0,103	-0,125
Beyzade x Ribera	3,184	0,109	0,567	0,139
Beyzade x Albeni	-2,168	-0,077	0,386	0,069
Beyzade x Garrafol	-1,693	5,595*	0,888	1,771*
Ribera x Lida	4,790**	-0,725	-1,116	-0,413
Ribera x Beyzade	-2,513	0,158	0,095	0,074
Ribera x Albeni	1,541	-1,791	0,215	-0,385
Ribera x Garrafol	-1,840	1,435	-0,675	0,235
Albeni x Lida	-2,094	1,245	-0,183	0,318
Albeni x Beyzade	1,067	0,192	-0,159	0,005
Albeni x Ribera	2,410	1,023	0,316	0,353
Albeni x Garrafol	-4,524*	0,785	0,886	0,453
Garrafol x Lida	4,265*	3,762	1,129	1,415**
Garrafol x Beyzade	6,157**	-4,708**	0,214	-1,229*
Garrafol x Ribera	1,827	4,080	0,783	1,321**
Garrafol x Albeni	1,048	6,710**	-0,042	1,872**
Gi	0,858	0,964	0,012	0,076
Sij	3,647	4,096	0,052	0,325
Rij	5,364	6,024	0,076	0,478

G_i : GCA, S_{ij}: SCA; R_{ij}: Reciprocal effect, ** : significant at 1% level; * : significant at 5% level

Heritability in the broad sense was 0.74, and heritability in the narrow sense was 0.26 (Table 6). In the narrow sense, it was seen that the contribution of the environment was high in the inheritance of a hundred-seed weight trait. For this reason, it may be more appropriate to start the selection process in terms of hundred-seed weight in later generations.

3.7. Protein content

In terms of protein ratio, the parental values were found to be between 24.31% (Albeni) and 28.12% (Lida), while the values of the F_2 generation ranged between 26.39% (Riberia x Garrafol) and 29.12% (Lida x Ribera) (Table 6). Ceyhan (2006), Ülker and Ceyhan (2008b), Iqbal et al. (2010), Varankaya and Ceyhan (2012), Ceyhan et al. (2014a), and Kepildek and Ceyhan (2021) had similar results with results of this research.

For the protein ratio, which was at the top of the quality factors, υ^2GCA / υ^2SCA ratios less than 1 and $(H/D)^{1/2}$ ratio of more than 1 revealed the non-additive gene effect and superior dominance (Table 6). Iqbal et al. (2010), Ceyhan et al. (2014a), and Kepildek and Ceyhan (2021) determined in their studies that non-additive genes were effective in the inheritance of protein ratio in beans.

When the parental GCA was examined, the Lida variety, which had an important and positive value at the 5% level, had been determined to be preferred in breeding studies to increase the protein ratio (Table 7). Iqbal et al. (2010), Ceyhan et al. (2014a), and Kepildek and Ceyhan (2021) reported that there were genotypes with significant and positive GCA and SCA values in terms of protein ratio.

The mean heterosis value was 4.69% and the mean heterobeltiosis value was 0.70%. Heterosis values varied between -3.92% (Ribera x Lida) and 9.72% (Albeni x Garrafol), while heterobeltiosis values varied between -8.66% (Ribera x Lida) and 5.16% (Albeni x Beyzade) (Table 8 and 9). Kepildek and Ceyhan (2021) found positive and negative heterosis and heterobeltiosis values in their study.

Broad and narrow-sense heritability was determined as 0.92 and 0.13, respectively (Table 6). The high level of heritability in the narrow sense indicated that the contribution of the environment was high. Considering the importance of non-additive gene effects in the inheritance of protein ratio, it may be more appropriate to start selection in late generations.

Table 8
Heterosis (%) values for investigated traits in full-diallel hybrid set

F_2 Populations	Seed Yield	Hundred Seed Weight	Protein Ration	Protein Yield
Lida x Beyzade	23,51	20,44	1,69**	22,68*
Lida x Ribera	-22,08	0,50	4,05**	4,88
Lida x Albeni	11,33	-19,92	7,17**	-13,78
Lida x Garrafol	4,56	26,32	-2,16**	23,60*
Beyzade x Lida	23,55	15,08	2,47**	18,44
Beyzade x Ribera	16,15	12,53	4,77**	18,16
Beyzade x Albeni	-9,91	3,75	9,54**	13,13
Beyzade x Garrafol	-17,51	80,20**	7,59**	93,20**
Ribera x Lida	5,09	-7,34	-3,92**	-11,05
Ribera x Beyzade	3,35	14,08	5,49**	20,90
Ribera x Albeni	-9,73	-21,28	3,65**	-17,24
Ribera x Garrafol	-13,17	12,68	-3,38**	9,53
Albeni x Lida	-1,20	-10,02	5,77**	-4,02
Albeni x Beyzade	-4,21	5,17	8,25**	13,29
Albeni x Ribera	2,63	-12,57	6,07**	-5,53
Albeni x Garrafol	-20,52	-7,69	9,72**	1,43
Garrafol x Lida	25,54	60,97*	6,06**	71,07**
Garrafol x Beyzade	10,07	40,39	9,24**	53,20**
Garrafol x Ribera	-5,26	53,31	2,35**	57,94**
Garrafol x Albeni	-15,80	42,50	9,39**	56,46**
Mean	0,32	15,46	4,69	21,32

** : significant at 1% level; * : significant at 5% level

3.8. Protein yield

It was determined that the parent values in terms of protein yield were between 4.68 g/plant (Ribera) and 7.37 g/plant (Albeni), and the protein yields in the F_2 generation ranged between 4.61 g/plant (Ribera x Lida) and 11.87 g/plant (Beyzade x Garrafol) (Table 6). The data of this research were in harmony with each other (Ülker and Ceyhan 2008b; Varankaya and Ceyhan 2012; Kepildek and Ceyhan 2021).

It had been stated that for protein yield, $\upsilon^2GCA/\upsilon^2SCA$ ratios were less than 1, and $(H/D)^{1/2}$ ratio was more than 1, non-additive gene effect and superior dominance were effectively found in the inheritance of protein yield trait (Table 6). Studies have also determined that the non-additive gene effect was effective in the inheritance of protein yield (Ceyhan et al., 2014a; Kepildek and Ceyhan, 2021).

When the GCA value was examined, it was determined that the Garrafol variety, which had a 5% positive and significant GCA value, had parents that can be preferred in breeding studies to be made in terms of protein yield. When the SCA values of the hybrids were examined, it was determined that the "Beyzade x Garrafol" hybrid, which showed a positive and significant SCC value at the 5% level, is the genotype that can be preferred in breeding studies in terms of protein yield (Table 7). Ceyhan et al. (2014a), and Kepildek and Ceyhan (2021) found the effect of GCA and SCA important in their studies.

Table 9
Heterobeltiosis (%) values for investigated traits in full-diallel hybrid set

F_2 Populations	Seed Yield	Hundred Seed Weight	Protein Ration	Protein Yield
Lida x Beyzade	10,41	10,72	-3,95*	18,94
Lida x Ribera	-32,72**	-7,65	3,55*	-4,34
Lida x Albeni	0,14	-33,25*	-0,11	-23,66
Lida x Garrafol	-17,65*	17,71	-4,52*	18,11
Beyzade x Lida	10,45	5,79	-3,21	14,82
Beyzade x Ribera	11,64	-4,21	-0,58	4,80
Beyzade x Albeni	-10,54	-6,86	8,01**	3,00
Beyzade x Garrafol	-28,65**	77,60**	4,04*	90,34**
Ribera x Lida	-9,27	-14,86	-4,39*	-18,87
Ribera x Beyzade	-0,67	-2,89	0,10	7,24
Ribera x Albeni	-13,82	-38,64*	-2,95	-32,33*
Ribera x Garrafol	-22,22**	-2,90	-5,25**	-4,10
Albeni x Lida	-11,13	-25,00	-1,41	-15,02
Albeni x Beyzade	-4,88	-5,59	6,74**	3,14
Albeni x Ribera	-2,02	-31,86*	-0,68	-22,76
Albeni x Garrafol	-31,66**	-18,19	4,67*	-6,39
Garrafol x Lida	-1,12	49,99*	3,51*	63,48**
Garrafol x Beyzade	-4,79	38,36*	5,64**	50,93*
Garrafol x Ribera	-15,14	32,11	0,37	38,28*
Garrafol x Albeni	-27,59**	26,28	4,36*	44,39*
Mean	-10,06	3,33	0,70	11,750

** : significant at 1% level; * : significant at 5% level

The mean heterosis value was 21.32% and the mean heterobeltiosis value was 11.75%. Heterosis values were found to be between -17.24% (Ribera x Allure) and 93.20% (Beyzade x Garrafol), and heterobeltiosis values were found to be between -32.33% (Ribera x Albeni) and 90.34% (Beyzade x Garrafol) (Table 8 and 9).

Broad and narrow sense heritability was found to be 0.66 and 0.25, respectively (Table 6). The fact that the heritability was low in the narrow sense indicated that the contribution of the environment was highly effective in the emergence of the protein yield feature. Considering the importance of non-additive gene effects in the inheritance of protein yield, selection should be started in later generations.

4. Conclusions

As a result of this research, a sufficient level of genetic variation has been determined in the population when we consider it in terms of the evaluated agricultural characteristics. Non-additive genes and dominant genes were determined to be more effective in terms of the features evaluated in this study. Evaluating the selection based on seed yield in this population and starting it in future generations will increase the level of success.

5. Acknowledgements

This research was supported by the Selcuk University BAP project, number 21211002. This paper was summarized from Ph. D. Thesis of the first author (Şemsi TAMÜSEK). The authors would like to acknowledge the financial support of Selcuk University.

6. References

Al-Mukhtar FA, Coyne DP (1981). Inheritance and association of flower ovule, seed, pod and maturity characters in dry edible beans (*Phaseolus vulgaris* L.). *Journal of the American Society for Horticultural Science* 106(6): 713-719.

Arunga EE, Van Rheenen HA, Owuoche JO (2010). Diallel analysis of snap bean (*Phaseolus Vulgaris* L.), varieties for important traits. *African Journal of Agricultural Research* 5(15): 1951-1957.

Barelli MAA, Goncalves Vidigal MC, Amaral Junior ATD, Vidigal Filho PS, Scapim CA, Sagrilo E (2000). Diallel analysis for grain yield and yield components in *Phaseolus vulgaris* L.. *Acta Scientiarum* 22(4): 883-887.

Bozoğlu H (1995). Kuru fasulyede (*Phaseolus vulgaris* L.) bazı tarımsal özelliklerin genotip x çevre interaksiyonu ve kalıtım derecelerinin belirlenmesi üzerine bir araştırma. Doktora Tezi, Ondokuz Mayıs Üniversitesi Fen Bilimleri Enstitüsü, Samsun.

Ceyhan E (2004a). Bezelye ebeveyn ve melezlerinde bazı tarımsal özelliklerin ve kalıtımlarının çoklu dizi analiz metoduyla belirlenmesi. Doktora Tezi, *Selçuk Üniversites*i *Fen Bilimleri Enstitüsü*, Konya.

Ceyhan E (2004b). Effects of sowing dates on some yield components and yield of dry bean (*Phaseolus Vulgaris* L.) cultivars. *Turkish Journal of Field Crops* 9(2): 87-95.

Ceyhan E (2006). Varitaions in grain properties of dry bean (*Phaseolus vulgaris* L.). *International Journal of Agricultural Research* 1(2): 116-121.

Ceyhan E, Harmankaya M, Kahraman A (2014a). Combining ability and heterosis for concentration of mineral elements and protein in common bean (*Phaseolus vulgaris* L.). *Turkish Journal of Agriculture and Forestry* 38(5): 581-590.

Ceyhan E, Kahraman A, Avcı MA, Dalgıç H (2014b). Combining ability of bean genotypes estimated by line x tester analysis under highly-calcareous soils. *The Journal of Animal and Plant Sciences* 24(29): 579-584.

Ceyhan E, Şimşek D (2021). Fasulyede tarımsal özelliklerin kalıtımlarının çoklu dizi analiz metoduyla belirlenmesi. *Türk Tarım ve Doğa Bilimleri Dergisi* 8(1): 215-225.

Chung JH, Stevenson E (1973). Diallel analysis of the genetic variation in some quantitative traits in dry beans. *New Zealand Journal of Agricultural Research* 16 (2): 223-231.

Falconer DS (1980). Introduction to Quantitative Genetics. London. Oliver and Boyd Ltd, 365.

FAO (2022). Production and trade statistics. https://www.fao.org/faostat/en/#home(02.06.2022).

Genchev D (1995). Assessment of tolerance to stress factors in breeding material of kidney beans (*Phaseolus vulgaris* L.). *Bulgarian Journal of Agricultural Science* 1(4): 415-422.

Griffing B (1956). Concept of general and specific combining ability relation to diallel crossing systems. *Australian Journal of Biological Sciences* 9: 463-493.

Iqbal AM, Nehvi F, Wani SA, Qadri H, Dar Z, Lone AA (2010). Combining ability studies over environments in Rajmash (*Phaseolus vulgaris* L.) in Jammu and Kashmir, India. *Journal of Plant Breeding and Crop Science* 2 (11): 333-338.

Kepildek R, Ceyhan E (2021). Determination of some agronomic traits of fresh bean parents and hybrids and their heritability with diallel analysis method. *Selcuk Journal of Agriculture and Food Sciences* 35 (2): 71-82.

Kwaye R, Hussein S, Mashela WP (2008). Combining ability analysis and association of yield and yield components among selected cowpea lines. *Euphytica* 162 (2): 205-210.

Mutari B, Sibiya J, Gasura E, Matova PM, Simango K, Kondwakwenda A (2022). Genetic analysis of grain yield and yield-attributing traits in navy bean (*Phaseolus vulgaris* L.) under drought stress. *Euphytica* 218 (5): 1-20.

Oliveira Junior A, Miranda GV, Cruz CD (1997). Evaluation of the combining ability of dry bean cultivars based on unbalanced circulating and partial diallel crossing systems. *Revista Ceres* 44(252): 215-229.

Açıkgöz N, Özcan K (1999). TARPOPGEN: Populasyon Genetiği için bir istatistik paket programı. 3. *Ulusal Tarımda Bilgisayar Uygulamaları Simpozyum bildirisi*, 28-30.

Rodrigues R, Leal NR, Pereira MG (1998). Diallel analysis of six agronomic traits in *Phaseolus vulgaris* L. *Bragantia* 57(2): 241-250.

Senbetay T, Tesfaye A, Jimma E (2015). Diallel analysis of white pea bean (*Phaseolus vulgaris* L.) varieties for yield and yield components. *Journal of Biology, Agriculture and Healthcare* 5(5): 75-81.

Singh SP, Urrea CA (1994). Selection for seed yield and other traits among early generations of intra- and interracial populations of the common bean, Brasileira de Genetica 17(3): 299-303.

Turhan Ş (2005). Tarnmda sürdürülebilirlik ve organik tarım. *Tarım Ekonomisi Dergisi* 11 (1 ve 2): 13-24.

Ülker M, Ceyhan E (2008a). Orta Anadolu ekolojik şartlarında yetiştirilen fasulye (*Phaseolus vulgaris* L.) genotiplerinin bazı tarımsal özelliklerinin belirlenmesi. *Selçuk Üniversitesi Ziraat Fakültesi Dergisi* 22(46): 77-89.

Ülker M, Ceyhan E (2008b). Orta Anadolu ekolojik şartlarında yetiştirilen fasulye (*Phaseolus vulgaris* L.) genotiplerinin protein ve bazı mineral oranlarının belirlenmesi. *Selcuk Journal of Agriculture and Food Sciences* 22 (46): 90-97.

Varankaya S, Ceyhan E (2012). Yozgat ekolojik şartlarında yetiştirilen fasulye (*Phaseolus vulgaris* L.) genotiplerinin bazı tarımsal özelliklerinin belirlenmesi. *Selçuk Tarım ve Gıda Bilimleri Dergisi* 26(1): 27-33.

Zimmerman MJO, Rosielle AA, Foster KW, Waines JG (1985). Gene action for grain yield and harvest index of common bean grown as sole crop and in intercrop with maize. *Field Crops Research* 12: 319-329.

Selcuk Journal of Agriculture and Food Sciences

SJAFS

http://sjafs.selcuk.edu.tr/sjafs/index

(2022) 36 (3), 342-348

e-ISSN: 2458-8377

Research Article

DOI:10.15316/SJAFS.2022.043

Pharmacognostic, Physicochemical, Phytochemical Screening and *In-Vitro* Antioxidant Activity of *Chrysochamela Noeana*, Endemic in Türkiye

Nuraniye ERUYGUR[1,*], Mehmet TEKIN[2], Erol DÖNMEZ[3]

[1]Selçuk University, Faculty of Pharmacy, Department of Pharmacognosy, Konya, Türkiye
[2] Trakya University, Faculty of Pharmacy, Department of Pharmaceutical Botany, Edirne, Türkiye
[3]Sivas Cumhuriyet University, Faculty of Science, Department of Biology, Sivas, Türkiye

ARTICLE INFO

Article history:
Received date: 06.04.2022
Accepted date: 23.08.2022

Keywords:
Chrysochamela noeana
Cruciferae
Phytochemical screening
Physicochemical
Antioxidant

ABSTRACT

The aim of the present study was to conduct preliminary phytochemical screening, pharmacognostical and physicochemical investigation of *Chrysochamela noeana*. The fresh and dried herbs were studied by morphology, microscopy, preliminary phytochemical screening and florescence analysis of powdered drug. In addition, physicochemical parameters were studied according to WHO guidelines. Physicochemical parameters and florescence analysis were also performed. The preliminary phytochemical analysis revealed the presence of steroids, flavonoids, phenolic compounds, and carbohydrates. Total phenol contents were 245 ± 3.67, 179.06 ± 4.52, and 15.98 ± 3.32 (GAE mg/g extract) and total flavonoid content were 180.85 ± 8.21, 146.41 ± 2.56, and 13.46 ± 0.23 (QUE mg/g extract) for ethyl acetate, methanol, and water extracts, respectively. The IC_{50} value for extracts in ABTS radical scavenging activity were calculated in order of 51.47, 122.26 and 1146.91 µg/mL, and in DPPH for 1.55, 1.13 and 27.16 mg/mL for ethyl acetate, methanol, and water extracts, respectively. The results of these studies could be useful for correct identification and detection of adulterants from this plant material.

1. Introduction

Microscopic characterization of medicinal plants is very important procedure for identity and quality assessment of herbal ingredients (Rajan et al 2011). It is well accepted by all national and international regulatory authorities as one of the four primary methodologies for the identification of crude drug materials including macroscopic appearance, organoleptic characters, microscopic characteristics and the phytochemical analysis (Manayi et al 2012).

The family Brassicaceae (= *Cruciferae*) consists of 350 genera and about 3500 species (Cartea et al 2011). This family includes important oilseed, forage, condiment and vegetable crops, are consumed by wild population from all over the world. The wild usage and beneficial effect of *Cruciferae* plants may be attributed to their phenolic compounds and flavonoids which are possessing antioxidant activity (Pereira et al 2009). Flavonoids are the most commonly found phenolic compounds, playing an important role in UV protection, pigmentation, and disease resistance (Crozier et al 2006).

The genus *Chrysochamela* (Fenzl) Boiss., includs four species, which are distributed in Turkey, Russia, Lebanon and Syria (Appel and Al-Shehbaz 2003). In flora of Turkey, *Chrysochamela* Boiss. species which has only three varieties so far identified, including *C. elliptica* (Boiss.) Boiss., *C. velutina* (DC.) Boiss. and *C. noeana* (Boiss.) Boiss (Sevindik et al 2019). Among them, two of these species are endemic to Turkey and show a very narrow spread. The third species is only spreads in the Syrian Desert except for Turkey (Davis 1965). *Chrysochamela noeana* (Boiss.) Boiss. (Brassicaceae) is an annual herb only grown in Sivas province of Turkey. The micromorphological and anatomical properties of three species of the *Chrysochamela* genus have been comperatively presented in a former study (Çakilcioğlu et al 2017). Literature review revealed that there is no report about the phytochemical investigations and biological activity studies except for phylogenetic and floristic studies. Despite the vast variety of the Brassicaceae family, only a few species, mostly from the Brassica genus, are consumed. The Brassicaceae plant are high in vitamins, minerals, and fiber while being low in fat. They are also rich in phytochemicals like as isothiocyanates and

* Corresponding author email: *nuraniye.eruygur@selcuk.edu.tr*

phenolic compounds, which play an essential role in the prevention of chronic illnesses (Fusari et al 2020).

Pharmacognosic investigation have not been reported for this plant. Therefore, the aim of the present work is to study the macro, microscopic and some other physicochemical and pharmacognostic characters of *C. noeana*, which could be used to prepare a monograph for the suitable identification of the plant

2. Materials and Methods

The plant materials of *C. noeana* were collected from Sivas (B6 Sivas: Center, gypsum areas in Seyfe Beli location, 1350-1400m, 1.5.2018) and identified by the Botanist XXX SJAFS. Each part of the plant dried under shade and powdered by laboratory-type blender. All the chemicals and reagents were analytical grade, obtained from Sigma chemical co. (St Türkiye) and Merck (Darmstadt, Germany)

Figure 1
Habitat image of *Chrysochamela noeana* (Photographed by E. Dönmez)

Pharmacognostic studies: Each plant parts have been morphologically observed by a microscope (Leica S6E) with magnifying lens for shape, size, surface characteristics, color, and odor etc. For anatomical investigation, parts of the fresh material were stored in %70 alcohol-water solution. Hand-made cross section of basal leaf, cauline leaf, stem, and root were taken by razor blade, then stained in 1% Alcian blue (Sigma) and 1% Safranin O (Sigma), in a ratio of 3/2. The sections were kept about 5 min in the dye. Semi-permanent slides were mounded using glycerin–gelatin (Tekin & Eruygur 2016). Photomicrographs were taken using Olympus BX51 light microscopy equipped with Olympus DP70 digital camera.

Quantitative investigation: The moisture content, total ash, acid-insoluble ash, water-soluble ash and hexane, chloroform, ethyl acetate, methanol and water extractive values of the powdered samples were determined by the method as described in WHO guidelines (WHO 1998).

Phytochemical screening: The phytochemical investigation of different extracts of *Chrysochamela noeana* herbs was carried out by the standard chemical tests.

Physicochemical investigation: Physicochemical parameters such as moisture content, loss on drying, water soluble ash, acid insoluble ash, and hexane, chloroform, ethyl acetate, methanol and water-soluble extractive values of the powdered samples were determined by the method as described in WHO guidelines (WHO 1998).

Fluorescence analysis**:** Fluorescence study of plant powder was conducted as previously reported procedure. The appropriate amount of powder is placed in a porcelain capsule, then a few drops of freshly prepared reagent is added and then allowed to stand for 2 minutes. Then the fluorescence properties were examined in day light and in the UV cabinet at 254nm and 366nm.

Antioxidant activity: For the DPPH radical scavenging assay, 20 µL of extract diluted appropriately in Dimethyl sulfoxide (DMSO) was mixed with 180 µL of DPPH in methanol (40 µg mL^{-1}) in wells of a 96-well plate. The plate was kept in the dark for 15 min, after which the absorbance of the solution was measured at 540 nm in a Microplate-reader. Appropriate blanks (only include the DMSO with free sample) and standards (quercetin solutions in DMSO) were run simultaneously. Extracts were first tested at a single concentration of 4 mg/mL, and those showing good evidence of antioxidant activity were tested over a range of concentrations to establish the EC_{50} (the concentration reducing DPPH absorbance by 50%) (Clarke et al 2013).

The ABTS radical scavenging activity assay was carried out via the ABTS cation radical decolorization with minor modifications (Chun et al 2005). The samples were prepared in the same procedure as the DPPH assay. The ABTS cation radical was prepared by reacting 7 mM aqueous solution of ABTS (15 mL) with 140 mM potassium persulphate (264 µL). The mixture was allowed to stand in dark at room temperature for 16 h before use. Prior to assay, the ABTS working reagent was diluted with methanol to give an absorbance of 0.70 ± 0.02 at 734 nm and was equilibrated at room temperature. The reaction mixtures in the 96-well plates consisted of 50 mL of sample and 100 µL of ABTS working solution in methanol. The mixture was stirred and left to stand for 10 min in dark, then the absorbance was taken at 734 nm against a blank. All determinations were performed in triplicate. The percentage scavenging effect was calculated as:

Scavenging activity %= $(Ac - A_s) / Ac \times 100\%$

Where Ac is the absorbance of the control (without sample) and As is the absorbance in the presence of the sample

The scavenging ability of the samples was expressed as IC_{50} value, which is the effective concentration at which 50% of ABTS radicals were scavenged. The IC_{50} value was calculated from the scavenging activities (%) versus concentrations of respective sample curve.

The total phenolics content of the extracts was determined by reaction with F-C reagent. Thus, 10 µL of extract diluted appropriately in DMSO was mixed with 100 µL F-C reagent freshly diluted 1/10 with distilled water. After five minutes, the solution was mixed with

100 µL 7.5% Na_2CO_3 solution, and the whole left for 60 min, before measurement of absorbance at 650 nm in a Multiscan plate-reader. Appropriate blanks (DMSO) and standard (quercetin in DMSO) were run simultaneously, after which the total phenolics content was calculated as mg gallic acid equivalents per g extract (Clarke et al 2013).

Total flavonoid contents were determined by Aluminum colorimetric method, using quercetin as the reference standard. Briefly, 150 µL of the test sample dissolved in ethanol was mixed with equal volume of 2% (w/v) $AlCl_3$ in 96-well plates. After 15 min of incubation at 25ºC, the absorbance was measured at 435 nm by spectrometer. All determinations were performed in triplicates. The content of total flavonoids was expressed as mg of quercetin equivalent per g of dry weight of the sample, using an equation obtained from the standard quercetin calibration (Jhade et al 2011; Yang et al 2011).

The chelation of ferrous ions by extracts was estimated by method of Dinis et al. (1994) and Ebrahimzadeh et al., (2008). Briefly, 50 µL of 2 mM $FeCl_2$ was added to 1 mL of different concentrations of the extract (0.2, 0.4, 0.8, 1.6 and 3.2 mg mL^{-1}). The reaction was initiated by the addition of 0.2 mL of 5 mM ferrozine solution. The mixture was vigorously shaken and left to stand at room temperature for 10 min. The absorbance of the solution was thereafter measured at 562 nm. The percentage inhibition of ferrozine–Fe^{2+} complex formation was calculated as $[(A_0- A_s)/ A_s] \times 100$, where A_0 was the absorbance of the control, and A_s was the absorbance of the extract/ standard. EDTA was used as positive control.

3. Results and Discussion

Macroscopic characteristics: The basal leaves are 0.5-3.5mm in length and 0.3-1.2mm in width. The leaf shape broadly or narrowly ovate, apex obtuse or acute, margin is entire, venation is pinnate, green above and beneath, leaf surface is covered with branched hairs or glabrous, leaf base is sessile, phyllotaxis is rosette. The stem leaves were 5-20mm in length and 3-5mm in width. The leaf is oblong-shaped, apex are acute to subobtuse, margin entire, venation pinnate, the bottom and top of leaf is green, leaf surface glabrous, leaf base is sessile and auriculate, phyllotaxy was alternate, have mustards' odor (Figure 2. a). Sepals are ovate, 1-1.5 × 0.4-0.6mm, entire or scarious, apex acute, bifurcate hairy. Petals are 1-1.5 × 0.4-0.6 mm, usually ovate-shaped, entire, obtuse, and glabrous. Inflorescence is zigzag, and the branches are glabrous or pubescent. The number of stamen is 6. Filaments are ascendens, adhered to the base and expanded in the lower part. Ovary elliptic, stilus is short, 0.1-0.15mm. Fruits are latiseptate siliqua, dehiscent, obovoid, outer side slightly reticulate, 3.5-4.5 × 1.8-2.5 mm, fruit stilus are 0.4-0.5 mm, reptum are 3.5-4.5mm. Seeds are 0.5-0.3mm.

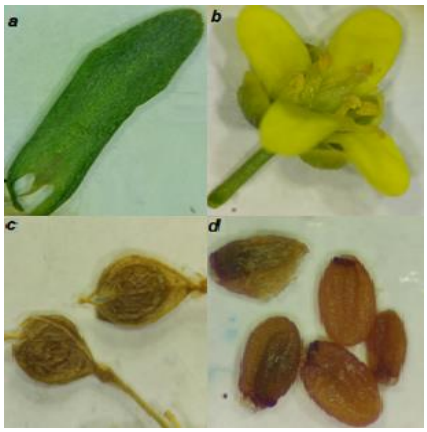


Figure 2

Macroscopic characteristics of *Chrysochamela noeana* leaf (a), flower (b), fruits (c) and seeds (d) (Photographed by E. Dönmez)

Anatomical Characteristics of roots: Transverse section of root is round shaped and in secondary structure. Outermost layer consists of 1–3 layered phellem cells. The phellem cells are 15–45 × 5–23 µm, depressed and sometimes fall broken. Phellogen is indistinguishable. There is 3–6 layered parenchymatic cortex under the periderm. Cortex cells are 15–63 × 5–38 µm, slightly depressed rectangular, oval, or irregular shaped and there are intercellular gaps between these cells. There is stele beneath the cortex. Phloem is located narrow, and xylem is located wide area of root cross section in stele. Cambium is indistinguishable between phloem and xylem. Xylem is composed of lignified xylem elements in close to cortex and unlignified xylem element in and around pith (Fig. 3 A, B).

Anatomical Characteristics of Stem: In the cross section of the stem, the epidermis is single layered, and cells have thin cuticle. The epidermis cells are 7–18 × 4–15 µm, squarish, rectangular, or oval shaped. The parenchymatous cortex with chloroplast is made up of 5–8 layers and placed just beneath epidermis. Cortex cells are 10–58 × 7–23 µm and oval or rounded shaped. The endodermis, innermost layer of the cortex, is distinct by having larger cells and thicker cell wall than other cortex cells. There is 7–14 layered pericycle which consist of sclerenchymatic fibers beneath endosperm. The stem has collateral type vascular bundles. Phloem is located between endodermis and sclerenchymatic fibers, and xylem is located in and under sclerenchymatic fibers surrounded by sclerenchyma fibers. The vascular cambium is indistinguishable between xylem and phloem. The pith is occupied by large, oval and thick-walled parenchyma cells and these cells are 12–63 × 5–50 µm in diameter (Fig. 3 C, D).

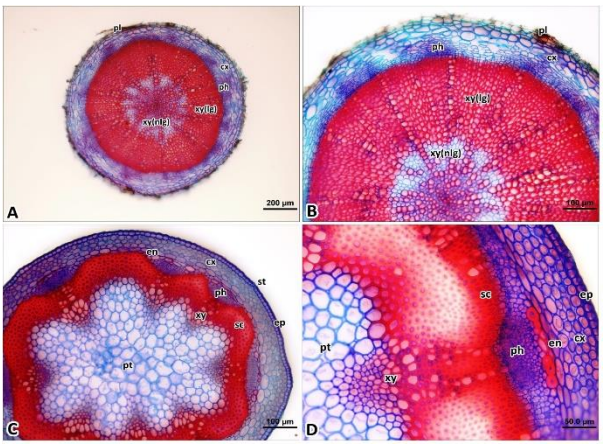


Figure 3

Photomicrographs of root cross section (A, B) and stem cross section (C, D) of *Chrysochamela noeana*. Abreviations: cx: cortex, en: endodermis, ep: epidermis, ph: phloem, pl: phellem, pt: pith region, sc: sclerenchymatic fibers, st: stoma, xy: xylem, xy(lg): lignified xylem, xy(nlg): non-lignified xylem.

Anatomical Characteristics of basal leaf: In cross section of basal leaf, adaxial and abaxial epidermises are uniseriate. The stomata are present on both epidermises. The stoma cells are located on the same level with the epidermal cells. Adaxial epidermis cells are 12–68 × 10–40 µm, oval or rectangular oval occasionally sguarish oval shaped. The mesophyll is bifacial and has 3–4 cell layers of palisade parenchyma below adaxial epidermis and 4–5 layers of spongy parenchyma below abaxial epidermis. Palisade parenchyma cells are 25–63 × 15–30 µm, and usually cylindrical, occasionally squarish shaped. Spongy parenchyma cells are 10–50 × 7–40 µm, usually oval, rarely roundish, or rectangular oval shaped. Mesophyll thickness is 180–230 µm. The midrib is obviously larger than the other bundles. Abaxial epidermis cells are 10-43 × 7-30 µm, oval or rectangular oval occasionally squarish oval shaped. (Fig 4 A, B).

Anatomical Characteristics of cauline leaf: In cross section of cauline leaf, adaxial and abaxial epidermises are uniseriate. The stomata are present on both epidermises. The stoma cells are mesomorphic. Adaxial epidermis cells are 7–43 × 7–28 µm, usually depressed rectangular, occasionally squarish shaped. The mesophyll is bifacial and has 3–4 cell layers of palisade parenchyma below adaxial epidermis and 4–5 layers of spongy parenchyma below abaxial epidermis. Palisade parenchyma cells are 22–58 × 17–30 µm, and usually cylindrical or oval rarely squarish shaped. Spongy parenchyma cells are 20–68 × 15–48 µm oval or rounded shaped. Mesophyll thickness is 210–300 µm. The midrib is obviously larger than the other bundles. Abaxial epidermis cells are 12–45 × 10–33 µm, oval or rectangular oval occasionally squarish oval shaped (Fig. 4 C, D).

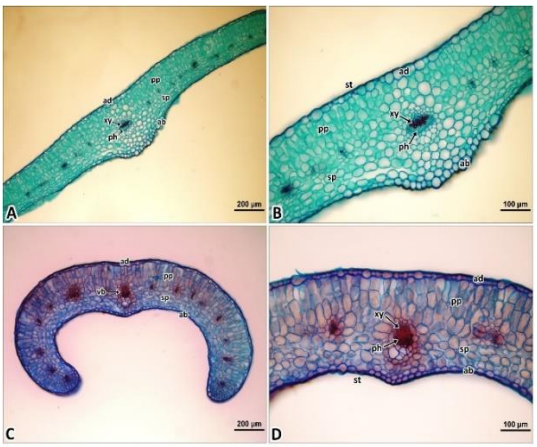


Figure 4

Photomicrographs of basal leaf cross section (A, B) and cauline leaf cross section (C, D) of *Chrysochamela noeana*. Abreviations: ab: abaxial epidermis, ad: adaxial epidermis ph: phloem, pp: palisade parenchyma, sp: spongy parenchyma, st: stoma, xy: xylem.

The physico-chemical evaluation of drugs is play an important role in detecting adulteration or improper handling of drugs. The total ash is indication of impurities in herbal drugs, to detect the presence or absence of inorganic components mixed with plant during harvesting coming from soil. The amount of moisture content effecting the stability of herbal drugs, it means that the lower moisture content, the lower proliferation of living microorganisms, therefore, the higher stability of the drug. Loos on drying is more used for detecting moisture content among the methods. The loose on drying value of the plant is not high, thus it is easy to storage or handling of the drug and lower than the general requirement for moisture content in drugs described in herbal pharmacopoeia. The total ash value is 18.7% (w/w), acid – insoluble ash value is 1.90% (w/w), most of the total ash is soluble in acid, therefore acid –insoluble ash value is low. In addition, the water-soluble extractive is higher in this plant, water extractive value is 8.75% (w/w) while other solvent extractive value is low, hexane, chloroform, ethyl acetate, methanol extractive value is, 0.65% (w/w), 1.03% (w/w), 0.38% (w/w), and 3.37 % (w/w), respectively. The results are presented in Table 1.

Table 1
Proximate parameters of *Chrysochamela noeana* herbs

Parameters	Value %
Loss on drying	5.2 ± 0.025
Total ash	18.7 ± 0.033
Acid insoluble ash	1.90 ± 0.020
Water soluble ash	3.72 ± 0.031
Hexane soluble extractive	0.65
Chloroform soluble extractive	1.03
Ethyl acetate soluble extractive	0.38
Methanol soluble extractive	3.37
Water soluble extractive	8.75

The results of fluorescence analysis of the entire plant powder of *C. noenana* are presented in Table 2, the fluorescence in day light, UV light (254 nm), and UV

light (365 nm) are different from each other when various chemical reagents are added. It indicates that there are various phytochemical groups existing in the plant.

Preliminary phytochemical screening: A different polarity solvent extracts namely hexane, chloroform, Ethyl acetate, methanol and water extracts prepared from herbs of *C. noeana* were investigated for their phytochemical profile. The extracts were subjected to various qualitative chemical tests for the identification of various plant constituents. The herbs show the presence of carbohydrates, steroid, flavonoid, phenolic compounds (Table 3).

Table 2
Fluorescence analysis of aerial part powder of *Chrysochamela noeana*

Reagents	Day light	UV light (254 nm)	UV light (365 nm)
Powder	Light yellow	Light yellow	Yellow
Powder + Water	Whitish yellow	Yellowish green	Light yellow
Powder + Methanol	Whitish yellow	Dark green	Bluish green
Powder + chloroform	Whitish yellow	Bluish yellow	Golden yellow
Powder + n-hexane	Whitish yellow	Bluish brown	Light yellow
Powder + CCI_4	Whitish yellow	Bluish green	Golden yellow
Powder + Xylene	Brown	Bluish brown	Whitish blue
Powder + Conc. Sulfuric acid	Dark Coffee	Golden yellow	Golden yellow
Powder + dil. Sulfuric acid	Pink	Whitish bluish green	Whitish blue
Powder + Conc. Hydrochloric acid	Yellowish orange	Brown	Dark yellow
Powder + dil. Hydrochloric acid	Pink	Greenish yellow	Whitish greenish yellow
Powder + Conc. Nitric acid	Reddish orange	Dark brown	Greenish brown
Powder + dil. Nitric acid	Light orange	Dark green	Dark green
Powder + Acetic acid	Light yellow	Whitish green	Whitish blue
Powder + Picric acid	Green	Greenish blue	Greenish brown
Powder + 1N NaOH	Greenish yellow	Brown	Greenish yellow
Powder + Ammonia	Green	Brownish yellow	Gold yellow
Powder + %5 Iodine	Dark	Brown	Dark brown
Powder + %5 $FeCI_3$	Dark green	Dark green	Dark green

Table 1
Phytochemical investigations of various extracts prepared from *Chrysochamela noeana**

Compounds	Tests	Hexane	Chloroform	Ethyl acetate	Methanol	Water
Alkaloids	Mayer	-	-	-	-	-
	Marquis	-	-	-	-	-
	Dragendorff	-	-	-	-	-
Carbohydrates	Molisch	+	+	+ +	+ + +	+
Saponins	Foam test	-	-	-	-	-
Coumarins	NaOH+ UV	-	+	-	-	-
Flavonoids	NaOH	-	+	+	-	-
	Shinoda	-	+ +	+	-	-
Lipids		-	-	-	-	-
Tannins, Phenolics	$FeCI_3$	+ + +	+ +	+	-	-
Anthraquinones	Borntrager's	-	-	-	-	-
Volatile oil	Sudan III	+	+	-	-	-
Protein	Ninhydrin	-	-	-	-	-
Steroids	Lieberman	-	+	+	+	-

* (+) means positive results, (++) is strongly positive results, (-) means negative results

Antioxidant activity: Figure 4 shows the dose-response curve of DPPH radical scavenging activity of the three different extracts of aerial parts of *C. noeana*, compared with quercetin. It was observed that the ethyl acetate and methanol extract have higher activity than water extract, the scavenging activity of the methanol extract was higher than ethyl acetate extract at the same concentration. However, ethyl acetate extract was more effective in ABTS radical scavenging than methanol extract, its activity was comparable with Trolox, water extract was lower than other extract. Results obtained in the present work revealed that the level of these phenolic compounds in ethyl acetate and methanol extract were considerable than water extract of *C. noeana*. In addition, the same extracts possessed higher level of total flavonoids (Figure 7). Excessive free irons have been implicated in biological system; it is used for antioxidant evaluation of plant extracts. In metal chelating assay, the extracts were tested in the concentration range of 10-1000 µg/mL, ethyl acetate and methanol extracts demonstrated strong chelating activities in concentration- dependent manners, while the lowest activity was detected in water extract.

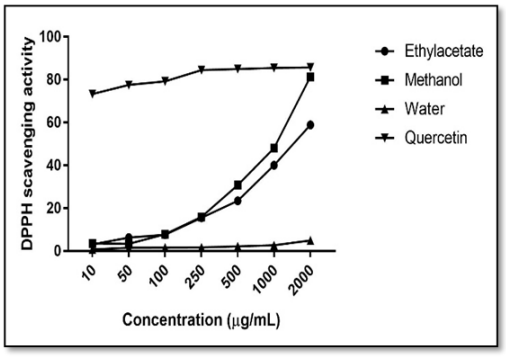


Figure 5
DPPH scavenging activity of different extracts of aerial parts of *C. noeana* and standard quercetin

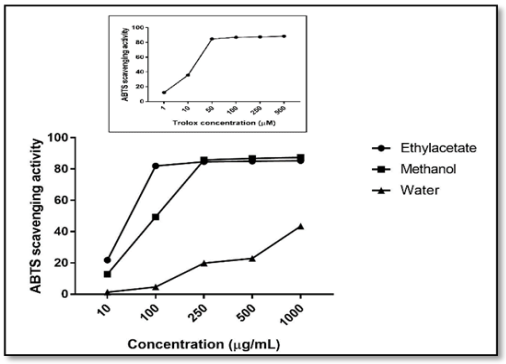


Figure 6
ABTS radical scavenging activity of different extracts of aerial parts of *C. noeana* and standard Trolox

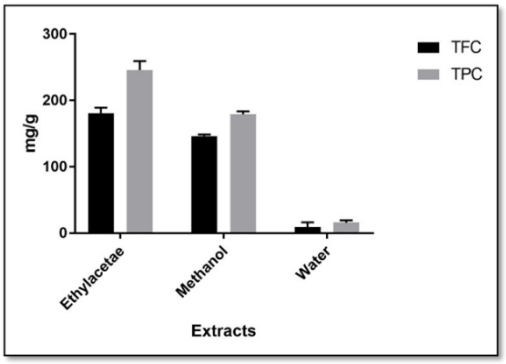


Figure 7
Total flavonoid (TFC) and total phenol content (TPC) of different extracts of aerial parts of *C. noeana*

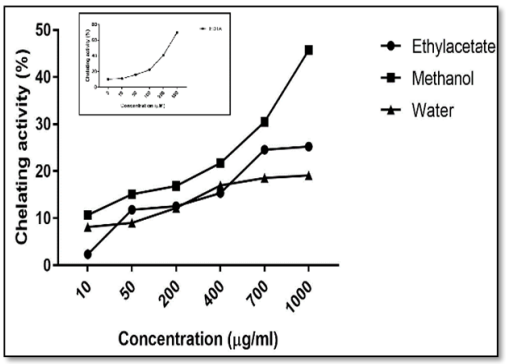


Figure 8
Metal chelating activity of extract prepared from herbs of *C. noeana*

The present study on the physico- and phytochemical investigation and pharmacognostic standardization of the *C. noeana* herbs can be useful to provide information in terms of its identification parameters. They can be used as proper quality control measures to ensure the quality, safety, and efficacy of this herbal drugs as well as may be helpful to preparation of monograph and herbal pharmacopeia standards of this plant.

The macroscopic and microscopic examinations revealed crucial traits that would aid in the identification of this plant. They are assumed significant for the acceptability of herbal drugs in the present case in Türkiye, which lacks regulatory laws to control the quality of herbal drugs. To the best of our knowledge, this is first study of its kind on *C. noeana*, therefore, it will be important to review and research on this plant. Despite the aforementioned research, more studies in phytochemical separation and in vitro screening in different enzymes and cell lines are needed to investigate the pharmacology of the uncommon growing indigenous species.

4. Acknowledgements

This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

5. References

Appel O and Al-Shehbaz IA (2003). Cruciferae. In: Kubitzki, K. (Ed.). The Families and genera of Vascular Plants, Springer, Berlin Heidelberg, pp. 75-174.

Cartea ME, Francisco M, Soengas P, Velasco P (2011). Phenolic compounds in Brassica Vegetables. *Molecules*, Vol; 16, 251-280. doi:10.3390/molecules16010251

Çakilcioğlu U, Paksoy MY, Babacan E and Polat R (2017). Comparative micromorphology and anatomy of *Chrysochamela* (Fenzl) Boiss.(Cruciferae) growing in Turkey. *Bangladesh J. Bot*. 2017; 46(2), 549-558.

Chun SS, Vattem DA, Lin YT, Shetty K (2005). Phenolic antioxidants from clonal oregano (*Origanum vulgare*) with antimicrobial activity against Helicobacter pylori. *Process Biochem*.2005; 40, 809-816.

Clarke G, Ting KN, Wiart C, Fry J (2013). High Correlation of 2,2-diphenyl-1-picrylhydrazyl (DPPH) Radical Scavenging, Ferric Reducing Activity Potential and Total Phenolics Content Indicates Redundancy in Use of All Three Assays to Screen for Antioxidant Activity of Extracts of Plants from the Malaysian Rainforest. *Antioxidants* 2013; 2, 1-10.

Crozier A, Clifford M, Ashihara H, Eds (2006). Blackwell: Plant Secondary Metabolites: Occurrence, Structure and Role in the Human Diet; Oxford, UK, 2006; pp. 1-24.

Davis PH (1965). Flora of Turkey and the East Aegean Islands (Vol:1). Edinburgh: Edinburgh University Press, pp.494-495.

Dinis TCP, Madeira VMC, Almeida MLM (1994). Action of phenolic derivates (acetoaminophen, salycilate and 5-aminosalycilate) as inhibitors of membrane lipid peroxidation and as peroxyl radical scavengers. *Arch. Biochem. Biophys.* 1994; 315:161-169.

Ebrahimzadeh MA, Pourmorad F, Bekhradnia AR (2008). Iron chelating activity, phenol and flavonoid content of some medicinal plants from Iran. *African Journal of Biotechnology* 2008; 7 (18): 3188-3192.

Fusari C M, Nazareno MA, Locatelli DA, Fontana A, Beretta V and Camargo AB (2020). Phytochemical profile and functionality of Brassicaceae species. *Food Bioscience* 2020; 36, 100606.

Jhade D, Ahirwar D, Jain R, Sharma N, Gupta S (2011). Pharmacognostic standardization, physico- and phytochemical evaluation of *Amaranthus spinosus* linn. Root. *J Young Pharm*. 2011; 3 (3): 221-225.

Manayi A, Mirnezami T, Saeidnia S, Ajani Y (2012). Pharmacognostical Evaluation, Phytochemical Analysis and Antioxidant Activity of the Roots of *Achillea tenuifolia* LAM. *Pharmacognosy Journal*, 2012; 4 (30): 14-20.

Pereira DM, Valentao P, Pereira JA, Andrade PB (2009). Phenolics: From Chemistry to Biology. *Molecules* 2009; 14, 2202-2211.

Rajan M, Kishor Kumar V, Satheesh Kumar P, Venkatachalam T, Anbarasan V (2011). Pharmacognostical and phytochemical studies of the leaves of *Albizia Odoratissima* (L.F) Benth. *Int J Pharmacog Phytochem Res*. 2011; 3(3):47-55.

Sevindik E, Paksoy MY, Topseçer F and Aydoğan M (2019). Phylogenetic Analysis of *Chrysochamela* (Fenzl) Boiss.(Brassicaceae) Taxa Growing in Turkey. *Turkish Journal of Agriculture-Food Science and Technology*, 2019; 7(12): 2129-2132.

Tekin M, Eruygur N (2016). Structural studies on the medicinal plant *Haplophyllum telephioides*. *Revista Brasileira de Farmacognosia* 2016; 26: 544-552.

WHO (1998). Quality Control Methods for Medicinal Plant Materials. Geneva: World Health Organization.

Yang H, Dong Y, Du H, Shi H, Peng Y, Li X (2011). Antioxidant Compounds from Propolis Collected in Anhui, China. *Molecules* 2011; 16: 3444-3455.

Selcuk Journal of Agriculture and Food Sciences

http://sjafs.selcuk.edu.tr/sjafs/index

Research Article

SJAFS
(2022) 36 (3), 349-355
e-ISSN: 2458-8377
DOI:10.15316/SJAFS.2022.044

Importance of Priming Application Times on Growth, Relative Water Content and Photosynthetic Pigments of Rapeseed (*Brassica napus* ssp. *oleifera* L.) Cultivars Under Salinity Stress

Mehtap GÜRSOY*
*Aksaray University, Güzelyurt Vocational School, Department of Plant and Animal Production, Aksaray, Türkiye

ARTICLE INFO

Article history:
Received date: 11.05.2022
Accepted date: 23.08.2022

Keywords:
Carotenoid
Chitosan
Chlorophyll
Germination
Time of priming application

ABSTRACT

Environmental stress factors affect plant production more and more every day. One of these stress factors is salinity. The use of biostimulants is increasing day by day and gaining importance in order to reduce the effects of stress factors and increase the yield and quality in plant production. Chitosan (Ch) is one of the biostimulants whose use in agriculture is increasing day by day. Seeds of rapeseed cultivars were used in this study, and it is an important oil plant. In this study, the times of priming applications with Ch (3 times) [0 (control) (Ch1), 12 hour (Ch2), 24 hour (Ch3)] and different doses of salt stress (S) [0 (control) (S1), 50 mM L^{-1} (S2), 100mM L^{-1} (S3)] in rapeseed cultivars (NK Caravel (C1), Elvis (C2), Champlain (C3) under laboratory conditions were investigated. Germination percentage (GP), seedling length (SL), root length (RL), seedling fresh weight (SFW), root fresh weight (RFW), relative water content (RWC), total chlorophyll (Total Chl), carotenoid (Crt) parameters were examined. As a result of the research, with Ch applications, GP (84.67% to 86.67), SL (7.83 cm to 8.12), RL (6.42 cm to 6.50), SFW (0.10 g to 0.53), RFW (0.02 g to 0.06), RWC (62.84% to 63.30), Total Chl (1.60 mg g^{-1} to 1.90), and Crt (1.60 mg g^{-1} to 1.89) has increased. It has been determined that Ch application times play an important role in reducing salt stress in the investigated parameters.

1. Introduction

Salinity is becoming one of the most important stress factor all over the world (Gürsoy 2020; Mushtaq et al. 2021; Gürsoy 2022a). The salinity causes adverse effects of physiological parameters and a decrease in the yield of a crop (Zahra et al. 2018; Iqbal et al. 2019). Crops can be exposed to salt stress at all stages of development from germination to maturity, but stress is known to be more sensitive for many plant species during the germination and early seedling growth phase (Ali et al. 2020). Today, seed priming methods are widely used to increase the tolerance of plant varieties against abiotic stresses (Lal et al. 2018). Seed priming is an alternative, inexpensive and feasible technique as compared with other agronomical applications for mitigate salt stress (Elsiddig et al. 2022). Rapid and uniform germination is of vital importance in plant production, and it can affect the viability of seedlings as well as yield and quality (Palve et al. 2022). Today, the application of some biopolymers such as chitosan, which has many advantages such as safe, cheap and easy production, is widely used all over the world (Hajihashemi and Kazemi 2022). Chitosan is a natural modified from chitins which act as a potential biostimulant in agriculture (Gürsoy 2020; Gürsoy 2022b; Zhang 2022). Application of chitosan in agriculture due to its biodegradability, antimicrobial activity and plant growth promotion, with seed priming plant defense mechanism, chlorophyll content can be increased (Ahmed et al. 2020; Chouhan and Mandal 2021). The application of exogenous chitosan increases plants tolerance to several forms of stress, such as drought, salt, osmotic, and low-temperature stress (Jabeen and Ahmad 2013; Pongprayoon et al. 2013; Li et al. 2017).

Rapeseed is a very important plant in the production of oil crops due to its high oil content and oil quality (Gürsoy and Kolsarıcı, 2017; Gürsoy 2019; Arslan and Culpan, 2022).

The aim of this study is to determine the effect of priming with chitosan at different times on rapeseed cultivars under salt stress conditions on the germination properties, relative water content and photosynthetic pigments of the cultivars.

* Corresponding author email: *mehtapgrsoy@gmail.com*

2. Materials and Methods

The research was carried out at the Aksaray University Scientific and Technological Research Laboratory (ASÜBTAM). Rapeseed cultivars [NK Caravel (C1), Elvis(C2), Champlain (C3)] were used in this study. Before starting the study, the seeds were weighed and then kept in sodium hypochlorite solution for 5 minutes for sterilization. After this process, they were washed several times with distilled water. They were left to dry at room conditions until they reached their initial weight. Uniform and healthy looking seeds (to decrease errors in seed germination) were selected from each of the 3 cultivars and the seeds subjected to in 3 different time periods [0 (control) (Ch1), 12 hour (Ch2), 24 hour (Ch3)] of chitosan priming treatment were applied to rapeseed cultivars at room temperature. Untreated seeds were used as control. For each chitosan priming application, 50 seeds of all varieties were placed in sterile petri dishes on Whatman No:1 blotting papers and 10 ml of different doses of salt [0 (control) (S1), 50 mM L^{-1} (S2), 100mM L^{-1} (S3)] concentrations were added. Only water was added to the control petri dish. In order to prevent evaporation the petri dishes are wrapped with parafilm. The petri dishes were left to germinate at room temperature. Filter papers were changed every 2 days and 10 ml of salt containing solutions were added. The research randomized plots experimental design were made with 3 replication according to the trial pattern. All germination processes were carried out according to ISTA rules (ISTA 2003). In the study; germination percentage (GP), seedling length (SL), root length (RL), seedling fresh weight (SFW), root fresh weight (RFW), relative water content (RWC), total chlorophyll (Total Chl), carotenoid (Crt) parameters were examined.

Germination percentage (%)

Germination% = (number of germinated seeds/total number of seeds)×100 (Siddiqi et al. 2007)

Determination of relative water contents

In order to determine the relative water content in the leaf samples taken from plants belonging to the rapeseed cultivars in the control and stress groups were weighed and their fresh weight was determined, then they were placed in glass tubes containing 5 ml of distilled water and kept in the light for 24 hours. At the end of this period, the hydrated leaf samples were weighed again and their weight in turgor condition was determined. Later, these leaf samples will be dried in the oven at 80°C for 48 hours and their dry weight will be determined again. Finally, the relative water contents will be found according to the formulas below (Ritchie et al. 1990).

RWC(%) = (FW − DW)/(TW − DW) x 100 (Relative water content)

FW: fresh weight, TW: turgor weight, DW: dry weight

Chlorophyll (mg g^{-1})

Fresh samples (0.25g) were taken from the leaves of the rapeseed seedling and homogenized with 80% acetone. It was filtered and made up to 25 ml with acetone. These samples were read in the spectrometer at 663 and 645 nm wavelengths spectrophotometer. Chlorophyll was calculated with the following formula (Lichtenthaler and Welburn 1983).

Chlorophyll a (mg g^{-1}) = (12.7*663 nm)-(2.69*645 nm)*V/W*10000

Chlorophyll b (mg g^{-1}) = (22.91*645 nm)-(4.68*663 nm)*V/W*10000

Total Chlorophyll = Chlorophyll a + Chlorophyll b

V = volume leaf extract in 80% Acetone

W = fresh weight of leaf material

Carotenoid (mg g^{-1})

Fresh samples (0.25g) taken from young leaves of rapeseed seedlings were homogenized in 80% acetone in a place not directly exposed to light, and then filtered. The amount of carotenoid will be determined according to the following formula by completing the obtained filtered extract with acetone to 25 ml and reading it at 450 nm wavelength (Lichtenthaler and Welburn 1983).

Carotenoid (mg g^{-1})= (4.07 x A450-(0.0435 x Chlorophyll a+0.367 x Chlorophyll b)

Statistical analysis

The data obtained in the research were subjected to statistical analysis using the MSTAT-C program. Duncan test was used to describe the degree of significance between the means.

3.Results and Discussion

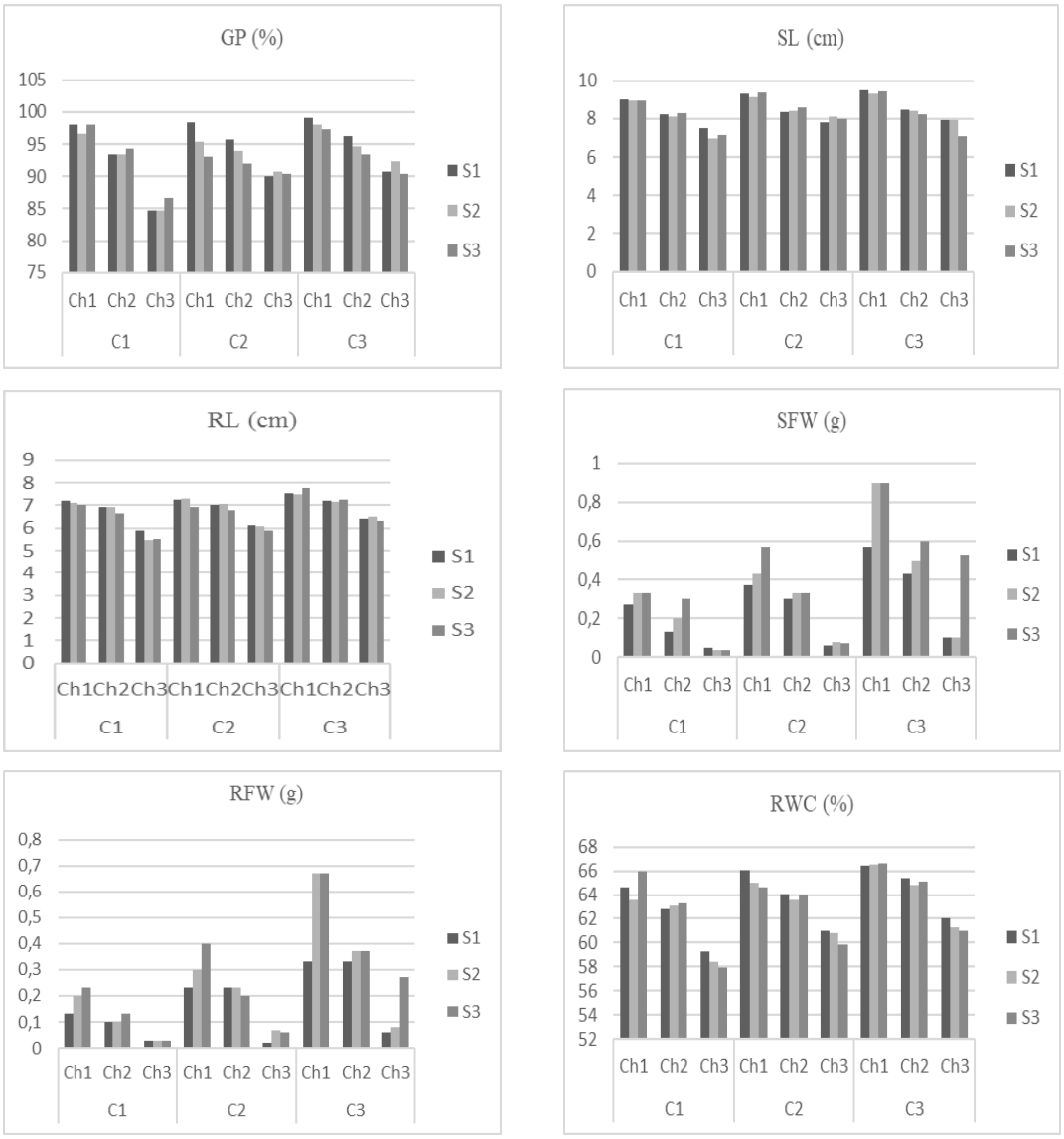
The variance analysis results of this study, which was conducted to determine the effects of chitosan priming application times on the germination parameters, seedling growth, total chlorophyll, relative water content and carotenoid of rapeseed varieties under salt stress, are given in Table 1. When Table 1 is examined, Cultivars × Ch times × S Doses triple interaction is seen to be significant at the level of 5% for root length and 1% for other parameters examined. Besides, Cultivars, Ch times and Salt doses are important at the 1% level. On the other hand, it was determined that the bilateral interactions were statistically significant at the level of 1%. In the RL parameter, the Cultivars × S doses bilateral interaction was found to be statistically insignificant.

Table 1
Analysis of variance on the investigated parameters in rapeseed cultivars of chitosan application times and salt stress

V.S.	D.F.	GP	SL	RL	SFW	RFW	RWC	Total Chl	Crt
		F Value							
Cultivars	2	40.07**	7.83**	10.63**	44.72**	40.07**	8.22**	6.66**	66.36**
Ch times	2	244.04**	112.10**	130.66**	204.09**	244.04**	147.54**	121.16**	330.91**
Cultivars ×Ch times	4	10.14**	17.83**	6.38**	11.15**	10.14**	3.69**	28.60**	21.08**
S Doses	2	331.68**	1441.33**	731.05**	291.82**	331.68**	891.35**	374.07**	100.99**
Cultivars ×S Doses	4	15.44**	11.31**	1.23^{ns}	4.16**	15.44**	6.72**	68.91**	7.47**
Ch times ×S Doses	4	25.13**	21.89**	7.86**	12.61**	25.13**	6.74**	27.85**	59.57**
Cultivars ×Ch times ×S Doses	8	5.84**	9.32**	2.25*	6.81**	5.84**	3.82**	19.17**	11.74**
Error	54	0.53	0.01	0.01	0.004	0.002	0.228	0.001	0.003
CV%		0.78	1.31	1.88	1.54	1.5	0.75	1.62	5.63

**: significance level at $p<0.01$, *: significance level at $p<0.05$. ns: non significant, VS: Variation source, DF: Degrees of Freedom, GP: Germination Percentage, SL: Seedling Length, RL: Root Length, SFW: Seedling Fresh Weight, RFW: Root Fresh Weight, RWC: Relative Water Content, Total Chl: Total Chlorophyll, Crt: Carotenoid

Duncan test results according to variance analysis results are given below as both figures and tables.



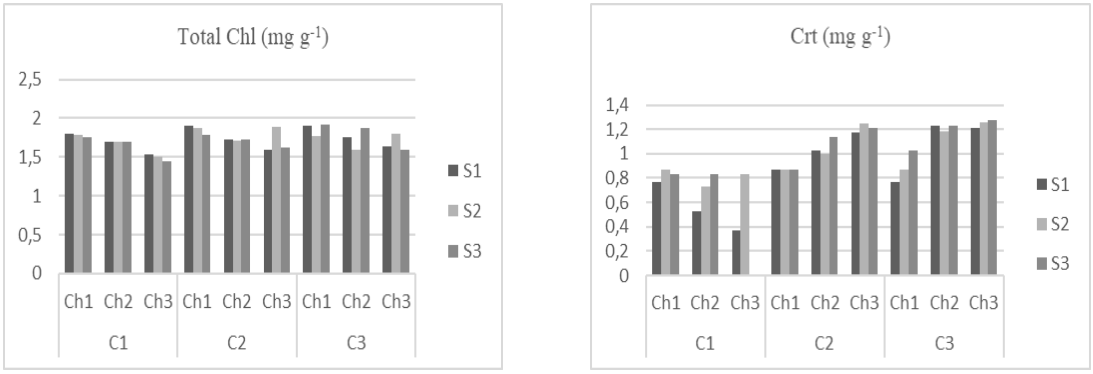


Figure 1
Mean values of germination characteristics, RWC and carotenoid content of rapeseed cultivars under salt stress of different chitosan priming times (triple interaction).

When Figure 1 and Table 1 of the results of the averages is examined, it is seen that the germination is decreased with S applications in the GP feature, but it is seen that Ch priming applications are effective in reducing the negative effects of salt stress. It is seen that the highest germination percentage was obtained from the C3 varieties from the Ch2 priming time application. Guan et al. (2009) reported that chitosan priming resulted in development maize germination and seedling growth under low temperature stress. Mahdavi et al. (2011) reported that germination decreased in high chitosan dose applications in their study in which they applied osmotic stress to safflower seeds. However, they reported that the germination percentage increased up to 0.4% chitosan dose. Hameed et al. (2013) reported that seed priming with chitosan enhanced the germination rate compared with non-primed seeds. Chitosan priming under stress resulted in very developed germination index and decreased germination time to promote early seedling establishment in maize. Jabeen and Ahmad (2013) applied chitosan to safflower and sunflower cultivars under salinity stress and they reported that small dose of chitosan application caused boost in germination parameters of both cultivars. When the SL feature is examined (Figure 1, Table 1), it is seen that the seedling length is prolonged in C1 and C2 varieties in Ch2 application at the S3 dose, where salt stress is the highest. Ch application seems to be effective in suppressing salt stress and increasing seedling height. On the other hand, in the RL feature, it is seen that especially Ch2 application in S2 application is effective in extending the root length (Figure 1). Guan et al. (2009) primed corn seeds with chitosan at low temperatures. As a result of the study, all priming treatments with chitosan significantly increased the shoot height and root length as compared with the control. Sheikha and Al-Malki (2011) chitosan application developed growth characteristics such as shoot and root length in case of bean. Hasanah and Sembiring (2018) found that application of salicylic acid and chitosan to leaves of soybean cultivars increased plant height, seedling, and root dry weights. Bakhoum et al. (2020) applied chitosan to reduce salt stress in sunflower plant. As a result of the study, they determined that chitosan applications increased seedling height, fresh weight and dry weight. When the SFW parameter is examined, it is seen that the effect of Ch application (Ch2 and Ch3) is clearly revealed as the doses of salt stress increase. Therefore, it was determined that chitosan application times were effective in reducing the effect of salt stress. Seraj et al. (2021) applied chitosan and salicylic acid to the seeds of the milk thistle plant under water stress conditions. As a result of the study, they reported that when chitosan applications were compared with the control, especially 200 mg L^{-1}application was important in increasing fresh and dry weight. Zhang et al. (2021) applied chitosan to lettuce seeds under salt stress. At the end of the application, they reported that chitosan increased the seedling fresh weight. Although the salt stress increased in the RFW parameter, it was observed that the root length increased with Ch applications. This is particularly evident in the C3 variety. Even at the highest salt application, root length increased 4 times in Ch3 application compared to control (Figure 1). Sen and Mandal (2016) reported that chitosan application to mung bean plant under salt stress increased the root length with Ch application. Harfoush et al. (2017) reported that the application of humic acid and chitosan to the potato plant caused significant increases in the growth parameters of the plant. It was determined that the RWC parameter (Figure 1) increased at the S3 dose, where the salt stress was the highest, especially in the Ch2 application. Abdelaal et al. (2021) reported that the RWC increased by 36.8% in the study they applied chitosan and yeast extract to the garlic plant under water stress conditions. Mazrou et al. (2021) reported that in a 2 year study in which they applied chitosan nanoparticles to *Matricaria chamomilla* plant, they provided an increase in RWC compared to the control, and they achieved the maximum value especially in the application of 300 mg L^{-1}. Photosynthesis is the most important process affected in plants under saline conditions (Zayed et al. 2017). Chlorophyll content in plants exposed to abiotic stress is an important feature in determining the tolerance of plants to stress. When plants are exposed to stresses such as salinity, their chlorophyll content decreases and growth retards (Safikhan et al. 2018).

Table 1
Mean values of germination characteristics, RWC and carotenoid content of rapeseed cultivars under salt stress of different chitosan priming times

Cultivars × Ch times S doses	GP(%)									
	NK Caravel			Elvis			Champlain			Mean
S1	98.00 abc	93.33 hıj	84.67 n	98.33 ab	95.67d-g	90.00 l	99.00 a	96.33 c-f	90.67 kl	97.07 A
S2	96.67 b-e	93.33 hıj	84.67 n	95.33 efg	94.00 ghı	90.67 kl	98.00 abc	94.67 fgh	92.33 ıj	94.11 B
S3	98.00 abc	94.33 gh	86.67 m	93.00 hıj	92.00 jk	90.33 kl	97.33 a-d	93.33 hıj	90.33 kl	88.93 C
Mean	92.00 C	94.67 A	95.33 A	91.56 C	93.33 B	95.00 A	93.00 B	91.78 C	93.67 B	
LSD%1	1.589									
Cultivars × Ch times S doses	SL (cm)									
	NK Caravel			Elvis			Champlain			Mean
S1	9.03 c	8.25 ef	7.517 ı	9.29 ab	8.36 def	7.83 h	9.48 a	8.460 de	7.950 gh	9.213 A
S2	8.97 c	8.14 fg	6.953 j	9.11 bc	8.43 de	8.12 fg	9.297 ab	8.417 de	7.923 gh	8.358 B
S3	8.95 c	8.29 ef	7.163 j	9.35 ab	8.61 d	7.99 gh	9.453 a	8.263 ef	7.067 j	7.612 C
Mean	8.26 C	8.49 B	8.63 AB	8.02 D	8.55 AB	8.55 AB	8.136 CD	8.649 A	8.261 C	
LSD%1	0.2388									
Cultivars × Ch times S doses	RL (cm)									
	NK Caravel			Elvis			Champlain			Mean
S1	7.20 cde	6.90 fg	5.90 l	7.25 cd	7.00 efg	6.11 kl	7.55 b	7.20 cde	6.42 j	7.29 A
S2	7.10 c-f	6.92 fg	5.49 m	7.28 c	7.07 c-f	6.07 l	7.50 b	7.17 cde	6.50 ıj	6.99 B
S3	7.03 d-g	6.65 hı	5.52 m	6.92 fg	6.80 gh	5.89 l	7.77 a	7.24 cde	6.32 jk	6.02 C
Mean	6.67 BC	6.79 B	7.06 A	6.50 CD	6.81 B	7.06 A	6.40 D	6.53 CD	7.11 A	
LSD%5	0.2071									
Cultivars × Ch times S doses	SFW (g)									
	NK Caravel			Elvis			Champlain			Mean
S1	0.27 fgh	0.13 hıj	0.05 ıj	0.37 def	0.30 efg	0.06 ıj	0.57 bc	0.43 cde	0.10 ıj	0.52 A
S2	0.33 efg	0.20 ghı	0.04 j	0.43 cde	0.33 efg	0.08 ıj	0.90 a	0.50 bcd	0.10 ıj	0.35 B
S3	0.33 efg	0.00 efg	0.04 ıj	0.57 bc	0.33 efg	0.07 ıj	0.90 a	0.60 b	0.53 bc	0.12 C
Mean	0.15 G	0.23 DEF	0.37 C	0.19 FG	0.28 CDE	0.50 B	0.23 EFG	0.32 CD	0.68 A	
LSD%1	0.1379									
Cultivars × Ch times S doses	RFW (g)									
	NK Caravel			Elvis			Champlain			Mean
S1	0.13 fgh	0.10 gh	0.03 h	0.23 def	0.23 def	0.02 h	0.33 bcd	0.33 bcd	0.06 h	0.35 A
S2	0.20 efg	0.10 gh	0.03 h	0.30 b-e	0.23 def	0.07 h	0.67 a	0.37 bc	0.06 h	0.23 B
S3	0.23 def	0.13 fgh	0.03 h	0.40 b	0.20 efg	0.06 h	0.67 a	0.37 bc	0.27 cde	0.07 C
Mean	0.09 F	0.16 DE	0.24 C	0.11 EF	0.20 CD	0.37 B	0.13 EF	0.22 CD	0.43 A	
LSD%1	0.09749									
Cultivars × Ch times S doses	RWC (%)									
	NK Caravel			Elvis			Champlain			Mean
S1	64.67 d-g	62.84 ıj	59.30 no	66.07 abc	64.07 e-h	61.00 kl	66.40 ab	65.37 bcd	62.03 jk	65.50 A
S2	63.57 ghı	63.07 hıj	58.45 op	65.00 c-f	63.60 ghı	60.83 lm	66.50 ab	64.85 def	61.28 kl	64.01 B
S3	66.00 abc	63.30 hı	57.92 p	64.67 d-g	63.92 f-ı	59.85 mn	66.63 a	65.12 cde	61.00 kl	60.19 C
Mean	62.27 EF	63.71 BC	64.60 A	61.69 F	63.14 CD	64.21 AB	62.41 E	62.81 DE	64.25 AB	
LSD%1	1.041									
Cultivars × Ch times S doses	Total Chlorophyll (mg g^{-1})									
	Maximus			Sirena			Reyna			Mean
S1	1.80 bcd	1.70 fgh	1.53 jk	1.90 a	1.73 d-g	1.60 ıj	1.90 a	1.76 c-g	1.64 hı	1.834 A
S2	1.78 cde	1.70 fgh	1.51 kl	1.88 a	1.71 e-h	1.89 a	1.77 c-f	1.59 ıj	1.81 bc	1.72 B
S3	1.76 c-g	1.69 gh	1.45 l	1.78 cde	1.73 d-g	1.62 ı	1.92 a	1.87 ab	1.59 ıj	1.63 C
Mean	1.68 EF	1.74 CD	1.77 BC	1.66 FG	1.83 A	1.72 D	1.63 G	1.71 DE	1.79 AB	
LSD%1	0.06894									
Cultivars × Ch times S doses	Crt (mg g^{-1})									
	Maximus			Sirena			Reyna			Mean
S1	0.77 e	0.53 f	0.37 g	0.87 e	1.03 cd	1.17 ab	0.77 e	1.23 ab	1.21 ab	0.86 C
S2	0.87 e	0.73 e	0.83 e	0.87 e	1.00 d	1.25ab	0.87 e	1.18 ab	1.26 ab	0.99 B
S3	0.83 e	0.83 e	1.03 cd	0.87 e	1.14 bc	1.21ab	1.03 cd	1.23 ab	1.28 a	1.07 A
Mean	0.56 F	1.02 C	1.07 BC	0.81 E	1.04 BC	1.10 B	0.90 D	1.07 BC	1.18 A*	
LSD%1	0.1194									

* Dissimilar letters in the column show different groups

In this study, despite the increase in S stress, increases in chlorophyll content were determined, especially in C2 and C3 varieties, with Ch2 application. On the other hand, chlorophyll content increased in the same cultivars with S2 salt dose in Ch3 application compared to the control (Figure 1). When the carotenoid parameter was examined, with the increase in salt strain in all cultivars, increases in Crt were also observed with the Ch application times. Zayed et al. (2017) applied Ch to bean plant under salt stress. They reported that they found increases in the relative water index and chlorophyll content as a result of the study. Gerami et al. (2020) reported an increase in the chlorophyll and carotenoid content of the plant in their study where they applied chitosan to the stevia plant.

4.Conclusion

In this study, the effects of chitosan priming application times on the germination parameters, RWC, chlorophyll and carotenoid content of rapeseed cultivars under salt stress conditions were investigated. Chitosan application times, especially Ch2, and C3 cultivar gave more positive results in terms of the parameters examined. Besides, applications should be made in other plants and under various stress conditions and their results should be evaluated.

5. References

Abdelaal K, Attia KA, Niedbała G, Wojciechowski T, Hafez Y, Alamery S, Alateeq TK, Arafa SA (2021). Mitigation of Drought Damages by Exogenous Chitosan and Yeast Extract with Modulating the Photosynthetic Pigments, Antioxidant Defense System and Improving the Productivity of Garlic Plants. *Horticulturae* 7, 510. https://doi.org/10.3390/ horticulturae7110510.

Ahmed KBM, Khan MMA, Siddiqui H, Jahan A (2020). Chitosan and its oligosaccharides, a promising option for sustainable crop production- a review. *Carbohydrate Polymers* 227: 115331. https://doi.org/10.1016/j.carbpol.2019.115331.

Ali AYA, Ibrahim MEH, Zhou G, Nimir NEA, Jiao X, Zhu G, Elsiddig AMI, Suliman MSE, Elradi SBM, Yue W (2020). Exogenous jasmonic acid and humic acid increased salinity tolerance of sorghum. *Agronomy Journal* 112:871–884.https://doi.org/10.1002/agj2.20072

Arslan B, Culpan E (2022). Innovative Agricultural Practises in Soil, Plant and Environment. *Kolza (Kanola) Yetiştiriciliği ve Islah Amaçları.* Chapter:14 437-458. ISBN: 978-625-8377-02-6, İksad Yayınevi.

Bakhoum GS, Sadak MS, Badr EAEM (2020). Mitigation of adverse effects of salinity stress on sunflower plant (*Helianthus annuus* L.) by exogenous application of chitosan. *Bulletin of the National Research Centre* 44(1):1-11. https://doi.org/10.1186/s42269-020-00343-7.

Chouhan D, Mandal P (2021). Applications of chitosan and chitosan based metallic nanoparticles in agrosciences-A review. *Int J Biol Macrolmol.* https://doi.org/10.1016/j.ijbiomac.2020.11.035.

Elsiddig AMI, Zhou G, Nimir NEA, Ali AYA (2022). Effect of exogenous ascorbic acid on two sorghum varieties under different types of salt stress. *Chilean Journal of Agricultural Research* 82(1):10-20. doi:10.4067/S0718-58392022000100010

Gerami M, Majidian P, Ghorbanpour A, Alipour Z (2020). Stevia rebaudiana bertoni responses to salt stress and chitosan elicitor. *Physiology and Molecular Biology of Plants* 26(5): 965–974. https://doi.org/10.1007/s12298-020-00788-0.

Guan Y, Hu J, Wang X, Shao C (2009). Seed priming with chitosan improves maize germination and seedling growth in relation to physiological changes under low temperature stress. *J Zhejiang Univ Sci B* 10(6):427-433.

Gürsoy M, Kolsarıcı Ö (2017). Effects of Different Humic Acid Doses in Leonardite Environment on the Oil Percentage and Fatty Acids of Summer Rapeseed (*Brassica napus* ssp. *oleifera* L.) under Central Anatolian Conditions. *Fresenius Environmental Bulletin* 26(11):6447-6456.

Gürsoy M (2019). Importance of some oil crops in human nutrition. *Turkish Journal of Agriculture-Food Science and Technology* 7(12): 2154-2158. Doi:https://doi.org/10.24925/turjaf.v7i12.2154-2158.2916

Gürsoy M (2020). Effect of chitosan pretreatment on seedling growth and antioxidant enzyme activity of safflower (*Carthamus tinctorius* L.) cultivars under saline conditions. *Applied Ecology and Environmental Research* 18(5):6589-6603. Doi: http://dx.doi.org/10.15666/aeer/1805_65896603.

Gursoy M (2022a). Effect of foliar aminopolysaccharide chitosan applications under saline conditions on seedling growth characteristics antioxidant enzyme activity, chlorophyll and carotenoid contents of safflower (*Carthamus tinctorius* L.) cultivars. *Pak. J. Bot.*, 54(5): DOI: http://dx.doi.org/10.30848/PJB2022-5(15).

Gürsoy (2022b). Biostimulant Applications in Agriculture. *7th International Zeugma Conference on Scientific Researches Congress*. January 21-23 2022, Gaziantep/Turkey, The Book of Full Papers pp:41-47.

Hajihashemi S, Kazemi S (2022). The potential of foliar application of nano-chitosan-encapsulated nano-silicon donor in amelioration the adverse efect of salinity in the wheat plant. *BMC Plant Biology* 22:148, 1-15. https://doi.org/10.1186/s12870-022-03531-x.

Hameed A, Sheikh MA, Farooq T, Basra S, Jamil A (2013). Chitosan priming enhances the seed germination, antioxidants, hydrolytic enzymes, soluble proteins and sugars in wheat seeds. *Agrochimica-Pisa* 57(2): 97-110.

Harfoush EA, Abdel-Razzek AH, El-Adgham FI, El-Sharkawy AM (2017). Effects of Humic Acid and Chitosan under Different Levels of Nitrogen and Potassium Fertilizers on Growth and Yield Potential of Potato Plants (*Solanum tuberosum* L.). *Alex. J. Agric. Sci.* 62(1): 135-148.

Hasanah Y, Sembiring M (2018). Effect of foliar application of chitosan and salicylic acid on the growth of soybean (*Glycine max* (L.) Merr.) cultivars. *IOP Conf.Ser:Earth Environ.Sci.* 122 012027. Doi: 10.1088/1755-1315/122/1/012027

Iqbal S, Basra SM, Afzal I, Wahid A, Saddiq MS, Hafeez MB, Jacobsen SE (2019). Yield potential and

salt tolerance of quinoa on salt-degraded soils of Pakistan. *J Agron Crop Sci.* 205(1): 13-21. https://doi.org/10.1111/jac.12290.

ISTA (2003). International rules for seed testing. ınternational seed testing association, Bassersdorf, Switzerland.

Jabeen N, Ahmad R (2013). The activity of antioxidant enzymes in response to salt stress in safflower (*Carthamus tinctorius* L.) and sunflower (*Helianthus annuus* L.) seedlings raised from seed treated with chitosan. *J. Sci. Food Agric.* 93, 1699–1705.

Lal SK, Kumar S, Sheri V, Mehta S, Varakumar P, Ram B, Borphukan B, James D, Fartyal D, Reddy MK (2018). Advances in Seed Priming. *Seed Priming: An Emerging Technology to Impart Abiotic Stress Tolerance in Crop Plants.* Chapter3:41-50. https://doi.org/10.1007/978-981-13-0032-5_3.

Li Z, Zhang Y, Zhang X, Merewitz E, Peng Y, Ma X, Huang L, Yan Y (2017). Metabolic pathways regulated by chitosan contributing to drought resistance in white clover. *J. Proteome Res.* 16: 3039–3052.

Lichtenthaler HK, Wellburn AR (1983). Determinations of total careteonids and chlorophylls a and b of leaf extracts in different solvents. *Biomchem. Soc. Transac.*, 11: 591-592.

Mahdavi B, Modarres SS, Aghaalikhani M, Sharifi M, Dolatabadian A (2011). Chitosan İmproves Osmotic Potential Tolerance in Safflower (*Carthamus tinctorius* L.) Seedlings. *J Crop Improve* 25(6): 728-741.

Mazrou R, Ali EF, Hassan S, Hassan FAS (2021). A Pivotal Role of Chitosan Nanoparticles in Enhancing the Essential Oil Productivity and Antioxidant Capacity in Matricaria chamomilla L.. *Horticulturae* 7, 574. https:// doi.org/10.3390/horticulturae7120574.

Mushtaq Z, Faizan S, Gulzar B, Mushtaq H, Bushra S, Hussain A, Hakeem KR (2021). Changes in Growth, Photosynthetic Pigments, Cell Viability, Lipid Peroxidationand Antioxidant Defense System in Two Varieties of Chickpea (Cicer arietinumL.) Subjected to Salinity Stress. *Phyton – International Journal of Experimental Botany* DOI: 10.32604/phyton.2021.016231.

Palve S, Ahire D, Gahile Y (2022). Salicylic acid pretreatment effects on *Beta vulgaris* L. multigerm germination and germination indices. *International Journal of Biosciences*. 20(1): 59-71.

Pongprayoon W, Roytrakul S, Pichayangkura R, Chadchawan S (2013). The role of hydrogen peroxide in chitosan-induced resistance to osmotic stress in rice (*Oryza sativa* L.). *Plant Growth Regul*. 70: 159–173.

Ritchie SW, Nguyen HT, Haloday AS (1990). Leaf water content and gas exchange parameters of two wheat genotypes differing in drought resistance. *Crop Science* 30: 105-111.

Safikhan S, Khoshbakht K, Chaichi MR, Moteshareza-deh AAB (2018). Role of chitosan on the growth, physiological parameters and enzymatic activity of milk thistle (*Silybum marianum* (L.) Gaertn.)in a pot experiment. – *Journal of Applied Research on Medicinal and Aromatic Plants* 10: 49-58. DOI:10.1016/j.jarmap.2018.06.002.

Sen SK, Mandal P (2016). Solid matrix priming with chitosan enhances seed germination and seedling invigoration in mung bean under salinity stress. *Journal of Central European Agriculture*, 17(3): 749–762. DOI: 10.5513/JCEA01/17.3.1773.

Seraj RGM, Behnamian M, Ahmadikhah A, Shariati V, Dezhsetan S (2021). Chitosan and salicylic acid regulate morpho-physiologicaland phytochemical parameters and improve water-deficit tolerancein milk thistle (*Silybum marianum* L.). *Acta Physiologiae Plantarum* 43:101https://doi.org/10.1007/s11738-021-03264-8.

Sheikha SA, Al-Malki FM (2011). Growth and chlorophyll responses of bean plants to the chitosan applications. *European Journal of Scientific Research* 50(1): 124-134.

Siddiqi E, Ashraf M, Aisha AN (2007). Variation in seed germination and seedling growth in some diverse line of safflower (*Carthamus tinctorius* L.) under salt stress. *Pak J Bot.*, 39: 1937-1944.

Zahra N, Mahmood S, Raza ZA (2018). Salinity stress on various physiological and biochemical attributes of two distinct maize (*Zea mays* L.) genotypes. *J Plant Nutr*. 41(11): 1368-80.

Zayed MM, Elkafafi SH, Zedan AMG, Dawoud SFM (2017). Effect of Nano chitosan on growth, physiological and biochemical parameters of *Phaseolus vulgaris* under salt stress. *Journal of Plant Production*, 8(5): 577–585. https://doi.org/10.21608/jpp.2017.40468.

Zhang G, Wang Y, Wu K, Zhang Q, Feng Y, Miao Y, Yan Z (2021). Exogenous Application of Chitosan Alleviate Salinity Stress in Lettuce (*Lactuca sativa* L.). *Horticulturae* 7, 342. https://doi.org/10.3390/horticulturae7100342.

Zhang M, Zhang F, Li C, An H, Wan T, Zhang P (2022). Application of Chitosan and Its Derivative Polymers in Clinical Medicine and Agriculture. *Polymers*, 14, 958. https://doi.org/10.3390/polym14050958.

Selcuk Journal of Agriculture and Food Sciences
SJAFS
http://sjafs.selcuk.edu.tr/sjafs/index
(2022) 36 (3), 356-361
Research Article
e-ISSN: 2458-8377
DOI:10.15316/SJAFS.2022.045

Factors Affective on The Consumers' Purchasing Behaviours in Shopping Malls: A Case Study in Eastern Part of Turkey

Muhsin YEŞİLYOL[1], Mustafa TERİN[2], İbrahim YILDIRIM[2*]

[1] T.C. Ziraat Bank, Van, Türkiye
[2] Yüzüncüyıl University, Faculty of Agriculture, Department of Agricultural Economics, Van, Türkiye

ARTICLE INFO

Article history:
Received date: 31.05.2022
Accepted date: 23.08.2022

Keywords:
Comsumers' Purchasing Behaviours
Shopping Malls
Van Province
Turkey

ABSTRACT

The study aims at determining the purchase behaviours of consumers in shopping malls in Center town of Van Province. The data of this study was collected from 268 consumers of shopping malls through a structured questionnaire in Van Province, Turkey. The represented sample size was determined using finite population sampling method with 90 % confidence interval and a 5 % margin of error. Pearson Chi square test and Kruskal-Wallis Khi-Square techniques and Five-point likert-scale was used in analysing the data. The major factors affective on the preference of the shopping malls were service quality (average likert scale score 3.18) followed by new product opportunities (average likert scale score 3.17), product design (average likert scale score 3.08) and confidence for products (average likert scale score 3.05). Income level was not statistically significant on the consumers' preferences of choosing the shopping malls as regards service quality, new product opportunities, product design and confidence for products ($P>0.05$). The test results showed that the groups with different income level had the same opinion regarding above-cited factors. On the other hand, there existed statistically significant difference between education level and service quality, new product opportunities, product design and confidence for products ($P<0.05$).

1. Introduction

Economics has a long track record in the study of consumer behaviour. The invisible hand of the market is used to achieve a distribution of products in accordance with consumer choice. Markets are influenced by people's characteristics, their perceptions of wants and tastes, their purchasing power and their relative position in the market as buyers and sellers (Martins 2012).

Globalization has tremendously modified and conditioned the purchase behaviour of consumers. Today's consumers have access to tremendous amounts of information, they have a tremendous number of alternatives they can choose from, and they have incomparably more access to products and services that they can buy (Samli 2013).

A Shopping Mall can be considered as collection or congregation of various stores offering different brands, products or services at one place. Shopping malls are a noteworthy financial environment and a global phenomenon in a city and they drive monetary and social advancement (Nasim & Shamsir 2018; Katrodia et al. 2018).

The uprisings of shopping malls have a tremendous impact on the shopping behaviours of customers. The convenience to easily hop from a retail shop to another retail shop in a comfortable, covered environment, has led to a huge preference of consumers for shopping malls over the hassle of visiting single, not connected shops, having the inconvenience to reach them one by one in different locations (Mok Kim Man & Cai Qian Qiu 2021; Manandhar 2020).

There exit a great deal of research on the preference of consumers regarding choosing the shopping malls namely; Assortment of the products (Katrodia et al. 2018; Nasim & Shamshir 2018; Güzel et al. 2017; Khare 2011; Dinçer & Dinçer 2011; Twari 2010; Akgün 2010; Azaboğlu & Dursun 2008; Zafar et al. 2007), service and product quality (Katrodia et al. 2018; Kushwaha et al 2017; Akat et al. 2006), new product opportunities (Katrodia et al. 2018; Khare 2011), product and shopping mall design (Khare 2011; Twari 2010; Arslan, 2009; Zafar et al. 2007), confidence for the product and shopping

* Corresponding author email: *iyildirim@yyu.edu.tr*

mall (Nasim & Shamshir 2018; Cengiz & Özden 2002), convenience (Nasim & Shamshir 2018; Kushwaha et al. 2017; Khare 2011; Patel & Sharma, 2009), location and proximity (Mok Kim Man & Cai Qian Qiu 2021; Kushwaha et al. 2017; Azabagaoğlu and Dursun 2008; Akat et al. 2006), social factors such as family outing, meeting friends and entertainment (Mok Kim Man & Cai Qian Qiu 2021; Çakmak 2012), parking facilities and playground for kids (Nasim & Shamshir 2018; Arslan, 2009; Akat et al. 2006), cheapness (Akgün 2010; Arslan 2009; İbicioğlu 2005).

There exists a great deal of the researches conducted in different countries in the world including Turkey as regards of the preferences of customers/consumers who visit shopping malls. The following are some represented researches related to the subject matter.

Mok Kim Man & Cai Qian Qiu (2021), who interviewed with 200 consumers in four shopping malls in Klang Valley-Kuala Lumpur area, Malaysia, indicated that the consumers' preference of a shopping mall depended on various factors, such as location, variety of shops, convenience, as well as other social factors, such as family outing, meeting friends or just self-entertainment, like window-shopping or socializing with other visitors.

In a study, in which 1,168 visiting customers involved; shyness, emotional stability, materialism and collectivism were cited as major factors effective on the impulsive buying tendency. The results also revealed that there was a significant and positive relationship between credit card use, time availability, and shopping enjoyment on the impulsive buying tendency. Women consumers, young consumers and low-income consumers had greater tendency to impulsive buying in comparison to men consumers, old consumers and high-income consumers, respectively (Baraka 2019).

Tirmizi et al. (2009), who interviewed with 165 respondents with higher income group in the area of Rawalpindi and Islamabad, Pakistan, reported that young people had generally greater tendency of impulse buying behaviour, but showed no association of impulse buying in higher income group of young people.

Nasim & Shamshir (2018), reviewed 100, article on the consumers' purchasing behaviours in shopping malls and concluded that shopping environment, ease of shopping, availability of different products, showbiz offered at malls, parking facility, good product quality, discount and sales promotion were the factors that convince the shoppers to visit shopping malls with entertainment.

Katrodia et al. (2018), who collected the data from 700 respondents through a questionnaire in Durban, South Africa, stated that there were notable gender differences, which shape shopping behaviour among men and women. Time and money spent at the mall was significantly high among female consumers as compared to male consumers. The study revealed that personal attributes and shopping mall attractiveness factors played a crucial role in influencing customer shopping behaviour amongst the mall shoppers. The other factors effective on the preference of the consumers on the shopping malls were cited as ambiance, services, and assortment of products.

In a survey conducted with 162 active mall shoppers in İndia, it was reported that utilitarian shopping motivations (convenient shopping, economic shopping and achievement shopping) and hedonic shopping motivations (shopping enjoyment, gratification shopping, idea shopping, shopping for aesthetic ambiance, roll shopping, and social shopping) were effective on the preference of shoppers in shopping malls. (Patel & Sharma 2009).

Guzel et al. (2017), who interviewed with 385 customers visiting Erzurum shopping mall and Palerium shopping mall in Erzurum Province, Turkey through a questionnaire reported that entertainment facilities, assortment of products, gracious and comfort places for resting were the major factors effected on the preferences of customers towards shopping malls.

The results of a study, spanning eight cities of India and involving 3,026 mall consumers, indicated that the heavy shoppers was significantly different from the other groups along multiple demographic and socioeconomic variables, behavioural variables, attitude and shopping orientation (Kuruvilla & Joshi 2010).

Akat et al. (2006), who interviewed 600 people in the range of 15-69 age interval indicated that the preference of international shopping malls was generally more common among people who were within 25-44 age range, had nuclear family and with relatively high education level of various professional groups. Proximity, ambience, product quality, confidence and parking facilities were cited as the major factors towards preferring the shopping malls.

Akgün (2010), who made a survey on 524 consumers in Konya, Turkey, reported that one-third of consumers (33%) preferred the shopping malls because of assortment of the products followed by proximity and cheapness with 21.0 and 16.0%, respectively. Demographic characteristics were also affected on the types and degrees of the preferences.

In a study conducted with 700 customers visiting four shopping malls in İstanbul; store features, cultural events, entertainment opportunities and price/quality relationship were cited as major factors on consumers' loyalty to the shopping malls (Arslan, 2009).

In research conducted through face to face interviews with 385 consumers in towns of İstanbul province, Turkey; proximity, assortment of the products and service qualities were reported as the major factors on the preference of consumers in choosing the shopping malls. (Azabagaoğlu & Dursun 2008).

In a study comprised nine closed shopping malls in İstanbul province, Turkey and made with 830 consumers; conscious shopping, reliable shopping opportunities, pleasant shopping, having fun and free time were cited as the major factors effective on the preference of shopping malls (Cengiz & Özden 2011).

Dinçer & Dinçer (2011) interviewed with 284 students who visited a shopping mall near their university

named İstanbul Commerce University and reported that sole aim of visiting was not only entertainment facilities but also the assortment of products.

The study could be considered as unique for eastern part of Turkey considering the scarcity of the research in the field. Furthermore, no such research is available in the research area.

2. Materials and Methods

Total number of households in in Center district of Van Province, Turkey (84,187) in 2014 year constituted the population of study (TÜİK 2015). The sample size, which represents the population, was determined as 268 households by using the finite population sampling method. The sample volume representing the main population was determined as 268 households by using the finite population sampling method below. A 90% confidence interval and a 5 % margin of error were used in finding the sample size (Miran 2003). Data were collected randomly from consumers with different incomes, education levels and occupations face-to face by the researcher between November 2013 and February 2014.

$$n = \frac{N_p * (1-p)}{(N-1) * \partial^2 px + p * (1-p)}$$

N= Population Number

p= the probability of event in the eopulation (0.5 is taken)

∂^2px= Variance of Ratio

Households were classified into four groups taken into account their monthly average income. The households who had average monthly income up to $500 constituted low-income group (27.6% of the households). The households with medium ($501-1000), upper medium ($1001-1500) and high income (more than $1500) made up 27.2, 33.2 and 11.9 % of the households. Thus, number of low-income, medium- income, upper medium-income and high-income households surveyed were 74, 73, 89 and 32, respectively.

Before analysing the data, the Outlier Test was conducted to determine the extreme values. Pearson Chi square test and Kruskal-Wallis Khi-Square techniques were used in determination of significant differences among the income group as regards some variables. Five-point likert-scale was applied to determine the relative average score of purchasing behaviours of consumers in shopping malls.

3. Research Findings and Discussion

3.1. Socio-Demographic Characteristics of the Households

The male and female population constituted 61.57 and 38.43%, respectively. The average age of the consumers was 33.30 year. The consumers below 35 years old were 60.82% followed by 35-50 years old range and above 50 years old with 31.72 and 7.46%, respectively. The household size was 4.95 people and the average child per household was 3.04 people (Table 1). In a similar study conducted in Bursa, it was determined that 65.3%of consumers were male and 34.7% were female while 45 % were below the age of 35 (Akat et al. 2006).

The average income per households was 3,000 TL and 6,553 TL for high-income households. The rate of the married and single consumers was 57.46 and 41.79%, respectively. More than half of the consumers (58.21%) had a bachelor diploma or was continuing their education at the university level followed by high school and secondary school graduates with 16.42 and 12.69%, respectively (Table 1). In a study conducted in Istanbul, 29.6% of consumers were married, 10.9% had a child and 49.4% graduated from universities (Arslan and Bakır 2009). In a study conducted in Karabük, it was reported that 58.37% of the consumers graduated from universities and 23.73% of them were married (Çakmak 2012).

More than half of the consumers (53.36 %) were civil servant and workers followed by students, and freelancers with 16.42 and 10.82 %, respectively. The households with children constituted more than half of sample size (56.7%) followed by single consumers and consumers who had married children with 23.51 and 10.07 %, respectively. The spouse of more than one-third (39.4%) of surveyed consumers had primary education level followed by high school, university and secondary school levels with 25.17, 21.09 and 10.20 %, respectively (Table 1). In a similar study conducted in Konya, 26.3% of the consumers were students, and 55.5% were civil servants (Akgün 2010). In a study conducted in Nepal, 44.8% of the consumers were students and 33.6% of them were workers (Manandhar 2020).

The consumers' expenditure for local and national shopping mall was $219 and $173, respectively. The expenditure of income groups in local shopping mall ranged from $108 for low-income groups and $325 for high-income group. The expenditure of income groups in national shopping mall ranged from $60 for low-income group to $295 for high-income group (Table 1).

Krushal-Wallis h test results showed that there existed a statistically significant difference between income groups and monthly average expenditure in both local and national shopping malls ($P<0.05$). Likewise, a statistically significant difference was found between education levels and monthly average expenditure in local and national shopping malls ($P<0.05$).

Table 1
Socio-demographic characteristics of consumers'

	Income Groups				Overall
	Up to $ 500	$ 501-1000	$ 1001-1500	Above $ 1500	
Gender (%)					
Female	59.5	26.03	32.58	34.38	38.43
Male	40.5	73.97	67.42	65.62	61.57
Age groups (%)					
15-27	60.80	27.40	33.71	9.38	36.57
28- 34	12.20	21.92	32.58	34.38	24.25
35-50	17.60	38.36	30.34	53.12	31.72
Average number of members	5.79	5.04	4.43	4.17	4.95
Average number of children	3.75	3.45	2.38	2.73	3.04
Average monthly income (TL)	617.68	1745.21	2623.03	6553.12	2299.47
Marital status (%)					
Married	39.19	65.75	60.67	71.88	57.46
Bachelor	59.46	32.88	39.33	28.12	41.79
Education (%)					
Primary school	17.57	21.92	3.37	6.25	12.69
Secondary school	6.76	12.33	2.25	6.25	6.72
High school	25.68	15.07	11.24	12.50	16.42
Bachelor	50.00	45.21	76.40	56.25	58.21
Occupation (%)					
Government officer	1.35	19.18	65.17	37.50	31.72
Worker	22.97	35.62	13.48	9.38	21.64
Student	55.41	1.37	2.25	0.00	16.42
Freelenacer	4.05	8.22	12.36	28.12	10.82
Family structure (%)					
Single	20.27	24.66	26.97	18.75	23.51
Family with children	63.51	49.32	53.93	65.62	56.72
Average Expenditure per month ($)					
Local shopping mall	108	224	270	325	219
National shopping mall	60	131	207	295	173

3.2. Consumers' Preference of Shopping Malls in Terms of Percentages

More than one-third (36.57%) of the consumers preferred the shopping malls because of service quality followed by the location and cheapness with 29.10 and 21.64%, respectively. The percentage of consumers increased in line with income groups, being the lowest with 18.92 for low-income group and the highest with 62.50% for high-income group.

On the other hand, low-income group attributed more importance to the cheapness compared to medium and high-income groups. The proportion of the consumers who preferred shopping malls because of the cheapness was the highest with 41.89 % in low-income group and the lowest with only 3.12 % in high-income group (Table 2).

Table 2
The Major Reasons of Consumers' Preference of Shopping Malls as Percentages

	Income Groups				Overall
	Up to $ 500	$ 501-1000	$ 1001-1500	Above $ 1500	
Service Quality	18.92	31.51	46.07	62.50	36.57
Location	29.73	31.51	28.09	25.00	29.10
Cheapness	41.89	20.55	12.36	3.12	21.64
Parking Facilities	-	1.37	6.74	-	2.61
Playground for Kids	-	4.11	1.12	-	1.49

3.3. Consumers' Preference of Shopping Malls in Terms of Likert Scale Scores

The major factors affective on the preference of the shopping malls were service quality (average likert scale score 3.18) followed by new product opportunities (average likert scale score 3.17), product design (average likert scale score 3.08) and confidence for products (average likert scale score 3.05). On the other hand, average likert scale score of the parking facilities and playgrounds for kids was 2.48 and 2.22, respectively (Table 3).

Income level was not statistically significant on the consumers' preferences of choosing the shopping malls as regards service quality, new product opportunities, product design and confidence for products (P>0.05).

The test results showed that the groups with different income level had the same opinion regarding above-cited factors.

On the other hand, consumers with different education levels had distinct opinion on the preference of choosing the shopping malls. There existed statistically significant difference between education level and service quality, new product opportunities, product design and confidence for products (P<0.05).

A great deal of researches cited the service quality as the major factor for consumers' preference of the shopping malls, which was in line with our results (Katrodia et al. 2018; Kushwaha et al. 2017; Azaboğlu & Dursun 2008; Akat et al. 2006). New product opportunity which was a major factor of the consumers' preference of choosing the shopping mall was also in concord with the other research results conducted in different countries (Katrodia et al. 2018; Khare 2011; Cengiz & Özden 2011; İbicioğlu 2005). Many research result cited the product design (Khare 2011; Twari 2010; Arslan 2009; Zafar et al. 2007) and confidence for the products (Nasim and Shamshir 2018; Cengiz and Özden, 2011; Akat et.al. 2006; Dorukan et.al. 2005), which were consistent with our results.

Table 3
Opinion of the Consumers as Regards of Preference of Choosing Shopping Malls (Likert Scale Score)*

	Income Groups				Overall
	Up to $ 500	$ 501-1000	$ 1001-1500	Above $ 1500	
Service Quality	3.00	3.13	3.39	3.18	3.18
New Product Opportunities	3.03	3.13	3.31	3.19	3.17
Product Design	3.01	3.00	3.19	3.09	3.08
Confidence for Products	3.06	2.79	3.22	3.19	3.05
Parking Facilities	2.24	2.32	2.68	2.88	2.48
Playground for Kids	2.26	2.00	2.41	2.12	2.22

**(1=Not at all, 2= Little, 3=A little, 4=A lot, 5= Definitely)*

5. Conclusions

A modern marketing approach based on consumer satisfaction is a necessity for shopping malls to survive in an intensely competitive environment. For sustainable profitability and new investment opportunities, the owners and managers of shopping malls need to know and constantly monitor consumers' purchasing behaviour. For this reason, they should benefit from research results on consumers' purchasing behaviour patterns in their shopping malls. This will contribute to deliver the right products to the consumers at the right prices at right time and place, and to provide after-sales services with a competitive approach. The consumers purchasing behaviour will also help the managers to determine the right type and size of meeting and entertainment venues for the consumers.

On the other hand, the findings of such studies will help the consumers in deciding on the purchasing of the right products with right price and quantity in right time, and manage the budget, especially in term of the credit card use and time in terms of the entertainment duration.

6. References

Akat Ö, Taşkın Ç, Özdemir A (2006). Uluslararası alışveriş merkezi tüketicilerinin satın alma davranışları ve Bursa ilinde bir uygulama. Uludağ Üniversitesi Sosyal Bilimler Dergisi, 2(26- 27): 13-30.

Akgün OV (2010). Modern alışveriş merkezlerinin tüketici davranışları üzerindeki etkisi ve Konya ilinde bir uygulama. KMÜ Sosyal ve Ekonomik Araştırmalar Dergisi, 12 (19): 153-163.

Arslan M (2009). Tüketicilerin alışveriş merkezlerini tercih etme nedenleri ve sadakate etkisi üzerine bir araştırma. Marmara Üniversitesi Sosyal Bilimler Enstitüsü Dergisi, 32(8):39-49.

Azabağaoğlu MÖ, Dursun E (2008). Tüketicilerin modern gıda perakendecilerine karşı dayanışma analizi. Tekirdağ Ziraat Fakültesi Dergisi,5(1):1-12.

Baraka MA (2019). A Proposed model for factors affecting consumers ' impulsive buying tendency in shopping malls. Journal of Marketing Management, 7(1): 120-134.

Çakmak AÇ (2012). Ziyaretçilerin alışveriş merkezlerini tercih etme nedenlerinin araştırılması karabük şehir merkezinde bir uygulama. Niğde Üniversitesi İİBF Dergisi, 5 (2): 195-215.

Cengiz E, Özden B (2002). Perakendecilikte büyük alışveriş merkezleri ve tüketicilerin büyük alışveriş merkezleri ile ilgili tutumlarını tespit etmeye yönelik bir araştırma. Ege Akademik Bakış Dergisi, 2(1):1-13.

Dincer B, Dincer C (2011). Tüketicilerin alışveriş merkezi ziyareti ve davranışı: üniversite öğrencileri üzerine bir araştırma. Süleyman Demirel Üniversitesi, İİBF Sosyal ve Ekonomik Araştırmalar Dergisi, 13(3): 317-331.

El-Adly Mİ (2007). Shopping malls attractiveness: A segmentation approach. international Journal of Retail & Distribution Management, 35(11): 936 – 950.

Güzel D, Demir İK, Yazıcılar FG (2017). Determınatıon Of The Relatıonshıp Between The Consumers Reasons For Preference Of Shoppıng Centers: Applıcatıon Of Erzurum. PressAcademia Procedia, 3(1): 666-673.

İbicioğlu H (2005). Alışveriş merkezleri: Demografik etkenler ve tüketici tutumlarının incelenmesine yöneliş bir araştırma. Süleyman Demirel Üniversitesi İktisadi İdari Bilimler Fakültesi, 10 (1): 43-55.

Katrodia A, Naude MJ, Soni S (2018). Consumer Buying Behavior at Shopping Malls: Does Gender Matter? Journal of Economics and Behavioral Studies, 10(1): 125-134.

Khare A (2011). Mall shopping behaviour of Indians mall town consumers, Journal of Retailing and Consumer Services, 18:110–118.

Kuruvilla SJ, Joshi N (2010). Influence of demographics, psychographics, shopping orientation, mall shopping attitude and purchase patterns on mall patronage in India, Journal of Retailing and Consumer Services, 17(4): 259–269.

Kushwaha T, Ubeja S, Chatterjee AS (2017). Factors Influencing Selection of Shopping Malls: An Exploratory Study of Consumer Perception.Vision, 21(3): 274–283.

Manandhar RR (2020). Attitude Towards Shopping Mall Purchasing Behavior. Management Dynamics, 23(2): 207-228.

Martins JM, Farhat Y, Swanson DA (2012). Consumer Demographics and Behaviour: Markets are People. (Springer series on demographic methods and population analysis; Vol. 30). Springer, Springer Nature. https://doi.org/10.1007/978-94-007-1855-5.

Miran B (2003). Basic Statistics, Ege Uni. Press, Izmir.

Mok Kim Man M, Cai Qian Qiu R (2021.) An empirical study of factors influencing consumers' purchasing behaviours in shopping malls. International Journal of Marketing Studies, 13(1):14-25.

Nasim S, Shamshir M (2018). Consumer Behavior towards Shopping Malls: A Systematic Narrative Review. Journal of Business Studies, 14(1): 81-95.

Patel V, Sharma M (2009). Consumers' Motivations to shop in Shopping Malls: A Study of Indian Shoppers. Advances in Consumer Research, 8, 285-290.

Samli AC (2013). International Consumer Behaviour in the 21st Century Impact on Marketing Strategy Development. Springer Science+Business Media New York

Tirmizi MA, Ur-Rehman K, Saif MI (2009). An Empirical Study of Consumer Impulse Buying Behaviour in Local Markets. European Journal of Scientific Research, 28(4): 522-532.

TÜİK (2015). Adrese Dayalı Nüfus Kayıt Sistemi Sonuçları.

Twari RK (2010). Understandıng the consumer behavıor towards shoppıng malls in raipur city. International Journal of Management & Strategy, 1(1):1-14.

Zafar UA, Ghingold M, Dahari Z (2007). Malaysian Shopping Mall Behavior: An Exploratory Study, Asia Pacific. Journal of Marketing and Logistics, 19(4): 331 – 348.

Selcuk Journal of Agriculture and Food Sciences SJAFS

http://sjafs.selcuk.edu.tr/sjafs/index

(2022) 36 (3), 362-367

e-ISSN: 2458-8377

DOI:10.15316/SJAFS.2022.046

Research Article

Biochemical Effects of Drought Stress in Some Strawberry Cultivars

Mostafa Fraidoon Faaek ALİ[1] , Lütfi PIRLAK[1*] , Metin TURAN[3]

[1]Selçuk University, Faculty of Agriculture, Department of Horticulture, Konya, Türkiye
[2]Yeditepe University, Faculty of Engineering and Architecture, Department of Genetics and Bioengineering, İstanbul, Türkiye

ARTICLE INFO

Article history:
Received date: 04.07.2022
Accepted date: 25.08.2022

Keywords:
Strawberry
Biochemical properties
Drought Stress

ABSTRACT

This research was 2016-2017 years of the Selcuk University Faculty of Agriculture Department of Horticulture Research and practice was carried out in the greenhouse. Study the Yalova Atatürk Central Horticultural Research Institute, Ata-77, Bolverim-77, Doruk-77, Dorukhan-77, Eren-77, Erenoğlu-77, and Hilal-77 strawberry varieties. After the planted strawberry seedlings reached the 5-6 leaf stage, after irrigation at field capacity, drought application was made until the plants lost their leaf turgor. After implementation of the varieties of drought with healing and drought period of watering again losses to determine.
In strawberry cultivars, significant decreases occurred in the amount of protein with the application of drought. In the recovery period after the drought, the protein amounts increased again. Drought treatment significantly increased proline content in all cultivars compared to control. Catalase enzyme activity in all strawberry cultivars increased significantly as a result of drought application in both years. There was a slight decrease in the recovery period. In both years of the study, drought application increased the peroxidase enzyme activity in all strawberry cultivars compared to the control, and a slight decrease occurred in the recovery period compared to the drought application. Drought application in cultivars increased the superoxide dismutase enzyme activity a little compared to the control, while the activity decreased to the control level during the recovery period.

1. Introduction

Strawberry is one of the important fruit species grown commercially in the world and Turkey, and this importance is increasing. Strawberry is a type of fruit that can easily adapt to different ecological conditions and climate types. It can be grown in different ecological conditions from Siberia to Ecuador, from places with high altitudes to places at sea level. Therefore, it can be grown in almost every region in Turkey (Geçer and Yılmaz, 2011). Strawberry is produced mostly in the Mediterranean and Aegean regions in Turkey, and it has started to be grown in the inner regions over time. In the world, China, the USA, Mexico, Turkey, Spain, and Egypt are important strawberry producer countries (FAO, 2021).

Today, drought has reached social and economic dimensions that threaten the environment with the increase in the world population, climate changes, deforestation, and global warming. Drought is one of the natural disasters that cause the most damage to people and the environment and cause great losses. It is predicted that Turkey is among the countries in a high-risk group in terms of the possible effects of global warming and that the Mediterranean and Central Anatolia regions will be more affected by climate change in the future. Climate elements and especially the precipitation factor, which has the greatest effect on production, show great changes in time and space. Although the annual precipitation average in Turkey is around 640 mm, water shortage and drought are experienced in many regions due to the irregularity of the precipitation distribution (Özcan et al., 2004). Atmospheric conditions, physical geography factors, and climatic conditions are among the main factors affecting drought in Turkey. Drought stress is caused by the lack of moisture required for the plant to grow normally and complete its life cycle; It is common in regions where rainfall is irregular and irrigation is insufficient (Sircelj et al., 2007). Drought is a meteorological phenomenon in general terms, and it is defined as the period when there is no precipitation until the water

* Corresponding author email: *pirlak@selcuk.edu.tr*
**This work is part of first author's MSc thesis

content of the soil and plant growth decreases significantly and the amount of water shortage is sufficient to suffer (Özcan et al., 2004). In drought conditions, the water potential of the soil and then the plant decreases. Then, low turgor pressure, closure of stomata, decrease in leaf growth, and decrease in photosynthesis occurs. Plants exposed to drought stress have limited growth, lower dry matter production, increased susceptibility to diseases and pests and decreased product quality and quantity (Monti, 1987).

Stress is defined as the force physically applied to an object. However, the calculation of the biological stress force is quite difficult. Because while biological conditions cause stress in one plant species, they may be normal for another species (Mahajan et al., 2005). Practically, biological stress is a counterforce and is defined as the inability of plants to perform their biological functions and the deterioration of their systems (Jones, 1990). Plants encounter many stress factors throughout their lives, and these factors can be examined into two groups abiotic and biotic. These stress factors reduce the biosynthetic capacity of plants and change their normal functions and cause damage that will lead to death (Ekmekçi and Kalefetoğlu, 2005). Abiotic stress contains significant threats in terms of aquaculture with the deterioration of environmental factors, and it causes negative changes in plants in morphological, physiological, biochemical, and molecular directions, leading to more than 50% of product loss and low yield in the world (Wang et al., 2003).

Plants to drought stress; respond with the changes they show in morphological, biochemical, and metabolic processes. As a result, plants exposed to water scarcity are more sensitive to other biotic and abiotic stresses (Caruso et al., 2008). Stress tolerance; is the ability of plants to survive against adverse environmental conditions. Plants form a two-way defense mechanism against drought stress, either by avoiding stress or by developing stress tolerance (Mundree et al., 2002). Drought tolerance is species-specific and even cultivar-specific, and it is of great importance to determine the stress avoidance abilities of plants in terms of determining their tolerance (Özcan et al., 2004). In addition to the tolerance mechanisms created against the mechanical damage caused by drought, plants have also developed various enzymatic defense systems against oxidative stress (Bian and Jiang, 2009). High levels of antioxidative enzyme activity in plants significantly reduce oxidative damage in drought tolerance (Sharma and Dubey, 2005). However, the activity of these antioxidant enzymes produced during drought varies according to the plant type, variety, stress intensity, and duration (Bian and Jiang, 2009).

Global warming, which is seen as a potential threat to agricultural production in the future and whose effect is felt increasingly day by day, will require the determination of drought tolerance of cultivated plants and the cultivation of new drought-tolerant genotypes. For this purpose, in this study, a total of 7 domestic short-day strawberry cultivars, Bolverim-77, Hilal-77, Doruk-77, Dorukhan-77, Ata-77, Eren-77, Erenoğlu-77, bred in Yalova Horticultural Institute.

2. Materials and Methods

Materials

This research was carried out in the greenhouse of the Department of Horticulture, Faculty of Agriculture, Selcuk University in 2016-2017. In the research, a total of 7 strawberry cultivars, Bolverim-77, Hilal-77, Doruk-77, Dorukhan-77, Ata-77, Eren-77, Erenoğlu-77, were bred in Yalova Horticultural Central Research Institute, were used.

Characteristics of the strawberry cultivars used

ATA-77: Tioga x Cruz hybrid. Its fruits are medium-sized, the outer color of the fruit is bright red, the fruit is easy to break from the stem, the fruit flesh is hard, the fruit is heart-shaped, and the taste and smell are very good. Although it is table quality, it is also suitable for the food industry (Anonymous, 2017).

BOLVERİM-77: Tioga x Yalova-104 hybrid. The fruits are large, the outer color of the fruit is bright light red, the fruit is easy to break from the stem, the hardness of the fruit is medium, the shape is flattened, and the fruit taste is medium. It is suitable for the food industry (Anonymous, 2017).

DORUK-77: Tufts x Cruz hybrid. The fruits are small, the outer color of the fruit is bright red, the fruit is easy to break from the stem, and the fruit flesh is quite hard. It is suitable for the food industry and can also be used as a table (Anonymous, 2017).

DORUKHAN-77: Tufts x Cruz hybrid. The fruits are medium-large, the outer color of the fruit is bright red, the fruit is easy to break from the stem, and the fruit flesh is hard. It is quite efficient. Although it is table quality, it is also suitable for the food industry (Anonymous, 2017).

EREN-77: Ottoman x Tufts hybrid. Its fruits are medium-large, conical in shape, the outer color of the fruit is bright red, the fruit is easy to break from the stem, the taste and smell are very good, the fruit quality is good and the fruit flesh is hard. It is a table variety suitable for the food industry (Anonymous, 2017).

ERENOĞLU-77: Cruz x Tioga hybrid. The fruits are large, the outer color of the fruit is bright red, the fruit is easy to break from the stem, and the fruit flesh is medium hard. The taste and smell are very good, heart-shaped, and very easy to break from the stem. Fruit quality is very good (Anonymous, 2017).

HİLAL-77: Ottoman x Tufts hybrid. The fruits are medium-large, heart-shaped, the outer color of the fruit is bright red, the fruit is very easy to break from the stem, the taste and smell are very good, the fruit quality is good and the fruit flesh is hard. It is a table variety suitable for the food industry (Anonymous, 2017).

Methods

In the study, frigo strawberry seedlings were planted in 3-liter pots filled with peat. After the seedlings are planted, the flowers and branches are plucked until they reach the 5-6 leaf stage, and vegetative development of the plants is ensured. The seedlings, whose development was completed, were irrigated at the field capacity level and no irrigation was applied until the first sign of drought (withering of the leaves). After the first sign of drought, the plants started to be watered again and the recovery vitality was maintained. In the research, biochemical analyzes were made on plants during the drought application and during the re-irrigation phase.

The study was planned on 7 strawberry cultivars with a 15-day drought application followed by re-irrigation and the recovery of the plants. In the study, there were 3 replications and 3 plants in each replication.

Biochemical Analysis

Protein determination in plants under stress was made according to the "Bradford" method using 0.5 g plant samples. Results were calculated in terms of "mg protein/g" of fresh tissue (Bradford, 1976).

Proline determination was made spectrophotometrically by the acid-ninhydrin method (Bates et al., 1973).

While the activity of catalase (CAT) enables the conversion of H2O2 to oxygen and water in the environment, it is based on monitoring the absorbance change at 240 nm (Havir and Mchale, 1987).

Peroxidase (POD) activity determination is based on monitoring the absorbance increase at 470 nm caused by the colored compound, which is the product of the reaction in which guaiacol and H_2O_2 are substrates (Angelini and Federico, 1989).

Superoxide dismutase activity is based on the spectrophotometric determination of the inhibition of the photochemical reduction reaction of nitro blue tetrazolium (NBT) with superoxide radicals to the blue-colored formazone by the SOD enzyme (Agarwal and Pandey, 2004).

3. Results and Discussion

Results

The effect of drought stress on the amount of soluble protein in strawberry cultivars

Significant decreases in protein amount occurred with the application of drought in strawberry cultivars. In the recovery period after the drought, the protein amounts increased again. However, with these increases, the protein level did not increase to the control level in cultivars except Doruk-77 (Table 1.).

Table 1
The effect of drought stress on the amount of soluble protein (μg g^{-1} TA) in strawberry cultivars.

Cultivars	2016				2017			
	0-day	15th-day control	15th-day drought	7th-day recovery	0-day	15th-day control	15th-day drought	7th-day recovery
Doruk-77	33.91 a	34.32 a	19.59 a	34.03 a	34.92 a	35.63 a	21.94 a	34.83 a
Dorukhan-77	29.36 b	31.10 b	14.74 bd	23.08 b	29.80 b	31.23 b	16.51 c	23.66 b
Hilal-77	22.86 d	23.33 c	13.15 d	16.59 e	23.56 e	23.00 e	15.10 e	17.00 e
Bolverim-77	28.62 b	29.00 b	14.28 c	18.17 d	28.91 b	29.42 b	15.70 d	18.62 d
Eren-77	26.00 c	25.45 c	15.22 b	19.44 c	26.39 c	26.12 c	17.50 b	19.93 c
Erenoğlu-77	24.50 cd	24.16 c	12.49 e	17.57 de	25.26 cd	24.75 d	14.36 f	18.00 de
Ata-77	23.17 d	22.84 c	13.25 d	17.56 de	23.63 de	24.36 de	14.57 ef	18.00 e
LSD	2.32	2.80	0.77	1.57	2.39	2.41	0.85	1.61

*: There is no difference between the averages shown with the same letter in the same column

As a result of our study, the amount of protein decreased with the application of drought in strawberry cultivars. This result was similar to Brito et al. (2003), olive, Behnamnia et al. (2009)'s tomato, and Zanjani et al. (2012) zucchini. Drought stress causes a disturbance in the protein metabolism of the plant. This disorder is seen as the breakdown of proteins and decreased protein synthesis (Kutlu, 2010).

Effect of drought stress on proline amount in strawberry cultivars

Drought treatment significantly increased proline content in all cultivars compared to control. In the recovery period after the drought, a decrease was detected in the proline content, but with these decreases, the proline content remained above the control level (Table 2).

Table 2
Effect of drought stress on proline amount (μg g^{-1} TA) in strawberry cultivars.

Cultivars	2016				2017			
	0-day	15th-day control	15th-day drought	7th-day recovery	0-day	15th-day control	15th-day drought	7th-day recovery
Doruk-77	15.86 e	15.78 d	29.16 d	19.35 f	19.23 e	21.63 e	26.10 f	22.63 e
Dorukhan-77	29.69 b	27.61 b	40.85 c	34.68 b	20.96 d	22.12 de	27.75 e	22.33 e
Hilal-77	23.14 cd	2300 c	44.94 b	31.38 c	28.43 b	31.24 b	38.47 b	31.08 b
Bolverim-77	29.63 b	27.56 b	28.35 de	38.81 a	24.39 c	26.86 c	38.97 b	28.94 c
Eren-77	31.11 a	32.18 a	27.30 e	34.79 b	23.43 c	24.50 cd	31.08 d	27.11 d
Erenoğlu-77	22.29 d	23.14 c	49.99 a	24.45 e	33.29 a	36.78 a	44.27 a	38.66 a
Ata-77	24.30 c	24.50 c	42.02 c	27.24 d	21.44 d	24.96 c	35.89 c	27.13 d
LSD	1.82	2.56	2.47	1.07	1.51	2.89	2.01	1.08

*: There is no difference between the averages shown with the same letter in the same column

Proline is a water-soluble amino acid that generally accumulates under stress conditions and acts as an indicator in terms of providing the plant's resistance ability (Bian et al., 1988). Besides serving as an osmolyte, it is an effective substance in stabilizing cells, adjusting cytosolic pH, and regulating hydroxyl radicals (Matysik et al., 2002). Plants exposed to stress accumulate various soluble substances in their cytoplasm and organelles to maintain osmotic balance. Apart from providing a positive effect on enzymes, these substances also play a role in ensuring osmotic regulation in plants under stress by maintaining membrane integrity (Ashraf and Foolad, 2007). In our study, it was determined that the amount of proline increased in strawberry plants under drought stress. Similarly, Alizadeh et al. (2011) on different apple rootstocks, Abbaspour et al. (2012) In Pistachio, Karimi et al. (2012) almond and GF 677 rootstock, Rostami and Rahemi (2013) fig, Bolat et al. (2014) found that drought stress caused increases in proline amounts in apple and pear, İpek (2015) on Myrobolan and Garnem rootstocks.

Effect of drought stress on Catalase (CAT) enzyme activity in strawberry cultivars

Catalase enzyme activity in all of the strawberry cultivars we used increased significantly as a result of drought application in both years. There was a slight decrease in the recovery period. In 2016, the cultivar with the highest catalase enzyme activity was Doruk-77, and in 2017, Erenoğlu-77 was found (Table 3).

Catalase is one of the most important enzymatic antioxidants that catalyzes the direct reduction of high concentration H_2O_2 to water and oxygen by using its 2 electrons (Dionisio-Sese and Tobita, 1998). When the genes encoding different catalase isozymes in many plants under stress were examined, it was observed that the expression levels of the genes encoding this enzyme increased in relation to stress (Millar et al., 2003). In our study, significant increases in catalase enzyme activity were detected in strawberry plants kept under drought stress. These results we obtained are also compatible with drought stress studies on different plants (Reddy et al., 2004; Chai et al., 2005; Moussa and Abdel-Aziz, 2008).

Effect of drought stress on peroxidase (POD) activity in strawberry cultivars

In both years of the study, drought application increased the peroxidase enzyme activity in all strawberry varieties compared to the control, and a slight decrease occurred in the recovery period compared to the drought application (Table 4).

Table 3
Effect of drought stress on Catalase (CAT) enzyme activity (EU g^{-1} TA) in strawberry cultivars

Cultivars	2016				2017			
	0-day	15th-day control	15th-day drought	7th-day recovery	0-day	15th-day control	15th-day drought	7th-day recovery
Doruk-77	161.83 e	155.01 g	783.83 a	402.17 e	143.00 e	145.36 f	508.17 b	377.83 c
Dorukhan-77	192.50 d	191.30 f	722.17 b	491.00 c	203.17 d	197.65 e	412.33 d	259.83 e
Hilal-77	362.51 a	345.36 a	442.62 d	426.33 d	387.67 a	380.25 b	476.00 c	401.00 b
Bolverim-77	204.50 d	200.36 e	701.83 b	477.50 c	266.17 c	271.36 d	358.83 e	327.50 d
Eren-77	231.33 c	236.75 d	691.67 b	528.17 b	294.17 b	290.01 c	425.67 d	329.00 d
Erenoğlu-77	301.33 b	289.63 b	466.67 d	391.17 e	398.00 a	387.34 a	624.33 a	455.67 a
Ata-77	248.00 c	255.02 c	575.33 c	555.17 a	383.33 a	380.69 b	470.67 c	450.67 a
LSD	31.24	2.80	54.19	24.76	38.34	2.38	25.93	26.29

*: There is no difference between the averages shown with the same letter in the same column

Table 4
Effect of drought stress on peroxidase (POD) enzyme activity (EU g^{-1} TA) in strawberry cultivars.

Cultivars	2016				2017			
	0-day	15th-day control	15th-day drought	7th-day recovery	0-day	15th-day control	15th-day drought	7th-day recovery
Doruk-77	406.53 a	400.01 a	1050.80 a	637.60 a	479.67 a	470.85 a	1054.80 a	623.33 a
Dorukhan-77	355.33 b	359.33 b	520.53 c	444.24 c	388.00 b	384.00 b	768.13 b	480.33 b
Hilal-77	192.53 g	187.44 g	333.33 e	316.80 f	288.00 e	280.01 e	488.80 d	374.33 c
Bolverim-77	294.40 c	290.18 c	637.86 b	549.87 b	323.00 c	326.00 c	467.46 e	362.33 c
Eren-77	225.20 e	229.66 e	358.13 e	313.80 f	245.66 f	240.23 f	492.80 d	301.66 d
Erenoğlu-77	266.13 d	270.61 d	475.60 d	347.60 e	301.33 d	299.63 d	595.73 c	387.33 c
Ata-77	207.80 f	205.36 f	440.00 d	276.40 d	233.33 g	230.14 g	426.80 f	383.33 c
LSD	5.02	2.44	55.73	10.19	4.21	3.08	6.54	54.82

*: There is no difference between the averages shown with the same letter in the same column

It has been reported that high POD activity is associated with drought tolerance of plants and an increase in POD activity may contribute to drought stress tolerance (Sairam and Srivastava, 2000). In our study, POD activity increased in all cultivars under drought stress. Similar results were obtained in studies on this subject. Tanaka et al. (1990) reported that POD activity increased in spinach leaves under water stress. Similarly, Bolat et al. (2014) in apple and pear, Patel et al. (2011) reported an increase in POD activity under drought stress in chickpea.

Effect of drought stress on superoxide dismutase (SOD) activity in strawberry cultivars

Drought application in cultivars increased the activity of superoxide dismutase enzyme slightly compared to the control, while the activity decreased to the control level during the recovery period (Table 5).

Table 5
Effect of drought stress on superoxide dismutase (SOD) enzyme activity (EU g^{-1} TA) in strawberry cultivars

Cultivars	2016				2017			
	0-day	15th-day control	15th-day drought	7th-day recovery	0-day	15th-day control	15th-day drought	7th-day recovery
Doruk-77	465.59 c	460.13 d	624.69 a	404.76 d	479.35 a	480.00 a	540.64 bc	418.06 f
Dorukhan-77	444.26 d	440.01 e	572.15 c	403.89 d	435.12 c	438.12 c	549.25 b	428.42 e
Hilal-77	503.51 a	505.78 a	542.54 f	463.65 a	423.51 c	420.00 f	483.44 e	443.44 d
Bolverim-77	423.08 f	420.52 g	567.56 d	418.35 c	438.99 c	443.11 b	514.84 d	484.98 b
Eren-77	434.55 e	436.99 f	557.69 e	423.62 b	435.91 b	428.36 d	529.60 cd	414.69 f
Erenoğlu-77	507.13 a	50.15 b	604.33 b	407.09 d	419.49 c	425.19 e	585.80 a	457.92 c
Ata-77	473.19 b	470.32 c	556.06 e	424.84 b	417.63 c	427.85 de	543.30 bc	505.59 a
LSD	7.72	3.20	4.49	6.34	30.16	2.92	22.76	7.06

*: There is no difference between the averages shown with the same letter in the same column

Oxidative stress in plants under drought stress causes the formation of reactive oxygen species (ROS) in cells. Plants use antioxidant enzymes such as SOD, GR, CAT, and APX to neutralize these radicals. The superoxide anion (O_2^-) is converted to H_2O_2 by the SOD enzyme in the cells and H_2O_2; other enzymatic systems are broken down into HO and O_2 by POD, APX, and GR (Bian and Jiang, 2009).

In our study, an increase in superoxide dismutase enzyme activity was detected in all cultivars during the drought period. Many researchers have examined the SOD enzyme activity in different plants under different stress conditions. Gong et al. (2005) stated that the SOD enzyme activity in wheat was lower under drought conditions, while Yediyıldız (2008) determined that there were no significant differences in SOD activity of wheat varieties under salt and drought stress compared to the control. Rahman et al. (2002) determined that the SOD enzyme reached the highest level at the end of the stress in tomato plants under drought stress. Reddy et al. (2004) mulberry, Cai et al. (2005) in coffee, and Sivritepe et al. (2008) found an increase in SOD enzyme activity in Gisela-5 rootstock grown in vitro.

4. Conclusions

It occurs when the usable water in the soil decreases in plants and water is lost by transpiration or evaporation due to atmospheric conditions (Jaleel et al., 2009). Drought stress is one of the most important stresses affecting plant growth and yield, and it affects many physiological, biochemical, and molecular properties in plants (Özfidan, 2010). Therefore, understanding the physiological and biochemical responses of plants in resistance to drought stress will be useful in identifying species and varieties that are resistant to drought conditions. In this context, in our study on newly bred strawberry cultivars, it was determined that the responses of cultivars to drought stress differ.

Factors that inhibit plant growth are called stress. Stress caused by drought, salinity, high and low temperatures, and heavy metals is common in many agricultural parts of the world. In recent years, with the effect of global warming, the importance of water has started to be felt more and more with agricultural drought. Plants develop tolerance mechanisms to adapt to environmental conditions like physiological, biochemical, and molecular responses to drought stress (İpek, 2015). Decrease in the amount of protein in varieties with drought application; proline, catalase, peroxidase, and superoxide dismutase enzyme activities increased.

In plants in general, the continuation of growth and development depends on the preservation of the water content of the cell. The lack of water in the plant causes a decrease in photosynthesis and thus a slowdown in development and, as a result, a decrease in yield and quality. As a result, irrigation systems have been emphasized and new methods have been developed to combat drought. However, nowadays, these are insufficient with the decrease in irrigation water. In this context, the selection of resistant plants should be emphasized to obtain drought-resistant plants. In addition, elucidating the drought stress mechanism will help to develop drought stress-resistant plants.

5.References

Abbaspour H, Saeidi-Sar S, Afshari H, Abdel-Wahhab MA (2012). Tolerance of mycorrhiza infected pistachio (Pistacia vera L.) seedling to drought stress under glasshouse conditions. *Journal of Plant Physiology* 169: 704 -709.

Agarwal A, Pandey R (2004). Antioxidant enzyme responses to NaCl stress in Cassia angustifolia. *Biol Plant*. 48:555-560.

Alizadeh A, Alizade V, Nassery L, Eivazi A (2011). Effect of drought stress on apple dwarf rootstocks. *Technical Journal of Engineering and Applied Science* 1: 86-94.

Angelini R, Federico M (1989). Histochemical evidence of polyamine oxidation and generation of hydrogen peroxide in the cell wall. *J. Plant Physiol*. 135: 212-217.

Anonymous (2017). https://arastirma. tarim.gov.tr /yalovabahce/Menu/34/Meyveler, Date of access: 19.06.2021.

Ashraf M, Foolad MR (2007). Roles of glycine betaine ve proline in improving plant abiotic stress resistance. *Environmental and Experimental Botany* 59: 206-216.

Bates A, Waldren W, Teare A (1973). Rapid determination of free proline for water-stress studies. *Plant and Soil,* 39: 205-207.

Behnamnia M, Kalantari KM, Rezanejad F (2009). Exogenous application of brassinosteroid alleviates drought-induced oxidative stress in Lycopersicon esculentum L. *General and Applied Plant Physioiogy*, 35: 22-34.

Bian YM, Chen SY, Xie MY (1988). Effects of HF on proline of some plants. *Plant Physiol. Commun.,* 6: 19-21.

Bian S, Jiang Y. (2009). Reactive oxygen species, antioxidant enzyme activities and gene expression patterns in leaves and roots of Kentucky bluegrass in response to drought stress and recovery. *Scientia Horticulturae*, 120: 264–270.

Bolat I, Dikilitas M, Ercisli S, Ikinci A, Tonkaz T (2014). The effect of water stress on some morphological, physiological, and biochemical characteristics and bud success on apple and quince rootstocks. *The Scientific World Journal*, 1-8.

Bradford C (1976). A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. Areproduction Research Laboratories, Department Of Biochemistry, University of Georgia, Athens, Georgia 30602 USA 942-951.

Brito G, Costa A, Fonseca H, Santos C (2003). Responses of Olea europea ssp. maderensis in vitro shoots exposed to osmotic stress. *Scientia Horticulturae*, 97: 411-417.

Caruso A, Chefdor F, Carpin S, Depierreux C, Delmotte FM, Kahlem GDM (2008). Physiological characterization and identification of genes differentially expressed in response to drought induced by PEG 6000 in Populus canadensis leaves. *Journal of Plant Physiology*, 165: 932-941.

Chai TT, Fadzillah NM, Kusnan M, Mahmood M (2005). Water stress-induced oxidative damage and antioxidant responses in micropropagated banana plantlets. *Biologia Plantarum*, 49: 153-156.

Dionisio-Sese ML, Tobita S (1998). Antioxidant responses of rice seedling to salinity stress. *Journal of Plant Science*, 135: 1-9.

Ekmekçi M, Kalefetoğlu S (2005). The effect of drought on plants and tolerance mechanisms. G. U. *Journal of Science*, 18: 723-740.

FAO (2021).https://fao.org. Date of access: 19.08.2021

Geçer MK, Yilmaz H (2011). Van ekolojik koşullarında çilek fidesi üretim olanaklarının belirlenmesi. *Yüzüncü Yıl Üniversitesi Tarım Bilimleri Dergisi*, 21: 28-34.

Gong H, Zhu X, Chen K, Wang S, Zhang C (2005). Silicon alleviates oxidative damage of wheat plants in pots under drought. *Plant Science*, 169: 313-321.

Havir K, Mchale A (1987). Biochemical and developmental characterization of multiple forms of catalase in tobacco leaves. *Plant Physiology*, 84: 1291-1294.

Ipek M, Pirlak L, Esitken A, Dönmez MF, Turan M, Sahin F (2014). Plant growth-promoting rhizobacteria (PGPR) increase yield, growth and nutrition of strawberry under high-calcareous soil conditions. *Journal of Plant Nutrition*, 37: 990-1015.

İpek M (2015). In vitro Şartlarda Garnem ve Myrobolan 29C Anaçlarının Kurak Stresine Karşı Tepkilerinin Belirlenmesi. Selçuk Üniversitesi Fen Bilimleri Enstitüsü Bahçe Bitkileri Anabilim Dalı, Doktora Tezi.

Jaleel CA, Manivannan P, Wahid A, Farooq M, Somasundaram R, Panneerselvam R (2009). Drought stress in plants: A review on morphological characteristics and pigments composition. *Int. J. Agric. Biol.*, 11: 100–105.

Jones GH (1990). Physiological aspects of the control of water status in horticultural crops. *HortScience*, 25: 19-26.

Karimi S, Yadollahi A, Nazari-Moghadam R, Imani A, Arzani K (2012). In vitro screening of almond (Prunus dulcis (Mill.)) genotypes for drought tolerance. *J. Biol. Environ. Sci.*, 18: 263-270.

Kutlu N (2010). Tahıllarda kuraklık stresi. *Türk Bilimsel Derlemeler Dergisi*, 3: 35-41.

Mahajan S, Tuteja N. (2005). Cold, salinity and drought stresses. *Biochemistry and Biophysics*, 444: 139-158.

Matysik J, Bhalu BA, Mohanty P (2002). Molecular mechanism of quenching of reactive oxygen species by proline under stress in plants. *Curr. Sci.*, 82: 525-532.

Millar AH, Mittova V, Kiddle G, Heazlewood JL, Bartoli C, Theodoulou FL (2003). Control of ascorbate synthesis by respiration and its implication for stress responses. *Plant Physiology*, 133: 443-447.

Monti LM. (1987). Breeding plants for drought resistance; the problem and its relevance, drought resistance in Plants. Meeting Held in Amalfi, Belgium, 1-8.

Moussa HR, Abdel-Aziz SM (2008). Comparative response of drought tolerant and drought sensitive maize genotypes to water stress. *Australian Journal of Crop Science*, 1: 31-36.

Mundree SG, Baker B, Mowla S, Peters S, Marais S, Willigen CV, Govender K, Maredza A, Muyanga S, Farrant JM, Thomson JA (2002). Physiological and molecular insights into drought tolerance, *African Journal of Biotechnology*, 1:23-38

Özcan S, Babaoğlu M, Gürel E (2004). *Bitki Biyoteknolojisi Genetik Mühendisliği ve Uygulamaları*. Selçuk Üniversitesi Vakfı Yayınları, Konya.

Özfidan C (2010). Ekzojen ABA Uygulamasının Kuraklık Stresi (PEG-6000) Altındaki Yabani ve ABA-eksik Arabidopsis Mutantları Üzerindeki Biyokimyasal Ve Fizyolojik Etkilerinin Araştırılması. Ege Üniversitesi Fen Bilimleri Enstitüsü, Doktora Tezi.

Patel PK, Hemantaranjan A, Sharma BK, Singh R (2011). Growth and antioxidant system under drought stress in chickpea (Cicer arietinum L.) as sustained by salicylic acid, *Journal of Stress Physiology & Biochemistry*, 7: 130-144.

Rahman SML, Mackay AW, Quebedeaux B, Nawata E, Sakuratani T, Mesbahuddin ASM (2002). Superoxide dismutase activity, leaf water potential, relative water content, growth and yield of a drought-tolerant and a drought-sensitive tomato (Lycopersicon esculentum Mill.) cultivars, *Subtropical Plant Science*, 54: 16-22.

Reddy AR., Chaitanya KV, Jutur PP, Sumithra K (2004). Differential antioxidative responses to water stress among five mulberry (Morus alba L.) cultivars, *Environ. Exp. Bot.*, 52: 33-42.

Rostami AA, Rahemi M (2013). Responses of capri fig genotypes to water stress and recovery, *J. Biol. Environ. Sci.*, 7: 131-139.

Sairam RK, Srivastava GC (2000). Induction of oxidative stress and antioxidant activity by hydrogen peroxide treatment in tolerant and susceptible wheat genotypes, *Biologia Plantarum*, 43: 381-386.

Sharma S, Dubey RS (2005). Drought induces oxidative stress and enhances the activities of antioxidant enzymes in growing rice seedlings. *Plant Growth Regulation*, 46: 209-221.

Sircelj H, Tausz M, Grill D, Batic F (2007). Detecting different levels of drought stress in apple trees (Malus domestica Borkh.) with selected biochemical and physiological parameters, *Scientia Horticulturae*, 113: 362-369.

Sivritepe N, Erturk U, Yerlikaya C, Turkan I, Bor M, Ozdemir F (2008). Response of the cherry rootstock to water stress induced in vitro, *Biologia Plantarum*, 52: 573-576.

Tanaka K, Masuda R, Sugimoto T, Omasa K (1990). Water deficiency-induced changes in the contents of defensive substances against active oxygen in spinach leaves, *Agricultural and Biological Chemistry*, 54: 2629-2634.

Wang W, Vinocur B, Altman A (2003). Plant responses to drought, salinity and extreme temperatures: towards genetic engineering for stress tolerance, *Planta*, 218: 1-14.

Yediyıldız AG (2008). Kuraklık ve Tuz Stresi Uygulanan Buğday (Triticum aestivum L.) Çeşitlerinde Antioksidant Enzim Aktivitesindeki Değişimlerin Belirlenmesi, Erciyes Üniversitesi Fen Bilimleri Enstitüsü,Yüksek Lisans Tezi.

Zanjani KE, Rad AHS, Naeemi M, Aghdam AM, Taherkhani T (20129. Effects of zeolite and selenium application on some physiological traits and oil 126 yield of medicinal pumpkin (Cucurbita pepo L.) under drought stress. *Current Research Journal of Biological Sciences*, 4: 462-470.

Selcuk Journal of Agriculture and Food Sciences SJAFS
http://sjafs.selcuk.edu.tr/sjafs/index
(2022) 36 (3), 368-374
e-ISSN: 2458-8377
DOI:10.15316/SJAFS.2022.047
Research Article

Food Expenditure Pattern of Household in Delta State, Nigeria: Economic Rationality Essentials

Osaihiomwan OJOGHO[1], Osagie Stephen IMADE[1]
[1]Benin University, Faculty of Agriculture, Department of Agricultural Economics, Benin, Nigeria.

ARTICLE INFO

Article history:
Received date: 18.03.2022
Accepted date: 26.08.2022

Keywords:
Preferences
AEI
GARP
SARP
SGARP
HARP
CM

ABSTRACT

Parametric utility model that predicts preferences of heterogeneous food consumers is flawed with issues of economic rationality. Using axiomatic preference indexes on food data from 459 household selected from a three-stage sampling procedure, the aim of the study was to check whether a finite set of price and demand observations made on household consumers in Delta state is rationalizable by some form of utility maximization that is common across all households. The study found heterogeneity in food consumption behaviour with evidence against rationality in utility maximization for food expenditure choices at Afriat Efficiency Index (AEI) of unity, violating the GARP, SARP, SGARP, HARP and CM outside the optimum AEI of between 0.536 and 0.982 inclusive. In three of the four food choice categories, households had below the Varian AEI threshold of 0.95. Particularly, household expenditure behavior in the State violated the GARP axiom of revealed preference at 0.018, 0.07, 0.104, 0.05, and 0.081 severity of violations for food in general, protein, carbohydrate, fats and oil, and fruits and vegetables sub-food categories respectively with 5-45% of inconsistent household behaviour. At AEI of unity, 14.30%, 4.79%, 11.43% and 45.04% of the households failed the zero tolerance in the carbohydrate, protein, fats and oil, and fruits and vegetables categories respectively. Only about three to six revealed preferences were found necessary to fully rationalise observed food expenditure choices in the State. Thus, not only are households irrational in utility maximization, there is unstable preference in food demand. Therefore, there is, in the aggregate, no exact one continuous, strictly increasing, piecewise strictly concave, skew-symmetric, and/or homothetic preference function that would completely rationalize households food consumption behaviour in Delta state.

1. Introduction

Consumption patterns contribute greatly to the social and economic policy of a country. For a developing country the consumption pattern is skewed towards food. In Nigeria, household expenditure on food and non-food items in 2019 was over ₦40 trillion with 56.5% spent on food items. Further analysis of food expenditure by households in 2019 showed that various foods consumed outside the home such as starchy roots, tubers and plantains, rice, vegetables, fish and sea food, grains and flours in that order were top in the list of household food items accounting for a combined 59.19% of food expenditure, 33.53% of total household expenditure on food and 24.8% of total household expenditure. Household expenditure on non-food items on the other hand were directed mostly at transport, health, education and services, rent and fuel and light, accounting for a combined 79.40% of non-food expenditure. Consumption pattern for Delta state in 2019 showed that the state had 48.08% non-food expenditure and 51.92% food expenditure in the total expenditure (National Bureau of Statistics, NBS, 2019). However, there are variations in the food consumption expenditure pattern across the country. These have been attributed to income of households and prices of food commodities subject to the demand that household food expenditure choice be explained by the theory only in terms of economic observables.

In Nigeria, household demand analyses have focused on functional forms which embody more general properties with respect to food prices and household income. Studies have determine household demand on the assumption of utility maximization that derived derivative-type conditions on demand functions that are implied by particular utility functional forms (Ojogho

* Corresponding author email: *o.ojogho@uniben.edu*

and Alufohai, 2010b; Ojogho and Ojo, 2017a; Ojogho and Ojo, 2017b; Colen *et.al*, 2018; Masters, *et.al*, 2018; Almås *et.al*, 2019) with the maintained hypotheses of integrability and global negative semi-definiteness of the Slutsky matrix. It is common practice that demand studies on micro-data may reject Slutsky symmetry either due to choice functional form, or no well-behaved form of preferences which rationalises the data on observed food prices and quantities. An important feature of demand data that calls for consideration is that it may be difficult to always find nice, well-behaved preferences that could have rationalised the observed choices (Polisson *et.al*, 2017).

In real life, preferences are not directly observable. Instead, they are discovered from observing household preferences behaviour on the assumption that those preferences remain unchanged while observing consume expenditure behaviour. Though, systematic aggregation of evidence is still surprisingly sparse, there is no reason to think *a priori* that observable economic parameters capture all the variation in preferences. Hence, the assumption that households are rational in the sense that they make choices as if they are maximizing some stable underlying utility function has been critically challenged over the last decades (Arkes *et al.*, 2016; Cason and Plott, 2014). So, modeling heterogeneity in food consumption behaviour, particularly with cycles of indifference, in a single utility maximization function that preserves theoretical consistency and tractability creates specification error. Here, the study asked how rational, in food expenditure choice-consistency, are households to revealed preference axioms, and how many canonical utility functions generate such choice behaviour in the event of inconsistency?

A nonparametric method presents an alternative way of providing answer to such question. The method has been used extensively in literatures (Varian, 1982; Fleissig and Whitney, 2007; Okrent and Alston, 2011). The essence of the approach consists in assessing the consistency of food expenditure data on price and quantity with the generalized (GARP), strong (SARP), and weak axioms of revealed preferences (WARP) (Bergtold *et.al*, 2004). Using nonparametric revealed preference analysis, it is possible to simultaneously test for both symmetry and negative semi-definiteness (Chambers and Echenique, 2016; Crawford and De Rock, 2014). The approach is in contrast to conventional econometric approaches, which typically adopt functional forms and restrict observed and unobserved heterogeneity *a priori*. Within the literature on demand for food in Nigeria, GARP and WARP have not been applied to demand for food. The study bridges the gap in empirical literature on food demand analysis in Nigeria.

Using household-level micro-data on observed food prices and quantities, the study examined household rationality consistency with utility maximization in Delta state, Nigeria. To achieve that, the study tested rationalizability of preferences for observed household food data on prices and quantities in the state, and determine minimum number of utility functions necessary to fully rationalise behaviour of households on food expenditure choices in the state. The purpose of the study is to test whether the food expenditure allocations selected by households are compatible with utility maximization taking a number of different forms without allowing any part of the consumer's expenditure to be "wasted". The results of the study shed light on caution in aggregating utility functions of household food demand in explaining household food data on price and quantity for policy thrust.

2. Methodology

The data for the study were drawn from a target population of households in Delta state, Nigeria. The State lies approximately between latitude 5°00' and 6°30' N and longitude 5°00' and 6°45' E with a total land-mass of 18,050 km^2 of which more than 60% is land. It is bounded in the north and west by Edo State, in the east by Anambra, Imo, and Rivers States, in the southeast by Bayelsa State, and on the southern extreme is the Bight of Benin that covers about 160 Km of its coastline. The state has three senatorial districts. The capital city is Asaba, located at the northern end of the state, with an estimated area of 762 km^2, while Warri is the economic nerve center of the state. The 2006 census puts the population of the state at 4,112,445 with males accounting for 50.3% of the population. Although the state is an oil producing State, yet agriculture dominates economic activities. The major agricultural food crops include cassava, rice, plantain, yam, sugar cane, groundnuts, and tomatoes, which are geared towards local and national markets.

A three-stage sampling procedure was used to select households in the state. The first stage used a simple random sampling to select two Local Government Areas (LGAs) from each senatorial district of the State. The LGAs were Ndokwa-east and Oshimili-South in Delta north, Warri-north and Bomadi in Delta-south, and Ethiope-west and Sapele in Delta-central. The second stage involved a simple random sampling three communities in each LGA from a sampling frame of communities in the respective LGAs. The communities were Aboh, Ibrede and Okapi-Oluchi in Ndokwa-east LGA, Asaba, Omeligbona and Ugbolu in Oshimili-South LGA of Delta-north senatorial district, Akugbene, Bomadi and Esanma in Bomadi LGA, Ogheye, Opuama and Tebu in Warri-north LGA of Delta-south senatorial district, and Irobe, Mosaga and Oghara in Ethiope-west LGA, Elume, Ikeransan and Oboba in Sapele LGA of Delta-central senatorial district of the state. The sample size for the study in each community was determined using the sample-size estimator of Krejcie and Morgan (1970) at 95% confidence interval and 0.05 degree of accuracy. The sample-size estimator is given as:

$$s_i = \frac{\chi^2 N_i P(1-P)}{d^2(N_i-1)+\chi^2 P(p-1)} \quad [1]$$

Where s_i is the sample size of the i^{th} community, N_i is the maximum target population proportion of the i^{th} community, ${\chi^2}_{0.05,1} = 3{,}841$, and $d = 0.05$. A household was identified by its household head which is usually the self-reported head. A simple random sample of households in each community was then taken from the target population developed from a pilot survey. The sample size were respectively 24 from Aboh, 28 from Ibrede and 19 from Okapi-Oluchi in Ndokwa-East LGA, 24 from Asaba, 28 from Omeligbona and 10 from Ugbolu in Oshimili-South LGA of Delta-North senatorial district, 52 from Akugbene, 36 from Bomadi and 28 from Esanma in Bomadi LGA, 32 from Ogheye, 48 from Opuama and 28 from Tebu in Warri-North LGA of Delta-South senatorial district, and 31 from Irobe, 26 from Mosaga and 23 from Oghara in Ethiope-West LGA, 31 from Elume, 39 from Ikeransan and 32 from Oboba in Sapele LGA of Delta-Central senatorial district of the state. That amounted to 133 households from Delta-North, 224 households from Delta-South, and 182 households from Delta-Central senatorial district of the state making a total sample size of 539 households for the study out of a target population of 588. Only 459 copies of questionnaire were retrieved from the households making a response rate of 85%. Household expenditure measures were on food, home grown food consumed, housing, clothing, education, health, transportation, communication, among other utilities, excluding irregular, one-time expenses arising from special occasions, repayments of loans other than house purchase mortgages, savings and taxes. Household consumption expenditure was scaled to take account of differing household size and composition (Donaldson and Pendakur, 2004; 2006), using a recommended scaling methods by Callan *et. al,* (1996) and O'Neill and Sweetman (1998). The weights were 1 for the first adult in the household, 0.7 for additional household members aged over 14 and 0.5 for household members aged less than 14. The prices of food commodities were measured as the sum of the transactions costs incurred by a household during purchase and the retail prices in naira equivalent per kg, while the quantity consumed of food commodities by a household was the quantities purchased at market price per kg. The study focused empirical analysis on consumption expenditure of households for carbohydrate, protein, fats and oil, fruit and vegetables aggregate food choices for the period of March-November 2018 in the state following the multistage budgeting.

Model Specification for Rationality Test

The study assumed that preferences of elementary food commodities within a sub-group are independent of the consumption of food items other than the sub-group. The empirical content of utility maximization model was particularly captured by $GARP$. The study tested whether observed household food data on prices and quantities, $(\boldsymbol{p}^t, \boldsymbol{x}^t)_{t=1,\dots T}$, satisfied the Generalized Axiom of Revealed Preference (GARP) on a revealed preference characterization of the utility maximization model under full Afriat Efficiency Index (AEI) of Varian (1982). The data were then considered to have been generated by a single utility function that rationalizes all observed demands, otherwise no single utility function exists which explain the choices of all of the households. Failure to satisfy the full Afriat efficiency Index, however, the optimum Afriat efficiency Index (AEI) on a revealed preference characterization of utility maximization model under partial efficiency, $e < 1$ of Halevy *et al.* (2018) was computed. The fraction of *wasted* household expenditure was computed as:$overall\ inefficiency\ Index = 1 - \mathrm{e},\ \forall e: 0 < e < 1$ [2]

If the observed household food data on prices and quantities, $(p^t, x^t)_{t=1,\dots T}$, for the set of household food items satisfy GARP at $0 < e < 1$, then it was concluded that $(p^t, x^t)_{t=1,\dots T}$ are approximately consistent with utility maximization, and that there is an approximate single utility function which explains the choices of all of the households after allowing for optimization error, $1 - e$. The maximal value of e at which the data satisfy $GARP$ gave the extent to which to relax household budget constraint in order for the observed data on prices and quantities to appear to be consistent with utility maximization. A critical cost efficiency index or AEI of unity denotes perfect consistency: The index approaches zero as the behavior becomes more inconsistent and the budget needs to be reduced starkly to eliminate inconsistency (Nitsch and Kalenscher, 2020a).

The analysis was repeated for Weak Generalized Axiom of Revealed Preference (WGARP), Strong Axiom of Revealed Preference (SARP), Weak Axiom of Revealed Preference (WARP), Symmetric Generalized Axiom of Revealed Preference (SGARP), Homothetic Axiom of Revealed Preference (HARP), and Cyclical Monotonicity (CM). The degree to which the consumer fails to minimize expenditure was measured by the *violation index*, given as:

Violation index, $i^t = \frac{p^t x^s}{p^t x^t},\ \ i^t < 1$ [3]

Revealed preference restrictions were used on the observed household food expenditure choice as guide to form two-sided bounds on the minimal exclusive exhaustive partitions of the data. Crawford and Pendakur (2012) affirms that in a framework where finding the minimum number of types is important, unobserved preference heterogeneity is vastly more important than observed demographic heterogeneity. The data were analysed using the *revealedPrefs* R-package of Boelaert (2019) and *rpaxioms* Stata-package of Demetry, *et al.* (2020).

3. Results and Discussion

The expenditure pattern of households in the State is presented in Table 1. The results showed that monthly household expenditure on food and non-food items were

₦42321.26 and ₦130082.90 with food accounting for only about 25%, on average. First, with food occupying a small part of the budget, the households may not be considered as poor but implies that their budgets are more diversified. It suggests that their budgets can be said to be non-food-intensive.

Table 1
Expenditure and Budget Shares Pattern of Households in Delta State

Commodity	Expenditure (₦)	Budget share
Food	42321.26	0.245
Protein	12288.00	0.290
Carbohydrate	15301.43	0.362
Fats and Oil	6936.57	0.164
Fruits	7795.26	0.184
Non-Food	130082.90	0.755
Education	21651.81	0.167
Transportation	7290.80	0.056
Clothing	33045.71	0.254
Housing	34646.00	0.266
Others	33448.57	0.257

Source: Computed from Field Survey, 2018.

This may be attributed to increase in industrialization, commercialization, awareness of the importance of such institutions as formal education, as well as increase in the cost of transportation. Unrelatedly, within the food budget, about 36% was spent on carbohydrate food items, 29% was spent on protein food, followed by fruits and vegetables and least with fats and oil category. This suggests that the composition of household food budget contain more necessities and fewer luxuries. Thus, cheaper, more starchy foods seem to predominant in household food expenditure in the State. This is supported by Ojogho and Ojo (2017a). However, the budget share on food categories are approximately the same amounting to seemly diversified diets. This may be due to the emphasis that nutritionists place on the role of a balanced, or diversified, diet for good health. A related reason is that greater diversity is usually thought to be a good thing in and of itself, which possibly reflects a basic concavity of the utility function as opined by Clements and Jiawei (2017).

The results of the degree to which a household in the State departs from full economic rationality are presented in Table 2. The results showed that, on GARP, the mean AEI for food choice consumption associated with utility maximization is 0.982, which means that the average household's monthly budget on food needs to be reduced by about 2% in order for the data on quantity and price to be exactly rationalizable by utility maximization. In other words, the household is, on average, wasting about 2% of its monthly budget on food in departing from rationality in the form of utility maximization. Thus, a household in the state could have obtained the same level of utility by spending only the fraction (0.982) of what it actually spent to attain current level of utility.

The mean AEI for protein food category consumption associated with utility maximization was 0.930, which means that the average household's monthly expenditure on protein food needs to be reduced by about 7% in order for the household food data on quantity and price in the protein food category to be exactly rationalizable by utility maximization. In other words, there is, on average, a 7% waste of monthly expenditure on protein food by households in departing from rationality with utility maximization. The pattern is similar for fruits and vegetables sub-food category where the mean AEI for utility maximization is 0.930. For fats and oil sub-food category, the mean AEI was about 0.950, which means that the average household's monthly budget on fat and oil sub-food category needs to be reduced by about 5% in order for the data on quantity and price in the fats and oil sub-food category to be exactly rationalizable by utility maximization. This is the only sub-food category where households had the Varian AEI threshold of 0.95. However, the average household is wasting about 5% of its monthly budget on fats and oil food category in departing from rationality with utility maximization. This means that the same level of utility can be achieved by the households through a different combination of food commodities which costs strictly less at the prevailing market prices. It potents a household innate inability to distinguish among similar food bundles as opined by Dziewulski (2020).

Table 2: Rationality Status of Households Food Choice in Delta State

Food Category	Afriat Efficiency Index (AEI)						
	SGARP	WGARP	SARP	WARP	HARP	CM	GARP
Food	0.536	0.917	0.917	0.917	0.891	0.789	0.982
Protein	0.515	0.931	0.931	0.931	0.848	0.715	0.930
Carbohydrate	0.550	0.896	0.896	0.896	0.693	0.773	0.896
Fats and Oil	0.589	0.950	0.950	0.950	0.868	0.776	0.950
Fruits and vegetables	0.610	0.911	0.912	0.910	0.856	0.810	0.919

Source: Computed from Field Survey, 2018; values in parentheses are the $overall\ inefficiency\ Index$

The results also showed that the mean AEI for carbohydrate food choice category associated with utility maximization was about 0.896, which means that the average household's monthly budget on food needs to be reduced by about 10.4% in order for the data on quantity and price on carbohydrate food category to be exactly rationalizable by utility maximization. Households in the State are spending about extra 10% of its monthly budget on carbohydrate food in departing from rationality with utility maximization. It implies that about 90% of food expenditure choice on carbohydrate sub-food category by households is rationalizable by utility maximization. The margin between their present optimization level and the level of perfect (100%) efficiency of optimization in the carbohydrate food category is too wide to be attributed to measurement errors, but to optimization error. The results showed that food expenditure choice behavior of households is below the Varian (1991) AEI threshold of 0.95. The optimization error resulting in departure from rationality in the form of utility maximization may be due to the heterogeneity in budget share and food choices.

Households were least rational in the carbohydrate food choice category, and most rational in the fats and oil food choice category in the State. The results imply that without allowing for optimization error, the household consumption data in the State on quantity and price for food and sub-food categories violate the GARP axiom of revealed preference with 0.018, 0.07, 0.104, 0.05, and 0.081 severity of violations for food in general, protein, carbohydrate, fats and oil, and fruits and vegetables food categories respectively. Similarly, households were cost inefficient at the above respective values. For household food consumption data on quantity and price, for example, in Delta State to be exactly rationalizable by a utility maximization function, about 2% minimal expenditure adjustment is required in order for the data to comply with GARP. This agrees with Dean and Martin (2016) that shows that the minimal cost to make a revealed preference relation acyclic can be relatively small.

Optimum Afriat Efficiency Index of less than unity implies that there were violations of GARP in the household food choice data on price and quantity. Households in the state can be said to have wasted money, as they did not obtain the maximum subjective value for their money. The results also showed that the household food expenditure choice violated the SARP, SGARP, HARP and CM at Afriat Efficiency Index of unity. These imply that household food expenditure choice data in the State cannot be exactly rationalized by a continuous, strictly increasing and concave utility function that is symmetric, homothetic, or quasilinear. Thus, household behaviour on food expenditure choice is not of the maximizing behaviour. It is implied that the household food data on price and quantity in the State are not generated by household with stable preferences who was always choosing the best food expenditure choice they could afford. The violation can only be attributed to households not choosing the most preferred food expenditure choice alternative that was affordable given their budget, rather selecting another less preferred option. An alternative plausible explanation for the non-rationalisation is either the theory of utility maximization is wrong for these households, or tastes, food prices and household income have changed for the households in the State. It suggests a symptom of unstable preference in food demand by households in the State. The unstable preference could also be predicated on changes in information about the health consequences of diet.

Table 3 presents the results of the partitions of households in the State by revealed preferences. The results showed that the number of types needed to completely explain all observed variation in consumption behaviour of households is quite small relative to the number of observations in household food expenditure data on quantity and price of household food consumption. The results showed that about three revealed preferences for food, four for the protein category, five each for the carbohydrate and fats and oil, and six revealed preferences for fruits and vegetables categories of food demand in the State are necessary to fully rationalize all observed choices in a data set with 459 observations of price and quantity vectors respectively. This implies that there are a minimum of three, four, and six groups which maximize different utility functions of food, protein, and fruits and vegetables demand in the State to completely rationalize all the observed variation in food expenditure choice behaviour. The results imply that there is unobserved preference heterogeneity in food expenditure choice among households in the state. Thus, there is no indifference curve that could be drawn, in aggregate, for household in the state that would make food choice bundles maximizing bundles. Instead, all of the households within each group of revealed preference can be modelled as having a common well-behaved utility function such that within-groups, a single utility function is sufficient to rationalize all the observed household food expenditure choice behaviour. Hence, modelling strategies with a small number of discrete types might be better in explaining food consumption expenditure behaviour of households in the State. This is in line with De Clippel and Rozen (2018) that having all possible observations is necessary for assessing some models while the results change when it is not the case.

Table 3
Partitioning of Households in the State by Revealed Preferences in Delta State

Food Category	Number of Preference Types	
	Lower Bounds	Upper Bounds
Food	1 [140]	3 [171, 3, 1]
Protein	1 [171]	4 [136, 29, 9, 1]
Carbohydrate	2 [156, 160]	5 [120, 24, 16, 10, 4]
Fats and Oil	2 [151, 153]	5 [123, 31, 14, 6, 1]
Fruits and Vegetables	1 [89]	6 [101, 35, 19, 9, 3, 8]

Source: Computed from Field Survey, 2018; the number of lower and upper bounds are reported at efficiency level, $e = 1$.

The results of the number and percentage of violators in food choice by preference indicators are presented in Table 4. The results showed that 14.30%, 4.79%, 11.43% and 45.04% of the households failed the 1.00 tolerance for Afriat efficiency in the carbohydrate, protein, fats and oil, and fruits and vegetables categories respectively. These suggest that between 5-45% of households were inconsistent with GARP, and had Afriat Efficiency below 0.98. The results also imply that households had the least number of violations of GARP in carbohydrate sub-food choice category but highest number of violations of GARP in the fruits and vegetables food choice category in the state. Of the four food choice categories, households violated one or more of the revealed preference axioms. Of these, households in all food categories had violations at Afriat efficiency indices of less than unity, and three of those were below the Varian (1991) Afriat efficiency index threshold of 0.95.

Table 4
Households Violations of Revealed Preference in Food Choice in Delta State by Preference Axioms

Variables	Number and Percentage of Violations						
	GARP	SGARP	WGARP	SARP	WARP	HARP	CM
Carbohydrate							
Number of violators	4304	18661	220	4312	220	174	174
Percentage of violators	(14.30)	(61.64)	(1.46)	(14.32)	(1.46)	(100.00)	(100.00)
Protein							
Number of violators	1442	17642	86	1597	124	174	174
Percentage of violators	(4.79)	(58.27)	(0.57)	(5.31)	(0.82)	(100.00)	(100.00)
Fats and Oil							
Number of violators	3442	14343	189	3514	204	174	174
Percentage of violators	(11.43)	(47.37)	(1.26)	(11.67)	(1.36)	(100.00)	(100.00)
Fruits and vegetables							
Number of violators	13559	18471	362	13571	363	174	174
Percentage of violators	(45.04)	(61.01)	(2.41)	(45.08)	(2.41)	(100.00)	(100.00)

Source: Computed from Field Survey, 2018; number and percentage of violators are reported at efficiency level, $e = 1$.

4. Conclusion

Using household-level micro-data on observed food prices and quantities, the study examined expenditure pattern of rural households in the light of economic rationality consistency with utility maximization in Delta state, Nigeria. To achieve that, the study tested rationalizability of preferences for observed food data on prices and quantities of household in the State, and determine minimum number of utility functions necessary to fully rationalize the behaviour of households on food expenditure choices in the State. The study found heterogeneity in food consumption behaviour with evidence against rationality in utility maximization for food expenditure choice at Afriat Efficiency Index (AEI) of unity. Food expenditure Households in the State violated the GARP, SARP, SGARP, HARP and CM outside the optimum AEI of between 0.536 and 0.982 inclusive. In three of the four food choice categories, households had below the Varian AEI threshold of 0.95. Particularly, observed expenditure for food and sub-food categories in the State violated the GARP axiom of revealed preference at 0.018, 0.07, 0.104, 0.05, and 0.081 severity of violations for food in general, protein, carbohydrate, fats and oil, and fruits and vegetables sub-food categories respectively with 5-45% of inconsistent households. At AEI of unity, 14.30%, 4.79%, 11.43% and 45.04% of the households failed the zero tolerance in the carbohydrate, protein, fats and oil, and fruits and vegetables categories respectively. About three to six revealed preferences were found necessary to fully rationalize the observed food expenditure choices in the state. Another essential observation from the study is that households are heterogeneous in food consumption behaviour. Accounting for this unobserved heterogeneity in food choice behaviour of households will be a necessary part of understanding food choices in the State. Following economic rationality essentials, households in the State are irrational in utility maximization with unstable preference in food demand. Thus, that it is indeed not possible to capture household

food expenditure choices in Delta state, in the aggregate, with exactly one continuous, strictly increasing, piecewise strictly concave, skew-symmetric, and/or homothetic preference function that completely rationalize households food consumption behaviour in Delta state.

Based on the findings, food suppliers should design their marketing strategies to address the needs of various consumer groups and food categories in the state. Similarly, policymakers should design policies that would achieve different effectiveness in different consumer segments *vis-a-vis* food categories rather than looking at food customers as a homogenous group. Food demand analysts would need to consider heterogeneity in empirical utility model specifications, both among food consumers and food categories, if they must use a single utility function.

5. References

Almås I, Haushofer J, Shapiro JP (2019).The Income Elasticity for Nutrition: Evidence from Unconditional Cash Transfers in Kenya. *NBER Working Paper* 25711.

Arkes HR Gigerenzer G, Hertwig R. (2016). How bad is incoherence? *Decision*, 3*(1)*: 20-39. https://doi.org/10.1037/dec0000043

Boelaert J (2019). revealedPrefs: Revealed Preferences and Microeconomic Rationality. R package version 0.4.1. https://CRAN.R-project.org/package=revealedPrefs

Bergtold J, Akobundu E, Peterson EB (2004). The Fast Method: Estimating Unconditional Demand Elasticities for Processed Foods in the Presence of Fixed Effects. *Journal of Agricultural and Resource Economics* 29*(2)*:276-295

Callan T, Nolan B, Whelan B, Whelan C, Williams T (1996). *Poverty in the 1990s: Evidence from the Living in Ireland Survey*, Dublin: Oak Tree Press.

Colen LPC, Melo Y, Abdul-salam D, Roberts SM, Paloma SGY (2018). Income Elasticities for Food, Calories and Nutrients across Africa: A meta-analysis. *Food Policy* 77:116-132.

Cason TN, Plott CR (2014). Misconceptions and Game Form Recognition: Challenges to Theories of Revealed Preference and Framing. *Journal of Political Economy*, 122*(6)*:1235-1270. https://doi.org/10.1086/677254

Clements KW, Jiawei SI (2017). Engel's Law, Diet Diversity, and the Quality of Food Consumption. *American Journal of Agricultural Economics,* 100 *(1*): 1-22

Crawford I, Pandakur K (2012). How Many Types Are There? *The Economic Journal*, 123: 77–95

Dean M, Martin D (2016). "Measuring Rationality with the Minimum Cost of Revealed Preference Violations". In: The Review of Economics and Statistics 98.3, pp. 524–534. doi: 10.1162/REST_a_00542.

Clippel G, Rozen K (2018). "Bounded rationality and limited datasets". In: mimeo. url: https://www.brown.edu/Departments/Economics/Faculty/Geoffroy%7B%5C_ %7DdeClippel/lim-data.pdf.

Demetry M, Hjertstrand P, Polisson M (2020). Testing Axioms of Revealed Preference in Stata. IFN Working Paper Number 1342.

Donaldson D, Pendakur K (2004). "Equivalent-Expenditure Functions and ExpenditureDependent Equivalence Scales", *Journal of Public Economics*, 88, 175-208.

Donaldson D, Pendakur K (2006). "The Identication of Fixed Costs >From Consumer Behaviour", *Journal of Business and Economic Statistics*, 24, 255-265.

Dziewulski P (2020). "Just-Noticeable Difference as a Behavioural Foundation of the Critical CostEfficiency Index." *Journal of Economic Theory*, 188: 105071.

Fleissig AR, Whitney GA (2007). Testing Additive Separability, *Economics Letters*, 96*(2):* 215-220.

Masters WA, Bai Y, Herforth A, Sarpong D, Mishili F, Kinabo J, Coates J (2018). Measuring the Affordability of Nutritious Diets in Africa: Price Indexes for Diet Diversity and the Cost of Nutrient Adequacy. *American Journal of Agricultural Economics* 100:1285-1301.

Nitsch FJ, Kalenscher T (2020a). *Keeping a cool head at all times. What determines choice consistency? https*://doi.org/10.31234/osf.io/etyhx

Okrent AM, Alston JA (2011). Demand for Food in the United States. Davis, CA: University of California, Davis, Giannini Foundation Monograph 48.

Ojogho O, Alufohai GO (2010b). Impact of Price and Total Expenditure on Food Demand in South-Western Nigeria. *African Journal of Food, Agriculture, Nutrition and Development*, 10 (11): 4350-4363.

Ojogho O, Ojo MP (2017a). Impact of Food Prices on the Welfare of Rural Households in South Eastern Nigeria, *Applied Tropical Agriculture*, 22*(1)*: 142-148

Ojogho O, Ojo MP (2017b). Linear Approximate Almost Ideal Demand System of Food Consumers Behaviour towards Price and Income in South-eastern Nigeria. *International Journal of Agricultural Economics and Rural Development*, 9*(1)*: 33-40

O'Neill D, Sweetman O. (1998). Poverty and Inequality in Ireland 1987-1994: A Comparison Using Measures of Income and Consumption, *mimeo*.

Polisson M, Quah JKH Renou L (2017). Revealed Preferences over Risk and Uncertainty," School of Economics and Finance Discussion Papers, 1706, University of St. Andrews.

Varian HR (1982). The Nonparametric Approach to Demand Analysis, *Econometrica*, 50*(4)*: 945-973.

Varian HR (1991). Goodness of Fit for Revealed Preference Tests, University of Michigan CREST Working Paper Number 13.

Selcuk Journal of Agriculture and Food Sciences SJAFS

http://sjafs.selcuk.edu.tr/sjafs/index

(2022) 36 (3), 375-379

e-ISSN: 2458-8377

DOI:10.15316/SJAFS.2022.048

Research Article

Identification and Technological Characterisation of Lactic Acid Bacteria Isolated From Traditional Algerian Cheese "J'ben"

Bounaama KHALIL*[,1], Dahou Abdelkader EL-AMINE[1], Meskini ZAKARIA[1], Abbad ABDERRAHMEN[1], Tahlaiti HAFIDA[1], Yerrou HOUARI[2], Homrani ABDELKADER[1]

[1] Mostaganem University, Faculty of Nature and Life Sciences, Department of Agriculture, Mostaganem, Algeria

[2] Mascara University, Faculty of Nature and Life Sciences, Department of Agriculture, Mascara, Algeria

ARTICLE INFO

Article history:
Received date: 28.02.2022
Accepted date: 30.08.2022

Keywords:
J'ben cheese
Identification
Lactic acid bacteria
Technological properties

ABSTRACT

Lactic acid bacteria have long been utilized in fermented foods and dairy products such as cheese, these bacteria play an important role in food bio-preservation, organoleptic properties development, and quality improvement. The purpose of this research was to determine and assess the biotechnological characteristics of lactic acid bacteria isolated from traditional Algerian cheese "J'ben". Fifteen lactic acid bacteria (gram positive, catalase negative) were molecularly identified according to their 16S rDNA sequences, six belonged to *Enterococcus durans*, three to *Enterococcus faecium*, three to *Lactococcus lactis*, and three to *Leuconostoc mesenteroide*. The strains were evaluated for proteolysis, lipolysis, antibacterial activity, exopolysaccharide synthesis, and safety (hemolytic activity). All studied strains had considerable proteolytic activity but no lipolysis potential, they were also all γ-hemolytic. The antimicrobial activity of the stains against three pathogenic bacteria (*Staphylococcus warneri, Serratia plymuthica, and Enterobacter aerogenes*) revealed that they were active against at least one of them. Finally, only three organisms produced exopolysaccharide in our study *Enterococcus durans* (KC1); *Leuconos-toc mesenteroide* (KC6); and *Lactococcus lactis* (KC15). These findings suggest that the lactic acid bacteria isolated from traditional cheese "J'ben" have significant technological propereties, making them suitable for use as starter culture in fermented dairy products.

1. Introduction

The microbial flora, particularly lactic acid bacteria (LAB), is an important element in the production of fermented foods and fermented milk products such as cheese. J'ben is a traditional soft white cheese prepared from non-pasteurised raw milk (sheep, goat, or cow). J'ben is a popular cheese in rural regions due to its unique taste and nutritional properties offered by its indigenous flora (Bousbia et al., 2018; Dahou et al., 2021). Lactic acid bacteria are a group of organisms that are Gram-positive, non-sporulating, cocci, coccobacilli, or rods, catalase negative, and have the common property of producing lactic acid. They are classified into the following genera: *Lactobacillus, Lactococcus, Streptococcus, Enterococcus, Oenococcus, Pediococcus, Leuconostoc, Tetragenococcus, Aerococcus, Carnobacterium, Weissella, Alloiococcus, Symbiobacterium, and Vagococcus* (Pfeiler & Klaenhammer, 2007; Kocková et al., 2011; Wedajo, 2015; Toe et al., 2019).

LAB are used to improve organoleptic properties, as probiotic organisms, and as bio-conservation agents (Wedajo, 2015; Karakas-Sen & Karakas, 2018; Göktepe & Elgün, 2020). The most isolated Lactic acid bacteria from raw milk and dairy products are *Enterococcus, Lactococcus, Leuconostoc, Lactobacillus, and Streptococcus* (Franciosi et al., 2009; Karakas-Sen & Karakas, 2018).

The biodiversity of LAB is a crucial factor that has a direct influence on the characteristics and quality of artisanal products (Franciosi et al., 2009). Many variables influence the formation of cheese's organoleptic features, including the milk source and its microbiota, the processes and techniques used to make the cheese, ripening circumstances and others. However, lactic acid bacteria have the most important role in cheese aroma, texture, and flavor by generating substances such as exopolysaccharides (EPS) (Dal Bello et al., 2001; Herreros et al., 2003; Gobbetti et al., 2018).

* Corresponding author email: *bounaamakhalilvet@gmail.com*

The LAB has the desired effect on cheese by developing sensory characteristics that are connected to metabolic processes like proteolysis and lipolysis during maturation (Bruno & Carvalho, 2009; Luiz et al., 2016; Farahani et al., 2017; Asensio-Vegas et al., 2018).

The LAB contributes to the proteolysis of cheese; they have proteinases enzymes associated with their cell wall that can degrade casein into peptides, and these free peptides of casein are hydrolyzed to free amino acids by the action of peptidases. The proteolysis process is important for the growth of LAB themselves, the development of flavor in dairy products, and the maturation of cheese (Tulini et al., 2016; Toe et al., 2019).

Lipolysis is the hydrolysis of triglycerides to produce glycerol, glycerides, and free fatty acids; this metabolic process aids in the development of texture, flavor, and aroma in products such as cheeses (Collins et al., 2003; Esteban-Torres et al., 2014).

Because of the many enzymatic reactions, fermentation is a complicated process (García-Cano, 2019). Furthermore, fermentation reduces pH and produces antimicrobial chemicals such as lactic acid, hydrogen peroxide, diacetyl, acetaldehyde, reuterin, and peptides as a result of lipolysis and proteolytic enzyme activity (Papagianni, 2012; Burgain et al., 2014).

The objectives of this paper were to characterize some technological properties of lactic acid bacteria isolated from traditional Algerian cheese, including their ability to produce exopolysaccharides as well as their proteolytic, lipolysis, haemolytic, and antimicrobial activity for use in fermented dairy products.

2. Materials and Methods

Samples collection and Bacterial isolation

Traditional cheese "J'ben" samples were collected from Naâma region, south-west of Algeria. Samples were aseptically obtained and transported to the laboratory in isotherm container at 4° C under sterile conditions.

For each sample, an initial dilution of 10g and 90 mL of physiological water was made, followed by homogenization in a Stomacher (seward STOMACHER, England). Successive decimal dilutions were made up to 10^{-7}, and 100 µL of each dilution was disseminated onto MRS and M17 agar (pH 6.5). The plates were incubated for 48 to 72 hours at 30°C under both aerobic and anaerobic conditions. Individual colonies with varied morphologies were transferred to M17 or MRS agar plates and were purified by sequential streaking into the same medium. The pure isolates of lactic acid bacteria were kept at 4°C and were renewed every month. The purified isolates were stored at -20°C in MRS or M17 broth containing 20% (v/v) glycerol.

Characterization and molecular identification of lactic acid bacteria isolates

The selected isolates were subjected to preliminary identification of morphology, gram staining, and catalase testing. The isolates that were gram positive, catalase negative, were considered to be lactic acid bacteria.

Fifteen isolates presenting characteristics of lactic acid bacteria were molecularly identified using 16S rDNA amplification and sequencing as fellow:

Extraction of total DNA of isolates.

Amplification of the DNA fragments by PCR in a Thermocycler (Biorad, USA) using specific primer (Qbiogene Research Service, Germany) for the 16S rDNA sequences, the following three steps were repeated for 35 cycles: denaturation at 94°C for 3 min, annealing at 53°C for 1 min, and extension at 72°C for 2 min. then the final extension at 72°C for 5 min. The amplified fragment was screened on agarose gel.

Sanger sequencing technique was used to sequence the amplified fragment of DNA. the resulting sequences were matched with GeneBank data, using the NCBI Blast (https://blast.ncbi.nlm.nih.gov/Blast.cgi)

Proteolytic and lipolysis activity

The selected stains were screened for their Proteolytic activity on skim milk agar medium (casein 0.5%, yeast extract 0.25%, dextrose 0.1%, and agar 1.5%) supple-mented with 10% of reconstituted skim milk. Wells of 6 mm diameter were prepared on the plate. Each well was inoculated by 50 µl of bacterial culture. Plates were then incubated at 30°C for overnight. The existence of a clear or opaque zone surrounding the wells indicated positive protease activity.

Tributyrin agar was used to assess lipolysis activity. Plates were incubated at 30°C for overnight. Positive activity was defined as the occurrence of clean zones surrounding the wells in Tributyrin Agar (Oliveira et al., 2020).

Exopolysaccharide production

Strains were cultivated on modified MRS medium with 50g/L of sucrose as described by Franciosi et al. (2009), and after 72 hours of incubation at 30°C, the plates were tested for the emergence of mucoid properties.

Haemolytic activity

Haemolysis was assessed using Columbia blood agar plates supplemented with 5% sheep blood and incubated for 48 hours at 30°C according to Jikang and Wenxiang, (2019). Plates were examined for the presence of β-haemolysis (clear zones around colonies), α- haemolysis (green-hued zones around colonies), or γ -haemolysis (no zones around colonies).

Antimicrobial activity

The antimicrobial activity of the isolates was tested against three subclinical mastitis pathogen germs (*Staphylococcus warneri, Serratia plymuthica and Enterobacter aerogenes*) isolated from subclinical cases of intramammary infection on LSTPA lab by Meskini et al. (2021). Agar well diffusion method was used to evaluate the antimicrobial activity of LAB according to Bhola &

Bhadekar, (2019). Briefly, a culture broth of 0.5 OD (McFarland Standard) of LAB and pathogen bacteria was prepared; indicator bacteria were swabbed on agar medium containing TSA+MRS (1:1). Wells of 6 mm diameter were prepared on these pre-swabbed medium and 50 µl of each LAB strain was inoculated in the corresponding well. The plates were then incubated overnight at 37°C then observed for the presence of inhibition halos.

3. Results and Discussion

After preliminary identification, fifteen isolates showing appearances with lactic acid bacteria were chosen at random for further testing. The isolates were all gram positive, catalase negative, and cocci shaped. The 16S rDNA gene sequence data revealed that isolates KC1, KC5, KC7, KC9, KC10, and KC13 were 100% similar to *Enterococcus durans*, whereas isolates KC2, KC3, and KC4 were 100% similar to *Enterococcus faecium*. KC6, KC8, and KC11 showed 100% resemblance to *Leuconostoc mesenteroide*, whereas KC12, KC14, and KC15 had 100% similarity to *Lactococcus lactis*.

Proteolytic and lipolytic activities are highly desired criteria in LAB for use as starter culture in cheese production since these two processes occur and play a vital role during cheese maturation (Bruno & Carvalho, 2009). The proteolytic system is critical in fermentation processes and allows the growth of LAB in milk; this system contains enzymes that hydrolyze casein and provide necessary amino acids to cells (Mayo et al., 2010; Balthazar et al., 2017; Farahani et al., 2017; Karakas-Sen & Karakas, 2018). The proteolytic system also contributes to the formation of several organoleptic properties in various fermented milk products (Savijoki et al., 2006; Gobbetti et al., 2018). In this study, lactic acid bacteria strains were tested for protease and lipolysis activities (Table 1). The results showed that all tested strains exhibited proteolytic activity by giving a proteolysis zone on milk agar plates, 06 strains of *E. durans* presented a diameter zone ranging between 14 and 23.5 mm, 03 strains of *E. faecium* between 12.5 to 14.5 mm, 03 strains of *Lc. lactis* between 12 to 12.5 mm, and 03 strains of *Leu.mesenteroides* between 17.5 to 22.5 mm, while none of the fifteen tested strains exhibited lipolysis activity in Tributyrin agar medium (Table1). Mechai et al. (2014) found that only two strains of the genus *Lactococcus* isolated from fermented milk and traditional cheese had strong proteolytic activity; however, Dahou et al (2021) found that ten *Lactococcus lactis* isolated from traditional Algerian cheese "J'ben" had proteolytic activity. Furthermore, another study found similar characteristic in one *Lactococcus* (*Lc. lactis subsp. Lactis*) and two *Enterococci* (*E. durans*) isolated from raw cow's milk (Franciosi, 2009). All of our selected LAB were classified as having significant proteolytic activity (halo size > 10mm), indicating their usefulness in the development of starter cultures for cheese maturation.

None of our strains had lipolysis activity; according to McSweeney and Sousa (2000) LAB are known to have limited lipolysis activity when compared to other microorganisms such as pseudomonas.

Table 1
Antimicrobial, proteolytic, and lipolysis activities of strains isolated from traditional cheese “J’ben”

Isolates	Activity zone (mm)				
	AA			PA	LA
	Serratia plymuthica	*Staphylococcus warneri*	*Enterobacter aerogenes*		
KC1	16 ± 0.00	12 ± 2.00	9 ± 4.50	15 ± 1.00	-
KC2	17.5± 7.50	13.5 ± 0.50	7 ± 1.50	14.5 ± 0.50	-
KC3	-	12.66 ± 0.94	7.33 ± 2.00	12.5 ± 0.50	-
KC4	-	15 ± 0.81	7.66 ± 2.50	13.5 ± 0.50	-
KC5	-	14 ± 1.41	7.66 ± 2.50	23.5 ± 1.50	-
KC6	22 ± 3.00	11 ± 1.00	-	11.5 ± 1.50	-
KC7	20 ± 5.00	15 ± 2 .00	-	17.5 ± 2.50	-
KC8	11 ± 1.00	14 ± 1.00	-	16.5 ± 2.50	-
KC9	20.5 ± 5.50	15.33 ± 0.47	-	18 ± 2.00	-
KC10	8 ± 0.00	12 ± 3.55	-	21.75 ± 0.25	-
KC11	9 ± 1.00	12.33 ± 3.29	-	15 ± 1.00	-
KC12	16 ± 8.00	15 ± 0.0	-	12 ± 0.00	-
KC13	17.5 ± 7.5	13.5 ± 0.5	-	14 ± 2.00	-
KC14	16 ± 1.00	13.5 ± 1.5	-	12.5 ± 0.50	-
KC15	7 ± 0.00	9.66 ± 0.47	-	12 ± 0.00	-

PA (proteolytic activity) and LA (lipolytic activity): Halo size >10 mm (very high), 3-10 mm (high) and <3 mm (low) for proteolytic and lipolytic activities (Alapont et al., 2015);
AA (Antimicrobial activity); (antimicrobial activity): Halo size >8 mm (strong), 4-8 mm (moderate) and 1-4 mm (weak) for antimicrobial activity (Akabanda et al., 2014);
- (absence of activity);
Enterococcus durans: KC1, KC5, KC7, KC9, KC10, KC13; *Enterococcus faecium*: KC2, KC3, KC4; *Leuconostoc mesenteroide*: KC6, KC8, KC11; *Lactococcus lactis*: KC12, KC14, KC15

In terms of exopolysaccharide synthesis, three strains (KC1, KC6, and KC15) were able to produce EPS when grown on MRS agar medium supplemented with sucrose (50g/L) as a carbon source. Because of the importance of this chemical compound in the pharma-

ceutical and food sectors, the production of exopolysaccharides is an important feather (Rajoka et al., 2018). EPS influences the stability and organoleptic properties of fermented foods including texture and flavor. Some EPS have a prebiotic impact on human health because they aid in adherence to human mucosa and also have antibiofilm and immunomodulation properties (Dal Bello et al., 2001; Liu et al., 2011; Russo et al., 2012; Rendueles et al., 2013). Three strains (KC1, KC6, and KC15) produced EPS, earning them the right to be chosen as the starter strains.

The existence of a lytic zone on blood agar plate was used to assess the haemolytic activity of our strains. The absence of haemolytic activity is an important indicator of safety when selecting a probiotic strain (Jikang & Wenxiang, 2019). Bacteria with haemolytic activity can produce haemolysin and harm human or animal red blood cells. Our finding was that none of the strains showed a β or α haemolytic activity; instead, they were all γ-haemolytic (no zones surrounding the colonies), showing safety for food or probiotic application.

In this work, antibacterial activity of the isolated LAB was investigated against subclinical mastitis pathogenic bacteria (Table 1).

The agar well diffusion technique exhibited results ranging from 0 to 17.5 mm for various bacteria. Antibacterial activity against *Serratia plymuthica* revealed that 8 LAB strains showed high inhibition (diameter ≥16 mm) whereas 3 strains showed no activity, and antibacterial activity against *Staphylococcus warneri* revealed that 14 strains presented inhibition zone (diameter ≥12 mm) except for KC15. Finally, for *Enterobacter aerogenes* 10 strains exhibited no inhibition activity, whereas 05 showed mild action.

Lactic acid bacteria are widely used in the food industry as starter cultures and bio-preservatives against pathogenic microorganisms by producing compounds with both technological and antimicrobial properties such as organic acids, diacetyl, reuterin, hydrogen peroxide, and bacteriocins. Many studies have proven the ability of LAB related the *Enterococci*, *Leuconostoc*, and *Lactococcus* groups to inhibit various microorganisms, as well as their utility in the food industry and medical sector. Karakas-Sen and Karakas (2018) revealed that two *Lactococcus lactis* had good antimicrobial activity, while López-Seijas et al. (2020) described an interesting inhibitory effect of *Lactococcus lactis* against the genus *Staphylococcus*; and Musikasang et al., (2009) isolated two *Enterococcus* (*E. faecium and E. durans*) that had important probiotic properties including a high antimicrobial activity. Finally, Morandi et al. (2013) identified 35 *Leuconostoc* strains isolated from North Italian typical cheese including *Ln. mesenteroid* with antimicrobial activity against several indicator bacteria.

The antibacterial activity of the traditional cheese J'ben LAB (at distinct genus *Enterococci*, *Leuconostoc*, and *Lactococcus*) identified in this study classified them as interesting bio-conservative agents. The antibacterial property may be necessary during cheese production to decrease the usage of chemical conservatives.

4. Conclusion

Traditional cheese "J'ben" is a good source with a wide diversity of lactic acid bacteria, and with interesting potential and technological properties. All strains displayed considerable proteolytic activity and their antagonistic effect varied depending on the LAB strain and pathogenic bacteria. LAB of our research exhibited no haemolysis classifying them safe to be used, which is an important characteristic for the selection of probiotic strains and starter cultures. The isolates KC1 and KC6, which exhibited high proteolytic and antibacterial activity, were able to create exopolysaccharides, and were safe (no haemolysis), and can be considered viable candidates for use in fermented dairy products, particularly in the production and maturation of cheese.

All strains had biotechnological properties and the potential for future application as a starter for fermented products or as a probiotic organism; however, more tests will be required to prove the efficiency of these strains and their effect on the quality of fermented products in order to confirm the beneficial role for human and animal health.

5. Acknowledgements

We would like to thank, the Directorate General of Scientific Research and Technological Development "DGRSDT".

6. References

Akabanda F, Owusu-Kwarteng J, Tano-Debrah K, Parkouda C, Jespersen L (2014). The use of lactic acid bacteria starter culture in the pro-duction of Nunu, a spontaneously fermented milk product in Ghana. *International Journal of Food Sciences*, 2014.

Alapont C, Martínez-Culebras P V, López-Mendoza, M C (2015). Determination of lipolytic and proteolytic activities of mycoflora isolated from dry-cured teruel ham. *Journal of Food Science and Technology*, 52(8), 5250-5256.

Asensio-Vegas C, Tiwari B, Gredilla A E, Bueno F, Delgado D, Martín-Diana A B (2018). Devel-opment of yoghurt from ovine milk with en-hanced texture and flavour properties. *Inter-national Journal of Dairy Technology* 71(1): 112–121.

Balthazar C F, Pimentel T C, Ferrão L L, Almada C N, Santillo A, Albenzio M, Mollakhalili N, Mor-tazavian A M, Nascimento J S, Silva M C, Freitas M Q, Sant'Ana A S, Granato D, Cruz A G (2017). Sheep Milk: Physicochemical Char-acteristics and Relevance for Functional Food Development. *In Comprehensive Reviews in Food Science and Food Safety* 16(2) :247-262.

Bhola J, Bhadekar R (2019). Invitro synergistic activity of lactic acid bacteria against multi-drug re-sistant staphylococci. *BMC Complementary and Alternative Medicine* 19(1): 1–8.

Bousbia A, Boudalia S, Guerioui Y, Belaize B, Meguelati S, Amrouchi M, Ghebache R, Belk-heir B, Benidir M (2018). Nutritional and hy-gienic quality of raw milk intended for consumption in the region of Guelma, Algeria. *Asian Journal of Dairy and Food Research*. 37(3): 192-196.

Bruno L M, & Carvalho J D C (2009). Microbiota láti-ca de queijos artesanais. *Brasília*: EMBRAPA.

Burgain J, Scher J, Francius G, Borges F, Corgneau M, Revol-Junelles AM, Cailliez-Grimal C, Gaiani C (2014) Lactic acid bacteria in dairy food: surface characterization and interactions with food matrix components. *Advances in colloid and interface science* 213:21–35.

Collins F, McSweeney P L H, Wilkinson M G (2003). Lipolysis and free fatty acid catabolism in cheese: a review of current knowledge. *Inter-national Dairy Journal*, 13(11), 841–866.

Dahou, A.A., Bekada, A.A. and Homrani, A. (2021). Identification of a Lactococcus lactis Isolated from a Fresh Local Cheese of the Western Al-gerian Steppe "J'ben of Naama". *Asian Jour-nal of Dairy and Food Research*, 40(1): 40-44.

Dal Bello F, Walter J, Hertel C, Hammes W P (2001). In vitro study of prebiotic properties of levan-type exopolysaccharides from Lactobacilli and non-digestible carbohydrates using dena-turing gradient gel electrophoresis. *Systematic and Applied Microbiology* 24(2): 232-237.

Esteban-Torres M, Mancheño JM, de Las Rivas B, Muñoz R (2014) Production and characteriza-tion of a tributyrin esterase from Lactobacillus plantarum suitable for cheese lipolysis. *Jour-nal of Dairy Science* 97(11):6737–6744.

Farahani Z, Rasooli I, Owlia P (2017). Isolation, identi-fication and characterization of indigenous lactic acid bacteria for flavour improvement. *International Food Research Journal* 24(1): 428-436.

Franciosi E, Settanni L, Cavazza A, Poznanski E (2009). Biodiversity and technological poten-tial of wild lactic acid bacteria from raw cows' milk. *International Dairy Journal*, 19(1): 3-11.

García-Cano I, Rocha-Mendoza D, Ortega-Anaya J, Wang K, Kosmerl E, Jiménez-Flores R (2019). Lactic acid bacteria isolated from dairy prod-ucts as potential producers of lipolytic, proteo-lytic and antibacterial proteins. *Applied Microbiology and Biotechnology* 103(13): 5243-5257.

Gobbetti M, Cagno, R. Di, Calasso, M, Neviani E, Fox, P. F, Angelis, M. De. (2018). Drivers that establish and assembly the lactic acid bacteria biota in cheeses. *Trends in Food Science & Technology*.

Göktepe Ç K, & Elgün A (2020). The Effect of Ex-opolysaccharide Producer Pediococcus Dam-nosus 2.6 and Yoghurt Starter Cultures on Ethanol Content, Some Physicochemical and Sensory Properties of Oat Boza. *Selcuk Jour-nal of Agriculture and Food Sciences* 34(2), 124-131.

Herreros M A, Fresno J M, González Prieto M J, Tor-nadijo M E (2003). Technological characteri-zation of lactic acid bacteria isolated from Armada cheese (a Spanish goats' milk cheese). *International Dairy Journal* 13(6): 469–479.

Jikang J, Wenxiang L (2019). Characterization and in vitro properties of Lactobacillus plantarum and Leuconostoc mesenteroides for probiotic potential and nitrite degradation. *Science Asia* 45(6): 555–561.

Karakas-Sen A, Karakas E (2018). Isolation, identifi-cation and technological properties of lactic acid bacteria from raw cow milk. *Bioscience Journal* 34(2): 985–999.

Kocková M, Gereková P, Petruláková Z, Hybenová E, Šturdík E (2011). Evaluation of fermentation properties of lactic acid bacteria isolated from sourdough. *Acta Chimica*. Vol 4: 78-87.

Liu CF, Tseng KC, Chiang SS, Lee BH, Hsu WH, Pan TM (2011). Immunomodulatory and anti-oxidant potential of Lactobacillus exopoly-saccharides. *Journal of the Science of Food and Agriculture*, 91(12), 2284-2291

López-Seijas J, García-Fraga B, da Silva A F, Sieiro C (2020). Wine Lactic Acid Bacteria with Anti-microbial Activity as Potential Biocontrol Agents against Fusarium oxysporum f. Sp. Lycopersici. *Agronomy* 10(1):31.

Luiz L M P, Castro R D, Sandes S H C, Silva J G, Oliveira L G, Sales G A, Nunes A C, Souza, M R (2016). Isolation and identification of lactic acid bacteria from Brazilian Minas artisanal cheese. CyTA - *Journal of Food*. 15(1), 125-128.

Mayo B, Aleksandrzak-Piekarczyk T, Fernndez M, Kowalczyk M, lvarez-Martn P, & Bardowski J (2010). Updates in the Metabolism of Lactic Acid Bacteria. *In: Biotechnology of Lactic Acid Bacteria: Novel Applications 3-33*.

McSweeney, P. L., & Sousa, M. J. (2000). Biochemical pathways for the production of flavour com-pounds in cheeses during ripening: A review. Le Lait, 80(3), 293-324.

Mechai A, Debabza M, & Kirane D (2014). Screening of technological and probiotic properties of lactic acid bacteria isolated from Algerian tra-ditional fermented milk products. *International Food Research Journal*, 21(6): 2451-2457.

Meskini Z, Rechidi-sidhoum N, Zouaoui K, Bounaama K. (2021). Infectious aetiologies of subclinical bovine mastitis and antimicro-bial susceptibility on northwest of Algeria. *Veterinaria* 70(3): 311–323.

Morandi S, Cremonesi P, Silvetti T, & Brasca M (2013). Technological characterisation, anti-biotic susceptibility and antimicrobial activity of wild-type Leuconostoc strains isolated from north Italian traditional cheeses. *Journal of Dairy Research* 80:457-466.

Musikasang H, Tani A, H-kittikun A, Maneerat S. (2009). Probiotic potential of lactic acid bacte-ria isolated from chicken gastrointestinal di-gestive tract. *World Journal of Microbiology and Biotechnology* 25(8): 1337-1345.

Oliveira E, Nespolo C R, Sehn C P. (2020). Lactic acid bacteria with antimicrobial, proteolytic and lipolytic activities isolated from ovine dairy products. *Food Science and Technology* 2061(June), 293–299.

Papagianni M (2012). Metabolic engineering of lactic acid bacteria for the production of industrially important compounds. *In Computational and Structural Biotechnology Journal* 3(4): e201210003.

Pfeiler E A, Klaenhammer T R (2007). The genomics of lactic acid bacteria. *In Trends in Microbi-ology* 15(12): 546-553.

Rajoka M S R, Hayat H F, Sarwar S, Mehwish H M, Ahmad F, Hussain N, Shah S Z H, Khurshid M, Siddiqu M, Shi J. (2018). Isolation and evaluation of probiotic potential of lactic acid bacteria isolated from poultry intestine. *Mi-crobiology* (Russian Federation) 87(1): 116-126.

Rendueles O, Kaplan J B, Ghigo J M (2013). Antibio-film polysaccharides. *In Environmental Mi-crobiology* 15(2) : 334-346.

Russo P, López P, Capozzi V, de Palencia P F, Dueñas M T, Spano G, Fiocco D (2012). Beta-glucans improve growth, viability and colonization of probiotic microorganisms. *International Jour-nal of Molecular Sciences* 13(5): 6026-6039.

Savijoki K, Ingmer H, Varmanen P (2006). Proteolytic systems of lactic acid bacteria. *In Applied Mi-crobiology and Biotechnology* 71(4): 394-406.

Toe, C. J., Foo, H. L., Loh, T. C., & Mohamad, R. (2019). Extracellular Proteolytic Activity and Amino Acid Production by Lactic Acid Bacteria Isolated from Malaysian Foods. *International Journal of Molecular sciences.*

Tulini F L, Hymery N, Haertlé T, Le Blay G, De Marti-nis E C P (2016). Screening for antimicrobial and proteolytic activities of lactic acid bacte-ria isolated from cow, buffalo and goat milk and cheeses marketed in the southeast region of Brazil. *Journal of Dairy Research* 83(1): 115-124.

Wedajo B (2015). Lactic Acid Bacteria: Benefits, Se-lection Criteria and Probiotic Potential in Fer-mented Food. *Journal of Probiotics & Health* 03(02):129.

Selcuk Journal of Agriculture and Food Sciences
SJAFS
http://sjafs.selcuk.edu.tr/sjafs/index
(2022) 36 (3), 380-386
Research Article
e-ISSN: 2458-8377
DOI:10.15316/SJAFS.2022.049

Natural Alternative Curing Agent in Fermented Sucuk Production: Sugar beet (*Beta vulgaris* var. *saccharifera* L.) Molasses

Nazik Meziyet DİLEK[1,*], Mustafa KARAKAYA[2]

[1] Selçuk University, Akşehir Kadir Yallagöz School of Health, Department of Nutrition and Dietetics, Konya, Türkiye
[2] Selçuk University, Faculty of Agriculture, Department of Food Engineering, Konya, Türkiye

ARTICLE INFO

Article history:
Received date: 21.06.2022
Accepted date: 30.08.2022

Keywords:
Fermented sucuk
Sugar beet molasse
Natural alternative curing
Nitrate
Nitrite

ABSTRACT

In this study, sugar beet molasses was used as a natural curing agent in the production of fermented Turkish sucuk in two different forms (molasse and powdered molasse). Some quality characteristics were determined during the ripening (1st, 3rd and 5th days) period of the obtained sucuk dough (0. day) and sucuk samples. In order to compare the effects of molasses and powdered molasses used as natural curing agents in sucuk, a control (K) group containing sodium nitrate was formed. The application of the natural alternative curing process in sucuk production causes a decrease in the pH value and TBA content of the samples during the ripening period, whereas causes an increase in the nitrosomyoglobin and residual nitrate contents. As a result of microbiological analysis, it was determined that the natural alternative curing process increased ($P<0.05$) the number of LABs and there was no significant change in the total yeast-mold numbers during the ripening period ($p>0.05$). In addition, total Coliform group bacteria were not detected in sucuk dough and sucuk samples on the last day (5th day) of the ripening period. Consequently, it has been determined that it is possible to reduce the amount of chemical nitrate by using sugar beet molasses and powdered molasses as natural curing agents in sucuk formulation.

1. Introduction

Meat and meat products are of highly rich resources in terms of nutritional elements such as protein, mineral, vitamin and mineral substance and have an important place in terms of healthy and balanced diet and meeting the need of food material of the increasing world population. In meeting dietary needs of society, nutritive features of foods as well as its effects of human health and microbiological quality are quite important. Therefore, like all other groups of food, also in the production of meat and meat products, it is necessary to provide a food safety at the top level.

When considered that meat is an expensive food material, to both prevent economic losses that may form and provide food safety, meat should be conserved with the suitable methods. While salt addition, one of the oldest methods used in meat conservation, decreases water activity, and impedes development of microorganisms, it accelerates the fat and pigment oxidation (Öztan, 2003; Honikel, 2008). The use of salt may cause these effects as well as negativities such as undesirable color formation in internal parts of muscular tissue, when used in high concentrations (Shahidi and Pegg, 2017). In order to prevent the negativities that form as a result of salt addition and bring a characteristic taste in meat products, curing process is applied (Gökalp et al 2004; Doğruer and Güner, 2005). In curing process, it is reported that curing agents such as the salt, nitrate, nitrite and spices, according to the sort of product, are added, and that it is aimed to improve in the specifications such as color, texture, taste, aroma and flavor of the product obtained and increase its durability over shelf life (Aksu and Kaya, 2002).

It is reported that nitrite, more active form of nitrate that is a passive curing agent, is reduced by bacteria with nitrate reductase activity and can be realized curing reactions in meat products (Terns et al., 2011). Nitrite is an important additive performing highly important functions such as developing characteristic cured meat color in meat products, flavor and tissue features and, especially *Clostridium botulinum*, impeding growth of microorganism and formation of oxidative rancidity (Ahn et al., 2004; Liu ve ark., 2010).

It is reported that, in addition to that nitrate/nitrite has positive effects, nitrite has also negative effects such

* Corresponding author email: *meziyettemel@hotmail.com*
This study was part of the pHD thesis of Nazik Meziyet DİLEK.

as N-nitroso compounds, which were identified that they are both carcinogenic, teratogenic and mutagenic, reacting with secondary amines in meat, and forming methemoglobin not having oxygen carrying feature, combining with hemoglobin pigments in blood (Pourazrang et al 2002; Zanardi et al., 2002; Cemek et al., 2007; Zarringhalami et al., 2009). In the recent years, consumer demands natural meat products, in which any chemical additive is not used. However, since nitrate and nitrite have a wide activity on characteristic properties of meat products, any alternative effective additive could not be identified at the level that can realize all functions of these additives. In recent studies, the various methods such as using new technological methods, microorganisms themselves, metabolites, natural additives and/or their combinations are tried to be developed, in order to reduce the use of nitrate and nitrite in formulations of meat products and content of residual nitrate and nitrite in product.

Sugar beet (*Beta vulgaris* var. *saccharifera* L.) is first of all a crop, which is grown to produce sugar and which belongs in *Chenopodiaceae* family (Yardımcı et al. 2012). Since sugar beet, a rich resource in terms of nitrate and betalain pigments, contains bioactive phytochemicals like phenolic compounds, it can be used as natural antioxidant and color substance (Georgiev et al., 2010; Račkauskienė et al., 2015). It was reported that nitrite content in sugar beet was quite less but as a result of reduction of nitrate to nitrite by means of thermophilic bacteria, that nitrite content may increase (Hoffmann and Märländer, 2005). Molasses, a byproduct of sugar industry, is a non-crystallized syrup released during sugar production. Molasses naturally forms during sugar production and is easily obtainable product having relatively low cost (Roukas, 1996).

Molasses of beet sugar was previously used for the purposes such as animal feeding, alcohol production and providing fermentation medium (Paturau, 1989; Ahmedna et al. 2000). However, in the studies carried out, it was identified that molasses of beet sugar had antioxidant (Koprivica et al., 2009; Filipčev et al., 2010; Valli et al., 2012), anti-inflammatory (Miles et al., 2005), and anti-microbial activity (Onbaşlı and Aslim, 2008) and showed effects repairing DNA damage (Asikin et al., 2013). It was reported that molasses of sugar beet was used by microorganisms as sugar source for xanthan fermentation and, consequently, that xanthan obtained was used as a thickener in food industry (Moosavi and Karbassi, 2010). It was also reported that molasses of beet sugar was used in the production of various products for human nutrition such as cookie (Filipčev et al., 2012; Filipčev et al., 2014), bread (Filipčev et al., 2010; Filipčev, 2011), gluten-free product (Šimurina et al., 2008; Filipčev et al., 2016; Šimurina et al., 2017), ice-cream (Acan et al., 2020), probiotic yoghurt (Ghazal and Atallah, 2016). However, as a result of the literature reviews carried out, it was not met any studies regarding that molasses of beet sugar was used in meat products.

In this study, sucuk was produced by means of natural alternative curing process. For this purpose, using sodium nitrate, molasses and molasses powder, 5 different sucuk formulation was prepared. In sucuk batters obtained and in ripening period of sucuks, residual nitrate/nitrite contents and some physicochemical and microbiological properties of sucuks were determined. With this study, using molasses and molasses powder, alternative curing process was applied in the direction of consumer demands, and it was identified that production of sucuk may be possible.

2. Materials and Methods

Fresh boneless beef and beef fat were obtained from a local meat plant (Panagro Meat Plant) in Konya, Turkey. The meats and fats taken from the various sections of carcasses in such a way that they will represent all body were used in sucuk production. Beef meat and fat were initially ground through a 9-mm plate. The sucuk production was conducted in Panagro Meat Plant in Konya, Turkey.

The other additives used in sucuk production (garlic, cumin, pepper, black pepper, salt) were supplied from the various local markets in Konya. In the production of control group, chemical sodium nitrate (Merck, Germany), was used. In the production of sucuks, as starter culture, mixture of *Pediococcus pentosaceus* and *Staphylococcus carnosus* (BFL-T03, Christian Hansen, Hoers Holm, Denmark) was used at a level of 10^7 CFU/kg of sucuk batter. For sucuk casing, 38 mm collagen casings were used (Vemag, Maschinenbau, Germany). Molasses, used as alternative curing agent, was supplied from Pankobirlik Konya-Çumra. Molasses was used in sucuk formulations in two different forms as molasses and molasses powder.

Sucuk batters were divided into five different groups in terms of the nitrate sources used for curing. K: the control group contains 100 ppm chemical sodium nitrate. In the other groups (A, B, C and D), appropriate amounts of chemical sodium nitrate, molasses or powder molasses were added to contain 100 ppm nitrate.

Preparation of Sucuk Sample

Formulation and ripening conditions of sucuk samples are given in Table 1 and Table 2, respectively.

Analyses of pH, residual nitrate and nitrite, microbiological analyses (TMAB, LAB, total Coliform and total yeast-mold were performed on days 0, 1, 3 and 5. Additionally, TBARS and nitrosomyoglobin content were determined on days 1 and 5. Tests were made double iteratively and, in each iterate, carried out in such a way that each iteration includes three parallelism. So, each parameter for routine analyses was realized on $5 \times 3 \times 2 = 30$ samples according to factorial test design.

pH Analysis

pH values of sucuk batter and sucuk samples was determined by pH meter (WTW Series pH 720, Weilheim, Germany).

Thiobarbituric Acid (TBARS) Numbers Analysis

In sucuk samples, thiobarbituric acid (TBARS) number was determined (Gökalp et al., 1999). For this purpose, sample absorbance obtained after certain extraction stages was read in spectrophotometry in 530 nm and absorbance values obtained later were multiplied by 7.03 coefficient and TBARS contents were calculated as mg malonaldehyde/kg sample (mg MA/ kg sample).

Nitrosomyoglobin (NOMb) Analysis

In order to determine nitrosomyoglobin content, cured pigment and total pigment analyses were performed (Zaika et al., 1976). For the analysis of cured pigment, adding 40 ml of acetone and 3 ml of pure water onto sucuk samples of 10 g, in brown glass bottles, after quickly over 5 minutes, they were filtered. From the filtrates obtained, on spectrophotometry, a value of 540 nm was read against blind sample containing 40 ml of acetone and 3 ml of pure water. Absorbance values read, multiplying by the coefficient 290, the content of cured pigment was determined as ppm.

For total pigment analysis, similarly, 10 g of samples is weighed in brown glass bottles and 40 ml of acetone, 2 ml of pure water and 1 ml of concentrated HCL were added to it. The samples, slowly agitated, were kept in a dark medium over 1 hour and then filtered. From the filtrate obtained, a value of 640 nm was read in spectrophotometry against the sample containing 40 ml of acetone + 2 ml of pure water +1 ml of concentrated HCl. Absorbance values read was multiplied by coefficient 680, and total pigment content was determined as ppm.

Table 1
The formulations of the sucuk samples

Ingredients	Treatments				
	K	A	B	C	D
Meat (kg)	10	10	10	10	10
Fat (kg)	2.75	2.75	2.75	2.75	2.75
Garlic powder (g)	386	386	386	386	386
Salt (g)	135	135	135	135	135
Curing agent*(g)	1	189.9	305.6	0.5+95	0.5+152.8
Spices (g)	439	439	439	439	439
Ascorbic acid (g)	3	3	3	3	3
Starter culture**(g)	5	5	5	5	5

The curing agent used for each group was calculated to contain 100 ppm nitrate. K: sample with sodium nitrate (1 g); A: sample with molasse (189.9 g); B: sample with molasse powder (305.6 g); C: sample with 50% sodium nitrate + 50% molasse (0.5 g + 95 g); D: sample with 50% sodium nitrate + 50% molasse powder (0.5 g + 152.8 g).

Table 2
Fermentation conditions in sucuk production

Ripening period	Temperature (°C)	Relative humidity (%)	Air flow velocity (m/sn)	Time (hour)
Fermentation	0	90	0.5	4-5
Fermantation	18-19	85	0.5	12
Fermantation	22	80	0.5	24 (until the pH reached 5.2-5.3)
Drying	16	70	0.5	30
Drying	10	50	0.5	42 (until the water content of samples reached 33-34%)

Residual Nitrate and Nitrate Analysis

In sucuk batters and sucuk samples, the analyses of residual nitrate and nitrite were performed according to the method given by Cortesi et al (2015) and, as a result of analysis, the residual nitrate content was expressed as mg $NaNO_3$ per kg sample (mg/kg) and residual nitrite content, as $NaNO_2$ per kg sample (mg/kg).

Microbiological Analyses

In sucuk batters and sucuk samples, while the counts of total mesophilic aerobic bacteria (TMAB), lactic acid bacteria (LAB) and total yeast-mold were conducted according to the method given by Gökalp et al (1999), in the count of total *Coliform* bacteria, the method given by Sağdıç et al. (2011) was used. The results were expressed as $\log^{10}$ colony forming units per gram sucuk ($\log^{10}$ CFU/g).

Statistical Analyses

This study was performed in two replicates with triplicate sampling and a completely randomized design was employed. One way analysis of variance (ANOVA) was performed for all variables by using MINITAB release 18.0 programme. Differences among the mean values were compared with Tukey Multiple Comparison Test. The curing treatments (K, A, B, C, D) and ripening days (0, 1, 3 and 5) were analyzed as a fixed factor while the replicate was considered as a random factor (Snedecor and Cochran, 1980).

3. Results and Discussion

pH values

The average pH values of sucuks decreased to 5.145 in 5th day from 6.146 that is the beginning value. As a result of that starter cultures (lactic acid bacteria) added to sucuk batters ferments carbohydrates in medium and that lactic acid concentration increases, it was reported that sucuk pHs would decrease (Incze, 1992; Bover-Cid et al., 2001; Ammor and Mayo, 2007). In the first days of ripening, it was reported that the decreases occurring

in pH was necessary to inactivate microorganisms causing spoilage, form and protect the desired color and flavor and produce high quality and safe products (Ilıkkan et al 2009).

While the highest pH (5.698) was determined in K group, it was identified that group A and D had the lowest pH (respectively, 5.440 and 5.446). Due to the fact that molasses and molasses powder were used in sucuk production as alternative curing agent, in the formulation, another carbohydrate resource was not used (Table 1). In the formulation of group K, since there is not any carbohydrate resource, as a result of fermentation, lactic acid formation decreased, and this case led group K to have the highest pH values.

Thiobarbiturik Acid (TBARS) Number

TBARS number of sucuk samples in 1st and 5th days were given in Table 3. At the beginning of ripening period, TBARS number of samples vary between 0.44-0.57 mg MA.kg sample^{-1} ($P<0.05$). When ripening period is completed, in ready-to-eat sucuks, the highest TBARS number (0.50 mg MA.kg sample^{-1}) was determined in group K. When TBARS number of sucuk samples, to which alternative curing methods were applied, are compared to control group, they were found statistically significant low ($P<0.05$). It is thought that molasses or molasses powder, used as alternative curing agent, showed an effect slowing oxidation. This case may be resulted from antioxidant activity of sugar beet molasses (Koprivica et al., 2009; Filipčev et al., 2010; Valli et al., 2012). A similar result was obtained from the study by Yıldız Turp et al. (2016) and, as a result of the study, it was reported that oxidation forming in the samples, in which red beet powder is used in sausage formulation, proceeded more slowly compared to control group, and that this case may be associated with antioxidant activity of red beet powder.

Nitrosomyoglobin (NOMb) values

According to Table 3, at the end of ripening period, it was identified that group B had the highest nitrosomyoglobin content (30.56%) and group K had the lowest nitrosomyoglobin content (17.50%). It was determined that addition of molasses or molasses powder into sucuk samples as alternative curing agent caused more nitrosomyoglobin formation. It was reported that nitrate/nitrite, obtained from natural resources, enabled the values of nitroslhemochrome, total pigment and color formation to be higher (Choi ve ark., 2020; Jeong et al., 2020).

Residual Nitrate and Nitrite content

The residual nitrate and residual nitrite contents of sucuk batters and sucuk samples were shown in Table 4. Residual nitrate contents of sucuk batters were determined in the range of 69.42-77.15 ppm. In the 1st day of ripening period, residual nitrate content, which was in the range of 66.88-75.50 ppm in all sample groups, decreased to 16.97-41.72 ppm range in 5th day. As a result of the activities of starter culture used in sucuk production, nitrate was reduced to nitrite and nitrite, to nitric oxide and, thus, residual nitrate content largely decreased in ripening period ($P<0.05$).

In the group K, the amount of residual nitrate decreased more than the alternative cured sucuk samples, and they contained a lower amount of residual nitrate at the end of the ripening period (in the ready-to-eat product). This can be explained by the fact that the pH value of the group K is higher than the other groups. In the groups, in which molasses and molasses powder were used, it is thought that lower pH values impeded that starter cultures with nitrate reductase activity and, thus, may have led to higher residual nitrate contents. Eisinaitė et al (2020) obtained similar results. Independently the resource of nitrate used (synthetic or freeze-dried powdered celeriac), they reported that the bacteria reducing nitrate in low pH values was inhibited and that there was high residual nitrate in final product.

Over ripening period, fluctuations occurred in the residual nitrite contents of all groups but these fluctuations was not found statistically significant in group K ($P>0.05$). It is thought that these increases and decreases resulted from continuing of reduction of the residual nitrate to nitrite in sucuks. It was reported that *Staphylococcus carnosus*, among starter cultures used in sucuk production, and those having nitrate reductase enzyme, among microorganisms included in natural flora, played role in reduction of nitrate to nitrite. (Macdougall et al., 1975; Sebranek, 1979; Pinotti et al., 2002). In the only 3rd day of ripening period, the difference between the residual nitrite contents of the samples was found significant ($P<0.05$) and it was determined that group K contained the lowest residual nitrite (3.49 ppm). This result is complied with literature findings, and it was reported that the samples containing natural nitrate/nitrite had higher residual nitrite content than the samples of control group containing chemical nitrate/ nitrite (Bertol et al., 2012; Sullivan et al., 2012; Horsch et al., 2014). Myers et al. (2013) expressed that this case was associated with quicker decomposition of chemical nitrite.

For cured meat products to be able to conserve the existing cure color, it was reported that it was useful for final product to contain residual nitrite at the level of 10-15 ppm (Sindelar and Milkowski, 2011). It is thought that the residual nitrite contents we determined in the study were lower may be resulted from the difference of the formulation, raw material and ripening conditions.

Table 3
TBARS and NOMb values of sucuk samples (Mean ± Standard error)

Analyses	Ripening Period (Day)	Treatments				
		K	A	B	C	D
TBARS*	1	0.44±0.02^{Ba}	0.57±0.03^{Aa}	0.46±0.03^{Ba}	0.46±0.00^{Ba}	0.51±0.00^{ABa}
	5	0.50±0.00^{Aa}	0.37±0.00^{Eb}	0.41±0.00^{Ca}	0.44±0.00^{Ba}	0.39±0.00^{Db}
NOMb**	1	17.82±0.25^{Ca}	19.78±0.44^{Ba}	14.81±0.13^{Db}	19.15±0.33^{BCb}	22.49±0.53^{Ab}
	5	17.50±0.11^{Ca}	24.48±2.05^{Ba}	30.56±1.71^{Aa}	27.99±0.27^{ABa}	29.29±0.06^{ABa}

Within the same row, values with different uppercase superscript letters indicate significant differences (P<0.05) Within the same column, values with different lowercase superscript letters indicate significant differences (P<0.05) K: sample with sodium nitrate (1 g); A: sample with molasse (189.9 g); B: sample with molasse powder (305.6 g); C: sample with 50% sodium nitrate + 50% molasse (0.5 g + 95 g); D: sample with 50% sodium nitrate + 50% molasse powder (0.5 g + 152.8 g). *mg MA/kg sample, **(%).

Table 4
Residual nitrate and residual nitrite content of sucuk batters and sucuk samples (ppm) (Mean ± standard error)

Analyses	Ripening Period (Day)	Treatments				
		K	A	B	C	D
Residual nitrate	0	72.05±1.42^{BCa}	69.42±0.76^{Ca}	73.87±0.96^{ABa}	77.15±1.03^{Aa}	73.16±0.05^{BCa}
	1	68.25±0.28^{Bb}	66.88±3.19^{Ba}	70.38±1.97^{ABa}	75.50±0.11^{Aa}	73.03±0.08^{ABa}
	3	17.82±0.62^{Dc}	49.16±2.43^{Ab}	48.50±1.66^{Ab}	38.84±1.41^{Bb}	27.15±1.13^{Cb}
	5	16.97±0.72^{Bc}	41.72±1.16^{Ab}	40.31±0.84^{Ac}	36.25±3.15^{Ab}	25.87±3.51^{Bb}
Residual nitrite	0	3.00±0.06	2.90±0.07^{c}	2.90±0.07^{c}	2.95±0.06^{d}	3.00±0.07^{d}
	1	4.10±0.04	4.13±0.08^{b}	4.14±0.09^{b}	4.10±0.04^{c}	4.36±0.04^{c}
	3	3.49±0.07^{C}	5.22±0.05^{Ba}	5.21±0.14^{Ba}	5.25±0.11^{Bb}	6.04±0.18^{Aa}
	5	4.73±0.93	4.82±0.33^{ab}	5.06±0.09^{a}	5.61±0.04^{a}	5.19±0.28^{b}

Within the same row, values with different uppercase superscript letters indicate significant differences (P<0.05) Within the same column, values with different lowercase superscript letters indicate significant differences (P<0.05) K: sample with sodium nitrate (1 g); A: sample with molasse (189.9 g); B: sample with molasse powder (305.6 g); C: sample with 50% sodium nitrate + 50% molasse (0.5 g + 95 g); D: sample with 50% sodium nitrate + 50% molasse powder (0.5 g + 152.8 g).

Microbiological quality

Microbiological counts of sucuk batters and sucuk samples were presented in Table 5. Curing process did not affect the TMAB (P>0.05). Ripening period increased of TMAB counts of sample in only group K and A (P>0.05). Aksu and Kaya (2004) reported that the TMAB counts of sucuks varied between 7.49-8.63 log CFU/g and, in the 3rd day of ripening period, that this number increased to 8.39 log CFU/g.

In the 1st day of ripening period, the LAB counts, which was in the range 5.80-6.48 log CFU/g, increased by fermentation conditions and, in 5th day, increased to the levels of 8.83-9.28 log CFU/g'. However, there was no different between the LAB counts of samples in 3rd and 5th days (P>0.05). This case is accounted for adaptation of starter cultures added to sucuk formulation to the fermentation conditions (Bover-Cid ve ark., 2001; Kaban and Kaya, 2006).

At the end of ripening period, it was determined that group D had the highest LAB counts (9.28 log CFU/g). Additionally, it was observed that group A, B and C (containing molasses, molasses powder, 50 % sodium nitrate + 50% molasses), had more LAB counts, when compared to group K. It is thought that this situation may be resulted from the addition of molasses and molasses powder used as curing agent and that it was additional resource of carbohydrate for lactic acid bacteria. The results are complied with literature data. Similarly, Ekici ve ark. (2015) reported that the LAB counts of sucuks were between 8.36-8.71 log CFU/g.

Table 5
Microbiological counts of sucuk samples (log CFU/g) (Mean ± standard error)

Analyses	Ripening Period (Day)	Treatments				
		K	A	B	C	D
TMAB	0	5.11±0.10^{b}	4.99±0.17^{b}	5.18±0.28	5.18±0.04	5.34±0.11
	1	6.00±0.05^{a}	5.96±0.04^{a}	5.91±0.35	6.15±0.00	6.04±0.23
	3	5.88±0.28^{a}	5.48±0.14^{ab}	4.86±1.00	5.54±0.65	5.64±0.26
	5	5.94±0.21^{a}	5.72±0.13^{a}	5.69±0.28	5.79±0.04	5.51±0.08
LAB	0	6.05±0.02^{ABb}	5.80±0.08^{Bb}	6.13±0.18^{ABb}	6.25±0.01^{Ab}	6.08±0.02^{ABc}
	1	6.00±0.19^{ABb}	5.80±0.00^{Bb}	6.00±0.06^{ABb}	6.17±0.18^{ABb}	6.48±0.10^{Ab}
	3	9.06±0.00^{Aa}	9.09±0.06^{Aa}	9.05±0.01^{Aa}	9.16±0.01^{Aa}	9.03±0.08^{Aa}
	5	8.83±0.04^{Ca}	9.18±0.04^{ABa}	9.15±0.03^{Ba}	9.17±0.03^{ABa}	9.28±0.01^{Aa}
Total yeast-mold	0	3.30±0.45	2.99±0.16	3.59±0.11	3.23±0.28	2.76±0.52
	1	3.65±0.06	3.47±0.33	3.68±0.10	3.41±0.00	3.44±0.35
	3	3.42±0.04	3.22±0.13	3.62±0.23	3.21±0.15	3.51±0.00
	5	3.42±0.04	3.22±0.13	3.62±0.23	3.21±0.15	3.51±0.00
Total *Coliform* bacteria	0	ndg	ndg^{b}	ndg^{b}	ndg	ndg^{c}
	1	1.27±1.80	2.46±0.19^{a}	2.57±0.11^{a}	2.52±0.00	2.56±0.23^{a}
	3	1.73±0.18	0.00±0.00^{b}	0.00±0.00^{b}	0.89±1.26	1.97±0.10^{b}
	5	ndg	ndg^{b}	ndg^{b}	ndg	ndg^{c}

Within the same row, values with different uppercase superscript letters indicate significant differences (P<0.05) Within the same column, values with different lowercase superscript letters indicate significant differences (P<0.05) K: sample with sodium nitrate (1 g); A: sample with molasse (189.9 g); B: sample with molasse powder (305.6 g); C: sample with 50% sodium nitrate + 50% molasse (0.5 g + 95 g); D: sample with 50% sodium nitrate + 50% molasse powder (0.5 g + 152.8 g). ndg: No detectable growth

When Table 5 is examined, it is seen that the curing process and ripening period did not affect the total yeast-mold counts of samples (8.36-8.71 log CFU/g) (P>0.05).

No significant difference (P>0.05) is found between each sample groups for total *Coliform* bacteria in terms of curing process. While total *Coliform* bacteria were not determined in sucuk batters, the total *Coliform* bacteria counts were between in the range of 1.27-2.57 log CFU/g at the beginning of ripening period (1st day) decreased to the range of 00-1.97 log CFU/g in the 3rd day. In the last day of ripening period (5th day) the total *Coliform* bacteria could not be determined in samples. It can be account for this case with the that lactic acid bacteria, which prevail to the medium in fermentation process reduce medium pH and thus, inhibe the total coliform bacteria. Also, it is thought that the total *Coliform* bacteria are inhibited as a result of the antimicrobial effects of nitrite, which is formed with the reduction of nitrate obtained from chemical or molasses.

4. Acknowledgements

This study was funded by the Scientific Research Projects (SUBAP- Konya, Turkey) of Selçuk University Coordinating Office (Project Number 20111002). This study was part of the pHD thesis of Nazik Meziyet Dilek.

5. References

Acan BG, Toker OS, Aktar T, Tamturk F, Palabiyik I, Konar N (2020). Using spray-dried sugar beet molasses in ice cream as a novel bulking agent. *International Journal of Food Science & Technology* 55 (3): 1298-1310.

Ahmedna M, Marshall W, Rao R (2000). Production of granular activated carbons from select agricultural by-products and evaluation of their physical, chemical and adsorption properties. *Bioresource Technology* 71 (2): 113-123.

Ahn HJ, Kim J H, Jo C, Lee JW, Yook HS, Kim HY, Byun MW (2004). Combined effects of gamma irradiation and a modified atmospheric packaging on the physicochemical characteristics of sausage. *Radiation Physics and Chemistry* 71 (1-2): 53-56.

Aksu Mİ, Kaya M (2002). Farklı kürleme yöntemleri ve starter kültür kullanılarak pastırma üretimi. *Turkish Journal of Veterinary and Animal Sciences* 26 (4): 909-916.

Aksu Mİ, Kaya M (2004). Effect of usage *Urtica dioica* l. on microbiological properties of sucuk, a turkish dry-fermented sausage. *Food Control* 15 (8): 591-595.

Ammor MS, Mayo B (2007). Selection criteria for lactic acid bacteria to be used as functional starter cultures in dry sausage production: An update. *Meat Science* 76 (1): 138-146.

Asikin Y, Takahashi M, Mishima T, Mizu M, Takara K, Wada K (2013). Antioxidant activity of sugarcane molasses against 2, 2′-azobis (2-Amidinopropane) dihydrochloride-induced peroxyl radicals. *Food Chemistry* 141 (1): 466-472.

Bertol TM, Fiorentini AM, Santos MJH, Sawitzki MC, Kawski VL, Agnes IBL, Costa CD, Coldebella A, Lopes LS (2012). Rosemary extract and celery-based products used as natural quality enhancers for colonial type salami with different ripening times. *Food Science and Technology* 32 (4): 783-792.

Bover-Cid S, Izquierdo-Pulido M, Vidal-Carou MC (2001). Effect of the interaction between a low tyramine-producing *lactobacillus* and proteolytic staphylococci on biogenic amine production during ripening and storage of dry sausages. *International Journal of Food Microbiology* 65 (1-2): 113-123.

Cemek M, Akkaya L, Birdane YO, Seyrek K, Bulut S, Konuk M (2007). Nitrate and nitrite levels in fruity and natural mineral waters marketed in Western Turkey. *Journal of Food Composition and Analysis* 20 (3-4): 236-240.

Choi JH, Bae SM, Jeong JY (2020). Effects of the addition levels of white kimchi powder and acerola juice powder on the qualities of indirectly cured meat products. *Food Science of Animal Resources* 40 (4): 636-648.

Cortesi ML, Vollano L, Peruzy MF, Marrone R, Mercogliano R (2015). Determination of nitrate and nitrite levels in infant foods marketed in Southern Italy. *CyTA-Journal of Food* 13 (4): 629-634.

Doğruer Y, Güner A (2005). Effect of using sodium and potassium nitrate on degrading and residue level of nitrate and nitrite contents of pastirma during the storage period. *Acta Alimentaria* 34 (2): 141-144.

Eisinaitė V, Tamkutė L, Vinauskienė R, Leskauskaitė D (2020). Freeze-dried celery as an indirect source of nitrate in cold-smoked sausages: Effect on safety and color formation. *LWT-Food Science and Technology*, 109586.

Ekici L, Ozturk I, Karaman S, Caliskan O, Tornuk F, Sagdic O, Yetim H (2015). Effects of black carrot concentrate on some physicochemical, textural, bioactive, aroma and sensory properties of sucuk, a traditional Turkish dry-fermented sausage. *LWT-Food Science and Technology* 62 (1): 718-726.

Filipčev B, Lević L, Bodroža-Solarov M, Mišljenović N, Koprivica G (2010). Quality characteristics and antioxidant properties of breads supplemented with sugar beet molasses-based ingredients. *International Journal of Food Properties* 13 (5): 1035-1053.

Filipčev B, Bodroža-Solarov M, Šimurina O, Cvetković B (2012). Use of sugar beet molasses in processing of gingerbread type biscuits: Effect on quality characteristics, nutritional profile, and bioavailability of calcium and iron. *Acta Alimentaria* 41 (4): 494-505.

Filipčev B, Šimurina O, Bodroža-Solarov M (2014). Quality of gingernut type biscuits as affected by varying fat content and partial replacement of honey with molasses. *Journal of Food Science and Technology* 51 (11): 3163-3171.

Filipčev B, Mišan A, Šarić B, Šimurina O (2016). Sugar beet molasses as an ingredient to enhance the nutritional and functional properties of gluten-free cookies. *International Journal of Food Sciences and Nutrition* 67 (3): 249-256.

Filipčev BV (2011). Sugar beet molasses: An ingredient to enhance micronutrients and functionality in bread. *Food and Feed Research* 38 (2): 101-108.

Georgiev VG, Weber J, Kneschke EM, Denev PN, Bley T, Pavlov AI (2010). Antioxidant activity and phenolic content of betalain extracts from intact plants and hairy root cultures of the red beetroot *Beta vulgaris* cv. detroit dark red. *Plant Foods for Human Nutrition* 65 (2): 105-111.

Ghazal G, Atallah A (2016). Production of probiotic yoghurt fortified with bee honey, beet molasses, date honey and pomegranate honey. *Egyptian Informatics Journal* 44: 102-117.

Gökalp H, Kaya M, Zorba Ö, Tulek Y (1999). Et ve Ürünlerinde Kalite Kontrolü ve Laboratuar Uygulama Kılavuzu. Atatürk Üniversitesi Ziraat Fakültesi Yayınları, No: 318, Erzurum-Türkiye.

Gökalp YG, Kaya M, Zorba Ö (2004). Et Ürunleri İşleme Mühendisliği, Atatürk Üniversitesi Ziraat Fakültesi Ofset Tesisi, Erzurum-Türkiye.

Hoffmann CM, Märländer B (2005). Composition of harmful nitrogen in sugar beet (*Beta vulgaris* L.)-amino acids, betaine, nitrate-as affected by genotype and environment. *European Journal of Agronomy* 22 (3): 255-265.

Honikel KO (2008). The use and control of nitrate and nitrite for the processing of meat products. *Meat Science* 78 (1-2): 68-76.

Horsch A, Sebranek J, Dickson J, Niebuhr S, Larson E, Lavieri N, Ruther B, Wilson L (2014). The effect of ph and nitrite concentration on the antimicrobial impact of celery juice concentrate compared with conventional sodium nitrite on *Listeria monocytogenes*. *Meat Science* 96 (1): 400-407.

Ilıkkan H, Ercoşkun H, Vural H, Şahin E (2009). The effect of addition of hazelnut oil on some quality characteristics of Turkish fermented sausage (sucuk). *Journal of Muscle Foods* 20 (1): 117-127.

Incze K (1992). Roh fermentierte, getrocknete fleischerzeugnisse. *Fleischwirtschaft (Frankfurt)* 72 (1): 8-19.

Jeong JY, Bae SM, Yoon J, Gwak SH (2020). Investigating the effects of chinese cabbage powder as an alternative nitrate source on cured color development of ground pork sausages. *Food Science of Animal Resources* 40 (6): 990-1000.

Kaban G, Kaya M (2006). Effect of starter culture on growth of *Staphylococcus aureus* in sucuk. *Food Control* 17 (10): 797-801.

Koprivica G, Misljenovic N, Levic L, Kuljanin T (2009). Influence of the nutrients present in sugar beet molasses and saccharose solutions on the quality of osmodehydrated carrot. *The National Society of Processing and Energy in Agriculture (PTEP) (Serbia)* 13 (2): 184-187.

Liu DC, Wu SW, Tan FJ (2010). Effects of addition of anka rice on the qualities of low-nitrite chinese sausages. *Food Chemistry* 118 (2): 245-250.

Macdougall DB, Mottram DS, Rhodes DN (1975). Contribution of nitrite and nitrate to the colour and flavour of cured meats. *Journal of the Science of Food and Agriculture* 26 (11): 1743-1754.

Miles EA, Zoubouli P, Calder PC (2005). Differential anti-inflammatory effects of phenolic compounds from extra virgin olive oil identified in human whole blood cultures. *Nutrition* 21 (3): 389-394.

Moosavi A, Karbassi A (2010). Bioconversion of sugar-beet molasses into xanthan gum. *Journal of Food Processing and Preservation* 34 (2): 316-322.

Myers K, Montoya D, Cannon J, Dickson J, Sebranek J (2013). The effect of high hydrostatic pressure, sodium nitrite and salt concentration on the growth of *Listeria monocytogenes* on RTE ham and turkey. *Meat Science* 93 (2): 263-268.

Onbaşlı D, Aslim B (2008). Determination of antimicrobial activity and production of some metabolites by *Pseudomonas Aeruginosa* B1 and B2 in sugar beet molasses. *African Journal of Biotechnology* 7 (24): 4614-4619.

Öztan A (2003). Et Bilimi ve Teknolojisi. TMMOB Gıda Mühendisleri Odası Yayınları.

Paturau JM (1989). By-products of the cane sugar industry. An introduction to their industrial utilization. *The Journal Sugar Series* 11.

Pinotti A, Graiver N, Califano A, Zaritzky N (2002). Diffusion of nitrite and nitrate salts in pork tissue in the presence of sodium chloride. *Journal of Food Science* 67 (6): 2165-2171.

Pourazrang H, Moazzami A, Bazzaz BF (2002). Inhibition of mutagenic n-nitroso compound formation in sausage samples by using L-ascorbic acid and α-tocopherol. *Meat Science* 62 (4): 479-483.

Račkauskienė I, Pukalskas A, Venskutonis PR, Fiore A, Troise AD, Fogliano V (2015). Effects of beetroot (*Beta vulgaris*) preparations on the maillard reaction products in milk and meat-protein model systems. *Food Research International* 70: 31-39.

Roukas T (1996). Ethanol production from non-sterilized beet molasses by free and immobilized *Saccharomyces cerevisiae* cells using fed-batch culture. *Journal of Food Engineering* 27 (1): 87-96.

Sağdıç O, Öztürk İ, Yılmaz MT, Yetim H (2011). Effect of grape pomace extracts obtained from different grape varieties on microbial quality of beef patty. *Journal of Food Science* 76 (7): 515-521.

Sebranek JG (1979). Advances in the technology of nitrite use and consideration of alternatives. *Food Technology (USA)* 33 (7): 58-62.

Shahidi F, Pegg RB (2017). Processing of nitrite-free cured meats. *Advanced Technologies for Meat Processing*, CRC Press, pp. 513-534.

Šimurina OD, Filipčev BV, Lević LB, Pribiš VD (2008). Application of sugar beet molasses in the production of tea biscuits. *Food and Feed Research* 35 (4): 201-205.

Šimurina OD, Radunović AZ, Filipčev BV, Jevtić-Mučibabić R, Šarić LĆ, Šoronja-Simović DM (2017). Quality improvement of gluten-free bread based on soybean and enriched with sugar beet molasses. *Food and Feed Research* 44 (1): 65-72.

Sindelar JJ, Milkowski AL (2011). Sodium nitrite in processed meat and poultry meats: A review of curing and examining the risk/benefit of its use. *American Meat Science Association White Paper Series* 3: 1-14.

Snedecor G, Cochran W (1980). Statistical Methods. Iowa State University Press, Iowa, USA.

Sullivan GA, Jackson-Davis AL, Niebuhr SE, Xi Y, Schrader KD, Sebranek JG, Dickson JS (2012). Inhibition of *Listeria monocytogenes* using natural antimicrobials in no-nitrate-or-nitrite-added ham. *Journal of Food Protection* 75 (6): 1071-1076.

Terns MJ, Milkowski AL, Claus JR, Sindelar JJ (2011). Investigating the effect of incubation time and starter culture addition level on quality attributes of indirectly cured, emulsified cooked sausages. *Meat Science* 88 (3): 454-461.

Valli V, Gómez-Caravaca AM, Di Nunzio M, Danesi F, Caboni MF, Bordoni A (2012). Sugar cane and sugar beet molasses, antioxidant-rich alternatives to refined sugar. *Journal of Agricultural and Food Chemistry* 60 (51): 12508-12515.

Yardımcı N, Kılıç HÇ, Ürgen G (2012). Eskişehir ili şeker pancarı üretim alanlarında görülen bazı virüs hastalıklarının DAS-ELISA yöntemiyle belirlenmesi. *Süleyman Demirel Üniversitesi, Ziraat Fakültesi Dergisi* 7 (1).

Yıldız Turp G, Kazan H, Ünübol H (2016). Sosis üretiminde doğal renk maddesi ve antioksidan olarak kırmızı pancar tozu kullanımı, *Celal Bayar Üniversitesi Fen Bilimleri Dergisi* 12 (2).

Zaika LL, Zell TE, Smith J, Palumbo S, Kissinger J (1976). The role of nitrite and nitrate in Lebanon bologna, a fermented sausage. *Journal of Food Science* 41 (6): 1457-1460.

Zanardi E, Dazzi G, Madarena G, Chizzolini R (2002). Comparative study on nitrite and nitrate ions determination. *Annali della Facolta di Medicina Veterinaria, Universita di Parma* 22: 70-86.

Zarringhalami S, Sahari M, Hamidi-Esfehani Z (2009). Partial replacement of nitrite by annatto as a colour additive in sausage. *Meat Science* 81 (1): 281-284.

Selcuk Journal of Agriculture and Food Sciences SJAFS

http://sjafs.selcuk.edu.tr/sjafs/index

(2022) 36 (3), 387-392

e-ISSN: 2458-8377

DOI:10.15316/SJAFS.2022.050

Research Article

Effects of Rhizobacteria on Plant Growth and Fruit Quality of Blackberry in Alkaline Soil

Muzaffer İPEK[1,*], Ahmet EŞİTKEN[1]

[1]Selçuk University, Faculty of Agriculture, Department of Horticulture, Konya, Türkiye

ARTICLE INFO

Article history:
Received date: 30.05.2022
Accepted date: 02.09.2022

Keywords:
Alkaline Soil
Blackberry
Fruit Quality
Plant Growth

ABSTRACT

This study was conducted at Selçuk University, Faculty of Agriculture, Department of Horticulture, Research and Application Orchard. In the study, the effects of different bacterial strains on yield, fruit quality, plant growth, and nutrition issues in Jumbo and Chester blackberry varieties in calcareous soil conditions were determined. As a result of the measurements and analyzes, it was determined that bacterial applications are effective on plant growth, yield, and fruit quality criteria. Shoot length values were found longer in the Chester variety, while the Jumbo variety had higher values in the number of shoots per plant. There was no statistically significant difference in terms of yield per decare of varieties. When the efficiency is examined in terms of applications, 637Ca application in Chester variety and 637Ca + SY 55 application in Jumbo variety have the highest efficiency values. With the data obtained, it was determined that bacterial applications made positive contributions to blackberry plants grown in calcareous soil conditions.

1. Introduction

Blackberries are widespread perennial shrubs native to the temperate Northern hemisphere. Blackberry (*Rubus fruticosus* L.) is a popular berry species that is widely found in nature and widely cultivated throughout the world. In addition to the rapid increase in blackberry consumption in recent years, it is also processed into various foods, such as fresh, frozen, or commercially, and products such as jam, wine, tea, ink, food coloring, ice cream, and cake. Blackberry berries have a pleasant taste, high nutritional value, and important health benefits (Wu et al., 2007). Although blackberries do well in most soils, deep, well-drained soils are ideal. Blackberries perform best at a soil pH between 5.5 and 6.5. Blackberry roots are located close to the surface, and excess fertilizer can burn leaves or even kill plants. Lime soils cover more than 30% of the world's land. These soils are mostly characterized as soils with low availability of nutrients due to poor solubility of microelements such as Fe, Mn, Zn, Cu, and B at high pH and also because P forms complex compounds with Ca (Marschner, 2011). Except for the Black Sea Region in Turkey, all of the other regions show high calcareous soil characteristics. Significant yield losses occur due to chlorosis caused by high lime content, and the producer increases input costs by using excess fertilizer to solve the problem. Nowadays, there is a global challenge to find alternatives to reduce the massive use of chemical fertilizers and agrochemical products. In this sense, PGPR is an eco-friendly alternative that may be used to replace or reduce the use of these chemical products. Some of the beneficial bacteria bind the free nitrogen in the air asymbiotically to the soil and make it available to the plants. In addition, some beneficial bacteria can promote phosphorus nutrition of plants by increasing the solubility of organic and inorganic phosphorus that plants cannot benefit from in the soil. In addition, it lowers the soil pH through the organic acids they secrete and can increase the availability of microelements such as Fe, Zn, Mn, Cu, and B in particular. Some bacteria can help plants uptake Fe by producing siderophores. PGPR is widely used to promote growth and development in different plant species (Glick et al., 2001). In addition, especially in recent years, intensive studies have been conducted to determine the effects of these bacteria to promote plant growth under abiotic stress conditions.

2. Materials and Methods

A two-year field study was conducted on the three-year-old plants of blackberry cultivars ‘Jumbo and Chester’ (*Rubus fruticosus* L.) propagated by *invitro* micropropagation technique of nodal segment. The soil has a high lime content (29.6%) with a 7.49 pH.

* Corresponding author email: *mipek@selcuk.edu.tr*

Alcaligenes feacalis 637Ca, *Microbacterium esteraromaticum* SY48, *Rhizobium radiobacter* SY55, and *Kocuria rhizophila* SY63 bacterial strains were used in the experiment. The 637Ca can dissolve $CaCO_3$, SY55 N-fixing, SY63 convert phosphorus, and SY48 convert potassium into forms that can be used by the plant.

Experiment, both as a single treatment such as;

1-Control,

2-637Ca,

3-SY55,

4-SY63,

5-SY48 and in a combination such as;

6-637Ca+SY55,

7-637Ca+SY63,

8-637Ca+SY48,

9-637Ca+SY55+SY63,

10-637Ca+SY55+SY48,

11-637Ca+SY63+ SY48,

12-637Ca+SY55+SY63+SY48 was performed.

The bacterial treatments with 637Ca, SY48, SY55, SY63 and their combinations (10^9 CFU·ml^{-1}) were given to the plant root via dipping 30 minutes before planting. The plants of the control were immersed in water. All bacteria used in the research were applied with irrigation in June, July, August, and September after planting. In the second year of the experiment, bacteria were applied with irrigation in May, June, July, August, and September. To determine the effect of the bacterial treatments on plant development, the number of the primocane shoot (shoot per plant), average primocane shoot length (cm), leaf area (cm^2), plant fresh and dried weight (g), root length (cm), and root fresh and dried weight (g) were measured and observed. Fruit number per plant, average fruit weight, yield per blackberry plant (g), and yield per decare (kg) were measured for the effect of bacterial treatments on yield. Also, the fruit quality was measured with total soluble solids (TSS) (%), pH of fruit juice, total acidity content (TAC) of fruit juice (%), and fruit color (L, C H.

All data were analyzed using one-way analysis of variance (ANOVA) and significant differences among the means were compared by Duncan's multiple range test at P = 0.05 level using SPSS 23.0 (SAS Inc., Cary, NC, USA).

3. Results and Discussion

Positive results of bacterial applications on plant growth, fruit quality characteristics, and yield have been reported in many fruit species such as apple, banana, cherry, peach, pear, quince, raspberry, sour cherry, and strawberry (Arıkan et al., 2013; Arikan and Pirlak, 2016; Aslantaş et al., 2007; Garcia-Seco et al., 2015; Ipek et al., 2014; Karakurt and Aslantas, 2010a; Karakurt et al., 2011; Karlidag et al., 2010; Kavino et al., 2010; Mia et al., 2005; Orhan et al., 2006; Pii et al., 2017; Seema et al., 2018). When all of the research data are considered, the results obtained are similar to the results of previous studies. In blackberry, the number of primocane shoots is an important criterion for yield. Primocane shoots become productive in their second year and take the name floricane. Some blackberry cultivars can have the floricane shoots that occurred from primocane shoots in the same year. Floricane shoots die within the same season after being harvested.

In this research, the primocane shoot number was counted in both years. A statistical difference was found between the effectiveness of the treatments and cultivars on the number of primocane shoots. In 2019, While the maximum number of primocane shoots were counted in the 637Ca, SY 48, SK 63, and 637Ca+SY 48+SY 55+SK 63 treatments in the Chester cultivar, SY 55, and 637Ca+SY 55 were found the maximum number of primocane shoots in the Jumbo cultivar. In 2020, the primocane shoot numbers decreased in both cultivars Chester and Jumbo because of the water deficiency and hot temperature in the Konya region. The maximum primocane shoot number was counted in all bacterial treatments in the Chester cultivar while the maximum primocane shoot number of Jumbo was counted in all treatments except for Control, 637Ca, and 637Ca+SY 48 treatments in 2020 (Table 1). Comparable results about shoot numbers of blackberry (García-Seco et al., 2013; Garcia-Seco et al., 2015; Robledo-Buriticá et al., 2018), apple (Aslantaş et al., 2007; Karakurt and Aslantas, 2010a), and raspberry (Orhan et al., 2006) have been reported.

Table 1
Primocane shoot number per blackberry plant

Treatments	2019		2020	
	Chester	Jumbo	Chester	Jumbo
Control	5.80 b	5.00 d	1.33 b	2.50 c
637Ca	6.00 ab	6.20 c	2.17 ab	2.50 c
SY 48	6.00 ab	6.80 bc	2.17 ab	2.83 ab
SY 55	4.60 c	8.20 a	1.83 ab	3.33 a
SK 63	5.40 ab	6.40 bc	1.67 ab	3.00 ab
637Ca+SY 48	5.00 c	7.00 b	2.17 ab	2.67 bc
637Ca+SY 55	4.00 d	8.20 a	2.00 ab	3.17 ab
637Ca+SK 63	4.40 c	6.80 bc	2.17 ab	3.33 a
637Ca+SY 48+SY 55	4.80 c	6.20 c	1.83 ab	2.83 ab
637Ca+SY 48+SK 63	3.80 d	7.00 b	1.83 ab	3.00 ab
637Ca+SY 55+SK 63	4.80 c	6.60 bc	2.00 ab	3.17 ab
637Ca+SY 48+SY55+SK 63	6.40 a	6.40 bc	2.33 a	3.33 a
Cultivars	5.80 b	6.73 a	1.95 b	2.97 a

The shoot length was measured longer in 2020 than in the 2019 year. While the water deficiency and hot temperature decreased the number of shoots in 2020, the other hand, it increased shoot length in the same year. According to the shoot length data in 2019, the longest shoot length had the highest value in all the remaining applications except Control, SK 63, 637Ca+SY 55, and 637Ca+SY 48+SY55+SK 63 in the Chester, while 637Ca+ SK 63 was the longest shoot length in the Jumbo cultivar. In 2020, while the number of shoots decreased, the length of shoots increased. The longest shoot length was measured in 637Ca+SY 48+SY 55, SK 63, 637Ca+SY 48+SK 63, 637Ca+SY 55, 637Ca+SY 48+SY 55+SK 63, and 637Ca treatments in the Chester, while the longest shoot length was obtained from 637Ca, 637Ca+SY 55, 637Ca+SY 48+SY 55, and 637Ca+SK

63 in Jumbo. In both years, Chester cultivars had a longer shoot length than the Jumbo cultivar (Table 2). In the previous studies about PGPR, shoot length have been increased by the PGPR treatments in some fruit species (Arikan and Pirlak, 2016; Aslantaş et al., 2007; Esitken et al., 2005; Esitken et al., 2006; Pérez Moncada et al., 2015; Robledo-Buriticá et al., 2018).

Table 2
Average primocane shoot length (cm) per blackberry plant

Treatments	2019		2020	
	Chester	Jumbo	Chester	Jumbo
Control	74 b	60 b	203 cd	175 b
637Ca	89 a	55 bc	316 ab	193 a
SY 48	83 a	50 c	178 d	141 c
SY 55	79 ab	52 bc	227 cd	145 c
SK 63	69 b	50 c	354 a	145 c
637Ca+SY 48	81 a	62 b	259 bcd	157 c
637Ca+SY 55	61 c	61 b	344 a	190 a
637Ca+SK 63	79 ab	70 a	280 bc	186 a
637Ca+SY 48+SY 55	77 ab	60 b	355 a	190 a
637Ca+SY 48+SK 63	76 ab	51 c	350 a	153 c
637Ca+SY 55+SK 63	85 a	59 b	198 cd	177 b
637Ca+SY 48+SY55+SK 63	72 b	55 bc	326 ab	170 b
Cultivars	77 a	57 b	283 a	168 b

The largest leaf area was measured in 637Ca+ SY 55 treatment in Chester while 637Ca, and 637Ca+ SY 55 treatments in the Jumbo cultivar in the 2019 year. The same result of the leaf area was obtained in both cultivars. The largest leaf area in 637Ca+ SY 55 treatment in Chester while 637Ca, and 637Ca+ SY 55 treatments in the Jumbo cultivar in the 2020 year (Table 3). The efficiency of the PGPR on leaf growing have been determined and reported by some researchers (Erturk et al., 2012; Karakurt and Aslantas, 2010b; Karakurt et al., 2011; Orhan et al., 2006; Pérez Moncada et al., 2015; Seema et al., 2018).

Table 3
Average leaf area (cm^2) per blackberry plant

Treatments	2019		2020	
	Chester	Jumbo	Chester	Jumbo
Control	52 d	58 e	56 d	59 d
637Ca	72 b	86 a	73 b	88 a
SY 48	74 b	60 e	74 b	60 d
SY 55	70 b	70 c	71 b	71 c
SK 63	62 cd	79 b	63 c	79 b
637Ca+SY 48	47 e	75 b	47 e	76 b
637Ca+SY 55	80 a	84 a	82 a	85 a
637Ca+SK 63	66 c	77 b	67 c	78 b
637Ca+SY 48+SY 55	55 d	76 b	56 d	77 b
637Ca+SY 48+SK 63	64 c	78 b	67 c	79 b
637Ca+SY 55+SK 63	73 b	73 bc	72 b	74 b
637Ca+SY 48+SY55+SK 63	63 c	65 d	66 c	66 c
Cultivars	65 b	74 a	66 b	74 a

In 2020, plant fresh and dry weight, root length, and root fresh, and dry weight were measured after uprooting the blackberry plants. The root length of the blackberry plants showed differences between cultivars and treatments. In the root length, plant fresh and dry weight, and root fresh and dry weight, the Chester highest value than the Jumbo cultivar. The longest root was measured in the cultivar of Chester. The treatments of 637Ca+SY 48+SY 55, 637Ca, 637Ca+SK 63, 637Ca+SY 48, SY 48, SK 63, and 637Ca+SY 48+SY 55+SK 63 had the longest root in Chester while the 637Ca+SY 48+SY 55, 637Ca, 637Ca+SY 55, 637Ca+SK 63, 637Ca+SY 55+SK 63, SK 63, and 637Ca+SY 48+SY 55+SK 63 treatments had the longest root length in Jumbo (Table 4). The fresh biomass weight of the Chester was found higher in SY 48, SK 63, 637Ca+SK 63, and 637Ca+SY 48+SY 55 than in other treatments. The fresh plant weight of the Jumbo cultivar was measured higher in the 637Ca, SK 63, 637Ca+SY 55, 637Ca+SK 63, 637Ca+SY 48+SY 55, 637Ca+SY 55+SK 63, and 637Ca+SY 48+SY55+SK 63 than the other treatments. All treatments except the Control, SY 55, 637Ca+SY 48, 637Ca+SY 55, 637Ca+SY 48+SK 63, and 637Ca+SY 55+SK 63 were found the highest plant dry weight in Chester. The highest plant dry weight of the Jumbo cultivar was measured in the 637Ca, 637Ca+SY 55, 637Ca+SK 63, and 637Ca+SY 48+SY 55 treatments (Table 4). The highest fresh root weight was measured in 637Ca, SY 48, SK 63, 637Ca+SK 63, 637Ca+SY 48+SY 55, and 637Ca+SY 48+SY 55+SK 63 in Chester while all remaining treatments except for Control, SY 48, SY 55 had the highest fresh root weight in Jumbo. Cultivar. In the Chester cultivar, the highest dry root weight was obtained from SK 63, 637Ca+SK 63, 637Ca+SY 48+SY 55, and 637Ca+SY 48+SY 55+SK 63 treatments. The 637Ca, SK 63, 637Ca+SY 55, 637Ca+SK 63, 637Ca+SY 48+SY 55, 637Ca+SY 55+SK 63, and 637Ca+SY 48+SY 55+SK 63 treatments was the highest in dry root weight in Jumbo (Table 4). In the plant root length and biomass measurements, PGPR treatments had increases in these plant features, and reports of some fruit species supported (Arıkan et al., 2013; Arikan and Pirlak, 2016; Erturk et al., 2012; Ipek et al., 2014; Karakurt and Aslantas, 2010b; Karakurt et al., 2011; Orhan et al., 2006; Pérez Moncada et al., 2015; Pırlak and Köse, 2009; Seema et al., 2018) the study results.

Table 4
Plant and Root Measurements

Treatments	Root Length (cm)		Fresh Plant Weight (g)		Dry Plant Weight (g)		Fresh Root Weight (g)		Dry Root Weight (g)	
	Chester	Jumbo	Chester	Jumbo	Chester	Jumbo	Chester	Jumbo	Chester	Jumbo
Control	88 d	88 c	346 e	313 c	277 d	271 b	158 d	145 c	85 c	77 b
637Ca	135 a	112 a	528 b	351 a	321 ab	279 a	191 a	160 a	84 c	86 a
SY 48	130 a	93 b	447 a	310 c	331 a	258 c	199 a	144 c	89 c	76 b
SY 55	100 c	97 c	363 d	313 c	287 c	259 c	165 c	145 c	89 c	77 b
SK 63	128 a	102 a	455 a	336 a	335 a	259 c	202 a	154 a	111 a	82 a
637Ca+SY 48	130 a	85 c	387 d	322 b	299 bc	264 c	174 b	149 ab	94 b	79 b
637Ca+SY 55	110 b	110 a	402 c	348 ab	307 b	278 a	180 b	159 a	98 b	85 a
637Ca+SK 63	131 a	105 a	452 a	345 a	333 a	276 a	200 a	158 a	110 a	84 a
637Ca+SY 48+SY 55	136 a	115 a	456 a	348 a	336 a	278 a	202 a	159 a	111 a	85 a
637Ca+SY 48+SK 63	89 d	90 b	328 e	319 b	268 e	262 c	151 d	147 ab	80 c	78 b
637Ca+SY 55+SK 63	92 d	103 a	343 e	338 ab	276 d	272 b	157 d	155 a	84 c	83 a
637Ca+SY 48+SY55+SK 63	128 a	100 a	435 b	333 ab	324 ab	270 b	194 a	153 a	106 a	81 a
Cultivars	116 a	100 b	411 a	331 b	307 a	269 b	181 a	152 b	95 a	81 b

The fruit quality criteria and yield value were observed in the 2020 year. In the first year of the study, the plant growth parameters were observed in blackberry plants. The Chester cultivar had a higher fruit number than the Jumbo cultivar. The highest fruit number per plant was counted in 637Ca in the Chester cultivar while the highest fruit number was found in the 637Ca+SY 55 treatment in the Jumbo cultivar (Table 5). The increase in the fruit number is supported by some reports (Kumar et al., 2020; Orhan et al., 2006; Seema et al., 2018).

Table 5
Average fruit number per blackberry plant

Treatments	Chester	Jumbo
Control	115 f	126 d
637Ca	323 a	168 c
SY 48	296 b	83 e
SY 55	209 c	125 d
SK 63	136 e	155 c
637Ca+SY 48	87 g	265 b
637Ca+SY 55	133 e	397 a
637Ca+SK 63	167 d	232 b
637Ca+SY 48+SY 55	157 d	120 d
637Ca+SY 48+SK 63	167 d	155 c
637Ca+SY 55+SK 63	141 e	78 e
637Ca+SY 48+SY55+SK 63	199 c	87 e
Cultivars	177 a	166 b

There was no statistical difference between the average fruit weight of the cultivars. While the 637Ca and SK 63 were found to have the highest average fruit weight in the Chester cultivar, the 637Ca+SY 55 was measured as the highest average fruit weight in the Jumbo cultivar (Table 6). Related results have been reported by other researchers in different fruit species (Arikan and Pirlak, 2016; Ipek et al., 2014; Orhan et al., 2006).

Table 6
Average fruit weight (g)

Treatments	Chester	Jumbo
Control	2.43 b	2.36 d
637Ca	3.05 a	2.62 bc
SY 48	2.49 b	2.40 d
SY 55	2.20 c	2.24 d
SK 63	3.04 a	2.62 bc
637Ca+SY 48	2.27 c	2.64 bc
637Ca+SY 55	2.49 b	3.21 a
637Ca+SK 63	2.52 b	2.80 b
637Ca+SY 48+SY 55	2.52 b	2.82 b
637Ca+SY 48+SK 63	2.43 b	2.75 b
637Ca+SY 55+SK 63	2.57 b	2.31 d
637Ca+SY 48+SY55+SK 63	2.45 b	2.51 c
Cultivars	2.54^{NS}	2.61^{NS}

It has been found no statistical difference between the yields per plant of the cultivars. In the Chester cultivar, 637Ca had the highest yield per plant while the 637Ca+SY 55 was found the highest yield per plant in the Jumbo cultivar (Table 7). In some fruit species, it has been reported that PGPR applications increased fruit weight (Esitken et al., 2005; García-Seco et al., 2013; Karlidag et al., 2010; Karlidag et al., 2007; Pirlak et al., 2007).

Table 7
Yield per blackberry plant (g)

Treatments	Chester	Jumbo
Control	280 g	297 e
637Ca	984 a	442 c
SY 48	737 b	199 g
SY 55	461 c	280 f
SK 63	416 d	408 c
637Ca+SY 48	197 h	699 b
637Ca+SY 55	331 f	1275 a
637Ca+SK 63	422 d	648 b
637Ca+SY 48+SY 55	395 e	339 d
637Ca+SY 48+SK 63	406 d	427 c
637Ca+SY 55+SK 63	364 e	181 g
637Ca+SY 48+SY55+SK 63	486 c	219 g
Cultivars	457^{NS}	452^{NS}

Although, it has been determined to be no statistical difference between the yields per decare of the cultivars, the highest yield per decare was harvested in the 637Ca treatment in the Chester cultivar and the 637Ca+SY 55 in the Jumbo (Table 8). It has been found that PGPR application increased the yield in some fruit species (Esitken et al., 2006; Karlidag et al., 2010; Karlidag et al., 2007).

Table 8
Yield per decare (kg)

Treatments	Chester	Jumbo
Control	93 f	99 d
637Ca	327 a	147 c
SY 48	245 b	66 e
SY 55	153 c	93 d
SK 63	138 d	135 c
637Ca+SY 48	65 g	233 b
637Ca+SY 55	110 e	424 a
637Ca+SK 63	140 d	215 b
637Ca+SY 48+SY 55	131 d	112 d
637Ca+SY 48+SK 63	135 d	142 c
637Ca+SY 55+SK 63	121 e	60 e
637Ca+SY 48+SY55+SK 63	162 c	73 e
Cultivars	152^{NS}	150^{NS}

While it was determined that there was no statistical difference between the cultivars on the TSS, the treatments showed differences in TSS value in each cultivar. The TSS value ranged from 9.3% to13.3% in Chester. The Jumbo TSS value showed to range from 9.7% to 13.9% (Table 9). The TTS value had been obtained previously in studies consistent with our results (Arikan and Pirlak, 2016; Erturk et al., 2012; Kumar et al., 2020; Seema et al., 2018)

Table 9
Total Soluble Solid (%)

Treatments	Chester	Jumbo
Control	11.6 b	11.9 c
637Ca	9.3 d	10.2 d
SY 48	12.6 a	12.9 b
SY 55	13.3 a	13.9 a
SK 63	10.0 c	11.3 c
637Ca+SY 48	13.0 a	12.7 b
637Ca+SY 55	13.1 a	11.5 c
637Ca+SK 63	10.2 c	10.6 d
637Ca+SY 48+SY 55	10.7 b	9.7 d
637Ca+SY 48+SK 63	10.3 c	11.6 c
637Ca+SY 55+SK 63	11.0 b	13.2 a
637Ca+SY 48+SY55+SK 63	12.7 a	12.6 b
Cultivars	11.4^{NS}	11.8^{NS}

The pH of the fruit juice value was not shown differences in both cultivars and treatments of the cultivars. The pH value ranged from 2.99 to 3.43 in treatments of both cultivars. Different studies have

shown consistent results for our study (Arikan and Pirlak, 2016; Kumar et al., 2020; Mia et al., 2005; Seema et al., 2018).

The TAC was not showed a difference between cultivars but there were statistical differences in TAC between treatments (Table 10). The total acidity content ranged from 0.55% to 0.80% and some reports showed equivalent results (Arikan and Pirlak, 2016; Kumar et al., 2020; Mia et al., 2005; Seema et al., 2018).

Table 10
Total Acidity Content (%)

Treatments	Chester	Jumbo
Control	0.76 a	0.64 ab
637Ca	0.72 a	0.63 ab
SY 48	0.65 ab	0.68 ab
SY 55	0.64 ab	0.58 b
SK 63	0.69 ab	0.65 ab
637Ca+SY 48	0.64 ab	0.67 ab
637Ca+SY 55	0.56 b	0.55 b
637Ca+SK 63	0.74 a	0.71 ab
637Ca+SY 48+SY 55	0.63 ab	0.77 a
637Ca+SY 48+SK 63	0.69 ab	0.67 ab
637Ca+SY 55+SK 63	0.80 a	0.69 ab
637Ca+SY 48+SY55+SK 63	0.73 a	0.62 ab
Cultivars	11.4^{NS}	11.8 ^{NS}

The L, C, and H color values of the fruit harvested in treatments of the Chester and Jumbo cultivar were shown in table11 and table 12.

Table 11
Fruit color of the Chester cultivar

Treatments	L	C	H
Control	19.9 a	1.5 ab	45.1 b
637Ca	19.8 a	1.3 b	54.0 a
SY 48	18.9 ab	1.6 ab	43.8 b
SY 55	19.4 ab	1.9 a	39.7 c
SK 63	19.8 a	1.8 a	45.0 b
637Ca+SY 48	20.0 a	1.7 ab	44.3 b
637Ca+SY 55	19.9 a	1.3 b	43.6 b
637Ca+SK 63	20.0 a	1.3 b	48.5 ab
637Ca+SY 48+SY 55	20.5 a	1.8 a	38.5 c
637Ca+SY 48+SK 63	19.3 ab	1.7 ab	47.1 ab
637Ca+SY 55+SK 63	18.1 b	1.4 b	45.5 b
637Ca+SY 48+SY55+SK 63	19.5 ab	1.8 a	55.5 a

Table12
Fruit color of the Jumbo cultivar

Treatments	L	C	H
Control	19.6 a	2.0 ab	35.5 c
637Ca	18.2 ab	1.6 b	47.6 a
SY 48	19.7 a	2.3 ab	41.0 b
SY 55	19.6 a	1.3 c	45.5 ab
SK 63	20.2 a	1.3 c	45.2 ab
637Ca+SY 48	18.3 ab	1.6 b	47.9 a
637Ca+SY 55	17.8 b	1.7 b	48.1 a
637Ca+SK 63	19.9 a	2.9 a	47.9 a
637Ca+SY 48+SY 55	19.8 a	1.3 c	42.0 b
637Ca+SY 48+SK 63	19.8 a	1.9 ab	44.3 ab
637Ca+SY 55+SK 63	20.1 a	3.1 a	39.9 b
637Ca+SY 48+SY55+SK 63	20.9 a	1.5 b	47.9 a

The L, C, and H values of the fruits of the cultivars showed no differences in L and H values, while there was a statistical difference in the C value of the cultivars (Table 13).

Table 13
Fruit color of the cultivars

Cultivars	L	C	H
Chester	19.63	1.62 b	45.93
Jumbo	19.52	1.92 a	44.45

4. Acknowledgements

This research was supported by a grant from Selçuk University, Scientific Research Project Institute, Turkey (Project No: 18401003).

5. References

Arıkan Ş, İpek M, Pırlak L (2013). 'Effects of plant growth promoting rhizobacteria (PGPR) on yield and fruit quality of quince' 2013 International Conference on Agriculture and Biotechnology IPCBEE.

Arikan Ş, Pirlak L (2016). Effects of plant growth promoting rhizobacteria (PGPR) on growth, yield and fruit quality of sour cherry (Prunus cerasus L.). Erwerbs-obstbau58(4), 221-226.

Aslantaş R, Cakmakçi R, Şahin F (2007). Effect of plant growth promoting rhizobacteria on young apple tree growth and fruit yield under orchard conditions. Scientia Horticulturae111(4), 371-377.

Erturk Y, Ercisli S, Cakmakci R (2012). Yield and growth response of strawberry to plant growth-promoting rhizobacteria inoculation. Journal of plant nutrition35(6), 817-826.

Esitken A, Ercisli S, Karlidag H, Sahin F (2005). 'Potential use of plant growth promoting rhizobacteria (PGPR) in organic apricot production' Proceedings of the international scientific conference: Environmentally friendly fruit growing, Polli, Estonia, 7-9 September, 2005. Tartu University Press, pp. 90-97.

Esitken A, Pirlak L, Turan M, Sahin F (2006). Effects of floral and foliar application of plant growth promoting rhizobacteria (PGPR) on yield, growth and nutrition of sweet cherry. Scientia Horticulturae110(4), 324-327.

García-Seco D, Bonilla A, Algar E, García-Villaraco A, Mañero J, Ramos-Solano B (2013). Enhanced blackberry production using Pseudomonas fluorescens as elicitor. Agronomy for sustainable development33(2), 385-392.

Garcia-Seco D, Zhang Y, Gutierrez-Manero FJ, Martin C, Ramos-Solano B (2015). Application of Pseudomonas fluorescens to blackberry under field conditions improves fruit quality by modifying flavonoid metabolism. PLoS One10(11), e0142639.

Glick BR, Penrose DM, Ma W (2001). Bacterial promotion of plant growth. Biotechnology Advances19(2), 135-138.

Ipek M, Pirlak L, Esitken A, Figen Dönmez M, Turan M, Sahin F (2014). Plant growth-promoting rhizobacteria (PGPR) increase yield, growth and nutrition of strawberry under high-calcareous soil conditions. Journal of plant nutrition37(7), 990-1001.

Karakurt H, Aslantas R (2010a). Effects of some plant growth promoting rhizobacteria (PGPR) strains on

plant growth and leaf nutrient content of apple. Journal of Fruit and Ornamental Plant Research18(1), 101-110.

Karakurt H, Aslantas R (2010b). Effects of some plant growth promoting rhizobacteria treated twice on flower thining, fruit set and properties on apple. African Journal of Agricultural Research5(5), 384-388.

Karakurt H, Kotan R, Dadaşoğlu F, Aslantaş R, ŞAHİN F (2011). Effects of plant growth promoting rhizobacteria on fruit set, pomological and chemical characteristics, color values, and vegetative growth of sour cherry (Prunus cerasus cv. Kütahya). Turkish Journal of Biology35(3), 283-291.

Karlidag H, Esitken A, Ercisli S, Donmez M (2010). The use of PGPR (plant growth promoting rhizobacteria) in organic apricot production. Acta horticulturae862, 309-312.

Karlidag H, Esitken A, Turan M, Sahin F (2007). Effects of root inoculation of plant growth promoting rhizobacteria (PGPR) on yield, growth and nutrient element contents of leaves of apple. Scientia Horticulturae114(1), 16-20.

Kavino M, Harish S, Kumar N, Saravanakumar D, Samiyappan R (2010). Effect of chitinolytic PGPR on growth, yield and physiological attributes of banana (Musa spp.) under field conditions. Applied Soil Ecology45(2), 71-77.

Kumar P, Sharma N, Sharma S, Gupta R (2020). Rhizosphere stochiometry, fruit yield, quality attributes and growth response to PGPR transplant amendments in strawberry (Fragaria× ananassa Duch.) growing on solarized soils. Scientia Horticulturae265, 109215.

Marschner H. (2011). Marschner's mineral nutrition of higher plants. Academic press,

Mia MAB, Shamsuddin ZH, Wahab Z, Marziah M (2005). High-yielding and quality banana production through plant growth-promoting rhizobacterial (PGPR) inoculation. Fruits60(3), 179-185.

Orhan E, Esitken A, Ercisli S, Turan M, Sahin F (2006). Effects of plant growth promoting rhizobacteria (PGPR) on yield, growth and nutrient contents in organically growing raspberry. Scientia Horticulturae111(1), 38-43.

Pérez Moncada UA, Ramírez Gómez MM, Zapata Narváez YA, Córdoba Sánchez JM (2015). Effect of the single and combined inoculation with Arbuscular Mycorrhizal Fungi (AMF) and Plant Growth Promoting Rhizobacteria (PGPR) in micropropagated blackberry plants (Rubus glaucus L.). Ciencia y Tecnología Agropecuaria16(1), 95-103.

Pii Y, Graf H, Valentinuzzi F, Cesco S, Mimmo T (2017). 'The effects of plant growth-promoting rhizobacteria (PGPR) on the growth and quality of strawberries' VIII International Symposium on Mineral Nutrition of Fruit Crops 1217. pp. 231-238.

Pırlak L, Köse M (2009). Effects of plant growth promoting rhizobacteria on yield and some fruit properties of strawberry. Journal of plant nutrition32(7), 1173-1184.

Pirlak L, Turan M, Sahin F, Esitken A (2007). Floral and foliar application of plant growth promoting rhizobacteria (PGPR) to apples increases yield, growth, and nutrient element contents of leaves. Journal of Sustainable Agriculture30(4), 145-155.

Robledo-Buriticá J, Aristizábal-Loaiza JC, Ceballos-Aguirre N, Cabra-Cendales T (2018). Influence of plant growth-promoting rhizobacteria (PGPR) on blackberry (Rubus glaucus Benth. cv. thornless) growth under semi-cover and field conditions. Acta Agronómica67(2), 258-263.

Seema K, Mehta K, Singh N (2018). Studies on the effect of plant growth promoting rhizobacteria (PGPR) on growth, physiological parameters, yield and fruit quality of strawberry cv. chandler. Chandler. J. Pharmacogn. Phytochem7(2), 383-387.

Wu W, Li W, Lu L-f, Wang X-m (2007). Comparison of nutrient constituents in fresh fruit of different cultivars of blackberry. Journal of Plant Resources and Environment16(1), 58.

Selcuk Journal of Agriculture and Food Sciences SJAFS

http://sjafs.selcuk.edu.tr/sjafs/index

(2022) 36 (3), 393-398

e-ISSN: 2458-8377

DOI:10.15316/SJAFS.2022.051

Research Article

The Effects of Some Post-Harvest Organic Acid Treatments on the Storage Quality of Brussels Sprouts

Jale BİLGİN [1*], Rafet ASLANTAŞ [1]

[1] Eskişehir Osmangazi University, Faculty of Agriculture, Department of Horticulture, Eskişehir, Türkiye

ARTICLE INFO

Article history:
Received date: 06.06.2022
Accepted date: 12.09.2022

Keywords:
Ascorbic Acid
Folic Acid
Salicylic Acid
Post-harvest Storage
Brussels Sprout

ABSTRACT

In this study, the effects of ascorbic, folic, and salicylic acid treatments on post-harvest quality losses in 'Franklin F1' Brussels sprouts stored at 4°C for 28 days were investigated. Weight loss (%), color values (L*, a*, b*), total soluble solids (TSS, %), pH, titrable acidity (TA, %), total chlorophyll (mg/g), and CO_2 concentration (ppm) were all measured at 7-day intervals. When the findings were compared to the control group, it was observed that all of the treatments were effective in reducing losses in the examined properties. At the end of the storage period, it was found that ascorbic acid was the most effective in terms of weight loss, pH, and TA features, salicylic acid in terms of L* value, and folic acid in terms of preventing pH, total chlorophyll, and CO_2 concentration changes. There has been no research on the effects of ascorbic, folic, and salicylic acid treatments in Brussels sprouts during the post-harvest period, and the goal of this study is to fill in the gaps in the literature and give light on future research. It is thought that determining the appropriate doses of the treatments performed in future studies, as well as examining the efficiency of the treatments in more detail, will be beneficial.

1. Introduction

Brussels sprouts are a Brassica species that gets its name from the Belgian city of Brussels, where it was first grown and is now widely consumed. A mutation of *Brassica oleracea* capitata L. Sabuda, a form of winter curly sprout, is assumed to be the source of Brussels sprouts (Anonymous, 2022). Brussels sprouts are high in water and include a lot of vitamins and minerals (85 %). As a consequence, post-harvest respiration rates are high (40-60 mg CO_2/kg hour at 5°C), therefore it is critical to preserve the species in line with its post-harvest physiology. As with many other types of vegetables, the most common quality loss in Brussels sprouts is yellowing caused by chlorophyll breakdown. Furthermore, darkening and chilling injuries are common problems when storage conditions are not suitable (Anonymous, 2021). It has been determined that Brussels sprouts are stored at optimum 0°C for 3-5 weeks, but chilling injury occurs at -0.6°C (Cantwell and Suslow, 2002). According to Lyons et al., (1959) post-harvest quality losses in Brussels sprouts storage at 5°C were exhibited by yellowing of the leaves by the 15th day.

Many experiments have been conducted to date to minimize quality losses and extend the storage duration of Brussels sprouts post-harvest To that end, the effects of hormone mechanisms (Thomas, 1977), different temperatures, and modified atmosphere packaging (Kosiyachinda and Ketsa, 1983), different light intensity during storage (Kasım and Kasım, 2007), edible coatings (Viña et al, 2007), the effects of LED light (Haspaure et al., 2016; Castillejo et al., 2021), and essential oil treatments (Kraśnıewska et al., 2016) were used. When looking through the relevant literature, no literature was found in which the effects of organic acid treatments in the post-harvest period on Brussels sprouts were examined.

Ascorbic acid (AA), a water-soluble vitamin, plays many important roles in plant metabolism. Studies have shown that ascorbic acid plays an important physiological role in reactive oxygen species (ROS) that occur in plants under stress conditions. It has been reported that exogenous ascorbic acid treatments in the post-harvest period are effective in preventing quality losses during storage in many vegetable species such as spinach (Bergquist et al., 2006), beans (Sikora and Świeca, 2018) and broccoli (Bilgin, 2021). Furthermore, the efficacy of ascorbic acid added to edible coatings (Sun et al., 2010; Saleem et al., 2021) or mixed with natural ingredients such as *Aloe vera* has been studied (Sogvar et al., 2016).

* Corresponding author email: *jbilgin@ogu.edu.tr*

Salicylic acid (SA) is a natural compound in the plant metabolism, plant growth regulators group; plant growth (Tufail et al., 2013), stomatal conductivity (Hayat et al., 2010), photosynthesis mechanism (Khodary, 2004), seed germination (Babalar et al., 2007), disease resistance (Delaney et al., 1994). Dempsey et al., 1999) have been reported to be involved in the metabolism of resistance to different stressors (Hayat et al., 2008; Chavan and Sakhal, 2020). In addition, in many different studies, it has been stated that salicylic acid suppresses ethylene production and fungal rot in the post-harvest period in fruits, prevents chlorophyll deterioration, is effective in the preservation of color values in storage products, and prevents enzyme activities that cause quality losses in the post-harvest period in different species (Leslie and Romani, 1988; Romani et al., 1989; Zhang et al., 2003; Babalar et al., 2007; Sayyari et al., 2009; Wei et al., 2011).

Although the effects of folic acid (FA), a water-soluble vitamin, on human health are well known, its role in plants has only lately been discovered. Studies have reported that folic acid regulates gene expression in plants through the riboswitch mechanism, plays a role in chlorophyll biosynthesis, and plays a role in the oxidative stress tolerance mechanism (Al-Said and Kemal, 2008; Raeisi et al., 2017; Xu et al., 2021). Studies are reporting that exogenous folic acid treatment in the post-harvest period prevents quality losses in broccoli (Xu et al., 2021; Bilgin, 2021).

In this study, the effects of AA, SA, and FA treatments, each of which is a natural compound and their effects on post-harvest quality losses in different species, were investigated on post-harvest quality losses of 'Franklin F1' Brussels sprouts kept at 4°C for 28 days.

2. Materials and Methods

Material

The research material was Brussels sprouts of the 'Franklin F1' variety grown in the Sarcakaya district of Eskişehir. Brussels sprouts were harvested in March 2021, during the last harvest period for cultivated ecology. The average head weight of preserved Brussels sprouts at harvest was 10.61 g, the TSS content was 8.15 %, the pH was 7.21, the TEA content was 0.18 %, and the total chlorophyll content was 0.58 mg/g. Homogeneously selected Brussels sprouts were treated with salicylic acid (2 mM), ascorbic acid (2 mM), and folic acid (5 mg L^{-1}). Tween 80 (0.01%) were added to the prepared solution for better adhesive. Brussels sprouts were immersed in the prepared solutions for 10 minutes and left to dry for one hour after immersion in the solutions. The control group was treated with pure water. After all treatments, Brussels sprouts were kept in non-color plastic containers (11x12x7cm) at 4˚C for 28 days.

Method

After packaging the Brussels sprouts, the first and last weight values were measured by making weight measurements with a precision scale with a sensitivity of 0.01 on the first day of storage and in each storage period. Color values (L*, a*, and b*); It was measured with the aid of a digital colorimeter (Konica Minolta, Japan). Afterward, the preserved products were ground, and the samples were stored at -18°C. The amount of TSS (%) measured with a refractometer and the pH level was measured with a pH meter (HI9321, Hanna, USA) were by using a small amount of water obtained from the ground samples. Amount of TA was determined according to Kowalczyk et al., (2019). The total chlorophyll content was determined by the colorimetric method, taking into account the fresh weights of the frozen samples (Yuan et al., 2010). Measurements were made at wavelengths of 645 and 663nm and the total amount of chlorophyll was calculated according to the formula;

$$Total\ chlorophyll\ (mg\ /\ g) = (20.2 \times \mathrm{OD645\ nm} + 8.02 \times \mathrm{OD663\ nm} \times \mathrm{V})100 \times \mathrm{W}.$$

where OD is optical density, V is a volume of the extract (mL) and W is the weight of the sample (g). The carbon dioxide concentration was measured with the help of a carbon dioxide meter (Tartes-AZ 7752) by taking samples from the airtight packages with an injector.

Statistical analysis

The study was carried out with 3 replications and 4 parallels by designing random plots according to a 4x5 factorial experimental design. In the study; the statistical model, the treatment (Control, FA, SA, and AA), and storage time (0, 7, 14, 21, and 28 days) were taken as factors, and whether there was a statistical difference between the treatment and storage time averages and the presence of interaction were investigated. In the statistical analysis of the research, after applying √(X_i+3/8) square root transformation to the weight loss feature, which does not provide homogeneity of group variances, and √(X_i+3/8) to the marketable status feature, two-way analysis of variance (Two-way ANOVA) was applied to all other features (Zar, 2014). IBM SPSS 23 (IBM Corp. Released, 2015) statistical package program was used in statistical analyzes in terms of the features discussed to determine the effect of the treatment topics on the examined features. In the research, Tukey HSD multiple comparison tests at a 5% significance level were determined as statistically significant in terms of the existence of interaction and the differences between which treatment and storage time averages.

3. Results and Discussion

Weight loss occurs as a result of the continued metabolic activity and the water loss in crops during the post-harvest period. As a result, wrinkling, shrinkage, and quality losses occur in the tissues. Table 2 shows that, the effect of the treatments on the weight loss of Brussels sprouts during the 28-day storage period. According to the findings, the difference between the treatment averages was found to be statistically significant ($p<0.05$). At the end of the storage period, it was determined that the lowest weight loss average occurred in

folic acid treatment (3.3%). Similar to these findings, similar results were found in studies on broccoli, and it was determined that folic acid treatment had a preventive effect on weight loss (Xu et al., 2021; Bilgin, 2021). In addition, when compared with the relevant literature, it is seen that the average head weight is higher than the studies conducted on this country's ecology (Sönmez, 2007; Yılmaz and Sarı, 2019). At the end of the 28-day storage period, it was determined that the average weight loss ranged between 3.3% (FA) and 3.8% (AA). Depending on the treatment effect, Viña et al. (2007) reported 15-25% weight loss on the 30th day of storage, while Kasım and Kasım (2007) reported 3-10% weight loss at the end of the 21-day storage period. It is thought that the difference between the literature and the findings is due to differences in the treatments, variety, and ecological differences.

Changes in the color values of the goods occur in tandem with the loss of quality in the tissues during storage. In the study, day x treatment interaction was determined in all of the color values findings. The most fundamental indicator of decay in Brussels sprouts during the post-harvest period is yellowing, which is caused by the degradation of chlorophyll. Although there is a decrease in L* value during storage, especially in fruit species, when Table 1 is examined, it is seen that there is an increase in L* value in the study, similar to the relevant literature and as in many vegetable species (Bonasia et al., 2013; Jin et al., 2015; Hasperué et al., 2016). On the other hand, there are also kinds of literature stating that the L* value decreases as the storage time increases (Viña et al., 2007; Kraśniewska et al., 2016). When Table 1 is examined, it is seen that the difference between the treatment averages is statistically significant, and all the treatments compared to the control group have a preventive effect on color values. In addition, the existence of day x treatment interaction was determined ($p<0.01$). At the end of the 28-day storage period, it was determined that the SA treatment was the most effective to prevent the change in L* value. When the changes in a* and b* values in the storage period are examined in Table 1, it is seen that both values increase as a result of chlorophyll degradation as the storage time increases in both values, similar to the relevant literature. At the end of the storage period, the highest a* value was 0.35 and the b* value was 39.25 as a result of local decays in the control group. When the findings were examined, it was determined that the least changes in the a* and b* values at AA treatment, and the* and b* values were -3.68 and 34.80, respectively. These findings are similar to the findings that ascorbic acid preserves color values in the post-harvest period in different species (Gil et al., 1998; Terdbaramee et al., 2006; Lin et al., 2007; Liu et al., 2014; Sikora and Świeca, 2018).

The changes in visual sensory scores are shown in Table 1. When the findings were examined, it was determined that although folic acid treatment had a preventive effect on changes in features such as weight loss and total chlorophyll amount, the highest average in visual quality scores was in SA treatment. This is because the folic acid solution has a yellow/mustard color due to the color of the folic acid, and this color affects the storage Brussels sprouts. Considering this situation, it is thought that it would be beneficial to determine the effective post-harvest folic acid treatment doses for different species. The findings are in line with studies in different species where SA has been reported to affect the preservation of visual quality in the post-harvest period (Shafiee et al., 2010; Wei et al., 2011; Kant et al., 2013; Chavan and Sakhal, 2020).

Table 1
Physical features of 'Franklin F1' Brussels sprouts during post-harvest storage at 4 °C.

Features	Treatments	Storage Period (Day)					Mean[2]
		0	7	14	21	28	
Weight Loss (%)	Control	0.00±0.00	2.29±0.52	2.43±0.41	3.47±1.35	3.71±1.98	2.38±0.53
	FA	0.00±0.00	2.02±0.36	2.57±0.56	3.01±0.68	3.30±1.09	2.18±0.37
	SA	0.00±0.00	1.49±0.34	2.07±0.84	2.89±0.60	3.59±1.31	2.01±0.41
	AA	0.00±0.00	1.78±0.39	2.59±0.89	3.04±0.94	3.87±0.73	2.26±0.41
	Mean[1]	0.00±0.00w	1.90±0.20x	2.42±0.32xy	3.10±0.42xy	3.62±0.61y	
L*	Control	44.46±0.18Da	47.24±0.03Ca	49.30±0.07Ba	51.79±0.16Aa	60.02±0.89Aa	50.58±1.22
	FA	42.29±0.50Ec	46.26±0.07Db	47.52±0.10Cd	50.75±0.08Bc	54.47±0.44Ac	48.50±0.87
	SA	43.51±0.14Eb	45.67±0.06Dc	48.14±0.06Cc	50.06±0.21Bd	53.06±0.26Ad	47.85±0.85
	AA	44.55±0.30Ea	46.68±0.04Db	48.76±0.13Cb	51.34±0.048Bb	57.09±0.44Ab	49.67±1.00
	Mean[1]	43.70±0.27	46.46±0.15	48.43±0.18	50.98±0.18	56.16±0.73	
a	Control	-7.45±0.06Ea	-6.32±0.08Da	-5.27±0.08Ca	-4.16±0.05Ba	0.35±1.07Aa	-4.57±0.65
	FA	-7.62±0.29Eab	-7.13±0.02Dc	-5.93±0.06Cc	-4.71±0.05Bbc	-2.86±0.26Ac	-5.65±0.40
	SA	-7.95±0.07Eb	-6.887±0.03Dbc	-5.60±0.05Cb	-4.51±0.04Bb	-1.85±0.23Ab	-5.36±0.48
	AA	-7.80±0.03Eab	-6.627±0.05Db	-6.07±0.01Cc	-4.92±0.03Bc	-3.68±0.06Ad	-5.82±0.32
	Mean[1]	-7.71±0.08	-6.74±0.08	-5.72±0.08	-4.57±0.07	-2.01±0.47	
b	Control	27.01±0.27Ec	30.81±0.09Da	32.61±0.19Ca	34.36±0.05Ba	39.29±0.53Aa	32.82±0.94
	FA	27.25±0.33Ec	30.16±0.10Db	32.12±0.08Ca	34.01±0.05Bb	35.99±0.14Ab	31.91±0.70
	SA	28.05±0.30Ea	29.89±0.06Dc	31.73±0.02Cb	33.77±0.10Bc	35.38±0.09Ac	31.31±0.58
	AA	27.67±0.19Eb	29.56±0.05Dc	31.31±0.08Cc	33.22±0.05Bd	34.81±0.17Ad	31.31±0.58
	Mean[1]	27.49±0.16	30.11±0.12	31.94±0.13	33.84±0.11	36.37±0.47	
Sensory Score	Control	5.00±0.00Aa	4.50±0.00Bb	4.10±0.06Cb	3.43±0.07Db	2.60±0.06Ed	3.93±0.23
	FA	5.00±0.00Aa	4.47±0.03Bb	4.23±0.03Cb	4.03±0.03Da	3.00±0.00Ec	4.15±0.18
	SA	5.00±0.00Aa	4.67±0.03Ba	4.50±0.00Ba	4.07±0.07Ca	4.03±0.03Ca	4.45±0.10
	AA	5.00±0.00Aa	4.57±0.09Bab	4.53±0.03Ba	3.50±0.00Cb	3.47±0.03Cb	4.21±0.17
	Mean[1]	5.00±0.00	4.55±0.03	4.35±0.06	3.76±0.09	3.28±0.16	

* The comparison of days within each treatment is shown as A, AB, B, C, D, E, while the difference between treatments within each day is shown as a, ab, b, bc, c, d, and the comparison of storage period means are shown as x, xy, y and w.

During the post-harvest period, the stored products lose water, become concentrated, and increase in the amount of TSS. In the findings obtained from the study, it is seen in Table 2 that there is an increase in the amount of TSS during the storage period. It was determined that all of the treatments, the effects of which were examined in the study, were effective in preventing the change in the TSS compared to the control group. One of the biochemical changes that occur in stored products is the changes in the pH level of the juice. This feature is also important in varieties where it is common to be served frozen or minimally processed, such as Brussels sprouts. According to findings, the difference between the treatment averages is statistically significant, and all the treatments compared to the control group have a preventive effect on pH levels. In addition, the existence of day x treatment interaction was determined ($p<0.01$). When Table 2 is examined, it is seen that there is an increase in the pH level during the storage period. According to the findings, the most important effect in preventing changes in pH level is in the AA treatment.

The changes in TA are shown in Table 2, and it is seen that the amount of TA increases during storage, similar to the relevant literature (Kowalczyk et al., 2019). When the findings are examined, it is seen that the difference between the treatment averages is statistically significant ($p<0.05$). When the treatment averages were examined, it was determined that the lowest TA average was 0.17% in the SA group. In addition, although the day x treatment interaction is not statistically significant, it is seen that the lowest TA increase was observed in the SA-treated group at all storage periods. These findings are compatible with studies that reported the preventive effect of SA treatment on the change in the amount of TA in the post-harvest period in different species (Davarynejad et al., 2015; Baninaiem et al., 2016).

The most important symptom of quality loss in the post-harvest period in many vegetable species with green color is the loss of species-specific color as a result of chlorophyll degradation. It has been reported that chlorophyll content, which is essentially a color pigment, increases the anticarcinogenic impact of Brussels sprouts as well as its effects on aroma (Olivera et al., 2011). Therefore, the amount of chlorophyll is an important feature in terms of both visual quality and its effect on biochemical content, and it is of great importance to prevent its losses in the post-harvest period. In the study, it was observed that the total chlorophyll content decreased during storage.

According to findings, the difference between the treatment averages is statistically significant, and all the treatments compared to the control group have a preventive effect on total chlorophyll amount. In addition, the existence of day x treatment interaction was determined ($p<0.01$). When the findings are examined, the observation of the lowest total chlorophyll content in the control group at the end of the storage period shows the effectiveness of the treatments. At the end of the storage period, the highest total chlorophyll amount was observed in FA and SA treatments. The findings are compatible with studies reporting the preventive effect of FA (Xu et al., 2021; Bilgin, 2021) and SA treatments prevent chlorophyll degradation (Wei et al., 2011).

The respiration proceeds in the stored crops, as a result of which increases the CO_2 concentration in the storage atmosphere. For this reason, monitoring the CO_2 concentration of the storage atmosphere is of great importance to prevent quality losses. In the species such as Brussels sprouts, which are usually offered to the consumer in closed packages, the increased CO_2 concentration in the limited air mass in the package affects the storage quality negatively.

Table 2
Biochemical features and CO_2 concentration of 'Franklin F1' Brussels sprouts during post-harvest storage at 4 °C.

Features	Treatments	Storage Period (Day)					Mean[2]
		0	7	14	21	28	
TSS (%)	Control	7.12±0.12	7.50±0.00	8.00±0.00	9.00±0.00	9.87±0.12	8.30±0.23Y
	FA	7.00±0.00	7.25±0.14	7.75±0.14	8.37±0.12	9.62±0.12	8.00±0.22Z
	SA	7.25±0.14	7.12±0.12	7.50±0.29	8.62±0.12	9.62±0.12	7.97±0.24Z
	AA	7.00±0.20	7.25±0.14	7.50±0.20	8.37±0.12	9.75±0.14	7.97±0.24Z
	Mean[1]	7.09±0.07z	7.28±0.06z	7.69±0.10y	8.59±0.08x	9.72±0.06w	
pH	Control	5.82±0.11Cb	7.00±0.01Ba	7.21±0.00Ba	7.36±0.00Ba	7.57±0.06Aa	6.99±0.14
	FA	5.80±0.11Db	6.80±0.04Cab	7.14±0.01Ba	7.31±0.00ABa	7.41±0.00Aa	6.90±0.14
	SA	5.99±0.12Cab	6.95±0.01Ba	7.20±0.00ABa	7.34±0.00Aa	7.44±0.01Aa	6.99±0.12
	AA	6.05±0.09Da	6.69±0.03Cb	7.08±0.02Ba	7.27±0.02ABa	7.38±0.00Aa	6.89±0.11
	Mean[1]	5.92±0.06	6.86±0.03	7.16±0.01	7.32±0.01	7.45±0.02	
TA (%)	Control	0.16±0.00	0.17±0.00	0.18±0.00	0.20±0.00	0.22±0.00	0.19±0.00Y
	FA	0.17±0.00	0.17±0.01	0.18±0.00	0.19±0.00	0.20±0.00	0.18±0.00Y
	SA	0.16±0.00	0.16±0.00	0.17±0.00	0.17±0.00	0.20±0.00	0.17±0.00Z
	AA	0.17±0.00	0.17±0.00	0.18±0.00	0.18±0.00	0.21±0.00	0.18±0.00Y
	Mean[1]	0.16±0.00z	0.17±0.00z	0.18±0.00y	0.19±0.00y	0.21±0.00x	
Total Chlorophyll (mg/g)	Control	0.58±0.00Ab	0.57±0.00Bb	0.51±0.00Ca	0.37±0.004Dc	0.35±0.00Ec	0.47±0.02
	FA	0.58±0.01Aa	0.57±0.00Aab	0.51±0.00Ba	0.48±0.01Ca	0.46±0.00Da	0.52±0.01
	SA	0.57±0.00Ab	0.57±0.00Aab	0.51±0.00Ba	0.47±0.00Cb	0.47±0.00Da	0.52±0.01
	AA	0.58±0.01Aa	0.58±0.01Aa	0.51±0.00Ba	0.48±0.00Cab	0.37±0.01Db	0.50±0.02
	Mean[1]	0.58±0.00	0.57±0.00	0.51±0.00	0.45±0.01	0.41±0.01	
CO_2 concentration (ppm)	Control	0.00±0.00D	696.67±0.87Ca	695.00±1.15Ca	722.67±1.45Ba	845.00±2.08Aa	739.80±18.6
	FA	0.00±0.00C	660.67±1.20Bc	683.67±0.68Bb	683.67±0.85Bc	766.00±1.73Ad	698.50±12.1
	SA	0.00±0.00D	688.67±0.58Cb	674.67±0.88Cc	694.33±1.45Bb	774.33±1.20Ac	708.00±11.8
	AA	0.00±0.00E	683.33±0.67Bc	676.00±1.15Cc	670.33±1.76Dd	821.33±1.76Ab	712.80±19.0
	Mean[1]	0.00±0.00	682.33±4.05	682.33±2.48	692.75±5.84	801.67±9.89	

* The comparison of days within each treatment is given as A, AB, B, C, D, E, while the difference between treatments within each day is shown as a, ab, b, bc, c, d. Also, the comparison of the treatment means regardless of the storage time is expressed as Y and Z, while the comparison of the storage periods regardless of the treatment is shown as x, w, y and z.

The temporal variation of the CO_2 concentration detected in the storage atmosphere samples taken from the inside of the packages of storage Brussels sprouts is shown in Table 2. According to the findings, close CO_2 concentration values were obtained on the 7th, 14th, and 21st days of storage (between 684-722 ppm), but these values increased to 846 ppm on the 28th day of storage. The findings show that CO_2 concentration increases at the last period of storage (28th day). This situation can be interpreted as the respiratory rate of the storage crop increased during this period and therefore the quality losses accelerated. The significant increase in the changes in weight loss and total chlorophyll (Table 2) content on the 28th day of storage compared to the other days features the relationship between the properties.

4. Conclusion

In this study, the effects of AA, FA, and SA treatments, each of which are natural compounds, on the prevention of quality losses in the post-harvest period in Brussels sprouts were investigated. According to the findings, it was determined that quality losses occurred at a lower rate compared to the control group in all of the treatments, although it varies according to the day and features. With the study, information on the effectiveness of AA, FA, and SA treatments in Brussels sprouts, where studies on preservation are limited compared to many vegetable species, have been brought to the literature. In future studies, it is thought that it will be useful to determine the effective doses of the treatments in terms of species and to examine the effectiveness of the treatments more comprehensively.

5. Acknowledgements

Some of the findings from this study were presented orally at the 4th International Agriculture, Environment and Health Congress.

6. References

Anonymous (2022). https://www.newworldencyclopedia.org/entry/Brussels sprout Accessed on, January 4, 2022.

Anonymous (2021). https://www.postharvest.net.au/product-guides/brussels-sprout/ Accessed on, July 21, 2021.

Al-Said MA, Kamal AM (2008). Effect of foliar spray with folic acid and some amino acids on flowering, yield, and quality of sweet pepper. *Journal of Plant Production*, 33 (10), 7403-7412.

Babalar M, Asghari M, Talaei A, & Khosroshahi A (2007). Effect of pre-and postharvest salicylic acid treatment on ethylene production, fungal decay, and overall quality of Selva strawberry fruit. *Food Chemistry*, 105 (2), 449-453.

Baninaiem E, Mirzaaliandastjerdi AM, Rastegar S, Abbaszade KH (2016). Effect of pre-and postharvest salicylic acid treatment on quality characteristics of tomato during cold storage. *Advances in Horticultural Science*, 30 (3), 183-192.

Bilgin J (2021). The Effects of Salicylic, Folic, and Ascorbic Acid Treatment on Shelf Life Quality of Broccoli Florets. *Journal of Agricultural Production*, 2 (1), 7-15.

Bonasia A, Conversa G, Lazzizera C, Elia A (2013). Pre-harvest nitrogen and Azoxystrobin application enhances postharvest shelf-life in Butterhead lettuce. *Postharvest biology and technology*, 85 (2013), 67-76.

Castillejo N, Martínez-Zamora L, Gómez PA, Pennisi G, Crepaldi A, Fernández JA, Artés-Hernández F (2021). Postharvest LED lighting: effect of red, blue and far red on quality of minimally processed broccoli sprouts. *Journal of the Science of Food and Agriculture*, 101 (1), 44-53.

Cantwell M and Suslow T (2002). Lettuce, Crisphead: Recommendations for Maintaining Postharvest Quality. http://ucanr.edu/sites/Postharvest_Technology_Center_/Commodity_Resources/Fact_Sheets/Datastores/Vegetables_English/?uid=19&ds=799 Accessed on, January 18, 2022.

Chavan RF, Sakhal BK (2020). Studies on the effect of exogenous application of salicylic acid on post-harvest quality and shelf life of tomato fruit Cv. Abhinav. Food Res, 4 (5), 1444-1450.

Davarynejad GH, Zarei M, Nasrabadi ME, Ardakani E (2015). Effects of salicylic acid and putrescine on storability, quality attributes and antioxidant activity of plum cv.'Santa Rosa'. *Journal of food science and technology*, 52 (4), 2053-2062.

Delaney TP, Uknes S, Vernooij B, Friedrich L, Weymann K, Negrotto D, Gaffney T, Gut-Rella M, Kessmann H, Ward E, Ryals J (1994). A central role of salicylic acid in plant disease resistance. *Science*, 266 (5188), 1247-1250.

Dempsey DMA, Shah J, Klessig DF (1999). Salicylic acid and disease resistance in plants. *Critical Reviews in Plant Sciences*, 18 (4), 547-575.

Gil MI, Gorny JR, Kader AA (1998). Responses of'Fuji'apple slices to ascorbic acid treatments and low-oxygen atmospheres. HortScience: a publication of the American Society for Horticultural Science USA.

Hasperué JH, Rodoni LM, Guardianelli LM, Chaves A R, Martínez GA (2016). Use of LED light for Brussels sprouts postharvest conservation. *Scientia Horticulturae*, 213 (2016), 281-286.

Hayat S, Hasan SA, Fariduddin Q, Ahmad A (2008). Growth of tomato (Lycopersicon esculentum) in response to salicylic acid under water stress. *Journal of Plant Interactions*, 3 (4), 297-304.

Hayat Q, Hayat S, Irfan M, Ahmad A (2010). Effect of exogenous salicylic acid under changing environment: a review. *Environmental and Experimental Botany*, 68 (1), 14-25.

Jin P, Yao D, Xu F, Wang H, Zheng Y (2015). Effect of light on quality and bioactive compounds in postharvest broccoli florets. *Food chemistry*, 172 (2015), 705-709.

Kant K, Arora A, Singh VP, Kumar R (2013). Effect of exogenous application of salicylic acid and oxalic acid on post harvest shelf-life of tomato (Solanum lycopersicon L.). *Indian Journal of Plant Physiology*, 18 (1), 15-21.

Kasım R, Kasım MU (2007). Inhibition of yellowing in Brussels sprouts (B. oleraceae var. gemmifera) and broccoli (B. oleraceae var. italica) using light during storage. *Journal of Food Agriculture and Environment*, 5 (3/4), 126.

Kosiyachinda S, Ketsa S (1983). Effects of Temperature and Modified Atmosphere on Quality and Storage Life of Brussels Sprouts. *Agriculture and Natural Resources*, 17 (2), 9-13.

Kowalczyk D, Kordowska-Wiater M, Kałwa K, Skrzypek T, Sikora M, Łupina K (2019). Physiological, qualitative, and microbiological changes of minimally processed Brussels sprouts in response to coating with carboxymethyl cellulose/candelilla wax emulsion. *Journal of Food Processing and Preservation*, 43 (8), e14004.

Kraśniewska K, Gniewosz M, Kosakowska O, Cis A (2016). Preservation of brussels sprouts by pullulan coating containing oregano essential oil. *Journal of Food Protection*, 79 (3), 493-500.

Khodary SEA (2004). Effect of salicylic acid on the growth, photosynthesis and carbohydrate metabolism in salt stressed maize plants. International *Journal of Agriculture & Biology*, 6 (1), 5-8.

Leslie CA, Romani RJ (1988). Inhibition of ethylene biosynthesis by salicylic acid. *Plant Physiology*, 88 (3), 833-837.

Lichtenthaler HK (1987) Chlorophylls and Carotenoids: Pigments of Photosynthetic Biomembranes, *Methods in Enzymology*.

Lin L, Li QP, Wang BG, Cao JK, Jiang WB (2007). Inhibition of core browning in 'Yali'pear fruit by postharvest treatment with ascorbic acid. *The Journal of Horticultural Science and Biotechnology*, 82 (3), 397-402.

Liu K, Yuan C, Chen Y, Li H, Liu J (2014). Combined effects of ascorbic acid and chitosan on the quality maintenance and shelf life of plums. *Scientia Horticulturae*, 176 (2014), 45-53.

Lyons J, Rappaport L (1959). Quality of brussels sprouts: Low temperatures during handling operations retarded quality deterioration in study of temperature effects on respiration. *Calif Agr* 13 (6):11.

Olivera DF, Viña SZ, Ferreyra RM, Mugridge A, Mascheroni RH, Chaves AR (2011). Integration of an Objective Approach Focused on the Consumer to Evaluate the Quality of Processed Brussels Sprouts. *Food and Nutrition* 2 (2011): 1011-1017.

Raeisi J, Pakkish Z, Saffari VR (2017). Efficiency of folic acid in improving yield and fruit quality of strawberry. *Journal of Plant Physiology & Breeding*, 7 (1), 15-25.

Romani RJ, Hess BM, Leslie CA (1989). Salicylic acid inhibition of ethylene production by apple discs and other plant tissues. *Journal of Plant Growth Regulation*, 8 (1), 63-69.

Sayyari M, Babalar M, Kalantari S, Serrano M, Valero D (2009). Effect of salicylic acid treatment on reducing chilling injury in stored pomegranates. *Postharvest Biology and Technology*, 53 (3), 152-154.

Shafiee M, Taghavi TS, Babalar M (2010). Addition of salicylic acid to nutrient solution combined with postharvest treatments (hot water, salicylic acid, and calcium dipping) improved postharvest fruit quality of strawberry. *Scientia Horticulturae*, 124 (1), 40-45.

Sun D, Liang G, Xie J, Lei X, Mo Y (2010). Improved preservation effects of litchi fruit by combining chitosan coating with ascorbic acid treatment during postharvest storage. *African Journal of Biotechnology*, 9 (22), 3272-3279.

Sogvar OB, Saba MK, Emamifar A (2016). Aloe vera and ascorbic acid coatings maintain postharvest quality and reduce microbial load of strawberry fruit. *Postharvest biology and Technology*, 114 (2016), 29-35.

Bergquist SÅ, Gertsson UE, Olsson ME (2006). Influence of growth stage and postharvest storage on ascorbic acid and carotenoid content and visual quality of baby spinach (Spinacia oleracea L.). *Journal of the Science of Food and Agriculture*, 86 (3), 346-355.

Saleem MS, Anjum MA, Naz S, Ali S, Hussain S, Azam M, Ejaz S (2021). Incorporation of ascorbic acid in chitosan-based edible coating improves postharvest quality and storability of strawberry fruits. *International Journal of Biological Macromolecules*, 189, 160-169.

Sikora M, Świeca M (2018). Effect of ascorbic acid postharvest treatment on enzymatic browning, phenolics and antioxidant capacity of stored mung bean sprouts. *Food Chemistry*, 239, 1160-1166.

Sönmez U (2007). Farklı dikim zamanlarının Brüksel lahanası (Brassica oleracea var. gemmifera) çeşitlerinde gelişme ve verime etkisi. Atatürk Üniversitesi Fen Bilimleri Enstitüsü Bahçe Bitkileri Anabilim Dalı Yüksek Lisans Tezi.

Terdbaramee U, Ratanakhanokchai K, Kanlavanarat S (2006). Control of postharvest browning of lychee fruit using ascorbic acid. IV International Conference on Managing Quality in Chains-The Integrated View on Fruits and Vegetables Quality. Bangkok. https://doi.org/10.17660/ActaHortic.2006.712.86 Accesed on, December 20, 2021.

Thomas TH (1977). Hormonal control of Brussels sprout senescence. In Symposium on Vegetable Storage August 2, 1977, New York, USA.

Tufail A, Arfan M, Gurmani AR, Khan A, Bano A (2013). Salicylic acid induced salinity tolerance in maize (Zea mays). *Pakistan Journal of Botany*, 45 (S1), 75-82.

Xu D, Zuo J, Fang Y, Yan Z, Shi J, Gao L, Wang Q, Jiang A (2021). Effect of folic acid on the postharvest physiology of broccoli during storage. *Food Chemistry*, 339 (2021), 127981.

Yılmaz İB, Sarı N (2019). Plant Growth, Yield and Sprout Quality in Brussels Sprouts Sowed in Different Periods in Çukurova Region Conditions. *Turkish Journal of Agriculture-Food Science and Technology*, 7 (7), 1062-1068.

Yuan G, Sun B, Yuan J, Wang Q (2010). Effect of 1-methylcyclopropene on shelf life, visual quality, antioxidant enzymes and health-promoting compounds in broccoli florets. *Food Chemistry*, 118 (3), 774-781.

Zar J (2014). Biostatistical analysis fifth edition. Pearson Education India.

Zhang Y, Chen K, Zhang S, Ferguson I (2003). The role of salicylic acid in postharvest ripening of kiwifruit. *Postharvest Biology and Technology*, 28 (1), 67-74.

Wei Y, Liu Z, Su Y, Liu D, Ye X (2011). Effect of salicylic acid treatment on postharvest quality, antioxidant activities, and free polyamines of asparagus. *Journal of Food Science*, 76 (2), S126-S132.

Viña SZ, Mugridge A, García MA, Ferreyra RM, Martino MN, Chaves AR, Zaritzky NE (2007). Effects of polyvinylchloride films and edible starch coatings on quality aspects of refrigerated Brussels sprouts. *Food Chemistry*, 103 (3), 701-709.

Selcuk Journal of Agriculture and Food Sciences SJAFS

http://sjafs.selcuk.edu.tr/sjafs/index

(2022) 36 (3), 399-404

e-ISSN: 2458-8377

DOI:10.15316/SJAFS.2022.052

Research Article

Effects of Replacing Breadcrumbs with Buckwheat, Chickpea, Corn and Millet Flour in Gluten-Free Meatball Formulation

Ali Samet BABAOĞLU*

Selçuk University, Faculty of Agriculture, Department of Food Engineering, Konya, Türkiye

ARTICLE INFO

Article history:
Received date: 12.08.2022
Accepted date: 29.09.2022

Keywords:
Celiac disease
Cooking yield
Meat product
Meatball quality
Sensory

ABSTRACT

The objective of this study was to evaluate the effects of different gluten-free flours on the physicochemical, textural and sensory properties of meatballs. Five different groups of meatballs were produced: C: control meatballs with breadcrumbs, Gf1: meatballs with buckwheat flour, Gf2: meatballs with chickpea flour, Gf3: meatballs with corn flour and Gf4: meatballs with millet flour. The chickpea flour increased the protein content of raw meatballs ($P < 0.05$). The cooking yield results were higher in gluten-free meatballs than in control samples ($P < 0.05$). Chickpea flour (Gf2) and corn flour (Gf3) were the most effective flours for reducing the diameter of meatballs ($P < 0.05$). The highest antioxidant activity was found in the meatballs with buckwheat flour (Gf1) ($P < 0.05$). The chickpea flour improved the texture of the meatball samples ($P < 0.05$), while corn and millet flour increased the hardness and chewiness values of the meatballs ($P < 0.05$). Millet flour decreased the flavour score compared to the control ($P < 0.05$), whereas the other gluten-free flours had no significant effect on all sensory properties of the meatballs ($P > 0.05$). This study suggests that chickpea flour had a better effect on the quality characteristics of meatballs among gluten-free flours.

1. Introduction

Meatballs are one of the restructured meat products that can be made from ground beef, pork, chicken or fish. Meatballs are very popular in all walks of life around the world and are made in both domestic and commercial meat processing plants. A meatball is minced meat rolled into a small ball, usually together with other ingredients such as breadcrumbs or bread, chopped onions, eggs, butter and spices (Kartikawati & Purnomo 2019; Saba et al 2018). Breadcrumbs or bread are made from wheat flour, which contains about 60% gluten (Jackson et al 2006). Celiac disease is one of the most notable gluten-related diseases, affecting about 1% of the world's population (Cui et al 2017). Celiac disease is a genetically predisposed autoimmune problem. People with celiac disease often suffer adverse reactions to products containing gluten (Larrosa et al 2013). As the only treatment option for celiac disease is a lifelong gluten-free diet (Gobbetti et al 2018), it is of great importance to improve gluten-free food alternatives that can meet sensory and nutritional quality requirements (Kerimoğlu & Serdaroğlu 2019). Therefore, it is necessary to advance the new ingredients and formulations, especially to produce gluten-free meat products. Buckwheat (*Fagopyrum esculentum* Moench), a type of pseudo-cereal, has been suggested as a good alternative for celiac patients because it contains bio-quality proteins, high levels of dietary fiber, flavonoids and essential minerals (Park et al 2016). The chickpea (*Cicer aeritinum* L.) is the most consumed legume in the world. The chickpea is a cheap and gluten-free legume with nutritious components such as carbohydrates, proteins, lipids, vitamins and minerals, and with high protein digestibility and low glycaemic index properties (Gobbetti et al 2018; Sofi et al 2020). Corn (*Zea mays* subsp.) flour has proven to be one of the most suitable flours for developing gluten-free products. This could be due to its soft taste, easily digestible carbohydrate content, low prolamin content and hypoallergenic properties (Marco & Rossell 2008). Millet (*Panicum miliaceum*), a gluten-free cereal, is considered one of the most important crops. It is also considered a good source of carbohydrates and has a high protein content, which is a richer source of essential amino acids than wheat (Kalinova & Moudry 2006).

Limited studies on the use of different cereal and legume flours to produce of gluten-free meat products such as rice flour in chicken nugget (Jackson et al 2006),

* Corresponding author email: *asmtbb@gmail.com*

sorghum flour in chicken nugget (Devatkal et al 2011), millet flour in kibbeh (Brasil et al 2015), chickpea flour in chicken nugget (Öztürk et al 2018), corn flour in fish patty (Romero et al 2018), soy flour in meatball (Mastanjević et al 2014) and quinoa flour in meatball (Bağdatlı 2018) are available in the literature. However, there is a lack of comprehensive study in which gluten-free meatballs are made from buckwheat, chickpea, corn and millet flour as a substitute for breadcrumbs. Therefore, the aim of this study is to evaluate the physicochemical, textural and sensory characteristics of meatballs containing buckwheat, chickpea, corn or millet flour as a substitute for breadcrumbs in the formulation.

2. Materials and Methods

2.1. Materials

The beef (*Biceps femoris*) and beef fat were obtained from a butcher in Konya. The breadcrumbs, buckwheat flour (Rasayana, Konya, Turkey), chickpea flour (Doğalsan, Ankara, Turkey), corn flour (Bağdat, Ankara, Turkey) and millet flour (Rasayana, Konya, Turkey) were purchased from a market in Konya. The salt (Salina, Ankara, Turkey), onion powder (Bağdat, Ankara, Turkey) and black pepper (Bağdat, Ankara, Turkey) used in the production of meatballs were obtained from a market in Konya.

2.2. Preparation of meatballs

The beef and beef fat were minced twice in a meat grinder with a plate with 3 mm diameter holes (Kitchen Aid, Classic Model, USA) and then the minced meat was divided into five parts. As outlined in Table 1, five different meatball formulations were prepared as follows: C (control group-including breadcrumbs), Gf1 (including buckwheat flour), Gf2 (including chickpea flour), Gf3 (including corn flour) and Gf4 (including millet flour). In the formulation of meatball samples, the breadcrumb was replaced completely by gluten-free flours in the groups of Gf1, Gf2, Gf3 and Gf4. The minced meat and the other ingredients were weighed separately and then mixed for 7 min. This meatball dough was stored at 4 ^{o}C for 5 h and formed into meatballs in a petri dish (40 g per meatball) to obtain an average size (about 4 cm in diameter).

Table 1
Formulation of meatball samples

Formulation (%)	Meatball samples				
	C	Gf1	Gf2	Gf3	Gf4
Meat	75.00	75.00	75.00	75.00	75.00
Beef fat	15.00	15.00	15.00	15.00	15.00
Breadcrumb*	5.00	5.00	5.00	5.00	5.00
Water	2.00	2.00	2.00	2.00	2.00
Salt	1.50	1.50	1.50	1.50	1.50
Onion powder	1.00	1.00	1.00	1.00	1.00
Black pepper	0.25	0.25	0.25	0.25	0.25
Cumin	0.25	0.25	0.25	0.25	0.25

*Breadcrumb was substituted completely by gluten-free flours in the groups of Gf1, Gf2, Gf3 and Gf4. C: control sample including breadcrumbs, Gf1: gluten-free sample including buckwheat flour, Gf2: gluten-free sample including chickpea flour, Gf3: gluten-free sample including corn flour, Gf4: gluten-free sample including millet flour.

A total of 150 meatball samples were produced: ten meatballs for each treatment x five treatments (C, Gf1, Gf2, Gf3 and Gf4) x three independent replications. The samples were grilled for 15 minutes and turned over every 2.5 minutes to reach an internal temperature of 72°C. The temperature was measured with a thermometer (Digitale Bratengabel-TCM).

2.3. Proximate composition and pH measurement

Moisture (AOAC method 985.14), total protein (AOAC method 979.09), total fat (AOAC method 991.36), total ash (AOAC method 942.05) and pH (AOAC method 981.12) of the raw meatball samples were determined according to AOAC (2000).

2.4. Cooking yield

The cooking yield was calculated from the weight differences of the meatball samples before and after cooking. The cooked samples were cooled to room temperature for 30 minutes and then weighed (Murphy et al 1975). The cooking yield results were expressed as a percentage (%).

2.5. Reduction in diameter of meatball samples

The reduction in the diameter of the meatballs was determined by calculating the difference in the diameter of the samples before and after cooking (Yildiz Turp et al 2016). Measurements of the meatball samples were made with a digital micrometre (Mitutoyo, Japan). The reduction in diameter of meatballs was given as a percentage (%).

2.6. Determination of reduction in meatball volume

The reduction in volume of the meatballs was determined by calculating the difference in volume of the samples before and after cooking (Yildiz Turp et al 2016). The reduction in volume of the meatballs was expressed as a percentage (%).

2.7. Determination of antioxidant activity

The antioxidant activity of the cooked meatball samples was determined using DPPH (1,1-diphenyl-2-picrylhydrazyl) according to Brand-Williams et al (1995). The absorbance of the solutions was measured at 517 nm. DPPH antioxidant activity results were given as a percentage of free radical scavenging activity (%).

2.8. Colour measurements

The colour measurements of the raw and cooked meatball samples were made with a colourimeter (Konica, Minolta CR 400, Osaka, Japan). The *L** (lightness), *a** (redness) and *b** (yellowing) were determined according to Hunt et al (1991).

2.9. Texture profile analysis

Texture profile analysis (TPA) was carried out using the double compression method with a texture analyser (TA-HD Plus Texture Analyser, UK). A cylindrical plate with a diameter of 35 cm and a 50 kg load cell were used. The sample was compressed twice, with a delay of

0.1 s between the descents, a distance of 5 mm, a pre-test velocity of 1 mm/s, a test velocity of 5 mm/s, a post-test velocity of 5 mm/sec and a compression of 50%. The parameters of hardness, springiness, cohesiveness and chewiness were determined (Crehan et al 2000).

2.10. Sensory evaluation

A sensory panel consisting of 21 panellists conducted the sensory evaluations of the meatballs. Before the panel, the panellists were informed about the study. Samples were coded with three-digit numbers and randomly presented to the panellists. Along with the meatballs, the panellists were given water and bread. A 9-point hedonic scale was used for the sensory panel (9: very high acceptability value, 1: very low level of acceptability). The panellists were asked to rate the appearance, odour, flavour and texture of the meatball samples using the scale given to them.

2.11. Statistical analysis

This study was conducted in three independent replicates with double sampling and a completely randomised design was used. One-way analysis of variance (ANOVA) was performed for all analysis results using the Minitab version 16.0 programme. Tukey multiple comparison tests were performed to determine differences between means at a 5% significance level.

3. Results and Discussion

3.1. Proximate composition and pH

Proximate compositions and pH values of the raw meatball samples are shown in Table 2. As seen, the different flours did not affect the moisture content, total fat content, total ash content and pH values of the samples, while the total protein contents of the C and GF1 were lower than the other groups ($P < 0.05$). The highest total protein content was found in the meatball samples produced with chickpea flour. Buresova et al (2017) pointed out that chickpea flour had a higher protein content than buckwheat, corn and millet flour. Therefore, this is the reason for the higher protein content of the meatball samples containing chickpea flour. These results are consistent with those of Kurt and Kılıççeker (2012), who reported that different cereal and legume flours did not change the pH and moisture content of raw meat patties, and chickpea flour increased the protein content of the samples compared to wheat flour.

Table 2
Proximate compositions and pH values of raw meatball samples

Samples	Moisture (%)	Total protein (%)	Total fat (%)	Total ash (%)	pH
C	61.89 ± 0.08	17.15 ± 0.11^{b}	16.74 ± 0.30	2.29 ± 0.05	6.12 ± 0.01
Gf1	61.86 ± 0.18	17.11 ± 0.04^{b}	16.97 ± 0.30	2.23 ± 0.07	6.19 ± 0.01
Gf2	61.46 ± 0.28	17.93 ± 0.04^{a}	16.61 ± 0.38	2.35 ± 0.03	6.16 ± 0.03
Gf3	61.09 ± 0.09	17.58 ± 0.32^{ab}	16.98 ± 0.16	2.44 ± 0.04	6.12 ± 0.03
Gf4	61.55 ± 0.45	17.62 ± 0.26^{ab}	16.77 ± 0.12	2.19 ± 0.01	6.13 ± 0.04

Values with different lowercase superscript letters show significant differences ($P < 0.05$). C: control sample including breadcrumbs, Gf1: gluten-free sample including buckwheat flour, Gf2: gluten-free sample including chickpea flour, Gf3: gluten-free sample including corn flour, Gf4: gluten-free sample including millet flour.

3.2. Cooking characteristics

The cooking yield, reduction in diameter and in volume of the meatball samples are given in Table 3. Gluten-free flours increased the cooking yield of the meatball samples ($P < 0.05$). The lowest cooking yield was found in the control group ($P < 0.05$). The differences in the cooking yield results of samples with gluten-free flours were not significant ($P > 0.05$). It was reported that cooking characteristics of meat products were generally influenced by the ability to bind water and fat during cooking process (Salcedo-Sandoval et al 2014). The results of the current study indicate that the improvement in cooking yield by adding gluten-free starch-based flour to meatballs is mainly related to water retention. When the flour is heated, the starch gelatinises, and the flour fibres swell. The swollen starch and fibres can interact with the protein of the meatball matrix to prevent the migration of moisture from the product during cooking (Narayana et al 1982). Similarly, Makri and Douvi (2014) indicated that corn flour showed increased cooking yield in gilthead sea bream (*Sparus aurata*) patties. Alakali et al (2010) also stated that Bambara groundnut flour increased the cooking yield values of beef patties.

Table 3
Cooking characteristics of meatballs

Samples	Cooking yield (%)	Reduction in diameter (%)	Reduction in volume (%)
C	80.53 ± 0.44^{b}	16.73 ± 0.28^{a}	20.72 ± 3.99^{a}
Gf1	84.08 ± 0.61^{a}	14.50 ± 0.13^{b}	17.85 ± 4.62^{ab}
Gf2	84.66 ± 0.25^{a}	8.35 ± 0.98^{c}	12.63 ± 3.09^{ab}
Gf3	85.28 ± 1.29^{a}	9.29 ± 0.95^{c}	8.08 ± 0.58^{b}
Gf4	84.31 ± 0.41^{a}	13.37 ± 1.17^{b}	14.26 ± 5.70^{ab}

$^{a-c}$: Values with different lowercase superscript letters show significant differences ($P < 0.05$). C: control sample including breadcrumbs, Gf1: gluten-free sample including buckwheat flour, Gf2: gluten-free sample including chickpea flour, Gf3: gluten-free sample including corn flour, Gf4: gluten-free sample including millet flour

Gluten-free flours decreased the reduction in diameter of meatball samples compared to control group ($P < 0.05$). Chickpea flour (Gf2) and corn flour (Gf3) have been found to be the most effective flours for reducing the diameter of meatballs ($P < 0.05$). It was determined that corn flour was the most effective in volume reduction ($P < 0.05$). This effect of corn flour could be due to its starch, which plays an important role in improving reformed meat products, as well as its protein content and gelling properties (Berry 1997; Alakali et al 2010). Similarly, Kurt and Kılıççeker (2012) reported that corn flour decreased the diameter reduction values of beef patties.

3.3. Antioxidant activity

DPPH antioxidant activity results of the cooked meatball samples are shown in Figure 1. The highest antioxidant activity was determined in meatball samples with buckwheat flour (Gf1) ($P < 0.05$).

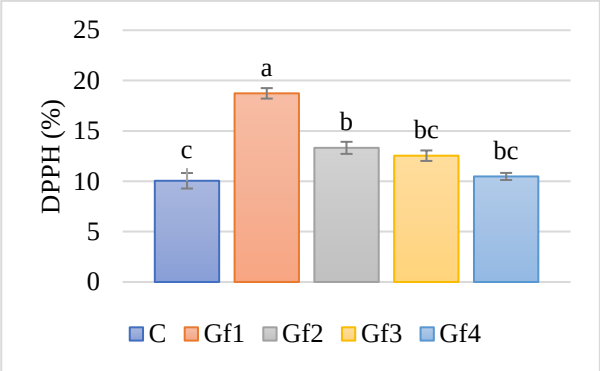


Figure 1
DPPH antioxidant activity of cooked meatball samples. Bar charts with different letters (a-c) indicate significant differences between the sample groups ($P < 0.05$). C: control sample including breadcrumbs, Gf1: gluten free sample including buckwheat flour, Gf2: gluten free sample including chickpea flour, Gf3: gluten free sample including corn flour, Gf4: gluten free sample including millet flour.

This group was followed by meatballs containing chickpea flour (Gf2) ($P < 0.05$). Control group meatballs had the lowest DPPH values. Similar results were obtained by Sedej et al (2010), who reported that buckwheat flour had higher polyphenols content and DPPH antioxidant activity than wheat flour. Beitane et al (2018) also pointed out that the content of phenols and antioxidant activity in buckwheat flour was higher than in wheat flour.

3.4. Colour properties

Colour is one of the most important quality parameters for meat products. The *L**, *a** and *b** values of the raw and cooked meatball samples can be seen in Table 4. Gluten-free flours did not change the colour parameters of either the raw or the cooked meatballs ($P > 0.05$). Similarly, Sanjeewa et al. (2010) found that the *L** and *a** values for the cooked bologna were not affected by the addition of chickpea flour. Also, Makri and Douvi (2014) reported that addition of 2.5% corn flour did not affect the colour properties of the sea bream (*Sparus aurata*) patties.

Table 4
Colour properties of raw and cooked meatball samples

Samples	Raw meatball samples			Cooked meatball samples		
	*L**	*a**	*b**	*L**	*a**	*b**
C	39.38 ± 1.13	12.78 ± 0.83	7.75 ± 0.38	34.40 ± 0.80	6.81 ± 0.31	5.78 ± 0.29
Gf1	44.09 ± 1.18	13.62 ± 0.80	9.65 ± 0.92	35.27 ± 0.65	7.78 ± 0.26	5.91 ± 0.26
Gf2	41.76 ± 1.43	13.42 ± 0.86	10.21 ± 0.88	35.62 ± 0.97	6.42 ± 0.47	6.28 ± 0.29
Gf3	39.22 ± 0.86	14.17 ± 1.17	9.71 ± 1.02	35.44 ± 1.34	7.70 ± 0.53	6.72 ± 0.30
Gf4	41.70 ± 1.64	12.19 ± 0.35	9.14 ± 1.05	34.34 ± 0.83	7.30 ± 0.22	5.77 ± 0.22

Values with different lowercase superscript letters show significant differences ($P < 0.05$). C: control sample including breadcrumbs, Gf1: gluten-free sample including buckwheat flour, Gf2: gluten-free sample including chickpea flour, Gf3: gluten-free sample including corn flour, Gf4: gluten-free sample including millet flour.

3.5. Textural properties

The values for hardness, springiness, cohesiveness and chewiness of the meatball samples are given in Table 5. The addition of gluten-free flours influenced all parameters of the texture analysis ($P < 0.05$). The lowest hardness and chewiness values were determined in the samples with breadcrumbs (C) and chickpea flour (Gf2), while corn, millet and buckwheat flour increased the hardness and chewiness values compared to the control ($P < 0.05$). In terms of springiness and cohesiveness, samples including gluten-free flours were simlar to control group ($P > 0.05$).

The change in textural properties due to the addition of gluten-free flour to meatballs is mainly related to water binding. In this study, improving the textural properties of meatballs with chickpea flour, which has the best cooking properties (Table 3), shows that the results are mutually supportive. Similar observations were reported for gilthead sea bream patties formulated with different concentrations of corn flour (Makri & Douvi 2014). Bahmanyar et al (2021) also reported that buckwheat flour increased the values of textural parameters in fried beef burgers compared to the control group.

Table 4
Textural characteristics of meatball samples

Samples	Hardness (N)	Springiness	Cohesiveness	Chewiness (N x mm)
C	168.30 ± 4.84^{c}	0.85 ± 0.01^{ab}	0.58 ± 0.02^{ab}	98.25 ± 3.57^{c}
Gf1	191.49 ± 4.99^{b}	0.86 ± 0.01^{a}	0.62 ± 0.01^{a}	118.47 ± 3.56^{b}
Gf2	163.15 ± 3.29^{c}	0.83 ± 0.00^{b}	0.54 ± 0.01^{b}	88.17 ± 2.33^{c}
Gf3	223.39 ± 7.36^{a}	0.87 ± 0.01^{a}	0.63 ± 0.01^{a}	140.22 ± 5.60^{a}
Gf4	207.70 ± 4.08^{ab}	0.86 ± 0.00^{a}	0.61 ± 0.01^{a}	127.54 ± 3.84^{ab}

Values with different lowercase superscript letters show significant differences ($P < 0.05$). C: control sample including breadcrumbs, Gf1: gluten-free sample including buckwheat flour, Gf2: gluten-free sample including chickpea flour, Gf3: gluten-free sample including corn flour, Gf4: gluten-free sample including millet flour.

3.6. Sensory scores

The sensory results of the cooked meatball samples are presented in Figure 2. The gluten-free flours had no effect on the appearance, odour and texture of the samples ($P > 0.05$), while the flavour scores of the meatballs were significantly different ($P < 0.05$). The samples including millet flour had the lowest flavour scores. The differences between the other groups were not significant for the flavour scores ($P > 0.05$). Although no difference was detected between the texture scores of the samples in the sensory panel, as seen in Table 5, the textural properties of the samples were different in the texture profile analysis. This inconsistency may be due to different temperatures of the test conditions. In the sensory panel, the samples were served at a temperature of about 40 °C, but texture profile analysis with the Texture Analyser was measured at room temperature (Bahmanyar et. al., 2021). Brasil et. al. (2015) reported that cooked kibbehs with millet flour did not differ in appearance, texture and flavour from the samples with wheat flour. Elhassan et al. (2019) indicated that sensory evaluation (appearance, taste, texture, juiciness and overall acceptability) significantly decreased by increasing the chickpea flour content in beef sausages. These different results between studies may be due to differences in treatments, levels of added flour, other additives, meat products and cooking process.

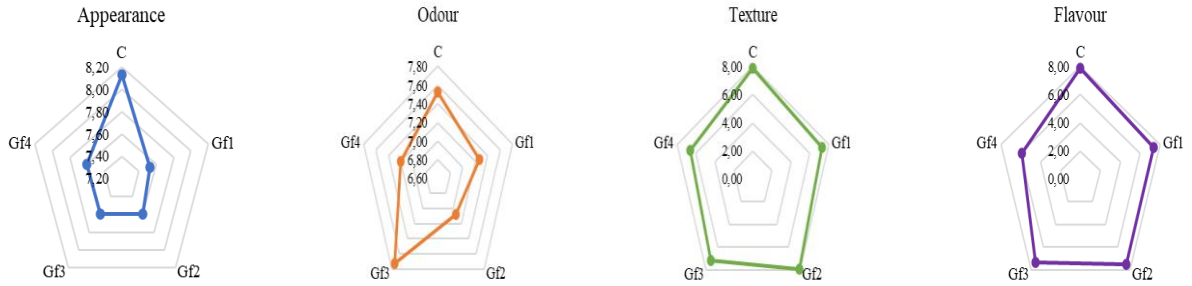


Figure 2
Spider web view of the sensory scores of meatball samples. (a): Appearance scores of the samples, (b): Odour scores of the samples, (c): Texture scores of the samples, (d): Flavour scores of the samples. C: control sample including breadcrumbs. Gf1: gluten free sample including buckwheat flour, Gf2: gluten free sample including chickpea flour, Gf3: gluten free sample including corn flour, Gf4: gluten free sample including millet flour.

4. Conclusion

The results of this study could be helpful in the production of gluten-free meatballs for celiac patients. The replacement of breadcrumb by buckwheat, chickpea, corn and millet flours in samples was found to be effective on characteristics of meatballs. The obtained results showed that the raw meatballs including chickpea flour had a higher protein content. Gluten-free flours increased the cooking yield of the samples and chickpea flour in particular improved the cooking properties of the meatballs. The cooked meatballs with buckwheat flour had the highest antioxidant activity. Gluten-free flours had no significant effect on the colour properties of raw and cooked meatballs. In terms of textural properties, the chickpea flour improved the texture of the meatball samples. Although millet flavour decreased the flavour score of the meatballs, the other gluten-free flours had no effect on the sensory properties of the samples compared to the control. In this respect, especially chickpea, corn and buckwheat flours could be used as substitutes for breadcrumbs in the meatball formulations.

5. Acknowledgements

This study received no external funding. The author would like to thank Rabia Duru, Rümeysa Durmuş and Şenay Can Bilgin for contributions to the analyses in the laboratory

6. References

Alakali JS, Irtwange SV, Mzer MT (2010). Quality evaluation of beef patties formulated with bambara groundnut (*Vigna subterranean* L.) seed flour. *Meat Science* 85(2): 215-223.

AOAC (2000). Official methods of analysis. In: AOAC. Gaithersburg, MD.

Bağdatlı A (2018). The influence of quinoa (*Chenopodium quinoa* willd.) flour on the physicochemical, textural and sensorial properties of beef meatball. *Italian Journal of Food Science* 30: 280-288.

Bahmanyar F, Hosseini SM, Mirmoghtadaie L, Shojaee-Aliabadi S (2021). Effects of replacing soy protein and bread crumb with quinoa and buckwheat flour in functional beef burger formulation. *Meat Science* 172: 108305.

Beitāne I, Krūmiņa-Zemture G, Krūma Z, Cinkmanis I (2018). Phenolics content in buckwheat flour. In Proceedings of the Latvian Academy of Sciences 72(2): 75-79 De Gruyter Poland.

Berry BW (1997). Sodium alginate plus modified tapioca starch improves properties of low-fat beef patties. *Journal of Food Science* 62(6): 1245-1249.

Brand-Williams W, Cuveier ME, Berset C (1995). Use of a free radical method to evaluate antioxidant activity. *LWT-Food Science & Technology* 28: 25-30.

Brasil TA, Capitani CD, Takeuchi KP, Ferreira TAPC (2015). Physical, chemical and sensory properties of gluten-free kibbeh formulated with millet flour (*Pennisetum glaucum* (L.) R. Br.). *Food Science and Technology* 35(2): 361-367.

Burešová I, Tokár M, Mareček J, Hřivna L, Faměra O, Šottníková V (2017). The comparison of the effect of added amaranth, buckwheat, chickpea, corn, millet and quinoa flour on rice dough rheological characteristics, textural and sensory quality of bread. *Journal of Cereal Science* 75: 158-164.

Crehan C, Hughes E, Troy D, Buckley D (2000). Effects of fat level and maltodextrin on the functional properties of frankfurters formulated with 5, 12 and 30% fat. *Meat Science* 55(4): 463-469.

Cui C, Basen T, Philipp AT, Yusin J, Krishnas-wamy G (2017). Celiac disease and nonceliac gluten sensitivity. *Annals of Allergy, Asthma and Immunology* 118(4): 389-393.

Devatkal SK, Kadam DM, Naik PK, Sahoo J (2011). Quality characteristics of gluten-free chicken nuggets extended with sorghum flour. *Journal of Food Quality* 34(2): 88-92.

Elhassan IH, Basheer EO, Ismaiel AE, Khalid AM, Alnor MA, Elrahman NAF, Babekir WS (2019). Quality characteristics of beef sausage incorporated with chickpea flour. *Journal of Academia and Industrial Research* 7(12): 169.

Gobbetti M, Pontonio E, Filannino P, Rizzello CG, De Angelis M, Di Cagno R (2018). How to improve the gluten-free diet: The state of the art from a food science perspective. *Food Research International* 110: 22-32.

Hunt M, Acton J, Benedict R, Calkins C, Cornforth D, Jeremiah L, Shivas S (1991). Guidelines for meat color evaluation. Paper presented at *the 44th Annual Reciprocal Meat Conference*.

Jackson V, Schilling MW, Coggins PC, Martin JM (2006). Utilization of rice starch in the formulation of low-fat, wheat-free chicken nuggets. *The Journal of Applied Poultry Research* 15(3): 417-424.

Kalinova J, Moudry J (2006). Content and quality of protein in proso millet (*Panicum miliaceum* L.) varieties. *Plant Foods for Human Nutrition* 61: 43-47.

Kartikawati M, Purnomo H (2019). Improving meatball quality using different varieties of rice bran as natural antioxidant. *Food Research* 3(1): 79-85.

Kerimoğlu BÖ, Serdaroğlu M (2019). Celiac disease and new attempts to develop gluten-free meat product formulations. *Food and Health* 5(4): 253-264.

Kurt Ş, Kilincceker O (2012). The effects of cereal and legume flours on the quality characteristics of beef patties. *Kafkas Üniversitesi Veteriner Fakültesi Dergisi* 18(5): 725-730.

Larrosa V, Lorenzo G, Zaritzky N, Califano A (2013). Optimization of rheological properties of gluten-free pasta dough using mixture design. *Journal of Cereal Science* 57: 520-526.

Makri M, Douvi X. (2014). Quality Evaluation of gilthead sea bream (*Sparus aurata*) patties formulated with corn flour. *British Journal of Applied Science & Technology* 4(19): 2684-2698.

Marco C, Rosell CM (2008). Breadmaking performance of protein enriched, gluten-free bread. *European Food Research and Technology* 227(4): 1205-1213.

Mastanjević K, Jukić M, Ugarčić Ž, Kovačević D, Kosović I, Koceva K, Daliborka KA, Keleković S (2014). Use of maize, soy and rice breadcrumbs in the formulation of the gluten free meatballs. Proceedings of the 7th International Congress Flour - Bread '13 [and] 9th Croatian Congress of Cereal Technologists, 218-223.

Narayana K, Narasinga Rao MS (1982). Functional properties of raw and heat processed winged bean (*Psophocarpus tetragonolobus*) flour. Journal of Food Science 47(5): 1534-1538.

Murphy EW, Criner PE, Grey BC (1975). Comparison of methods for calculating retentions of nutrients in cooked foods. *Journal of Agricultural and Food Chemistry* 23: 1153–1157.

Öztürk B, Serdaroğlu M, Karabıyıkoğlu M (2018). Quality attributes of chicken nuggets produced by using dif-ferent gluten-free ingredients. *International Poultry Science Congress WPSA Turkish Branch*, 09-12 May 2018, Cappadocia, Turkey, 192-196.

Park W, Kim J-H, Ju M-G, Yeon S-J, Hong G-E, Lee C-H (2016). Physicochemical and textural properties of pork patties as affected by buckwheat and fermented buckwheat. *Journal of Food Science and Technology* 53(1): 658–666.

Romero MC, Fogar RA, Rolhaiser F, Clavero VV, Romero AM, Judis MA (2018). Development of gluten-free fish (*Pseudoplatystoma corruscans*) patties by response surface methodology. *Journal of Food Science and Technology* 55(5): 1889-1902.

Saba N, Hashem M, Azad M, Hossain M, Khan M (2018). Effect of bottle gourd leaf (*Lagenaria siceraria*) extract on the quality of beef meatball: Bottle gourd leaf extract on meatball. *Bangladesh Journal of Animal Science* 47(2): 105–113.

Salcedo-Sandoval L, Cofrades S, Ruiz-Capillas C, Jiménez-Colmenero F (2014). Effect of cooking method on the fatty acid content of reduced-fat and PUFA-enriched pork patties formulated with a konjac-based oil bulking system. *Meat Science* 98: 795–803.

Sanjeewa WT, Wanasundara JP, Pietrasik Z, Shand PJ (2010). Characterization of chickpea (*Cicer arietinum* L.) flours and application in low-fat pork bologna as a model system. *Food Research International* 43(2): 617-626.

Sedej IJ, Sakač MB, Mišan AČ, Mandić AI (2010). Antioxidant activity of wheat and buckwheat flours. *Zbornik Matice srpske za prirodne nauke*, (118): 59-68.

Sofi S A, Singh J, Muzaffar K, Mir S A, Dar BN (2020). Effect of germination time on physico-chemical, functional, pasting, rheology and electrophoretic characteristics of chickpea flour. *Journal of Food Measurement and Characterization* 14(5): 2380-2392.

Yildiz Turp G, Icier F, Kor G (2016). Influence of infrared final cooking on color, texture and cooking characteristics of ohmically pre-cooked meatball. *Meat Science* 114: 46-53.

Selcuk Journal of Agriculture and Food Sciences SJAFS

http://sjafs.selcuk.edu.tr/sjafs/index

(2022) 36 (3), 405-409

e-ISSN: 2458-8377

DOI:10.15316/SJAFS.2022.053

Research Article

Relationships between Some Spectral Traits and Grain Yield in Bread Wheat under Rainfed Conditions

Seyfi TANER[1*], Şah İsmail CERİT[2], Enes YAKIŞIR [2], Musa TÜRKÖZ[2]

[1]Karamanoğlu Mehmetbey University, Vocational School of Technical, Karaman, Türkiye

[2] Bahri Dağdaş International Agricultural Research Institute, Konya, Türkiye

ARTICLE INFO

Article history:
Received date: 11.08.2022
Accepted date: 11.10.2022

Keywords:
Canopy Temperature,
Normalized Difference Vegetation Index,
Soil Plant Analysis Development,
Yield

ABSTRACT

Developing varieties adapted to dry conditions is one of the biggest targets for breeders. It is important to use inexpensive spectral sensing methods saving time in variety development. The aim of this study was to select bread wheat genotypes having high grain yield by using spectral sensing methods. Twenty-five bread wheat (*Triticum aestivum* L.) genotypes were evaluated under rainfed condition at three locations in Central Anatolia Region. The experiment was arranged in randomized complete block design with three replications. Grain yield (GY), Canopy Temperature (CT), Soil Plant Analysis Development (SPAD) and Normalized Difference Vegetation Index (NDVI) values were recorded. GY, CT, SPAD and NDVI were found to be statistically significant in terms of both genotype and environment. The relationship between grain yield and NDVI (R^2=0.321**) values was linear. The positive correlation of GY (0.5671**) and SPAD (0.1729*) with NDVI suggest that NDVI can be used as efficient and precise selection criteria for identifying high effiency wheat varieties under rainfed conditions.

1. Introduction

Wheat is one of the most cultivated crops in the world. Drought is one of the biggest factors limiting wheat yield. On the other hand, as in the province of Konya, the World's population is increasing and production areas are decreasing. For this reason, it is necessary to increase the yield per unit area in order to meet the increasing food demand. Therefore, breeders, physiologists and agronomists are trying to register variety and develop new agronomic techniques. For success of any breeding program, it is essential for the breeders to have a material with a wide array of diversity to be tested for desired characteristics in a variety of environments. The objectives including selection of suitable cross combinations, experimental design, location, nursery screening are essential elements for breeding programs. Existence of highly variable starting material and proper use of them are other key elements of breeding programs for positive achievement. Breeders want to select genotypes having desirable traits from early generation. Physiological characteristics of plants offer useful tools to breeders as indications of several desired characteristics. For this purpose, various indirect measurement methods such as Soil Plant Analysis Development (SPAD), Normalized Difference Vegetation Index (NDVI) and Canopy Temperature (CT), which can be applied easily, are used. Remote sensing technique has the advantage of being a cost-effective technique (Araus et al 2001).

SPAD meter which measures leaf chlorophyll content as indirect can be used in breeding programs (Giunta et al 2002; Shapiro et al 2013). Several studies suggest a positive correlation between SPAD value and chlorophyll content (Yadava 1986; Fischer 2001; Uddling et al 2007; Nemeskéri et al 2018).

NDVI is calculated by reflection of light in near infrared and red wavelengths. NDVI ranges from 0.1 to 1 and as it approaches to 1, it means the healthier plant (Usman et al 2013). It was reported that NDVI are used in many areas such as the indirect determination of plant biomass (Laidler et al 2008; Magney et al 2016), nitrogen level in the plant (Magney et al 2016), and leaf area index (Steltzer and Welker 2006; Fan et al 2009).

The relationship of leaf temperature with soil water content and transpiration is negatively linear (Pallas et al 1967). That is, as soil water content and transpiration

* Corresponding author email: *seyfitaner@yahoo.com*

increase, the CT decreases. Many researchers (Babar et al 2006; Mason and Singh 2014; Tahmasebi et al 2014; Thapa et al 2018; Sohail et al 2020) reported that CT can be used as a selection criterion.

The objective of the present study was to find out relationship between grain yield and some spectral traits in 25 bread wheat genotypes under rainfed conditions.

2. Materials and Methods

The experiment was carried out with 25 bread wheat (*Triticum aestivum* L.) variety, landraces and pure lines (Table 1) during 2009-2010 growing season at Center of Konya, İçeri Çumra and Gözlü locations of Central Anatolia under rainfed condition.

Table 1
Genotypes of the experiment

G.No	Genotypes
1	BAYRAKTAR 2000 (variety)
2	GEREK 79 (variety)
3	KARAHAN 99 (variety)
4	TOSUNBEY (variety)
5	BEZOSTAYA 1 (variety)
6	SYD/3/NAI60/HN//BUC/4/KEA/TOW/5/YAN7875.128 (breeding line)
7	ERAYBEY (variety)
8	AK 702/BDME 4 (breeding line)
9	EGL//BUC/PVN/3/KIRAÇ (breeding line)
10	ARG/R16//BEZ*2/3/AGRI/KSK/5/TRK13/6/494J6.1111/MNCH (breeding line)
11	IG42644/6/ZCL/3/PGFN//CNO67/SON64(ES86-8)/4/SERI/5/UA-2837/7/BJN 837/GRK//ES84.24 (breeding line)
12	LOV26//LFN/SDY(ES84-24)/3/SERI/4/SERI/5/SUL-TAN/6/TAST/SPRW//ZAR (breeding line)
13	90-1004a112/3/AU/CO652337//2*CA8055/6/PI/MZ//CNO67/3/LFN/4/ANT/5/AT-TILA (breeding line)
14	YEREL GENOTİP (landrace)
15	GV/4/D6301/NAİ//WRM/3/CNO*3/CHR/5/ BL2973/6/ LOVRİN6/SAMSUN (breeding line)
16	DAGDAS/KAROUS3//ALTAY (breeding line)
17	NAI60/3/1453/ODIN//CI1344/4/GRK/5/ATAY85/6/BAYRAKTAR (breeding line)
18	İKİZCE96/ EDECH28 (breeding line)
19	K340//FN/TH/3/NP"S"/4/093-44/4/ BAYRAKTAR (breeding line)
20	İKİZCE/TÜRKMEN (breeding line)
21	BEZOSTAYA-1/BAYRAKTAR (breeding line)
22	ABN/JUN//ATAY/3/ATAY (breeding line)
23	GUN/DRUM1//HKAYA (breeding line)
24	TAST/ANZA/3/F1.SU185/LTTO//BUG/BJY/4/KTK/5/SZN (breeding line)
25	TX69A330-1/3/NAD63/361-2-2//BEZ/4/KK/ITD//DAC/5/ID800994.W/VEE (breeding line)

The experiments were arranged in randomized complete block design with three replications. The plot dimensions are 1.2 by 5 m long (6 rows consist of 0.2 m space). Sowings were done at second week of October with 550 seeds m^{-2}. Harvesting was at second week of July. 70 kg N and 70 kg P ha^{-1} were applied. All of the phosphorus fertilizer was applied in DAP (Diammoniumphosphate) form with sowing. 27 kg ha^{-1} N (coming from DAP) was applied with sowing and 43 kg ha^{-1} N (ammonium nitrate) was applied in spring time.

SPAD was taken from flag leaf of main tiller by using SPAD-502 meter (Minolta Co.). Green-seeker (NTec Industries, Inc.) for NDVI and infrared thermometer (Testo 826-T2) for canopy temperature were used at Zadoks growth stage 50.

Data were analyzed with the "JMP" software. The significance of differences of means was checked by LSD test. A regression analysis was calculated using Microsoft Excel software. The regression graph between grain yield and NDVI was created using all locations and their replications.

3. Results and Discussion

Result of the analysis of variance show that there were significant differences ($p<0.01$) between all traits and locations. There also were significant differences among genotypes ($p<0.01$) with grain yield, canopy temperature and SPAD; between genotypes and NDVI values ($p<0.05$). Interaction between GY x L was significant $p<0.01$); between GY x SPAD & NDVI ($p<0.05$). There was no interaction between CT x L (Table 2).

Table 2
Analysis of variance for GY, CT, SPAD and NDVI

Source	Df	GY (kg ha^{-1})	CT (^oC)	SPAD value	NDVI value
Genotype (G)	24	**	**	**	*
Location (L)	2	**	**	**	**
G x L	48	**	NS	*	*
CV %	16	5	6	9	

NS, Non significant; P< 0.05 (* %5 significant level); P< 0.01(**%1 significant level)

Mean GY, CT, SPAD and NDVI values of the genotypes were given in Table 3; mean values for the same traits were given in Table 4.

Table 3
Mean values of traits measured at locations

Source	GY	CT	SPAD	NDVI
Center of Konya	4063 a	26.15 a	48.28 a	0.749 a
Gözlü	3656 b	17.35 c	44.13 b	0.697 a
İçeri Çumra	2759 c	23.03 b	47.36 a	0.608 b
LSD (%5)	320	1.83	1.42	0.05

3.1 Grain yield

Grain yield was elevated in terms of location and genotypes. While the highest grain yield (4063 kg ha^{-1}) was in Centre of Konya location, the least grain yield (2759 kg ha^{-1}) was İçeri Çumra location (Table 3). As for genotypes, while genotype 11 (2735 kg ha^{-1}) was the lowest, Karahan 99 variety (4352 kg ha^{-1}) was the highest (Table 4).

Table 4
Averages for GY, CT, SPAD and NDVI values

G.No	GY	CT	SPAD	NDVI
1	3258 e-j	23.25 ab	42.62 ıj	0.647 cd
2	3491 d-h	23.69 a	44.88 g-ı	0.667 b-d
3	4352 a	22.11 c-e	51.64 a	0.688 a-d
4	3567 c-h	23.61 a	45.40 f-h	0.661 cd
5	3677 b-e	21.86 de	49.93 a-c	0.727 ab
6	4043 a-c	22.72 a-d	49.56 a-c	0.744 a
7	4122 ab	22.05 c-e	50.20 ab	0.706 a-c
8	3593 b-g	21.80 de	45.74 e-g	0.692 a-d
9	3466 d-h	22.22 b-e	46.56 d-g	0.693 a-d
10	3657 b-f	22.00 c-e	45.72 e-g	0.675 b-d
11	2735 j	21.11 c-e	47.34 c-g	0.639 d
12	3869 a-d	21.52 e	42.95 h-j	0.661 cd
13	3062 h-j	21.36 e	45.84 e-g	0.689 a-d
14	2934 ıj	22.13 b-e	44.73 g-ı	0.670 b-d
15	3553 c-h	22.30 b-e	47.52 c-f	0.660 cd
16	3184 e-j	21.94 de	48.04 b-e	0.657 cd
17	3539 c-h	21.58 e	42.96 h-j	0.650 cd
18	3476 d-h	21.77 de	48.80 b-d	0.674 b-d
19	3137 f-j	21.97 c-e	41.91 j	0.748 a
20	3516 c-h	21.77 de	45.85 e-g	0.696 a-d
21	3856 a-d	21.91 de	44.87 g-ı	0.687 a-d
22	3086 g-j	22.16 b-e	49.28 a-c	0.678 b-d
23	3185 e-j	21.66 de	49.61 a-c	0.745 a
24	3673 b-e	23.08 a-c	47.57 b-f	0.672 b-d
25	3287 e-ı	21.83 de	45.16 f-ı	0.689 a-d
LSD(%5)	530.9	1.12	2.62	0.06

Grain yield is affected by genetic structure of genotype, soil characteristics, climate factors such as precipitation, temperature. Therefore, different results are obtained from different experiments. While Yağmur et al (2021) found the grain yield between 2395-3317 kg ha^{-1}, Aydoğan and Soylu (2017) obtained between 4474-7091 that they carried out their studies with different genotypes. Our results were similar to the results of Yağmur et al (2021). Keser et al (2017) reported that Karahan 99 had the highest grain yield with mean 3650 kg ha^{-1} obtained from 21 locations located in Central Anatolia. Similarly, Karahan 99 became prominent in our studies, too.

3.2. Canopy temperature

The highest canopy temperature was 26.15 ºC at centre of Konya location followed by İçeri Çumra (23.03 ºC) and Gözlü (17.35 ºC). In the present study, there were significant differences in terms of canopy temperature between locations (Table 3). It was determined that the canopy temperature was statistically significant according to the environment (Lopes et al 2012; Ohnishi et al 2021). While variety 2 (Gerek 79) gave the highest canopy temperature (23.69 ºC), genotype 11 had the lowest canopy temperature (21.11 ºC) (Table 4). As stated in the previous study (Sohail et al 2020), differences between genotypes may be resulted from genetic characteristic of genotypes. Although some researchers like Babar et al (2006), Mason and Singh (2014), Sohail et al (2020) stated that there was a significant relationship between CT and grain yield, the result of Araus et al (2001) was on the contrary. We also didn't determine any correlation between grain yield and CT, but canopy temperature was positively correlated with SPAD (0.4017**) (Table 5).

Table 5
Correlation matrix among grain yield, SPAD, NDVI and canopy temperature

	GY	SPAD	CT
NDVI	0.5671**	0.1729*	0.0478
CT	0.0164	0.4017**	-
SPAD	0.0966	-	-

3.3. Soil Plant Analysis Development

The highest SPAD values was obtained from centre of Konya followed by İçeri Çumra and Gözlü locations respectively with 48.28, 47.36 and 44.13 (Table 3). As reported by some researchers like Roy et al (2021), there were differences in SPAD values among genotypes. The highest SPAD (51.64) was recorded on Genotype 3 (Karahan 99) (Table 4). While positive relationship was detected between SPAD and grain yield in some studies (Spaner et al 2005; Yıldırım et al 2013; Islam et al 2014; Faisal and Al-Tahir 2014; Fotovat et al 2007; Ramya et al 2016), we didn't any relationship between them (Table 5).

3.4. Normalized Difference Vegetation Index

NDVI measured among locations were ranged between 0.608 (İçeri Çumra) and 0.749 (Konya) (Table 3). As for genotypes, it ranged between 0.639 (genotype 11) and 0.748 (genotype 19). Genotype 3 (Karahan 99), genotype 6, genotype 7 (Eraybey) and genotype 21 having high NDVI had high grain yield at the same time (Table 4). The NDVI were positively correlated with SPAD (0.1729*) and grain yield (0.5671**) (Table 5). These results show that NDVI can use to estimate the grain yield. Similar results were reported by Roy et al (2021), Marti et al (2007), Sultana et al (2014), Morgounov et al (2014), Mekliche et al (2015).

By considering the positive correlation between grain yield and NDVI, regression analysis was carried out (Fig. 1). This regression was positively linear (r^2=0.321**) formulated Y=5229X-87.45. Since the NDVI is an indirect measurement of biomass, this positive regression can be associated with plant density per unit area which is one of the yield components. Mekliche et al (2015) suggested that the grain yield could be predicted using a single regression with NDVI. In addition to grain yield, Hazratkulova et al (2012) also stated that NDVI can be used for identifying physiological superior.

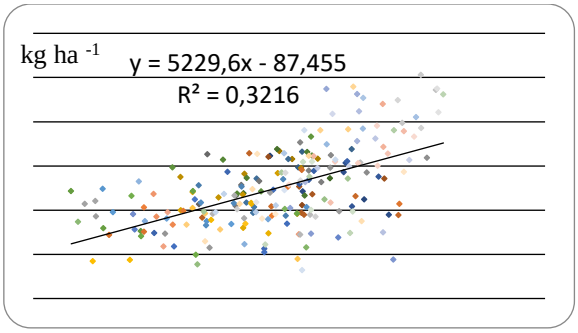


Figure 1
The regression between grain yield (kg ha^{-1}) and NDVI value

4. Conclusion

As a result, the significant and positive relationship between grain yield and NDVI show that NDVI can use to estimate the grain yield. This method is inexpensive, easy and useful. Many genotypes obtained from breeding program can be screened speedily in short time by using NDVI. NDVI also provides a useful tool to the breeders with utilization of physiological indirect selection criteria in drought conditions.

5. Acknowledgements

This research was funded by General Directorate of Agricultural Research and Policies (TAGEM) of the Ministry of Agriculture and Forestry of Türkiye; we are grateful for the financial support of TAGEM.

6. References

Araus JL, Casadesus J, Bort J (2001). Recent Tools for the Screening of Physiological trails Determining Yield. Application of Physiology in Wheat Breeding, Chapter 5: 59-77.

Aydoğan S, Soylu S (2017). Determination of Yield, Yield Components and Some Quality Properties of

Bread Wheat Varieties. Journal of Field Crops Central Research Institute, 26 (1):24-30. (In Turkish)

Babar MA, Reynolds MP, van Ginkel M, Klatt AR, Raun WR, Stone ML (2006). Spectral Reflectance to Estimate Genetic Variation for In-Season Biomass, Leaf Chlorophyll, and Canopy Temperature in Wheat. Crop Scicience, 46 (3): 1046–1057. Doi: 10.2135/cropsci 2005. 0211.

Faisal M, Al-Tahir M (2014). Flag leaf characteristics and relationship with grain yield and grain protein percentage for three cereals. Journal of Medicinal Plants Studies, 2 (5): 01-07.

Fan L, Gao Y, Brück H, Bernhofer CH (2009). Investigating the relationship between NDVI and LAI in semiarid grassland in Inner Mongolia using in-situ measurements. Theoretical and Applied Climatology, 95: 151–156 DOI 10.1007/s00704-007-0369-2.

Fischer RA (2001). Selection Traits for Improving Yield Potantial. Application of Physiology in Wheat Breeding, Chapter-13, p.148-159.

Fotovat R, Valizadeh M, Toorchi M (2007). Association between water-use efficiency components and total chlorophyll content (SPAD) in wheat (*Triticum aestivum* L.) under well-watered and drought stress conditions. Journal of Food, Agriculture & Environment Vol.5 (3&4): 225-227.

Giunta F, Motzo R, Deidda M (2002). SPAD readings and associated leaf traits in durum wheat, barley and triticale cultivars. Euphytica, 125: 197–205.

Hazratkulova S, Sharma RC, Alikulov S, Islomov S, Yuldashev T, Ziyaev Z, Khalikulov Z, Ziyadullaev Z, Turok J (2012). Analysis of genotypic variation for normalized difference vegetation index and its relationship with grain yield in winter wheat under terminal heat stress. Plant Breeding, 131: 716-72.

Islam MR, Haque KMS, Akter N, Karim MA (2014). Leaf chlorophyll dynamics in wheat based on SPAD meter reading its relationship with grain yield. Scientia Agriculture, 8 (1): 13-18.

Keser M, Gummadov N, Akin B, Belen B, Mert Z, Taner S, Topal A, Yazar S, Morgounov A, Sharma RC, Ozdemir F (2017). Genetic gains in wheat in Turkey: Winter heat for dry land conditions. The Crop Journal, 5 (6): 533-540.

Laidler GJ, Treitz PM, Atkinson DM (2008). Remote Sensing of Arctic Vegetation: Relations between the NDVI, Spatial Resolution and Vegetation Cover on Boothia Peninsula, Nunavut. ARCTIC Vol. 61, No. 1, P. 1–13.

Lopes MS, Reynolds MP, Manes Y, Singh RP, Crossa J, Braun HJ (2012). Genetic Yield Gains and Changes in Associated Traits of CIMMYT Spring Bread Wheat in a "Historic" Set Representing 30 Years of Breeding. Crop Science, 52:1123-1131. Doi: 10.2135/cropsci2011.09.0467.

Magney TS, Eitel JUH, Huggins DR, Vierling LA (2016). Proximal NDVI derived phenology improves in-season predictions of wheat quantity and quality. Agricultural and Forest Meteorology 217 (2016) 46–6. http://dx.doi.org/10.1016/j.agrform

Marti J, Bort J, Slafer GA, Araus JL (2007). Can wheat yield be assessed by early measurements of Normalized Difference Vegetation Index? Annals of Applied Biology, 150 (2): 253-257.

Mason RE, Singh RP (2014). Considerations When Deploying Canopy Temperature to Select High Yielding Wheat Breeding Lines under Drought and Heat Stress. Agronomy, 4 (2): 191-201. Doi: 10.3390/agronomy 4020191.

Morgounov A, Gummadov N, Belen S, Kaya Y, Keser M, Mursalova J (2014). Association of digital photo parameters and NDVI with winter wheat grain yield in variable environments. Turkish Journal of Agriculture and Forestry, 38: 624-632.

Mekliche A, Hanifi-Mekliche L, Aidaoui A, Gate P, Bouthier A, Monneveux P (2015). Grain yield and its components study and their association with normalized difference vegetation index (NDVI) under terminal water deficit and well-irrigated conditions in wheat (*Ttiticum durum* Desf. and *Triticum aestivum* L.). African Journal of Biotecnology, 14 (26): 2142-2148.

Nemeskéri E, Molnár K, Helyes L (2018). Relationships of spectral traits with yield and nutritional quality of snap beans (*Phaseolus vulgaris* L.) in dry seasons. Archives of Agronomy and Soil Science, 64 (9): 1222–1239. Doi.org/10.1080/03650340.2017.1420903.

Ohnishi S, Kasuya M, Sonoda T, Jinno H (2021). Normalization method for canopy temperature as an indirect indicator of yield potential in wheat breeding programs. Breeding Science 71: 520–527. doi: 10.1270/jsbbs.21011.

Pallas JE, Michel BE, Harris DG (1967). Photosynthesis, Transpiration, Leaf Temperature, and Stomatal Activity of Cotton Plants under Varying Water Potentials. Plant Physiol, 42, 76-88.

Ramya P, Singh GP, Jain N, Singh PK, Pandey MK, Sharma K, Kumar A, Harikrishna, Prabhu KV (2016). Effect of Recurrent Selection on Drought Tolerance and Related Morpho-Physiological Traits in Bread Wheat. PLOS ONE, 11(6): e0156869. doi:10.1371/journal.pone.0156869.

Roy C, Chattopadhyay T, Ranjan RD, Hasan WUI, Kumar A, De N (2021). Association of leaf chlorophyll content with the stay-green trait and grain yield in wheat grown under heat stress conditions. Czech J. Genet. Plant Breed., 57: 140−148. https://doi.org/10.17221/45/2021-CJGPB.

Sohail M, Hussain I, Qamar M, Tanveer SK, Abbas SH, Ali Z, Imtiaz M (2020). Evaluation of Spring Wheat Genotypes for Climatic Adaptability using Canopy

Temperature as Physiological Indicator. Pakistan Journal of Agricultural Research, Volume 33, Issue 1,Pages 89-96. DOI: http://dx.doi.org/10.17582/journal.pjar/2020/33.1.89.96.

Shapiro CA, Francis DD, Ferguson RB, Hergert GW, Shaver TM, Wortman CS (2013). Using a Chlorophll Meter to Improve N Management, Nebguide, GI632. University of Nebraska-Lincoin Extension, Institute of Agriculture and Natural Resources.

Spaner D, Todd AG, Navabi A, McKenzie DB, Goonewarddene LA (2005). Can Leaf Chlorophyll Measures at Different Growth Stage be used as an Indicator of Winter Wheat and Spring Barley Nitrogen Requirements in East Canada. J. Agronomy &Crop Science, vol. 191: 393-399.

Steltzer H, Welker JM (2006). Modeling The Effect of Photosynthetic Vegetation Properties on The NDVI–LAI Relationship. Ecology, 87(11), 2006, pp. 2765–2772.

Sultana SR, Ali A, Ahmad A, Mubeen M, Zia-Ul-Haq M, Ahmad S, Ercisli S, Jaafar HZE (2014). Normalized Difference Vegetation Index as a Tool for Wheat Yield Estimation: A Case Study from Faisalabad, Pakistan. Scientific World Journal, volume 2014: 1-8.

Tahmasebi S, Heidari B, Pakniyat H, Kamali MR (2014). Independent and combined effects of heat and drought stress in the Seri M82 3 Babax bread wheat population. Plant Breeding, 133, 702–711: doi:10.1111/pbr.12214.

Thapa S, Jessup KE, Pradhan GT, Rudd JC, Liu S, Mahan JR, Devkota RN, Baker JA, Xue Q (2018). Canopy temperature depression at grain filling correlates to winter wheat yield in the U.S. Southern High Plains. Field Crops Research 217; 11–19.

Uddling J, Gelang-Alfredsson J, Piikki K, Pleije H (2007). Evaluating the relationship between leaf chlorophyll concentration and SPAD-502 chlorophyll meter readings. Photosynthesis Ressearch, 91: 37–46. DOI 10.1007/s11120-006-9077-5.

Usman U, Yelwa SA, Gulumbe SU, Danbaba A (2013). Modelling Relationship between NDVI and Climatic Variables Using Geographically Weighted Regression. Journal of Mathematical Sciences and Applications, 2013, Vol. 1, No. 2, 24-28. DOI:10.12691/jmsa-1-2-2.

Yadava UL (1986). A rapid and nondestructive method to determine chlorophyll in intact leaves. Horticultural Science, 21: 1449–1450.

Yağmur M, Sözen Ö, Pala D (2021). The Effects of Different Seeding Rates on Grain Yield and Some Grain Yield Components in Two Bread Wheat (*Triticum aestivum* L.) Varieties under Semi-Arid Climate Conditions. Manas Journal of Agriculture Veterinary and Life Sciences. Volume 11 (Issue 1), Pages 10-20. (In Turkish)

Yıldırım M, Koç M, Akıncı C, Barutçular C (2013). Variations in morphological and physiological traits of bread wheat diallel crosses under timely and late sowing conditions. Field Crops Research, 140: 9-17.

Selcuk Journal of Agriculture and Food Sciences SJAFS

http://sjafs.selcuk.edu.tr/sjafs/index

(2022) 36 (3), 410-413

e-ISSN: 2458-8377

DOI:10.15316/SJAFS.2022.054

Research Article

The Incidence of Barley (*Hordeum vulgare* L.) Leaf Diseases in Central Anatolia Region of Turkey in 2020

Emine Burcu TURGAY*, Sibel BÜLBÜL, Merve Nur ERTAŞ ÖZ, Öztekin URLA

Republic Of Türkiye Ministry Of Agriculture And Forestry, Field Crops Central Research Institute, Ankara, Türkiye

ARTICLE INFO

Article history:
Received date: 10.02.2022
Accepted date: 12.10.2022

Keywords:
Barley
Barley leaf diseases
Central Anatolia
Survey
Region

ABSTRACT

Barley (*Hordeum vulgare* L.) is one of the most important cereal crops that their grains are used in the malt and forage industry. Yield and quality losses occur in barley due to abiotic and biotic stress factors. Barley leaf diseases caused by fungal pathogens are common diseases leading to significant yield losses. Within the framework of environmentally-friendly integrated crop management, disease-resistant varieties should be developed together with a breeding program for sustainable agriculture. In disease resistance studies, the first and the most important step is to monitor the situation of the disease agent(s) in a region or country. For this purpose; In 2019-2020, barley leaf diseases survey in 54 barley fields was done in 5 provinces of Central Anatolia (Ankara, Eskişehir, Kırıkkale, Kırşehir, Yozgat). As a result of the surveys, the most common disease was determined as barley stripe disease (*Drechslera graminea*). Barley net spot disease (*D. teres* f. *maculata*, *D. teres* f. *teres*), Barley leaf spot (*Rhynchosporium commune*) and Barley powdery mildew (*E. graminis* f. sp. *hordei*) diseases followed in this order.

1. Introduction

Barley is an important cereal group, which is among the most planted plants in the world and in Turkey (Kün 1996, Geçit et al., 2009). It is used in animal feed and in fields such as malting production, and it is also used in small amounts of human food (Geçit 2016). For 2020, barley cultivation areas in our country are 30971625 da and production is 8300000 tons (TUIK, 2022). It is predicted that barley cultivation was first practiced in the fertile crescent region in our country (Geçit et al. 2009, Harlan 1979) and there are various types of barley. There are important biotic and abiotic factors affecting production in barley grown areas. Fungal factors, especially among biotic factors, can cause serious yield and quality losses. The main barley fungal diseases in Central Anatolia are barley scald (*Rhynchosporium commune*), stripe disease (*Drechslera graminea*) and barley smut diseases (*Ustilago nuda* fs.p. *hordei*, *Ustilago hordei*, *Ustilago nigra*). In our country, among these diseases, smut causes yield loss of 3-5% in barley, while it causes yield losses of 12% and 20% in striped leaf spot and leaf spot (Aktaş 2001).

The occurrence and severity of many diseases seen in barley cultivation areas in Central Anatolia have been reported. In the study carried out in the Bala district of Ankara in 2018, a total of 50 barley fields were examined and leaf spot (*Rhynchosporium commune*), striped leaf spot (*Drechslera graminea*), two forms of net blotch (*Drechslera teres* f. *maculate* and *Drechslera teres* f. *teres*) and barley powdery mildew (*Erysiphe graminis* f. sp. *hordei*) was detected (Ertürk et al., 2018). In 2016, 17 different lands were examined in the Çubuk district of Ankara, and leaf spot, striped leaf spot, two forms of net blotch, powdery mildew, and also barley rust (*Puccinia hordei*) were reported (İlgen et al., 2017).

The first issue to be considered in disease resistant breeding studies is that the status of diseases in the region or country should be well determined. In this study, with the aim of contributing to future barley breeding programs, barley cultivation areas in the Central Anatolia Region were examined and the status of barley diseases was reported.

2. Materials and Methods

Survey of barley disease was conducted between April and May 2020 in cropping season in five important barley-producing provinces (Ankara, Eskişehir, Kırkkale, Kırşehir and Yozgat) and surrounding areas of the Central Anatolia Region of Turkey.

A total of 54 barley (*Hordeum vulgare*) fields were assessed in the Central Anatolia Region of Turkey.

The systematic sampling method was used in the survey program. The surveys were conducted following the main

*Corresponding author email: *cercospora79@gmail.com*

roads and accessible routes in each survey district, and examinations were done in every 1-20 kilometers stops. Each field was surveyed following a W pattern in the field and at least 100 plants were examined (Aktaş 2001). Samples were taken at every 1-20 kilometers. At least 100 barley plants were inspected in each field, and percentages of diseases were calculated. For determination of mean disease prevalence, both diseased fields and non-diseased fields were considered.

The determinations of diseases severity were assessed using a 1-9 scale developed by Saari and Prescott (Saari and Prescott, 1975)The following format is used for scoring severity: 10% coverage = 1, 20% coverage = 2, 30% coverage = 3, 40% coverage = 4, 50% coverage = 5, 60% coverage = 6, 70% coverage = 7, 80% coverage = 8, 90% coverage 9.

3. Results and Discussion

Fifty-four barley fields were examined as a result of the surveys conducted in Ankara, Eskişehir, Kırıkkale, Kırşehir and Yozgat. In these fields, stripe disease of barley (*Drechslera graminea*), barley scald disease (*Rhynchosporium commune*), two forms of net blotch disease, spot form net blotch (*Drechslera teres* f. *maculata*) and net form net blotch (*Drechslera teres* f. *teres*), and barley mildew (*Blumeria graminis* f. sp. *hordei*) were determined.

Table 1
Barley (*Hordeum vulgare* L.) leaf diseases observed in Ankara, Eskişehir, Kırıkkale, Kırşehir, Yozgat province of Central Anatolia Regions of Turkey

No	Province	District	Diseases situation	*Rc* Inc.(%)	*Rc*Sev.	Bgh Inc (%).	BghSev.	Dtm Inc. (%)	DtmSev.	Dtt Inc.(%)	DttSev.	Dg Inc.(%)
1	Ankara	Bala	present	15	3							
2	Ankara	Bala	present	7	3							
3	Ankara	Bala	present									15
4	Ankara	Bala	present							8	3	
5	Ankara	Bala	present									5
6	Ankara	Haymana	present									20
7	Ankara	Haymana	present									35
8	Ankara	Haymana	present									30
9	Ankara	Haymana	present									20
10	Ankara	Yenimahalle	present	5	3					2	3	10
11	Ankara	Gölbaşı/ikizce	present	30	7							10
12	Ankara	Elmadağ	present									5
13	Ankara	Elmadağ	present	5	3							
14	Ankara	Polatlı	present									5
15	Ankara	Polatlı	present	4	3							10
16	Ankara	Polatlı	present									5
17	Eskişehir	Mihalıççık	present	10	3	5	3					
18	Eskişehir	Mihalıççık	present	40	5	10	3			3	3	
19	Eskişehir	Mihalıççık	present					3	3	5	3	
20	Eskişehir	Mihalıççık	present	15	3							
21	Eskişehir	Mihalıççık	present							8	3	
22	Eskişehir	Yunusemre	present									15
23	Eskişehir	Yunusemre	present							5	3	
24	Eskişehir	Yunusemre	present							7	3	10
25	Eskişehir	Beylikova	present									5
26	Eskişehir	Beylikova	absent									
27	Eskişehir	Beylikova	present					10	5			8
28	Eskişehir	Beylikova	present									16
29	Eskişehir	Beylikova	present	13	5							20
30	Eskişehir	Çifteler	present									9
31	Eskişehir	Çifteler	present	2	3							10
32	Eskişehir	Doğanlar	present					3	3			17
33	Eskişehir	Mahmudiye	present									9
34	Eskişehir	Mahmudiye	present									4
35	Eskişehir	Sivrihisar	present					5	3			
36	Eskişehir	Sivrihisar	present					8	3			
37	Eskişehir	Sivrihisar	present									5
38	Eskişehir	Sivrihisar	present									6
39	Eskişehir	Sivrihisar	absent									
40	Kırıkkale	Kesikköprü	present	10	5					7	5	
41	Kırıkkale	Kesikköprü	present	10	5					6	5	
42	Kırıkkale	Kesikköprü	present									15
43	Kırıkkale	Kesikköprü	present	5	3							
44	Kırıkkale	Çelebi	present	8	3							4
45	Kırkkale	Çelebi	present									5
46	Kırkkale	Merkez	present			6	3					
47	Kırkkale	Merkez	present	10	5					10	3	
48	Kırşehir	Kaman	present									10
49	Kırşehir	Kaman	present	55	7							13
50	Kırşehir	Kaman	present									8
51	Yozgat	Yerköy	present							6	3	
52	Yozgat	Yerköy	present									10
53	Yozgat	Sekili	present									20
54	Yozgat	Sekili	present									15

(R. comm: *Rhynchosporium commune*, tm*: Drechslera teres* f. *maculata*, Dtt: *Drechslera teres* f. *teres*, Bgh: *Blumeria graminis* f. sp. *hordei*, *Drechslera graminea* : Dg, Inc.: Incidence, Sev.: Severity)

For Ankara, barley leaf diseases survey was carried out in 5 barley fields in Bala, 4 fields in Haymana, one field in Yenimahalle and Gölbaşı, 2 fields in Elmadağ and 3 fields in Polatlı. In total, 16 barley fields were examined. Stripe disease (*Drechslera graminea*) was detected in 12 fields of those. Barley scald disease (*Rhynchosporium commune*)

was detected in 6 fields out of 16 barley fields and the net form net blotch (*Drechslera teres* f. *teres*) in 2 barley fields (Table-1). In the examined areas, the rate of disease prevalence of stripe disease was detected as 5-35% (Tablo-1). On the other hand, the incidence for barley leaf spot disease in the examined areas was found between 4% and 30% and the severity of the disease was detected as 3 to 7 values. The area with the highest incidence of the disease was the 11th field of Gölbaşı, with a rate of 30% and severity of 7. Additionally, the percentage of net form of net blotch (*Drechslera teres* f. *teres*) was 8% in the 4th field of Bala. The incidence of the same disease in the 10th field of Yenimahalle was reported as 2% with the 3 out 9 severity.

For Eskisehir, 23 barley fields in total were surveyed, which 5 of them was in Mihalıççık, 3 fields in Yunusemre, 5 fields in Beylikova, 2 fields in Çifteler, one field in Doğanlar, 2 fields in Mahmudiye, and 5 fields in Sivrihisar. Stripe leaf disease (*Drechslera graminea*) was found in 13 of these fields whereas barley scald (*Rhynchosporium commune*) was recorded in 5 fields. As well, net form net blotch of barley (*Drechslera teres* f. *teres*), spot form net blotch (*Drechslera teres* f. *maculata*) and barley powdery mildew (*Blumeria graminis* f. sp. *hordei*) diseases was found in 5, 5 and 2 fields, respectively (Table-1). The incidence for stripe leaf disease in the examined areas was found between 4% and 20%. The highest rate of disease was reported in Beylikova, especially in the 29th field with 20%. The prevalence of barley scald was between 2-40% and the disease severity was between 3-5. Barley scald was observed in Mihalıççık in the 18th field with the maximum incidence of 40% and the severity was 5. Barley powdery mildew (*Blumeria graminis* f. sp. *hordei*) was observed with the rate of 5% and 10% in the 17th and 18th fields of Mihalıççık, respectively. Both fields had 3 out 9 in terms of severity. The net form net blotch disease (*Drechslera teres* f. *teres*) was detected in 3-8% prevalence with 3 disease severity. The incidence of the spot form net blotch disease (*Drechslera teres* f. *maculata*) was 3-10% and the severity of the disease was 3-5. The area where the spot form net blotch (*Drechslera teres* f. *maculata*) disease is the most common was determined as the 27th of Beylikova with 10% disease incidence and 5 disease severity.

For Kırıkkale, surveys were conducted in 8 barley fields where 4 of those were in Kesikköprü, 2 of those were in Çelebi and 2 of them were in the centre. Stripe leaf (*Drechslera graminea*) was detected in 3 areas, barley scald (*Rhyncosporium commune*) was found in 5 areas and net form net blotch disease (*Drechslera teres* f. *teres*) was seen 3 areas. Finally, barley powdery mildew (*Blumeria graminis* sp. *hordei*)was detected (Table 1). The incidence of stripe leaf disease was found between 4-15% in the areas examined. The incidence for barley scald was 5-10% with between 3 and 5 severity. The barley scald was seen the most the 40th and 41st region of Kesikköprü. The percentage of net form net blotch was stated as 6-10% and 3-5 severity. Finally, the incidence rate of mildew is determined as 6% with 3 severity in the 46th area in the centre of Kırıkkale.

In Kaman, the district of Kırşehir, barley stripe disease (Drechslera graminea) was founded in the 48th, 49th and 50th regions considering 3 barley survey. The incidences for this are 10%, 13% and 8 %, accordingly. Barley scald, with 55% incidence and 7 severity, was recorded in the 49th region of Kaman.

In Yozgat, 4 fields, two from Yerköy district and two from Sekili district, was examined and 3 of these fields was infected with stripe disease (*Drechslera graminea*). Net form net blotch (*Drechslera teres* f. *teres*) were found one of these areas. Barley stripe disease was observed in the 52nd region of Yerköy, the 53rd and 54th regions of Sekili with the rate of 10%, 20% and 15%, respectively. In the 51st area of Yerköy, net blotch disease, with the incidence of 6% and 3 out of 9 severity, was founded.

In the examination of 5 province in Central Anatolia region with 54 barley fields, 34 field of those was founded with barley stripe disease whereas 15 of those was recorded with the infection of barley scald. As well as, net blotch disease was determined in 16 fields, where 11 of them infected with net form net blotch and 5 of them was reported with spot form net blotch. The most prevalent disease was defined as barley stripe, following by net blotch and barley scald diseases, respectively.

This study shows the existence of barley leaf diseases in 5 provinces located in Central Anatolia Region (Ankara, Eskişehir, Kırşehir, Kırıkkale and Yozgat) with 54 barley (*Hordeum vulgare* L.) field. Stripe leaf disease was founded as the most widespread disease, and net blotch disease and barley scald was detected, respectively. Besides powdery mildew is the least common disease among the others.

These diseases are common diseases in our country and have been reported by other researchers from different regions of Turkey (Akan, 2006; Aktaş, 1987, 1997; Çelik ve Karakaya, 2015; Ertürk ve ark., 2018; İlgen ve ark., 2017; Karakaya ve ark., 2014, 2016; Mamluk ve ark., 1997; Özdemir ve ark., 2017; Yıldırım ve ark., 1999). Çelik and Karakaya (2015) in Eskişehir province Barley leaf in their survey in 2012 net spot (*Drechslera teres*),

Stripe diseases (*D. graminea*), scald disease (*Rhynchosporium secalis*), and powdery mildew (*Erysiphe graminis* f. sp. *hordei*) were determined. Of these diseases, net blotch and scald disease were found to be the most common diseases. Barley growing areas of Elazığ province of Turkey were surveyed during the period of May and June of 2018 and barley leaf diseases were determined. Spot form of net blotch and net form of net blotch were the most commonly encountered diseases (Saraç et.al., 2019) As a result, in this study, the same barley diseases (net blotch disease and barley scald) were found as the most widespread disease in the survey area.

Though there are chemical control methods for the detected diseases (seehttps://bku.tarimorman.gov.tr/), disease-resistant varieties must be developed and using the varieties need to be promoted since this is the most efficient and environmentally friendly approach. Using disease-free and certificated seeds, crop rotation, and cultural practices are the most effective methods in the control of these diseases. Furthermore, disease-resistant cultivars must be used for stripe disease (Ulus ve Karakaya 2007, Bayraktar ve Akan 2012, Çelik ve ark. 2016), net blotch, and barley scald (Mert and Karakaya 2004, Karakaya and Akyol 2006, Düşünceli et al. 2008, Taşkoparan and Karakaya 2009, Usta et al. 2014, Azamparsa et al. 2015, Yazıcı et al. 2015).

4. Acknowledgements

This study was suported by General Directorate of Agricultural Research and Policies (National Cereal Disease Research Project, TAGEM/TBAD/14/A12/P01/002)

5. References

Akan K, Çetin L, Albostan S, Düşünceli F, Mert Z (2006). İç Anadolu'da görülen önemli tahıl ve nohut hastalıkları. Tarla Bitkileri Merkez Araştırma Enstitüsü Dergisi 15 (1-2): 29-48

Aktaş H (1987). Untersuchungen Uber Die Physiologische Variationen von Drechslera teres (Sacc.) Shoemaker on der Mittelaratolien angebauten Gersten Und die Feststellung der Reaktionen der Gerstensorten gegen diesen Erreger. Journal of Turkish Phytopathology 16: 53-65.

Aktaş H (1997). Untersuchungen über die Netzfleckenkrankheiten (Drechslera teres Shoem. f. sp. teres Smedeg. D. teres Shoem. f. sp. maculata Smedeg.) an Gerste. Journal of Turkish Phytopathology 26 (1): 17-22

Aktaş H (2001). Önemli Hububat Hastalıkları ve Survey Yöntemleri Kitapçığı Tarım Ve Köyişleri Bakanlığı Tarımsal Araştırmalar Genel Müdürlüğü Bitki Sağlığı Araştırmaları Daire Başkanlığı, 80 sayfa, Ankara

Azamparsa MR, Mert Z, Karakaya A, Sayim İ, Ergün N, Aydoğan S (2015). Determination of the seedling reactions of some barley cultivars and advanced barley lines to Rhynchosporium commune. Bitki Koruma Bülteni 55(3): 247-252.

Balcı S, Karakaya A, Çelik Oğuz A, Ergün N, Sayım İ, Aydoğan S (2018). Bazı kavuzsuz arpa çeşit ve hatlarının Cochliobolus yaprak lekesi hastalığına karşı fide dönemi tepkilerinin değerlendirilmesi. *Plant Protection Bulletin* , 58 (4) , 221-226 . DOI: 10.16955/bitkorb.368467

Bayraktar H, Akan K (2012). Genetic characterization of Pyrenophora graminea isolates and the reactions of some barley cultivars to leaf stripe disease under greenhouse conditions. Turkish Journal Agriculture and Forestry 36: 329-339.

Çelik E, Karakaya A (2015). Eskişehir ili arpa ekim alanlarında görülen fungal yaprak ve başak hastalıklarının görülme sıklıklarının ve yoğunluklarının belirlenmesi. Bitki Koruma Bülteni 55 (2): 157-170.

Çelik Y, Karakaya A, Çelik Oğuz A, Mert Z, Akan K, Ergün N, Sayim İ (2016). Determination of the reactions of some barley landraces and cultivars to Drechslera graminea. Akdeniz Üniversitesi Ziraat Fakültesi Dergisi 29(2): 43-47.

Düşünceli F, Çetin L, Albustan S, Mert Z, Akan K, Karakaya A (2008). Determination of the reactions of some barley cultivars and genotypes to scald under greenhouse and field conditions, Tarım Bilimleri Dergisi (Journal of Agricultural Sciences) 14(1): 46-50.

Ertürk H, Karakaya A, Çelik Oğuz A (2018). Leaf diseases occurring on barley plants in Bala district of Ankara province, Turkey. *Ecological Life Sciences* 13 (4), 204-207. Retrieved from https://dergipark.org.tr/tr/pub/nwsaecolife/issue/39711/46147

Geçit HH Çiftçi CY, Emeklier Y, İkincikarakaya SÜ, Adak M S, Kolsarıcı Ö, Ekiz H, Altınok S, Sancak C, Sevimay CS Kendir H (2009). Tarla Bitkileri. Ankara Üniversitesi, Ziraat Fakültesi Yayınları, Yayın no: 1569, Ders Kitabı: 521.

Geçit HH (2016). Serin İklim Tahılları (Buğday, Arpa, Yulaf, Triticale). Ankara Üniversitesi Ziraat Fakül-tesi Yayınları, Yayın No:1640. Ankara.

Harlan JR (1979). On the origin of barley. In: Barley: origin, botany, culture, winter hardiness, genetics, utilization, pests. Agriculture Handbook 338, Unites States Department of Agriculture, Washington, D.C., 10-36 p

İlgen MZ, Karakaya A, Oğuz AÇ (2017). Leaf diseases occurring on barley and wheat fields in Çubuk District of Ankara, Turkey. Radovi Poljoprivrednog Fakulteta Univerziteta u Sarajevu\Works of the Faculty of Agriculture University of Sarajevo, 62(67 (2)), 210-215

Karakaya A, Mert Z, Çelik Oğuz A, Çetin L (2016). Distribution of barley stripe disease in Central Anatolia, Turkey. Selcuk Journal of Agriculture and Food Sciences 30 (2): 58-61.

Karakaya A, Mert Z, Çelik Oğuz A, Azamparsa MR, Çelik E, Akan K, Çetin L (2014). Current status of scald and net blotch diseases of barley in Turkey. IWBLD – 1st International Workshop on Barley Leaf Diseases. 03-06 June 2014, Salsomaggiore Terme, Italy.

Karakaya A, Akyol A (2006). Determination of the seedling reactions of some Turkish barley cultivars to net blotch. Plant Pathology Journal 5(1):113- 114.

Kün E (1996). Tahıllar - I (Serin iklim Tahılları). Ankara Üniversitesi, Ziraat Fakültesi Yayınları, Yayın No: 1451, Ankara, 332 s.

Mamluk OF, Çetin L, Braun HJ, Bolat N, Bertschinger L, Makkouk KM, Düşünceli F (1997). Current status of wheat and barley diseases in the Central Anatolia Plateau of Turkey. Phytopathologia Mediterranea 36: 167-181.

Mert Z, Karakaya A (2004). Assessment of the seedling reactions of Turkish barley cultivars to scald. Journal of Phytopathology 152(3): 190-192.

Özdemir HY, Karakaya A, Çelik Oğuz A (2017). Kırıkkale ilinde buğday ve arpa ekim alanlarında görülen fungal yaprak hastalıklarının belirlenmesi. Bitki Koruma Bülteni 57 (2): 89–112

Ulus C, Karakaya A (2007). Assessment of the seedling reactions of some Turkish barley cultivars to barley stripe. Tarım Bilimleri Dergisi (Journal of Agricultural Sciences) 13(4): 409-412.

Usta P, Karakaya A, Çelik Oğuz A, Mert Z, Akan K, Çetin L (2014). Determination of the seedling reactions of twenty barley cultivars to six isolates of *Drechslera teres* f. *maculata*. Anadolu Tarım Bilimleri Dergisi (Anadolu Journal of Agricultural Sciences) 29(1): 20-25.

Saari EE, Prescott JM (1975). A scale for appraising the foliar intensity of wheat diseases. Plant Disease Report, 59: 377-380.

Taşkoparan H, Karakaya A (2009). Assessment of the seedling reactions of some barley cultivars to *Drechslera teres* f. *maculata*. Selçuk Tarım ve Gıda Bilimleri Dergisi 23(50):60-62.

TÜİK (2022). Bitkisel Üretim İstatistikleri. https://data.tuik.gov.tr/Kategori/GetKategori?p=tarim-111&dil=1 (Erişim tarihi: 18.1.2022)

Yazıcı B, Karakaya A, Oğuz AÇ, Mert Z (2015). Determination of the seedling reactions of some barley cultivars to *Drechslera teres* f. *teres*. Bitki Koruma Bülteni 55(3): 239-245.

Yıldırım AF, Kınacı E, Hekimhan H, Çeri S (1999). Konya, Karaman, Niğde ve Aksaray yörelerinde tahıllarda önemli hastalıkların durumu ve bunlara dayanıklılık kaynaklarının araştırılması. Orta Anadolu'da Hububat Tarımının Sorunları ve Çözüm Yolları Sempozyumu. 8-11 Haziran 1999, Konya, 404-413.

Selcuk Journal of Agriculture and Food Sciences SJAFS

http://sjafs.selcuk.edu.tr/sjafs/index

(2022) 36 (3), 414-420

e-ISSN: 2458-8377

DOI:10.15316/SJAFS.2022.055

Research Article

Determination of Disease Severity and Anastomosis Groups of *Rhizoctonia solani* Isolates from Chickpea Plant in Konya Province

Fatma Rana BAYRAM[1], Nuh BOYRAZ[1*], Kemal KESENCİ[2]

[1]Selçuk University, Faculty of Agriculture, Department of Plant Protection, Konya, Turkiye
[2]Safa Tarım A.Ş., Konya, Türkiye

ARTICLE INFO

Article history:
Received date: 16.09.2022
Accepted date: 02.11.2022

Keywords:
Chickpea
Rhizoctonia solani
Disease severity
Anastomosis group

ABSTRACT

This study was carried out to determine the anastomosis groups and disease severity of *Rhizoctonia solani*, which causes root and root collar rot in chickpea production areas in Konya. A total of eleven isolates of *Rhizoctonia* spp. were obtained in the isolations made from the root and root collar of the plants in the surveys. *Rhizoctonia* isolates examined microscopically were grouped as multinucleate, binucleate according to the number of nuclei. Ten isolates were multinucleate and identified *R. solani*, while one isolate was binucleate *Rhizoctonia* sp. In pathogenicity tests, five multinucleate isolates were found to be pathogenic. Disease severity was determined as 100% for 1.2 A and 1.2 B coded isolates, 86% for 1.1 A coded isolate, 75% for 1.1 B coded isolate, and 44% for 1.2 C coded isolate. It was observed that the other multinucleate five isolates caused only a shortening in plant heights. No disease symptoms were observed in the plants inoculated by the binucleate isolate. All isolates were characterized using the internal transcribed spacer (ITS) region of ribosomal DNA, and two different anastomosis groups were defined accordingly. It was determined that 5 virulence multinucleate *R. solani* isolates belonged to AG-4 HGII and 5 other multinucleate isolates to AG BI anastomosis group.

1. Introduction

In terms of cultivation area in Turkiye, legumes take the most important place after cereals. Considering the total legume production, Turkey is among the largest producers in the world (FAOSTAT, 2021). Among nine legume species grown in Turkey, chickpea (*Cicer arietinum* L.) is the most common. While chickpea cultivation area constitutes 57.6% of the legume cultivation and 51.2% of the production amount area in the country (TURKSTAT, 2021).

Chickpea, which is an important source of vegetable protein, has been used in human and animal nutrition and green fertilization both in Turkiye and in the world, especially in the Near East, Far East, Mediterranean, South America, and Central America countries, which has been cultivated since ancient times (Eser, 1976; Reddy and Singh., 1984; Reddy and Kabbabeh, 1985). It can be grown in almost every region in Turkey, and the region where it grows most is the Central Anatolia region. Almost 65.7% (414 thousand tons) of the total chickpea production takes place in the Central Anatolia region. According to the provinces, Ankara ranks first with 93 thousand tons in chickpea production, Yozgat ranks second with 86 thousand tons, Kırşehir ranks third with 78 thousand tons, and Konya ranks fourth with 50 thousand tons (TURKSTAT 2021).

Although it has a large cultivation area, it is thought that the desired yield cannot be obtained for many reasons in chickpea. Abiotic and biotic stress factors are the most important factors limiting production, including diseases and pests. It is seen that the soil-borne agents also cause yield losses in recent years, together with the *Ascochyta rabiei* factor. *Fusarium oxysporum* f. sp. *ciceris F. solani F. acuminatum F.moniliforme F.sambucinum F. equiseti Rhizoctonia solani* Kühn., *Macrophomina phaseolina* (Tassi) Goid, and *Cylindrocarpon tonkinense* Bugnic. were determined as soil-borne agents that causing wilt and root rot in chickpea (Aydın and İnal, 2019; Dolar 1996; Dolar and Nirenberg, 1998; Soran, 1977; Yücel and Güncü, 1991).

In particular, fungi of the genus *Rhizoctonia* are common in many parts of the world and have a wide host range. It causes a variety of diseases in important cultivars worldwide, including species in Solanaceae, Fabaceae, Asteraceae, Poaceae, and Braccicaceae, as well as ornamental and forest trees (Ogoshi, 1975; Ogoshi et al., 1996).

* Corresponding author email:*nboyraz@selcuk.edu.tr*

Rhizoctonia fungi are a large and complex group that varies within itself (Carling and Summer, 1992). They are examined in three groups in terms of the number of nuclei in hyphae cells. These; They are called multinucleic (MN, multinucleated), binucleic (BN, binucleated) and uninucleic (UN, mononuclear) (Sneh et al.,1991). Within the multinucleic and binucleic *Rhizoctonia* genera, there are subgroups whose hyphae are compatible with each other and can fuse where they come into contact, and these fused subgroups are called the anastomosis group (AG) (Vilgays and Cubeta, 1994; Sneh et al., 1996). Anastomosis groupings are classically based on the characterization of hyphal anastomosis reactions of *Rhizoctonia* hyphae, and deoxyribonucleic acid (DNA)-based methods are increasingly used to identify anastomosis groups (AGs) (Vilgalys and Gonzalez, 1990; Cubeta et al., 1991; Carling et al.,2002).

R.solani, one of the important species in the *Rhizoctonia* genus, was named by Kühn in 1858 and the most important species of the *Rhizoctonia* genus was introduced to the scientific world with the naming of the telemorph *Thanatephorus cucumeris* (Frank) Donk and became the most studied and known species in the genus (Carling and Sumner, 1992). Matsumoto reported the presence of hyphal anastomosis for the first time in *R.solani* in 1921. Schulz, on the other hand, classified isolates according to their anastomosis abilities for the first time in 1936.There are 14 anastomosis groups of *R.solani*, AG 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13 and AGB1 (Carling et al. 1999; 2002; Yang and Li, 2012) groups detected in chickpea are AG-1, AG-2-2, AG-2-2LP, AG-2-3, AG-3, AG-4 and AG-5 (Dubey et al. 2011).AG-4 and AG-5 groups are also available in chickpea cultivation areas in our country (Tuncer and Erdiller,1990) Demirci et al.,1999; Başbağcı et al., 2019).

In this study, it was aimed to determine the anastomosis groups of *Rhizoctonia* isolates obtained from chickpea cultivation areas in and around Konya and to reveal their virulence.

2. Materials and Methods

2.1.Materials

2.1.1. Plant material

The main material of the study is the diseased plant samples collected from chickpea cultivation areas in Konya province of Turkey and the disease-sensitive ILC-482 chickpea variety used in the pathogenicity tests of *Rhizoctonia* isolates.

2.1.2. Fungal material

In the study,eleven isolates of *Rhizoctonia* isolates were obtained from diseased chickpea plants and grouped according to nucleus and anastomosis properties.

2.1.3.Plant growing media and chemicals

In pathogenicity experiments, peat, garden soil and perlite were used in the ratio of 2:1:1 as the growing medium. Potato dextrose agar (PDA) and water agar (WA) with the addition of streptomycin sulfate were used in fungal isolations and purifications. Safranin O solution was used to determine the number of nuclei of *Rhizoctonia* isolates. In the isolations, 1% NaOCl (Sodium hypochlorite) was used for surface disinfection, and barley seeds were used for long-term storage of the isolates.

2.2.Methods

2.2.1. Collection of diseased plant samples

Diseased plant samples were collected from April to July of 2019 according to guided sampling. Samples were taken considering typical disease symptoms with dark brown lesions on the root and root collar. The samples were labeled and brought to the laboratory in paper bags and stored at +4 °C until isolation.

2.2.2. Fungal ısolation from diseased plant samples

Collected plant samples were washed under tap water. The washed plant samples were allowed to dry on blotting papers.Then, with the help of a sterile scalpel, sections including the healthy part along with the area showing disease symptoms were taken on the root and root collar parts. The sections taken were disinfected superficially in 1% NaOCI for 1-2 minutes, passed through sterile distilled water three times and left to dry for 5-10 minutes in a laminar flow cabinet on sterile blotting papers.

Dried plant sections were placed in Potato Dextrose Agar (PDA, Merck + Streptomycin sulfate) medium, 5-6 pieces per petri dish. Petri dishes were incubated at 25 °C for 3-5 days. At the end of this period, mycelial discs with a diameter of 5 mm from the developing colonies were transferred back to fresh PDA medium and grown for 7-10 days at 25 °C to obtain pure cultures.

2.2.3. Storage of Rhizoctonia isolates

Barley seeds were taken into capped glass tubes and sterilized in an autoclave for one hour at 121 °C for two consecutive days. Then, mycelial discs with a diameter of five mm, taken from *Rhizoctonia* cultures that were previously grown in PDA medium for 5-6 days, were placed in these tubes and incubated at 24±1 °C for 20-30 days and the hyphae were expected to cover the seeds (Başbağcı., 2020). Then, *Rhizoctonia* isolates, which were wrapped in sterile barley seeds, were placed in a refrigerator operating at +4 °C for long-term storage (Olaya and Abawi, 1994).

2.2.4. Identification and determination of groups

Identification of fungal isolates obtained from diseased plants was made according to Carling and Summer (1993), taking into account the morphological characteristics of *Rhizoctonia* spp. In order to determine the number of nuclei of the isolates identified as *R. solani*, firstly nuclei were stained to determine multinucleate (multi-nucleated), binucleate (two-nucleated) isolates. To prepare 100 ml of safranin O solution; 6 ml of 0.5% saffron prepared with distilled water was poured into a

flask, and 10 ml of 3% KOH solution prepared with distilled water, 5 ml of glycerine and 79 ml of distilled water were added and mixed (Akın, 2001). A drop of Safranin O solution was dropped on a slide for examination. Hyphae tips taken from *Rhizoctonia* isolates developed by transferring to PDA and WA medium and incubating in the dark at 25 °C with the help of a coverslip were placed on the solution in the slide. In the preparations prepared in this way, the number of nuclei in the hyphae was determined by taking into account the number of nuclei in at least 20 cells (Ogoshi et al.,1990, Carling et al., 1994, Karaca et al., 2002).

Molecular characterization studies to determine the anastomosis groups of *Rhizoctonia solani* isolates were carried out in Bolu Abant İzzet Baysal University, Faculty of Agriculture, Department of Plant Protection Laboratory. In the relevant laboratory, anastomosis groups were determined by amplification of *R.solani* isolates using ITS gene region 1F (CTTGGTCATTTAGAGGAGTAA) and ITS 4B (CAGAGACTTGTACACGGTCCAG)primers (Gardens and Bruns 1993).

2.2.5. Pathogenicity Test

ILC-482 chickpea cultivar known to be sensitive to *R.solani* was used in the studies. For this, sterile growing media containing a 2:1:1 mixture of peat, garden soil, and perlite were filled into 2-liter sterile pots.After the sterile growing medium was filled into the pots, 3 pieces of chickpea seeds, which were pre-germinated under sterile conditions in a medium containing 1% WA, were planted in each pot.

The inoculation process was carried out by placing one each of the barley seeds on which *R.solani* hyphae had been wrapped before, next to each germinated chickpea seed sown. After the inoculation process, the seeds were covered with sterile growing medium. As a control, healthy chickpea seeds germinated in a medium containing 1% WA disinfected with 2% NaOCl for 5 minutes were planted in pots containing the same amount of sterile growing medium.3 pots were used for each isolate and one pot was considered a replicate. All pots were left to develop at 20-25 °C under controlled climate room conditions. Approximately 45 days after planting, the plants were removed and disease severity values were calculated according to the Townsend-Heuberger formula given below, taking into account the 0-4 scale given in Table 1 (Townsend-Heuberger, 1943; Kim et al., 1997; Demirci, 1998; Paulitz et al., 2000).

Table 1
0-4 scale used in the evaluation of disease severity of *Rhizoctonia* isolates

Scale degree	Symptom
0	No symptom (root and root collar)
1	Slight discoloration or roots emerging from seed less than 3 cm
2	One or more small lesions (< 0.5 cm) or roots emerging from the seed less than 2 cm
3	One or more small lesions (> 0.5 cm) or roots emerging from the seed less than 1 cm
4	Severe lesion, completely dead or rootless seedling

Disease Severity (%) = Σ(nxV/ZxN)x100

n: number of plants with different disease degrees on the scale

V: scale degree

Z: highest scale degree

N: total number of plants observed

2.2.6. Statistical analysis

The SPSS statistical program (SPSS Inc., version 17.0) was applied to disease severity values obtained from pathogenicity experiments. The difference between the applications was determined by the Tukey multiple comparison test (P≤0.01).

3. Results and Discussion

Eleven *Rhizoctonia* isolates were obtained as a result of isolations and identifications made from 204 diseased plant samples collected in 2019 by guided sampling method in chickpea cultivation areas of Konya Province.

R.solani isolates, which were examined microscopically using the prepared 0.5% Safranin O solution, were grouped as multinucleate (multinucleated), binucleate (binucleated) according to the number of nuclei, and 10 isolates were multinucleated (1.1A, 1.1B, 1.2A, 1.2B, 1.2C, KK-11E, KK-11A, KK-11B, KK-11C, KK-11D (Figure 1) and 1 isolate was binucleate (KAR-5 (Figure 2).

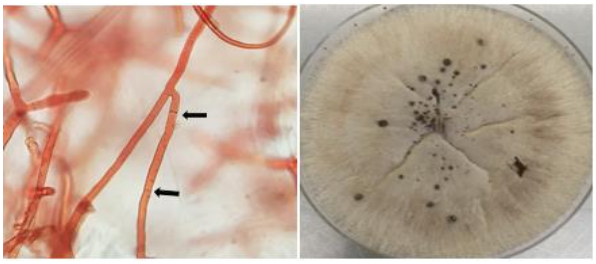


Figure 1
Microscopic view (left) and petri dish (right) of the multinucleate *Rhizoctonia solani* isolate

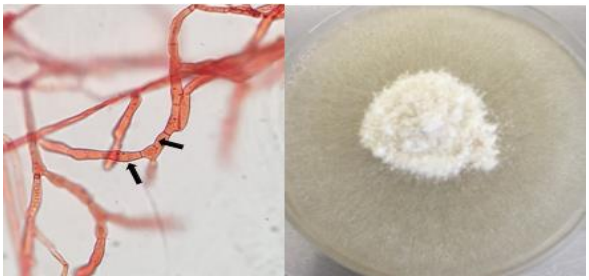


Figure 2
Microscopic view of binucleate *Rhizoctonia* sp. isolate (left) and petri dish (right)

As a result of the analyzes performed considering the molecular characterization of a total of eleven *Rhizcotonia* isolates, it was determined that the existing multinucleate isolates were included in the AG-4 HGII and AG-BI anastomosis groups. The anastomosis group of the binucleate isolate (KAR-5) could not be determined (Table 2).

Table 2
Anastomosis groups of *Rhizoctonia* isolates determined according to ITS primers

Isolate number	Isolate number	Anastomosis groups according to ITS primers
1	1.1 A	AG-4 HGII
2	1.1 B	AG-4 HGII
3	1.2 A	AG-4 HGII
4	1.2 B	AG-4 HGII
5	1.2 C	AG-4 HGII
6	KK-11 E	AG-BI
7	KK-11 A	AG-BI
8	KK-11 B	AG-BI
9	KK-11 C	AG-BI
10	KK-11 D	AG-BI
11	KAR-5	Not determined

The disease severity values of 11 *R.solani* isolates in chickpea plants as a result of the pathogenicity tests carried out with three replications are given in Table 3.

Table 3
Disease severity rates of *Rhizoctonia* isolates on chickpea plants (%)

Isolate number	Isolate number/anastomosis group	Disease severity (%)
1	1.1 A/AG-4 HGII	86.00 b
2	1.1 B/AG-4 HGII	75.00 c
3	1.2 A/AG-4 HGII	100.00 a
4	1.2 B/AG-4 HGII	100.00 a
5	1.2 C/AG-4 HGII	44.00 d
6	KK-11 E/AG-BI	0.00 e
7	KK-11 A/AG-BI	0.00 e
8	KK-11 B/AG-BI	0.00 e
9	KK-11 C/AG-BI	0.00 e
10	KK-11 D/AG-BI	0.00 e
11	KAR-5-/not determined	0.00 e
12	Control	0.00e

P≤0.01 (There is no statistical difference between the means expressed with the same letter in the same column)

While different rates of disease severity were observed in five isolates as a result of pathogenicity tests, no disease severity value could be calculated in six isolates. All of the isolates that were determined with different rates of disease belonged to the AG-4 HGII anastomosis group, with the highest disease severity in 1.2A and 1.2B isolates detected. These two isolates were followed by isolate 1.1A with a disease severity value of 86%. The lowest disease severity was obtained from isolate 1.2 C with 44% (Table 3). Symptoms such as seed infection, pre-emergence and post-emergence damping-off and visible lesions on the root and collar were observed in plants infected with the disease at different levels by inoculation of isolates in the AG-4 HGII anastomosis group (Figure 3, 4).

Figure 3
Infection of *R.solani* isolate no. 1.2 A on ILC-482 chickpea seeds

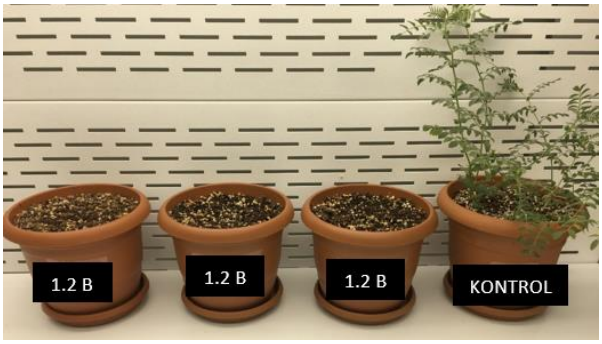


Figure 4
The image that emerged as a result of pre-emergence and post-emergence damping-off infection on chickpea plants(ILC-482 variety) in the pathogenicity test of isolate 1.2 B of *R.solani*

In the pathogenicity experiment, none of the symptom types described in the 0-4 scale given in Table 1 of the multinucleate five isolates with the AG BI anastomosis group were observed in plants, so numerical disease severity values could not be determined in them. Therefore, the disease severity of all of these isolates was evaluated as 0%.Although these isolates did not have the disease symptoms specified in the 0-4 scale, it was observed that they caused a partial shortening of the plant heights compared to the control. KAR-5 isolate, which was detected as a binucleate isolate among *R. solani* isolates, did not cause any disease symptoms on plants (Figure 5).

Figure 5
Image of inoculated chickpea plants(ILC-482 variety) with binucleate KAR-5 isolate of *Rhizoctonia* sp.

Although the morphological characters of the obtained isolates are variable, eleven *R. solani* isolates are divided into three categories according to colony color and most of them have light brown colony color (Ganeshamoorthi and Dubey, 2015).

Rhizoctonia spp. it is a complex group that includes pathogen species that cause economic loss in many agriculturally important plants, as well as non-pathogenic species (Sneh et al., 1996). There are disease records of different *Rhizoctonia* spp. in various legumes grown in various ecological conditions of the world. Generally, binucleated *Rhizoctonia solani* isolates are not considered pathogenic in plants, although some exceptions have been reported (Ogoshi, 1987; Hua et al., 2014; Türkkan et al., 2018). In our study, it was determined that the binucleate *Rhizoctonia solani* isolate that we obtained did not show any disease symptoms in plants, that is, it was not pathogenic. On the other hand, multinucleated *Rhizoctonia solani* isolates cause root and crown diseases in many plants, including legumes (Verma, 1996; Sneh et al., 1996; Erper et al., 2016; Yıldırım and Erper, 2017; Türkkan et al., 2020). In our study, among eleven *R. solani* isolates whose virulence was tested, five isolates in the multinucleate AG-4 HGII anastomosis group were found to cause pre-emergence damping-off at different percentages. It was observed that the other five isolates in the multinucleate, AG-BI anastomosis group caused only stature shortening in plants. *R.solani* AG-4 isolates isolated from chickpea were found to be moderately and highly virulent, and some of them caused pre-emergence damping-off (Hwang et al., 2003).

R.solani, which is in the AG-4 anastomosis group, has been reported many times worldwide to be pathogenic for chickpeas (Hwang et al.,2003; Dubey et al., 2011, 2014; Ganeshamoorthi and Dubey, 2013, 2015). Dubey et al. (2011) reported that the severity of disease caused by AG-4 isolates ranged from 11% to 100% in chickpea, and the highly virulent isolate level was 75.6% in chickpea. However, Ganeshamoorthi and Dubey (2015) found that 4 of 50 *R.solani* isolates of AG-4 were moderately virulent in chickpea. This information supports our findings, and it was found that our isolates in the AG-4 anastomosis group caused between 44% and 100% disease severity on chickpea plants.

Five AG groups (AG-1, AG-2, AG-3, AG-4 and AG-5) of *R.solani* have been determined on chickpea in the world (Hwang et al., 2003; Mikhail et al., 2010; Youssef et al., 2010; Dubey et al., 2011, 2014; Ganeshamoorthi and Dubey, 2013, 2015).

In Turkey, AG-4 of *Rhizoctonia solani* was found in barley (Demirci, 1998; Ünal and Kara, 2017), beans (Eken and Demirci, 2004; Kılıçoğlu and Özkoç, 2013), pepper (Tuncer and Eken, 2013), tomato (Yıldız and Döken, 2002), soybean (Erper et al., 2011), cotton (Kural et al., 1994), Johnson grass (Demirci et al., 2002), and wheat (Demirci, 1998; Ünal et al., 2015) has been reported. AG-4 and AG-5 groups with high virulence are present in chickpea cultivation areas in our country (Tuncer and Erdiller 1990; Demirci et al., 1999;Başbağcı et al., 2019). This study showed that the AG-BI anastomosis group of *R.solani* is also present in our country.

Since the isolates of *R.solani* fungus do not have the potential to cause disease at the same severity in chickpea plants, and some isolates do not have any effect on disease formation, not every *R.solani* isolate obtained is considered as a pathogen and considering the compatibility of such isolates with pathogenic isolates, they can be used as biological control agents. With the anastomosis that may occur between pathogen and apathogen *R.solani* isolates, it may be possible to determine effective biological control agents by activating the hypovirulence mechanism.

4. Acknowledgements

This study was carried out within the scope of the University-Industry Collaboration Support Program, with the code AR-TAR-0002, "Detection of National Biological Agents in Biopesticide Development" and It includes a part of Fatma Rana BAYRAM's master's thesis. The results of the study were presented orally at the 8th Plant Protection Congress with International Participation, and the abstract was published in the congress booklet.

We would like to thank Selcuk University Scientific Research Coordinatorship (BAP) and Safa Tarım A.Ş. for their support. At the same time, for his contributions to the detection of anastomosis groups of *Rhizoctonia* isolates, we would like to thank Doç.Dr.Göksel ÖZER.

5. Reference

Akın V (2001). Samsun ili Bafra, Ondokuzmayıs ve Tekkeköy ilçelerindeki tarım alanlarından izole edilen patojen *Rhizoctonia* izolatları üzerine patojen olmayan izolatların biyokontrol etkisi. Yüksek Lisans Tezi, Ondokuz Mayıs Üniversitesi, Fen Bilimleri Enstitüsü, Biyoloji Anabilim Dalı, 60, Samsun.

Aydın MH, İnal B (2019). Genetic characterization and virulence of *Fusarium* spp. isolated from chickpea. *Cellular and Molecular Biology*, 65 (1), 56-60.

Başbağcı G, Ünal F, Uysal A, Dolar FS (2019). Identification and pathogenicity of *Rhizoctonia solani* AG-4 causing root rot on chickpea in Turkey. *Spanish Journal of Agricultural Research*, 17 (2), 1007.

Başbağcı G (2020). Uşak, Kütahya, Isparta ve Denizli İllerinde Nohutta Sorun Olan *Rhizoctonia* Tür ve Anastomosis Gruplarının Karakterizasyonu ile Bazı Çeşitlerin Reaksiyonlarının Belirlenmesi. Ankara Üniversitesi Fen Bilimleri Enstitüsü, Bitki Koruma Anabilim Dalı, Doktora Tezi, Ankara, 90s.

Carling DE, Summer DR (1992). *Rhizoctonia* in: Singleton L.L., Mihail, J.D. ve Rush, C.M. (eds.) *Methods for research on soil-borne phytopathogenic fungi*. APS Press, 157-165, Minnesota.

Carling DE, Rothrock CS, MacNish GC, Sweetingham MW, Brainard KA, Winters SW(1994). Characterization of anastomosis group 11 (AG-11) of *Rhizoctonia solani*. *Phytopathology*, 84(12), 1387-1393.

Carling DE, Pope EJ Brainard KA, Carter D (1999). Characterization of mycorrhizal isolates of *Rhizoctonia solani* from an orchid, including AG-12, a new anastomosis group. *Phytopathology*. 89(10), 942-946.

Carling DE, Baird RE, Gitaitis RD, Brainard KA, Kuninaga S (2002). Characterization of AG-13, a newly reported anastomosis group of *Rhizoctonia solani*. *Phytopathology*, 92(8), 893-899.

Cubeta MA, Echandi E,Abernethy T, Vilgalys R (1991). Characterization of anastomosis groups of binucleate *Rhizoctonia* species using restriction analysis of an amplified ribosomal RNA gene. *Molecular Plant Pathology* 81: 1395-1400.

Demirci E (1998). *Rhizoctonia* species and anastomosis groups isolated from barley and wheat in Erzurum, Turkey, *Plant Pathology*, 47(1), 10-15.

Demirci E, Eken C, Kantar F (1999). Pathogenicity of wilt and root rot pathogens of chickpea cv. Aziziye-94. *The Journal of Turkish Phytopathology*, 28(l-2), 25-32.

Demirci E, Eken C, Zengin H (2002). First report of *Rhizoctonia solani* and binucleate *Rhizoctonia* from johnsongrass in Turkey. *Plant Pathology*, 51(3),391.

Dolar FS (1996). Determination of chickpea diseases in Ankara, Turkey. *International Chickpea and Pigeonpea Newsletter*, 3, 33-34.

Dolar F.S,Nirenberg HI (1998). *Cylindrocarpon tonkinense* Bugn.- A new pathogen of chickpea. *Journal of Phytopathology*, 146(10), 521-523.

Dubey SC, Tripathi A, Upadhyay BK, Thakur M (2011). Pathogenic behaviour of leguminous isolates of *Rhizoctonia solani* collected from different Indian agro-ecological regions. *Indian Journal of Agricultural Sciences*, 81(10), 948-953.

Dubey SC, Tripathi A, Upadhyay BK, Deka UK (2014). Diversity of *Rhizoctonia solani* associated with pulse crops in different agro-ecological regions of India. *World Journal of Microbiology and Biotechnology*, 30(6), 1699-1715.

Eken C, Demirci E (2004). Anastomosis groups and pathogenicity of *Rhizoctonia solani* and binucleate *Rhizoctonia* isolates from bean in Erzurum, Turkey. *Journal of Plant Pathology*, 86(1), 49-52.

Erper İ, Karaca GH, Özkoç İ (2011). Identification and pathogenicity of *Rhizoctonia* species isolated from bean and soybean plants in Samsun, Turkey. *Archives of Phytopatholgy and Plant Protection*, 44(1), 78-84.

Erper İ, Cebi-Kılıçoğlu M, Türkkan M, Onder H (2016). Characterization and pathogenicity of *Rhizoctonia* spp.isolated from winter squash in the Black Sea region of Turkey. *European Journal of Plant Pathology*, 146, 683-697.

Eser D (1976). Nohut (*Cicer arietinum* L.)'ta başlıca bitki özelliklerinin kalıtım değerleri, bu özellikler ile bitki verimi arasındaki ilişkiler ve *Ascochyta rabıei* (pass)'ye dayanıklılığın kalıtımı. *Ankara Üniversitesi, Ziraat Fakültesi yayınları*, yayın no:363, Ankara.

FAOSTAT D (2021). FAOSTAT D (2021) Food and agriculture organization of the United Nations. Statistical database. http://www.fao.org/faostat/en/#data/QC. (Erişim tarihi: 14.06.2021).

Ganeshamoorthi P, Dubey SC (2013). Anastomosis grouping and genetic diversity analysis of *Rhizoctonia solani* isolates causing wet root rot in chickpea. *African Journal of Biotechnology*, 12(43), 6159-6169.

Ganeshamoorthi P, Dubey SC, (2015). Morphological and pathogenic variability of *R. solani* isolates associated with wet root rot of chickpea in India. *Legume Research-An International Journal*, 38(3), 389-395.

Gardens M, Bruns TD (1993). ITS primers with enhanced specificity for basidiomyces-application to the identification of mycorrhizae and rusts. *Molecular Ecology*, 2(2), 113-118.

Hua GKH, Bertier L, Soltaninejad S, Höfte M (2014). Cropping systems and cultural practices determine the *Rhizoctonia* anastomosis groups associated with *Brassica* spp. in Vietnam. *PloS one*, 9(11), 111750.

Hwang SF, Gossen BD., Chang KF, Turnbull GD, Howard RJ, Blade SF (2003). Etiology and impact of *Rhizoctonia* seedling blight and root rot of chickpea on the Canadian prairies.*Canadian Journal of Plant Science*, 83(4), 959-967.

Karaca GH, Özkoç İ, Erper İ (2002). Determination of the anastomosis grouping and virulence of *Rhizoctonia solani* Kühn isolates associated with bean plants grown in Samsun/Turkey. *Pakistan Journal of Biological Sciences*, 5(4), 434-437.

Kılıçoğlu MÇ, Özkoç İ (2013). Phylogenetic analysis of *Rhizoctonia solani* AG-4 isolates from common beans in Black Sea coastal region, Turkey, based on ITS5.8S rDNA. *Turkish Journal of Biology*, 37(1), 18-24.

Kim DS, Cook RJ, Weller DM (1997). *Bacillus* sp. L324-92 for biological control of three root diseases of wheat grown with reduced tillage. *Phytopathology*, 87(5), 551-558.

Kural İ, Sağır A, Tatlı F (1994). Characterization and pathogenicity of anastomosis groups of *Rhizoctonia solani* isolated from cotton in Southeastern Turkey. *9th Congress of the Mediterranean Phytopathological Union*, 117-120, Aydın.

Mikhail MS, Sabet KK, Omar MR, Amal AA, Kasem KK (2010). Current *Rhizoctonia solani* anastomosis

groups in Egypt and their pathogenic relation to cotton seedlings. *African Journal of Microbiology Research*, 4(5), 386-395.

Ogoshi A (1975). Grouping of *Rhizoctonia solani* Kühn and their perfect stages. *Review Plant Protection Research*, 8, 93-103.

Ogoshi A (1987). Ecology and pathogenicity of anastomosis and intraspecific groups of *Rhizoctonia solani* Kühn. *Annual Review of Phytopathology*, 25(1), 125-143.

Ogoshi A, Cook RJ, Bassett EN (1990). *Rhizoctonia* species and anastomosis groups causing root rot of wheat and barley in the Pacific Northwest. *Phytopathology*, 80(9), 785-788.

Ogoshi A, Sneh B, Jabaji-Hare S, Neate S, Dijst G (1996). Introduction – the genus *Rhizoctonia*. In: *Rhizoctonia* species: *Taxonomy, Molecular Biology, Ecology, Pathology and Disease Control*. Kluwer Academic Publishers, Dordrecht, 1–9 p.

Olaya G, Abawi GS (1994). Characteristics of *Rhizoctonia solani* and binucleate *Rhizoctonia* species causing foliar blight and root rot on table beets in New York State. *Plant Disease*, 78(8), 800-804.

Paulitz TC, Smith JD, Kidwell KK (2003). Virulence of *Rhizoctonia oryzae* on wheat and barley cultuvars from the Pacific Northwest. *Plant Disease*, 87(1), 51-55.

Reddy MV, Singh KB (1984). Evaluation of a World Collection of Chickpea Germ Plas Accessions for Resistance to Ascochyta Blight. *Plant Disease*, 68, 900-901.

Reddy MV, Kabbabeh S (1985). Pathogenetic Variability in *Ascochyta rabiei* (Pass.) Lab. in Syria and Lebanon. *Phytopath.Medit*., 24, 265-266.

Sneh B, Burpee L, Ogoshi A (1991). Identification of *Rhizoctonia* species. *American Phytopathological Society Press*, p.133, Minnesota.

Sneh B, Jabaji-Hare S, Neate S, Dijst G (1996). *Rhizoctonia* species: Taxonomy, molecular biology, ecology, pathology and diseases control. Springer Science & Business Media, 578, Netherland.

Soran H (1977). The fungus disease situation of edible legumes in Turkey. *The Journal of Turkish Phytopathology*, 6(1), 1-7.

Townsend GK, Heuberger JW (1943). Methods for estimating losses caused by diseases in fungicide experiments. *Plant Disease Reporter*, 27, 340-343.

Tuncer G, Erdiller G (1990). The identification of *Rhizoctonia solani* Kuhn anastomosis groups isolated from potato and some other crops in Central Anatolia. *The Journal of Turkish Phytopathology*, 19 (2): 89-93.

Tuncer S, Eken C (2013). Anastomosis grouping of *Rhizoctonia solani* and binucleate *Rhizoctonia* spp. isolated from pepper in Erzincan, Turkey. *Plant Protection Science*, 49(3), 127-131.

TURKSTAT (2021). Turkish Statistical Institute. Crop Production Statistics. https://biruni.tuik.gov.tr/medas/?kn=92&locale=tr. (Erişim tarihi: 14.06.2021).

Türkkan M, Erper İ, Çebi Kılıçoğlu M, Yazıcıoğlu E, Özcan M (2018). Characterization and pathogenicity of *Rhizoctonia* spp. isolated from kiwifruit in the Middle and Eastern Black Sea region of Turkey. *Journal of Phytopathology*, 166, 761-774.

Türkkan M, Kılıçoğlu MÇ, Erper İ (2020). Characterization and pathogenicity of *Rhizoctonia* isolates collected from *Brassica oleracea* var. *acephala* in Ordu, Turkey, *Phytoparasitica*, 48, 273-286.

Ünal F, Bayraktar H, Yıldırım AF, Akan K, Dolar FS (2015). Kayseri, Kırşehir, Nevşehir ve Aksaray illeri buğday ekim alanlarındaki *Rhizoctonia* tür ve anastomosis gruplarının belirlenmesi.*Bitki Koruma Bülteni*, 55(2), 107-122.

Ünal F, Kara ME (2017). Molecular characterization of *Rhizoctonia* species and anastomosis groups in barley production areas in Ankara province. *The Journal of Turkish Phytopathology*, 46(2):61-67.

Verma PR (1996). Oilseed rape and canola diseases incited by *Rhizoctonia* species. In B. Sneh, S. Jabaj-Hare, S. Neate, & G. Dijst (Eds.), *Rhizoctonia* species: *Taxonomy, molecular biology, ecology, pathology, and disease control* (pp. 249–258). Dordrecht, The Netherlands: Kluwer Academic Press.

Vilgalys R, Gonzalez D (1990). Ribosomal DNA restriction fragment length polymorphisms in *Rhizoctonia solani. Molecular Plant Pathology,* 80, 151-158.

Vilgalys R, Cubeta MA (1994). Molecular systematics and population biology of *Rhizoctonia. Annual Review of Phytopathology*, 32(1), 135-155.

Yang G, Li C (2012). General description of *Rhizoctonia* species complex. In: *Plant Pathology*; Cumagun CJR (ed). InTech, 41-52, Croatia.

Yıldırım E, Erper İ (2017). Characterization and pathogenicity of *Rhizoctonia* spp. isolated from vegetable crops grown in greenhouses in Samsun province, Turkey. *Bioscience Journal*, 33(20), 257-267.

Yıldız A, Döken MT (2002).Anastomosis group determination of *Rhizoctonia solani* Kuhn (Teleomorph: *Thanatephorus cucumeris*) isolates from tomatoes grown in Aydın, Turkey and their disease reaction on various tomato cultivars. *Journal of Phytopathology* -Phytopathologısch Zeithschrift, 150(10), 526-528.

Yücel S, Güncü M (1991). Akdeniz bölgesi yemeklik baklagillerinde görülen fungal hastalıklar. *Bitki Koruma Bülteni*, 31(1-4), 19-30.

Youssef NOB, Rhouma A, Krid S,Kharrat M (2010). First report of *Rhizoctonia solani* AG 2-3 on chickpea in Tunisia. *Phytopathologia Mediterranea*, 49(2), 253-257.

Selcuk Journal of Agriculture and Food Sciences
SJAFS
http://sjafs.selcuk.edu.tr/sjafs/index
(2022) 36 (3), 421-427
Research Article
e-ISSN: 2458-8377
DOI:10.15316/SJAFS.2022.056

Enumeration Of *Bifidobacterium* Spp., *Lactobacillus Acidophilus* and Starter Cultures from Commercial Probiotic Yogurts and Freeze-Dried Yogurt Starter Mixes

Talha DEMIRCI[1,*]
[1]Selçuk University, Faculty of Agriculture, Department of Food Engineering, Konya, Türkiye

ARTICLE INFO

Article history:
Received date: 08.10.2022
Accepted date: 02.11.2022

Keywords:
yogurt
probiotic
enumeration
Bifidobacterium
L. acidophilus
starter

ABSTRACT

Probiotic yogurt is a popular functional food to deliver of probiotic cells for the health-enhancing effects worldwide. The viability of probiotics in yogurt before consumption is the most important factor to providing desired effects, however, probiotic microorganisms have occasionally inadequate viability in marketable food products. In this current study, *Bifidobacterium* spp., *L. acidophilus* and yoghurt starter bacteria enumerations were made in commercial probiotic yoghurt and freeze-dried yogurt mixes. RCA 5.3 and MRS 5.2 media were used for *L. delbrueckii* subsp. *bulgaricus* counting, ST Agar and M17 Agar were used for *Stretococcus thermophilus* counts. While using MRS-Bile Agar and RCA-Clindamycin Agar for *L. acidophilus* enumeration, *Bifidobacterium* spp. counts were performed using MRS-NNLP medium. 5 out of yoghurt samples (A, C, D, and E) did not reveal satisfactory recovery (< 5 log CFU/g) for *L. delbrueckii* subsp. *bulgaricus* colonies on MRS 5.2 Agar while *L. delbrueckii* subsp. *bulgaricus* colony counts on RCA 5.3 Agar below 5 log CFU/g for same tested 4 samples (A, C, D, and E). The recovery rates over 9 log CFU/g were obtained in the enumerations made for all yogurt samples on both ST and M17 media. The problem of insufficient recovery rates that occurred for *L. delbrueckii* subsp. *bulgaricus* in some yogurt samples was not valid for *S. thermophilus.* This work indicated that high amounts of *L. acidophilus* were detected on both media in both of the two yoghurt samples declared as *L. acidophilus* on the label (F and G). On the other hand, bifidobacteria was determined above 5 log CFU/g in only 1 yoghurt sample (B) out of 7 probiotic yoghurts claimed to be *Bifidobacterium* spp. This study reveals relevant information on probiotic and starter counts of commercial probiotic yogurts in Turkey and discusses in detail the possible reasons for the results obtained.

1. Introduction

Probiotics are defined as “live microorganisms that, when administered in adequate amounts, confer a health benefit on the host” by Hill et al. (2014). The probiotic microorganisms that occur in products as a single or mixed cultures have been generally sourced from the gut or from artisanal fermented foods, such as pickles, yoghurts, and cheeses. The majority of the probiotics used in commercial probiotic preparations, are mainly from the *Lactobacillus* genera like *Lactobacillus acidophilus*, *Lactobacillus casei*, *Lactobacillus johnsonii*, *Lactobacillus amylovorous* species and *Bifidobacterium* genera like *Bifidobacterium animalis*, *Bifidobacterium bifidum*, *Bifidobacterium infantis*, and *Bifidobacterium adolescentis* species which are accepted as Generally Regarded as Safe (GRAS) status in the United States http://accessdata.fda.gov or granting Qualified Presumption of Safety status by the European Food Safety Authority (EFSA) (O’Toole, Marchesi et al. 2017). Various therapeutic and health promoting effects have been attributed to probiotic microorganisms including antagonistic effects to pathogens in human gut, immune system booster effects, accelerating the growth of desirable microorganisms and strengthening the body’s defence mechanisms etc. (Meybodi et al., 2020).

Yogurt is one of the popular fermented dairy product which is fermented by *Lactobacillus bulgaricus* and *Streptococcus thermophilus* and it has a long history for health-promoting effects by means of being nutritionally rich in protein, calcium, riboflavin, vitamin B6, and vitamin B12 (Ashraf and Shah 2011). Yogurt bacteria could not survive through the gastric passage and colonise

* Corresponding author email: *talhademirci@selcuk.edu.tr*

within the gastrointestinal tract owing to acid and bile sensitivity, hence, the bacteria do not play a role for human gut health (Ashraf and Shah 2011; Meybodi et al., 2020). For a long time researchers and manufacturers have increasingly added probiotic microorganisms to improve the functional characteristics of yoghurt besides existing nutritional benefits. Indeed, probiotic yogurts regarded as one of the most popular functional foods worldwide, in a way that confirms probiotic yoghurt sales have grown at a Compound Annual Growth rate (CAGR) of 5.1 % between 2016 and 2020 according to market analysis http://futuremarketinsights.com. While it is predicted that bacteria that are very sensitive to oxygen and contain various technological barriers, defined as new generation probiotics such as *Akkermansia muciniphila* and *Faecalibacterium prausnitzii*, will dominate the probiotic marketplace in the coming years, it seems that *Lactobacillus* and *Bifidobacterium* species, which are traditional probiotics, will continue to be used in probiotic yoghurts for a long time.

On the other hand, according to recommendations, there should be minimum 10^6 CFU mL^{-1} of viable probiotic bacteria at the time of consumption to provide expected health benefits. However, many previous studies have revealed that probiotic bacteria are often below the recommended viability level in the products on the market (Meybodi et al., 2020; Shah 2000; Shah et al., 1995; Shima et al., 2012). Accordingly, it is important to observe that the viability and survival of the added probiotic microorganisms and interactions amongst probiotics and starter cultures in yogurt throughout the storage to ensure that it can provide the expected health-promoting benefits to the consumers. It is noteworthy that presence of both starter cultures and probiotic cultures in a same product matrix, can make it difficult to achieve a differential or selective colony count of probiotic bacteria (Van de Casteele et al., 2006). Nevertheless, various culture media have been developed for the selective enumeration and differentiation of concomitant probiotic and starter bacteria in yoghurt or other probiotic products (Ashraf and Shah 2011).

Based on all this information, the aim of this study was to determine survivability of starter and probiotic bacteria in local and global commercial probiotic yogurts and assess some previously suggested selective media.

2. Materials and Methods

2.1. Yogurt samples

Seven commercial probiotic yoghurt and two probiotic freeze-dried yogurt mixes were purchased from Turkish supermarkets. While five of these yoghurts were produced by local manufacturers, two of them were in the brands of global producers, and both freeze-dried probiotic culture mixes were products from local producers. Probiotic yogurt A, C, D, E, and I contained only bifidobacteria as a probiotic microorganism and apricot, date-chia, fig-oat, strawberry and apricot, respectively. Probiotic yogurt B possessed bifidobacteria and *Lactobacillus rhamnosus* without any ingredients whilst F freeze-dried and G liquid probiotic yogurt mixes included *L.acidophilus* as a probiotic microorganism. Overall, probiotic yogurt H contained only bifidobacteria without any ingredients. While these analyzes were performed 4 days before the expiration date of A, C and D yogurts, the analysis day of B yogurt was the last day of the expiration date. While the expiration date of E yogurt was 10 days, the expiration date of H and I yogurts were 20 and 22 days, respectively. As of the day of analysis, while the expiration date of F freeze-dried probiotic yogurt mix was 143 days, it was the last day of the expiry date of G liquid probiotic yogurt mix.

2.2. Selective and differential media

The media used for bifidobacteria enumeration was deMan Rogosa Sharpe (MRS) Agar supplemented with neomycin sulphate (100 mg/l), nalidixic acid (15 mg/l), paramomycin (200 mg/l), lithium chloride (3 g/l) (NNPL) according to Dave and Shah (1996). MRS Agar with Ox-bile (0.15 %, w/v) and Reinforced Clostridial Agar (RCA) supplemented with bromcresol green solution (0.2 %, w/v, autoclaved at 121 ºC for 15 min, 20 ml/l) and clindamycin (5 mg/100 ml, filter-sterilized, 2 ml/l) were chosen for selective and differential enumeration of *L. acidophilus* according to Darukaradhya et al., (2006). MRS 5.2 Agar (adjusted to 5.2 with filter-sterilized acetic acid) and RCA 5.3 Agar (adjusted to 5.3 with filter-sterilized acetic acid) were used for counting *L. delbrueckii* subsp. *bulgaricus* according to recommendation of Van de Casteele et al. (2006). M17 Agar and *Streptococcus thermophilus* (ST) Agar were used for enumeration of *S. thermophilus* according to recommendation of Ashraf and Shah (2011). The incubation conditions applied for each media are listed in Table 1.

2.3. Microbiological enumerations

Ten grams of each probiotic yogurt samples were suspended in 90 ml of sterile ringer solution and homogenized in a stomacher for 2 minutes. For freeze-dried probiotic yogurt mix, 1gr mix were suspended in 9 ml of sterile ringer solution. The homogenized suspension (1ml) was serially diluted in sterile 9 ml of ringer solution and 0.1 ml of the appropriate dilution was spread into the above-mentioned selective and differential media in triplicate. After incubation time, plates containing 25 to 250 colonies were enumerated and colony forming units (CFU) per gram of the yogurt samples was calculated. These numbers are described as viable counts ($\log_{10}$ CFU/g) (Talwalkar and Kailasapathy,2004).

Table 1
Selective and differential agar media used for probiotic and starter bacteria in yoghurt

Media	Incubation			Presumptive bacteria target	Reference
	Temp.	Time	Oxygen		
MRS-NNLP	45 ºC	72 h	anaerobic	Bifidobacteria	Dave and Shah (1996)
RCA-Clindamycin	37 ºC	72 h	microaerophilic	*L.acidophilus*	Darukaradhya et al. (2006)
MRS-Bile	37 ºC	72 h	microaerophilic	*L.acidophilus*	Darukaradhya et al. (2006)
MRS 5.2	45ºC	72 h	anaerobic	*L.delbrueckii* subsp. *bulgaricus*	Van de Casteele et al. (2006)
RCA 5.3	45ºC	72 h	anaerobic	*L.delbrueckii* subsp. *bulgaricus*	Van de Casteele et al. (2006)
M17	45ºC	24 h	aerobic	*S.thermophilus*	Ashraf and Shah (2011)
ST	37 ºC	24 h	aerobic	*S.thermophilus*	Ashraf and Shah (2011)

2.4. Statistical analysis

Statistical analysis of the data was performed by ANOVA and Tukey's mean comparison tests using the Minitab statistical package (version 18; Minitab Inc., State College, PA) to determine significant differences between the assessed responses.

3. Results and Discussion

3.1. Enumeration of Lactobacillus delbrueckii subsp. bulgaricus in commercial probiotic yogurt samples

In this current study, MRS Agar at pH 5.2 (MRS 5.2 Agar) and RCA Agar at pH 5.3 (RCA 5.3 Agar) were used for enumeration of *L. delbrueckii* subsp. *bulgaricus* when the incubation is carried out at 45ºC for 72 h under anaerobic incubation. These media were previously recommended by various researchers for *L. delbrueckii* subsp. *bulgaricus* (Ashraf & Shah, 2011; Lankaputhra & Shah, 1996; Van de Casteele et al., 2006). While Van de Casteele et al. (2006) stated that MRS 5.2 Agar was the most suitable media for enumeration of *L. delbrueckii* subsp. *bulgaricus*, RCA 5.3 Agar with anaerobic incubation at 45ºC for 72 h was found suitable media for selective recovery and enumeration by Dave and Shah (1996). On the other hand, some of the bifidobacteria strains also grew in these media, however, *L. delbrueckii* subsp. *bulgaricus* colonies were easily differentiated from those of bifidobacteria (Ashraf and Shah, 2011; Ashraf and Smith, 2015). Interestingly, in this present study 5 yoghurt samples (A, B, C, D, and E) did not reveal satisfactory recovery (< 5 log CFU/g) for *L. delbrueckii* subsp. *bulgaricus* colonies on MRS 5.2 Agar while *L. delbrueckii* subsp. *bulgaricus* colony counts on RCA 5.3 Agar below 5 log CFU/g for same tested 4 samples (A, C, D, and E).

This inadequate recovery of the *L. delbrueckii* subsp. *bulgaricus* in some yogurt samples may be attributed to declining in pH values towards the end of the storage period. Indeed, it was observed that the pH values of the samples with low numbers of *L. delbrueckii* subsp. *bulgaricus* were lower compared to the other yoghurt samples (data not shown). For example, pH values of A and E yoghurt samples were determined as 4.17 and 4.20, respectively, whilst mean pH values for B and H yoghurt samples were measured as 4.30 and 4.35, respectively, hence, it is estimated that this pH decrease has a negative impact on the number of *L. delbrueckii* subsp. *bulgaricus*. Similarly, in a previous study conducted by Mani-López et al. (2014), the researchers observed that the decrease in pH in probiotic yogurts during refrigeration storage caused a reduction in the viability of bacteria. In parallel with results obtained from this current work, Mani-López et al. (2014) observed the low recovery rates (about 5 log CFU/g) of *L. delbrueckii* subsp. *bulgaricus per contra S. thermophilus* recovery (about 7-8 log CFU/g) at the end of the 35 days storage in probiotic yogurt samples which separately produced with *L. casei* and *L. reuteri*. It is a fact that the post-acidification phenomenon in yoghurt increases with the addition of probiotic microorganisms and prebiotics, and the presence of *L. delbrueckii* subsp. *bulgaricus* is known to be one of the biggest causes of post-acidification in yoghurt, however, *L. delbrueckii* subsp. *bulgaricus* is expected to be more resistant to post acidification conditions than *S. thermophilus* (Deshwal et al. 2021). On the other hand, starter culture mixes with a high cocci/bacilli ratio or without *L. delbrueckii* subsp. *bulgaricus* are recommended so that post-acidification does not adversely affect the quality and consumer acceptability of yogurt taking into account that some of the basic flavor components of yogurt are lower (Deshwal et al., 2021; Pinto et al., 2009). In this present study, in probiotic yoghurt samples detected low recovery of *L. delbrueckii* subsp. *bulgaricus* (below 5 log CFU/g), this starter lactobacilli might be used in low amounts in the mix to prevent post acidification or the *L. delbrueckii* subsp. *bulgaricus* in these mixes may have been selected from strains that could not provide sufficient resistance in the face of decreasing pH. The same situation was previously revealed by Coeuret et al. (2004) who stated that some probiotic products included fewer lacatobacilli numbers than claimed or none, however, these researchers attributed this to disruption of the cold-chain or strain-dependent loss of viability. Again, the low recovery of *L. delbrueckii* subsp. *bulgaricus* in some probiotic yogurt samples can be explained by study of Temmerman et al. (2003) in which they stated that *L. delbrueckii* subsp. *bulgaricus* rapidly overgrown by the other bacteria in dairy products -especially in the presence of other lactobacilli- and its isolation becomes difficult. As for the comparison between the media, no statistical difference was observed between the numbers of *L. delbrueckii* subsp. *bulgaricus* obtained in MRS and RCA media. Previously, Van de Casteele et al. (2006) reported that higher recovery was obtained in MRS 5.2 medium compared to RCA medium in terms of *L. delbrueckii* subsp. *bulgaricus* LMG 6901 numbers when trying different media for enumeration of starter and probiotic bacteria

in probiotic dairy products. In addition, study of Ashraf and Smith (2015) demonstrated that a higher recovery rate of *L. delbrueckii* subsp. *bulgaricus* 11842 strain achieved in MRS 5.2 media (8.95±0.05) compared to RCA 6.1 and 6.8 media (8.56 and 8.26±0.06, respectively) when anaerobic incubations were carried out at 37 ºC for 72 h. It is noteworthy that the *L. delbrueckii* subsp. *bulgaricus* numbers obtained from both media for 5 yogurt samples (B, F, G, H, and I) varied from 6.19 log CFU/g to 8.82 log CFU/g and these counts are consistent with the literature (Lankaputhra and Shah, 1996; Van de Casteele et al., 2006).

Table 2
Viable counts of yogurt starter bacteria, *Bifidobacterium* spp. and *L.acidophilus* on different media (log CFU/g)

SAMPLE	M17	ST	RCA-Clindamycin	MRS-Bile	MRS 5.2	RCA 5.3	MRS-NNLP
A	9.97±0.22^{a}	10.07±0.08^{a}	-	-	<5	<5	<5
B	9.87±0.05^{ab}	10.19±0.65^{a}	-	-	7.88±0.01^{a}	8.05±0.06^{a}	7.85±0.07
C	9.54±0.10^{bcd}	9.45±0.05^{a}	-	-	<5	<5	<5
D	9.72±0.04^{abc}	9.73±0.02^{a}	-	-	<5	<5	<5
E	9.82±0.01^{ab}	9.81±0.13^{a}	-	-	<5	<5	<5
F	9.16±0.04^{e}	9.29±0.17^{a}	8.12±0.02^{b}	8.21±0.07^{b}	7.84±0.10^{a}	7.88±0.48^{a}	-
G	9.48±0.03^{cde}	9.44±0.18^{a}	10.08±0.02^{a}	9.40±0.35^{a}	8.82±0.12^{a}	8.44±0.52^{a}	-
H	9.27±0.01^{de}	9.34±0.00^{a}	-	-	8.22±0.11^{a}	7.60±0.17^{a}	<5
I	9.42±0.03^{cde}	9.83±0.02^{a}	-	-	6.19±0.34^{b}	6.41±0.21^{b}	<5

-: yogurt did not claim to possess those probiotic strains
^{a,b,c,d,e} Values in same column having different superscripts differ significantly ($p < 0.05$), means in media used for same targeted bacteria do not differ significantly

3.2. Enumeration of Streptococcus thermophilus in commercial probiotic yogurt samples

In this current study, M17 Agar and ST Agar were used for enumeration of *S. thermophilus* when the incubation is carried out at 45 ºC and 37 ºC for 72 h under aerobic incubation, respectively. M17 media were previously recommended by International Dairy Federation (IDF, 1981) for selective enumeration of *S. thermophilus* from yogurt and also various researchers reported that M17 medium was the most suitable media for enumeration and isolation of lactic streptococci (Ashraf and Shah, 2011; Van de Casteele et al., 2006). On the other hand, ST Agar at 37 ºC and aerobic conditions were recommended by Tharmaraj and Shah (2003) for *S. thermophilus* amongst the media tested. In this present work, the recovery rates over 9 log CFU/g were obtained in the enumerations made for all yogurt samples on both ST and M17 media. The problem of insufficient recovery rates that occurred for *L. delbrueckii* subsp. *bulgaricus* in some yogurt samples was not valid for *S. thermophilus.* These numbers from the present study were very similar to results reported by Mani-López et al. (2014) who reported that *S. thermophilus* counts from the commercial probiotic yogurts stored for 35 days varied from 9.48 log CFU/g to 10.34 log CFU/g. Moreover, in consistent with our results these researchers also revealed that *S. thermophilus* numbers in probiotic yogurts produced individually with *L. casei*, *L. reuteri* and *L. acidophilus* remained higher than *L. delbrueckii* subsp. *bulgaricus* and probiotic bacteria at the end of the storage. Similar results were obtained in the study of Gueimonde et al. (2004) where *S. thermophilus* counts from 10^{7} to 10^{9} CFU/ml obtained after 30 d of cold storage. In fact, in a previous study Mani-López et al. (2014) four culture mixtures were prepared to produce yogurts as follows: (1) *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus*, (2) *S. thermophilus, L. delbrueckii* subsp. *bulgaricus* and probiotic *L. acidophilus*, (3) *S. thermophilus, L. delbrueckii* subsp. *bulgaricus* and probiotic *L. casei*, (4) *S. thermophilus, L. delbrueckii* subsp. *bulgaricus* and probiotic *L. reuteri.* The researchers examining the viability of these bacteria during storage determined that when there was a probiotic bacteria in the mix, the numbers of *S. thermophilus* remained very high at the end of storage, but the viability of *L. delbrueckii* subsp. *bulgaricus* was affected very negatively, however, the number of *L. delbrueckii* subsp. *bulgaricus* in yoghurts prepared without probiotic bacteria was around 10 log CFU/ml at the end of the storage period and it was higher than *S. thermophilus.* Hence, the result of the aforementioned study supports the findings obtained from this present study and it is seen that *S. thermophilus* is not suppressed in the presence of probiotic bacteria, but *L. delbrueckii* subsp. *bulgaricus* is negatively affected. Again, in consistent with this current findings Rutella et al. (2016) reported that whilst *L. delbrueckii* subsp. *bulgaricus* declined from 8.52±0.16 to 6.50 log CFU/ml at the end of the 27 d. cold storage, there were minimal differences over storage time in the viability of *S. thermophilus* numbers. This situation has been attributed by the various researchers to the fact that *S. thermophilus* is more resistant to cold stress than *L. delbrueckii* subsp. *bulgaricus* (Donkor et al., 2007; Rutella et al., 2016).

3.3. Enumeration of Bifidobacterium spp. and L. acidophilus in commercial probiotic yogurt samples

In this current study, MRS-Bile Agar and RCA-Clindamycin Agar were used for enumeration of *L. acidophilus* when the incubation is carried out at 37ºC for 72 h under aerobic incubation. Besides, MRS-NNLP media was used for enumeration of *Bifidobacterium* spp. with anaerobic incubation at 45ºC for 72 h. MRS-Bile Agar were previously recommended by de Carvalho Lima et al. (2009) for *L. acidophilus,* while RCA-Clindamycin Agar was recommended by International Organization for Standardization (Organisation, 2006) for the enumeration of presumptive *L. acidophilus* in dairy products. MRS-NNLP Agar was found as suitable media for selective enumeration of bifidobacteria from probiotic yogurts (Ashraf and Shah, 2011; Van de Casteele et al., 2006). Actually, although there are studies suggesting the addition of 0.05% L-cysteine to this medium, de Carvalho Lima et al. (2009) could not detect a difference in performance between MRS-NNLP media containing cysteine and those without. Therefore, in this

study, MRS-NNLP media was used without the addition of cysteine. This work indicated that high amounts of *L. acidophilus* were detected on both media in both of the two yoghurt samples declared as *L. acidophilus* on the label (F and G). On the other hand, bifidobacteria was determined above 5 log CFU/g in only 1 yoghurt sample (B) out of 7 probiotic yoghurts claimed to be *Bifidobacterium* spp. In consistent with our results, Mani-López et al. (2014) observed that the numbers of *L. acidophilus* decreased 1-1.5 log during storage but remained at the level recommended by FAO/WHO in yogurt and fermented milk samples that contained *L. acidophilus*. Although the number of bifidobacteria was found to be above the minimum recommended number in this present study, Coeuret et al. (2004) stated that some commercial probiotic products contained fewer lactobacilli than claimed, or none, due to the disruption of the cold chain or strain-dependent loss of viability. It is not a very surprising finding that bifidobacteria were found to be above 5 log CFU/g in only 1 sample tested, because when the literature is examined, it has been seen that there are many similar results. Accordingly, inhibited probiotic contents (*Bifidobacterium lactis*, *L. rhamnosus* etc.) of the end product were detected when co-cultured with the fast-acidifying strain *S.thermophilus* (Oliveira et al. 2009). Indeed, many researchers have reached results reporting that the number of bifidobacteria in probiotic dairy products or supplements is much lower than it should be, or that it cannot be detected at all (De Vecchi et al., 2008; Lewis et al., 2016; Temmerman et al., 2003). In this regard, Marinova et al. (2019) reported that viable bacteria were not detected in 11.53 % of the tested probiotic dietary supplements in Bulgarian market and 7.69 % contained a minimal amount (about 10^2 CFU/g) while Zawistowska-Rojek et al. (2016) observed that only one medicinal product, two dietary supplement and two foods of all analyzed 25 different products revealed good quality in regard to number of probiotic cells. Previously, it is highlighted that the usage of some fruit juice or their pulps may negatively influence the viability of probiotic bacteria in yoghurt probably due to the high acidity or the antibacterial components (Meybodi et al., 2020; Shori, 2015). In this current study, the fact that 6 out of the 7 probiotic yogurt samples in which the bifidobacteria count was below 5 log CFU/g were fruit-supplemented, actually shows that one of the reasons for this result may be the effect of fruit additives. On the other hand, Van de Casteele et al. (2006) stated that the plating technique used whilst counting bifidobacteria from dairy products also affected the results, and the pour-plate method was more appropriate since these bacteria were oxygen sensitive. In fact, researchers who have encountered similar findings about absence or low recovery of bifidobacteria in probiotic commercial products have previously expressed many ideas about possible reasons for this: it might be due to (1) the lack of optimal selective/elective/differential enumeration media and MRS-NNLP agar not being a suitable medium for reliably counting this bacterial group (Talwalkar and Kailasapathy, 2004; Temmerman et al., 2003), the lack of differentiation and recovery amongst the strains of different LAB species and (2) possible antagonism between all strains used in producing the probiotic product (Oberg et al. 2011), (3) drawbacks of plating methods, which is still the most used approach, such as labor intensive, revealing variable results and not determining the viable but not culturable (VBNC) bifidobacteria cells (Di Lena et al., 2015; Fusco et al., 2021; Huys et al., 2013), (4) the dilution of the sample (the problem that these probiotic bacteria start to disappear as the dilutions increase) (Talwalkar and Kailasapathy 2004), (5) inability of some probiotic cells to form colony on solid medium through bacterial stress (Talwalkar and Kailasapathy 2004), (6) negatively influence of freeze-drying and encapsulation processes on probiotic bacteria recovery and/or freeze-dried probiotic products (Masco et al. 2005) (7) decrease of the bifidobacteria cells's survival rate over inappropriate transportation and storage conditions (Di Lena et al. 2015), and (8) voluntary or involuntary mislabeling of the bifidobacteria counts (Fusco et al. 2021). As a result, on the occasion of this study, it is worth remembering that whatever the reason - from the producer, from the culture supplier or from the impropriety of the analysis- in probiotic products the number of probiotic bacteria should be constantly monitored at the beginning of production and during storage in order to give the desired health benefits. Whatever the factors are, these should be tried to be corrected and new generation technologies such as molecular and omics approaches, flow cytometry etc. should be rapidly integrated into the applications for probiotic enumeration and monitoring in order to get rid of the disadvantages of the plate counting methods in solid media. It should also be noted that recently there have been studies with the concepts of 'postbiotic' and 'paraprobiotic' that these microorganisms are beneficial even if they are not alive and that VBNC probiotic cells also have health benefits (Aguilar-Toalá et al., 2018; Fiore et al., 2020; Fusco et al., 2021; Taverniti and Guglielmetti 2011), however, this does not change the minimum number that should be in a product manufactured with the claim of being a probiotic food or supplement. Hence, all these concepts should be evaluated differently.

4. References

Aguilar-Toalá J, Garcia-Varela R, Garcia H, Mata-Haro V, González-Córdova A, Vallejo-Cordoba, B, Hernández-Mendoza A (2018). Postbiotics: An evolving term within the functional foods field. *Trends in Food Science & Technology 75*: 105-114.

Ashraf R, Shah NP (2011). Selective and differential enumerations of Lactobacillus delbrueckii subsp. bulgaricus, Streptococcus thermophilus, Lactobacillus acidophilus, Lactobacillus casei and Bifidobacterium spp. in yoghurt—A review. *International Journal of Food Microbiology 149*: 194-208.

Ashraf R, Smith SC (2015). Selective enumeration of dairy based strains of probiotic and lactic acid

bacteria. *International Food Research Journal 22*: 2576-2586.

Coeuret V, Gueguen M, Vernoux JP (2004). Numbers and strains of lactobacilli in some probiotic products. *International Journal of Food Microbiology 97*: 147-156.

Darukaradhya J, Phillips M, Kailasapathy K (2006). Selective enumeration of Lactobacillus acidophilus, Bifidobacterium spp., starter lactic acid bacteria and non-starter lactic acid bacteria from Cheddar cheese. *International Dairy Journal 16*: 439-445.

Dave R, Shah N (1996). Evaluation of media for selective enumeration of Streptococcus thermophilus, Lactobacillus delbrueckii ssp. bulgaricus, Lactobacillus acidophilus, and bifidobacteria. *Journal of Dairy Science 79*: 1529-1536.

De Carvalho Lima KG, Kruger MF, Behrens J, Destro MT, Landgraf M, de Melo Franco BDG (2009). Evaluation of culture media for enumeration of Lactobacillus acidophilus, Lactobacillus casei and Bifidobacterium animalis in the presence of Lactobacillus delbrueckii subsp bulgaricus and Streptococcus thermophilus. *LWT-Food Science and Technology 42*: 491-495.

De Vecchi E, Nicola L, Zanini S, Drago L (2008). In vitro screening of probiotic characteristics of some Italian products. *Journal of Chemotherapy 20*: 341-347.

Deshwal GK, Tiwari S, Kumar A, Raman RK, Kadyan S (2021). Review on factors affecting and control of post-acidification in yoghurt and related products. *Trends in Food Science & Technology 109*: 499-512.

Di Lena M, Quero GM, Santovito E, Verran J, De Angelis M, Fusco V (2015). A selective medium for isolation and accurate enumeration of Lactobacillus casei-group members in probiotic milks and dairy products. *International Dairy Journal 47*: 27-36.

Donkor O, Henriksson A, Singh T, Vasiljevic T, Shah NP (2007) ACE-inhibitory activity of probiotic yoghurt. *International Dairy Journal 17*: 1321-1331.

FDA (2022). http://accessdata.fda.gov (date accessed: 26.09.2022)

FMI (2022). http://futuremarketinsights.com (date accessed: 26.09.2022)

Fiore W, Arioli S, Guglielmetti S (2020). The neglected microbial components of commercial probiotic formulations. *Microorganisms 8*: 1177.

Fusco V, Fanelli F, de Souza EL (2021). Authenticity of Probiotic Foods and Dietary Supplements. *Frontiers in Microbiology 12*.

Gueimonde M, Delgado S, Mayo B, Ruas-Madiedo P, Margolles A, De Los Reyes-Gavilán CG (2004). Viability and diversity of probiotic Lactobacillus and Bifidobacterium populations included in commercial fermented milks. *Food Research International 37*: 839-850.

Hill C, Guarner F, Reid G, Gibson GR, Merenstein DJ, Pot B, Morelli L, Canani RB, Flint HJ, Salminen S, Calder PC Sanders ME (2014). The International Scientific Association for Probiotics and Prebiotics consensus statement on the scope and appropriate use of the term probiotic. *Nature reviews Gastroenterology & Hepatology 11*: 506-514.

Huys G, Botteldoorn N, Delvigne F, De Vuyst L, Heyndrickx M, Pot B, Dubois JJ Daube G (2013). Microbial characterization of probiotics–Advisory report of the W orking G roup "8651 Probiotics" of the B elgian S uperior H ealth C ouncil (SHC). *Molecular Nutrition & Food Research 57*: 1479-1504.

IDF (1981). International Dairy Federation. *Isolation and identification of mastitis bacteria. IDF document 132*: 19-27.

Lankaputhra W, Shah N (1996). A simple method for selective enumeration of Lactobacillus acidophilus in yogurt supplemented with L. acidophilus and Bifidobacterium spp. *Milchwissenschaft*.

Lewis ZT, Shani G, Masarweh CF, Popovic M, Frese SA, Sela DA, Underwood MA, Mills DA (2016). Validating bifidobacterial species and subspecies identity in commercial probiotic products. *Pediatric Research 79*: 445-452.

Mani-López E, Palou E, López-Malo A (2014). Probiotic viability and storage stability of yogurts and fermented milks prepared with several mixtures of lactic acid bacteria. *Journal of Dairy Science 97*: 2578-2590.

Marinova VY, Rasheva IK, Kizheva YK, Dermenzhieva YD, Hristova PK (2019). Microbiological quality of probiotic dietary supplements. *Biotechnology & Biotechnological Equipment 33*: 834-841.

Masco L, Huys G, De Brandt E, Temmerman R, Swings J (2005). Culture-dependent and culture-independent qualitative analysis of probiotic products claimed to contain bifidobacteria. *International Journal of Food Microbiology 102*: 221-230.

Meybodi NM, Mortazavian AM, Arab M, Nematollahi A (2020). Probiotic viability in yoghurt: A review of influential factors. *International Dairy Journal 109*: 104793.

O'Toole PW, Marchesi JR, Hill C (2017). Next-generation probiotics: the spectrum from probiotics to live biotherapeutics. *Nature Microbiology 2*: 1-6.

Oberg C, Moyes L, Domek M, Brothersen C, McMahon DJ (2011). Survival of probiotic adjunct cultures in cheese and challenges in their enumeration using selective media. *Journal of Dairy Science 94*: 2220-2230.

Oliveira RPDS, Perego P, Converti A, De Oliveira MN (2009). Effect of inulin on growth and acidification performance of different probiotic bacteria in co-cultures and mixed culture with Streptococcus

thermophilus. *Journal of Food Engineering 91*: 133-139.

ISO (2006). Milk Products–Enumeration of Presumptive Lactobacillus acidophilus on a Selective Medium–Colony-Count Technique at 37 Degrees C. ISO 20128: 2006 (IDF 192: 2006). In: International Standards Organisation Geneva, Switzerland.

Pinto SM, Clemente MDG, De Abreu LR (2009). Behaviour of volatile compounds during the shelf life of yoghurt. *International Journal of Dairy Technology 62*: 215-223.

Rutella GS, Tagliazucchi D, Solieri L (2016). Survival and bioactivities of selected probiotic lactobacilli in yogurt fermentation and cold storage: New insights for developing a bi-functional dairy food. *Food Microbiology 60*: 54-61.

Shah NP (2000). Effects of milk-derived bioactives: an overview. *British Journal of Nutrition, 84*(S1), 3-10.

Shah NP, Lankaputhra WE, Britz ML, Kyle WS (1995). Survival of Lactobacillus acidophilus and Bifidobacterium bifidum in commercial yoghurt during refrigerated storage. *International Dairy Journal 5*: 515-521.

Shima AR, Salina HF, Masniza M, Atiqah AH (2012). Viability of lactic acid bacteria in home made yogurt containing sago starch oligosaccharides. *International Journal of Basic Applied Sciences 12*: 58-62.

Shori AB (2015). The potential applications of probiotics on dairy and non-dairy foods focusing on viability during storage. *Biocatalysis and Agricultural Biotechnology 4*: 423-431.

Talwalkar A, Kailasapathy K (2004). A review of oxygen toxicity in probiotic yogurts: influence on the survival of probiotic bacteria and protective techniques. *Comprehensive Reviews in Food Science and Food Safety, 3*(3): 117-124.

Taverniti V, Guglielmetti S (2011). The immunomodulatory properties of probiotic microorganisms beyond their viability (ghost probiotics: proposal of paraprobiotic concept). *Genes & Nutrition 6*: 261-274.

Temmerman R, Pot B, Huys G, Swings J (2003). Identification and antibiotic susceptibility of bacterial isolates from probiotic products. *International Journal of Food Microbiology 81*: 1-10.

Tharmaraj N, Shah N (2003). Selective enumeration of Lactobacillus delbrueckii ssp. bulgaricus, Streptococcus thermophilus, Lactobacillus acidophilus, bifidobacteria, Lactobacillus casei, Lactobacillus rhamnosus, and propionibacteria. *Journal of Dairy Science 86*: 2288-2296.

Van de Casteele S, Vanheuverzwijn T, Ruyssen T, Van Assche P, Swings J, Huys G (2006). Evaluation of culture media for selective enumeration of probiotic strains of lactobacilli and bifidobacteria in combination with yoghurt or cheese starters. *International Dairy Journal 16*: 1470-1476.

Zawistowska-Rojek A, Zaręba T, Mrowka A, Tyski S (2016). Assessment of the microbiological status of probiotic products. *Polish Journal of Microbiology 65*: 7.

Selcuk Journal of Agriculture and Food Sciences SJAFS
http://sjafs.selcuk.edu.tr/sjafs/index
(2022) 36 (3), 428-437
e-ISSN: 2458-8377
DOI:10.15316/SJAFS.2022.057
Research Article

Effect of Temperature and Storage Time on Germination in Forage Peas**

Cafer TÜRKER[1,*], Ahmet TAMKOÇ[2]
[1]Selçuk University, Faculty of Agriculture, Department of Field Crops, Konya, Türkiye
[2]Ilgaz Directorate of Provincial Agriculture and Forestry, Çankırı, Türkiye

ARTICLE INFO

Article history:
Received date: 01.11.2022
Accepted date: 11.11.2022

Keywords:
Field pea
Germination
Root length
Temperature
Thousand seed weight

ABSTRACT

This research was carried out in 2016 in order to determine the most suitable germination temperatures of the seeds of 4 different forage pea varieties (Furkan, Bilgehan, Özkaynak, Taşkent) stored in paper bags for 7 years and 8 years at room temperature, and the damage that may occur on plants at subzero temperatures after germination were carried out. In the experiment, which was established with 4 replications in the "Random Plots in Factorial Experiment Design" under laboratory conditions; Number of germinated seeds on the 4th, 7th and 10th days at 5°C, 15°C, 25°C and 35°C temperatures, and after the 10th day, frost damage in plants kept at -5°C for 3 days, root length, stem length, root length/stem length ratio was determined. According to the findings of the research, while the best germination took place at 25°C, the most suitable germination temperature was determined between 15°C and 25°C. In addition, it was revealed that the seeds produced in 2008 germinated more than the seeds produced in 2016. On the other hand, among the plants kept at -5°C, it was determined that the varieties named Taşkent and Özkaynak, which were germinated at 15°C and 25°C, suffered the least damage, and the highest average in the comparison of the total height values was found in the Taşkent variety germinated at 25°C.

1. Introduction

With the rapid increase in the world population, animal and vegetable products, which are of great importance in meeting the food needs of people, are of great value for our country's economy. Proteins, which have a very important place in nutrition, should be taken as approximately 1 gram per kilogram of human live weight. About half of this protein can be consumed as vegetable and the other half as animal. At this point, it is known that there are production problems in the consumption of animal protein and consumption problems arise along with it. The basis of these problems is animal production and we can list many sources of this situation.

In Turkey, agricultural areas planted every year are approximately 15.5 million hectares. Forage crops can be produced only in 2.7 million hectares of this. If forage crops are produced and applied to the rotation system every four years, it will be possible to increase the amount of forage crops planted area to approximately 4 million hectares. It may be possible to increase the yield of pastures by 25-50% on average with forage crops breeding and pasture improvement and method. In particular, abandoned agricultural areas are approximately 4 million hectares and should be reintroduced to agriculture through artificial pastures. In addition, artificial meadows and pastures should be encouraged and supported by the Ministry. Forage crops should be encouraged as a secondary crop in irrigated farming areas. Production supports should be given to feed crop producers and livestock enterprises that make contracts with feed producers. Approximately 8 million hectares of forest pastures and bushes, which are considered forest areas, should be transformed into goat pastures that make good use of the bushes. It is a fact that if the increase in our sheep and goat population continues at this rate and our pastures are not rehabilitated, we will face very heavy erosion damage (Tamkoç, 2017).

With this study, it is aimed to prevent our farmers from harming with the data that can minimize the problems experienced in the cultivation of forage crops. In the study, errors in agricultural practices can be minimized by determining the appropriate planting times and temperatures, and the appropriate stem length that can

*Corresponding author email: *caferturker0606@hotmail.com*
**This article is summarized from Cafer TÜRKER's master's thesis ("YÖK" thesis number: 577444).

be caused with the least damage from winter in the results obtained with the seeds of winter forage peas (field pea for feding animals) germinated at different temperatures. In addition, it is aimed to prevent seed waste by evaluating the usability of old seeds by considering the germination status of old and new seeds.

2. Materials and Methods

In this study, Bilgehan, Furkan, Özkaynak and Taşkent registered varieties of forage pea (*Pisum arvense* L.) obtained fromSelcuk University Faculty of Agriculture, Field Crops Department were used as plant material. Furkan, Özkaynak, and Taşkent forage pea seeds are a product of 2008 (old) and 2016 (new), and Bilgehan variety is a product of 2009 (old) and 2015 (new). These seeds were kept in paper bags and at room conditions. Agricultural perlite was used as germination medium. The density of the perlite used is 70-80 kg m3, the grain diameter is 0-6 mm, the melting point is 1200°C, the chemical composition is SiO_2 74%, Al_2O_3 14%, Na_2O 3%, K_2O 5%, MgO 0.5%, CaO 0.05%, Fe_2O_3 1% and pH value between 6.5-7.5. 5%. Sodium hypochlorite (bleach, chlorine-based bleach) was used as disinfectant and commercial packaged water with pH 7.57 was used as water.

This research was carried out in the Climate Rooms of the Department of Field Crops, Faculty of Agriculture, Selcuk University in 2016.

Seeds of forage pea varieties used as material in the study were divided into two groups as old and new seeds. Each replication of old and new seeds consisted of 25 seeds and was arranged as 4 replications in randomized plots design (Düzgüneş et al., 1987). Forage pea seeds were chosen randomly from healthy seeds. Then, the seeds that would form each replication were kept in 5% sodium hypochlorite solution for 5 minutes and rinsed with commercially packaged water. 100 ml of agricultural perlite and 80 ml of commercially packaged water were added into half-liter glass jars with lids to be planted. The disinfected seeds were sown evenly on the growing medium filled with agricultural perlite and water. 2 drops of 5% sodium hypochlorite were added to each liter of commercially packaged water used to saturate agricultural perlite. The lids of the cultivated jars were closed and placed in the germination cabinet.

Within the scope of the study, the germination test at four different temperatures, 5, 15, 25 and 35°C, and then the frost damage test was applied by applying -5°C and 0°C temperatures in different rows and times. The germination of the seeds left to germinate at 5°C, 15°C, 25°C and 35°C temperatures on the 4th, 7th and 10th days were counted. In germination studies, using materials such as paper and sand, they stated that germinated seeds of Hungarian vetch on the 5th and 10th days at 20°C, and on the 5th and 14th days of vetch and common vetch could be counted (Elçi and Açıkgöz, 1993). Those whose radicula (rootlet) and plumule (stemlet) have come off are considered as germinated (Çuhadar, 1997).

The following processes are applied to the seeds grown at each temperature:

Germination at 5°C: The seeds germinated on the 4th, 7th and 10th days were counted. Seedlings with frost damage after 10 days of storage at 0°C for 2 days, at -5°C for 3 days, at 0°C for 2 days, at 5°C for 3 days and at 15°C for 3 days.

Germination at 15°C: The seeds germinated on the 4th, 7th and 10th days were counted. After day 10 at 5°C for 2 days, at 0°C for 2 days, at -5°C for 3 days, at 0°C for 2 days, at 5°C for 3 days and at 15°C After waiting for 3 days, the seedlings with frost damage were counted.

Germination at 25°C: The seeds germinated on the 4th, 7th and 10th days were counted. After day 10 at 15°C for 2 days, at 5°C for 2 days, at 0°C for 2 days, at -5°C for 3 days, at 0°C for 2 days, at 5°C Seedlings with frost damage were counted after 3 days and 3 days at 15°C.

Germination at 35°C: The seeds germinated on the 4th, 7th and 10th days were counted. After day 10 at 25°C for 2 days, at 15°C for 2 days, at 5°C for 2 days, at 0°C for 2 days, at -5°C for 3 days, at 0°C Seedlings with frost damage were counted after being kept for 2 days, 3 days at 5°C and 3 days at 15°C.

In order to determine the reactions of the plants, which will be formed by keeping them at -5 ° C for 3 days, after the last 2 days at 15 ° C, the plants were taken out of the jars and the damage was evaluated. Root and stem lengths of the plants were also measured.

Frost damage was evaluated with a scale of 1, 3, 5, 7, 9. Accordingly, the scale values are as follows:

1- It is unharmed and has new shoots

3- Leaf tips damaged and mortality less than 5%

5- Damage greater than 25%, death of leaves, branches or roots

7- 50% dead

9- more than 90% dead

Data from observations and measurements were analyzed using the MSTAT-C statistical package program(Anonymous, 1982).

Tthis article is summarized from part of Cafer TURKER's thesis ("YÖK" thesis number: 577444).

3. Results and Discussion

The data of the observations and measurements taken as a result of the effects of different temperatures on the germination of forage pea varieties and the statistical analysis results of these data are given below under the headings.

Number of seeds germinated on the fourth day

It was understood from the examination of Table 1 that the interaction of variety x seed production x temperature was effective on germination at the 4th day count ($P<0.01$). In other words, when the number of seeds germinated on the 4th day was examined in terms

of variety, seed production year and temperature (triple interaction), the changes were not always in the same direction. While the lowest germination in Bilgehan variety was in old seeds at 35°C, it was in new seeds at 25°C in Furkan variety (Table 2). It has been observed that germination values increase relatively from 0°C to 25°C, and decrease at 35°C. According to the 4th day counts, germination did not occur at 5°C. A similar study on some leguminous species revealed a triple interaction (Çuhadar, 1997). No germination occurred at 0°C in the 4th day count. Leaving the plants at temperatures lower than the appropriate germination temperatures slows the growth of the plants (Açıkgöz, 1991).

Table 1
Variance analysis table of 4th day germination in forage peas

Sources of Variance	DF	SS	MS	F
Variety	3	207.648	69.216	10.7826**
Year	1	15.820	15.820	2.4645
Variety x Year	3	280.148	93.383	14.5473**
Temperature	3	8762.529	2920.843	455.0116**
Variety x Temperature	9	411.133	45.681	7.1163**
Year x Temperature	3	47.523	15.841	2.4677
Variety x Year x Temperature	9	167.133	18.570	2.8929**
Error	96	616.250	6.419	

**: P<0.01

Table 2
Varieties x seed production year x temperature interaction of forage pea seeds germination on the 4th day (number)

Variety	Year	Temperature							
		35°C		25°C		15°C		5°C	
Furkan	2008	21.5	ABCD	23.8	AB	19.5	BCDE	0.0	J
	2016	15.5	EFGH	16.0	EFGH	12.0	HI	0.0	J
Bilgehan	2009	8.8	I	22.8	ABC	11.3	HI	0.0	J
	2015	14.5	FGH	23.3	ABC	14.5	FGH	0.0	J
Özkaynak	2008	21.3	ABCD	24.0	AB	19.0	CDEF	0.0	J
	2016	20.8	ABCD	22.0	ABC	16.8	DEFG	0.0	J
Taşkent	2008	13.5	GHI	24.8	A	13.0	GHI	0.0	J
	2016	14.0	GH	22.8	ABC	19.8	BCDE	0.0	J

LSD (P<0.01): 4.708; CV (%): 18.65

In the study of Çaçan et al. (2016), mentioned that a temperature of 10°C to 24°C is suitable for adequate germination in 13 forage pea genotypes (lines and varieties). According to the results of this research and the sources, the germination rate of forage pea seeds varies depending on the germination factors.

Number of seeds germinated on the seventh day

In the germination counts made on the seventh day, the effect of the years of production of the seeds was effective on the germination at a significance level of P<0.05 (Table 3). In Table 4, the averages of the cultivar x temperature interaction, which were found to be important as a result of the analysis of variance, are given. As a result of the averages applied to the LSD test, the best germination was obtained from the old seeds with 67.2% in the germination count on the 7th day. This rate was 62.8% in new seeds. According to the results, even if the old seed is old, the germination results of these seeds, which are under the same storage conditions in forage pea germination, are good and this shows that the seeds harvested in the past years can be used economically.

Variety x temperature interaction was effective on the 7th day counts of germination. Varieties differed in response to germination temperatures. In other words, the responses of the cultivars to temperature were not in the same direction. However, it was observed that germination increased as the temperature increased from 5°C to 25°C, whereas germination was observed at a low rate at 35°C (Table 4). At 5°C, none of the cultivars germinated. In a similar study on vetch species, it was stated that the increase in temperature after a certain point causes a decrease in germination (Qin, 1992). It has been reported that there is an optimum temperature range for germination and growth in plants, an increase in temperatures will show a positive reaction in plants to a certain extent, and it may be harmful to plants when the upper limit of the optimum temperature is exceeded (Açıkgöz, 1991).

When it is planned to be cultivated, forage pea varieties to be planted in autumn, the decrease in temperatures in general causes the germination rates to decrease, revealing the possibility that the plant will enter the winter without germination.

Table 3
Variance analysis table of 7th day germination in forage peas

Sources of Variance	DF	SS	MS	F
Variety	3	132.937	44.312	7.4697**
Year	1	34.031	34.031	5.7366*
Variety x Year	3	42.531	14.177	2.3898
Temperature	3	11428.400	3809.467	642.1577**
Variety x Temperature	9	192.750	21.417	3.6102**
Year x Temperature	3	22.656	7.552	1.2730
Variety x Year x Temperature	9	34.031	3.781	0.6374
Error	96	569.500	5.932	

*: P<0.05; **: P<0.01

Table 4
Variety x temperature interaction of 7th day germination of forage pea seeds

Variety	Temperature							
	35°C		25°C		15°C		5°C	
Furkan	21.1	BCD	21.3	ABCD	20.3	CD	0.0	F
Bilgehan	16.0	E	24.3	AB	19.1	DE	0.0	F
Özkaynak	21.9	ABCD	24.3	AB	23.4	ABC	0.0	F
Taşkent	20.9	CD	24.4	A	22.8	ABC	0.0	F

LSD (P<0.01): 3.200; CV (%): 15.02

Number of seeds germinated on the tenth day

At the 10th day count on germination, the variety x year interaction was effective at the significance level of P<0.05 (Table 5). Table 6-7 shows the averages of the interactions that were found to be important as a result of the analysis of variance. In other words, the response of the cultivars differed with the old age of the seeds. While old seeds of Furkan variety gave high results on the 10th day germination count, new seeds of Furkan variety gave low results and this difference between them was statistically significant (Table 6).

As a result of the averages obtained as a result of the LSD test, while the old seeds of Özkaynak variety achieved high germination results in the 10th day germination, the new seeds of Bilgehan variety gave a lower germination average and the difference between them shows statistical significance. On the other hand, the differences between the germination of old and new seeds of Özkaynak and Taşkent cultivars and the germination of old seeds of Furkan cultivar were not found significant (Table 6).

Table 5
Variance analysis table of 10th day germination in forage peas

Sources of Variance	DF	SS	MS	F
Variety	3	98.531	32.844	5.6229**
Year	1	20.324	20.324	3.4794
Variety x Year	3	50.269	16.756	2.8687*
Temperature	3	11139.753	3713.251	635.7144**
Variety x Temperature	9	262.421	29.158	4.9919**
Year x Temperature	3	32.830	10.943	1.8735
Variety x Year x Temperature	9	23.445	2.605	0.4460
Error	96	560.743	5.841	

*: P<0.05; **: P<0.01

Table 6
Variety x seed production year interaction of 10th day germination of forage pea seeds

Variety	Year	
Furkan	2008	17.2 AB
	2016	14.6 C
Bilgehan	2009	15.2 C
	2015	16.1 BC
Özkaynak	2008	18.2 A
	2016	17.1 AB
Taşkent	2008	17.6 AB
	2016	17.1 AB

LSD (P<0.05): 1.696; CV (%): 14.52

Table 7
Variety x temperature interaction of 10th day germination of forage pea seeds (number)

Variety	Temperature							
	35°C		25°C		15°C		5°C	
Furkan	21.1	CD	21.4	BCD	21.1	CD	0.0	F
Bilgehan	16.1	E	24.3	AB	19.9	D	2.4	F
Özkaynak	22.3	ABCD	24.4	AB	23.9	ABC	0.1	F
Taşkent	21.5	ABCD	24.6	A	23.4	ABC	0.0	F

LSD (P<0.01): 3.176; CV (%): 14.52

In the analysis of variance, the difference between cultivars and germination temperatures on the 10th day germination was effective at P<0.01 significance level (Table 7). In other words, the reactions of the varieties to the temperatures were in different directions.

In a study, the importance of temperature in germination for field agriculture was mentioned and appropriate temperature ranges for seed germination were mentioned even if there is sufficient moisture in the soil (Ashby and Hellmers, 1955).

When the 10th day count variety x temperature interaction was examined in the LSD test, the relatively best germination average was determined at 25°C. Although the best results were obtained at 25°C, the difference between the number of seeds germinated at 25°C on the 10th day, and the number of seeds germinated on the 10th day of the Özkaynak cultivar and Bilgehan cultivar at 25°C were statistically insignificant (Table 7) . The difference between the number of germination of Taşkent variety at 25°C and the 10th day counts of Furkan variety germinated at 25°C was statistically significant.

As a result of the study, we can say that among the applied germination temperatures, the best germination temperature of forage peas is between 15°C and 25°C. It was observed that at 35°C, some of the seeds deteriorated without germination and at this high temperature, the seeds lost their seed properties and melted. It has been stated that at high temperatures, the enzyme and protein structures of plants deteriorate, cell divisions slow down at plant development points, thus growth slows down, and the nutrients produced are rapidly consumed (Açıkgöz, 1991). It was found that there was very little germination at 5°C and the germination differences between varieties at 5°C were not statistically significant. It was determined that the seeds that did not germinate remained viable. In the examinations made, since it was accepted that the complete germination count was the radicle and plumule exit, only the seeds that had grown rootlets were not considered to be germinated. In a germination study on peas and broad beans in Poland, germination was carried out at different temperatures, and it was found that low temperature greatly reduced the germination percentage and reduced the chance of survival of the seedlings (Gorecki et al., 1990). It has been reported that the germination temperature of forage legumes planted in winter is 1-4°C at the minimum level and 20°C at the optimum level (Açıkgöz, 1991). In another study, the lowest average germination at 5°C; It was observed that some pea and bean seeds grown in cold soil showed abnormal germination and produced multiple shoots (Gorecki et al., 1990).

Number of seeds germinated before -5°C

This observation was made for the detection of seeds that re-germinated before switching to -5°C after 2 days at 0°C. According to the analysis of variance, the difference between the seed production years was effective at the P<0.05 significance level in the germination count made when the temperature of all replications was reduced to -5°C (Table 8). In Table 9, the averages of the cultivar x temperature interaction, which were found to be important as a result of the analysis of variance, are given. As a result of the LSD test applied averages, the best germination was obtained from the old seeds with 70% in the germination count made when the temperature was reduced to -5°C. Germination of new seeds was determined as 66.5%.

According to the variance analysis, the response of the cultivars differed with temperature in the germination count made when all replications were reduced to -5°C by the 10th day count (Table 8). The variety x temperature interaction was effective at the P<0.01 significance level.

Table 8
Analysis of variance of germination in forage peas when the temperature decreases to -5°C

Sources of Variance	DF	SS	MS	F
Variety	3	51.530	17.177	2.9185*
Year	1	26.283	26.283	4.4658*
Variety x Year	3	37.032	12.344	2.0974
Temperature	3	9922.616	3307.539	561.9909**
Variety x Temperature	9	618.824	68.758	11.6828**
Year x Temperature	3	22.654	7.551	1.2830
Variety x Year x Temperature	9	16.531	1.837	0.3121
Error	96	564.998	5.885	

*: P<0.05; **: P<0.01

Table 9
Variety x temperature interaction of the germination status of forage pea seeds at -5°C (number)

Variety	Temperature							
	35°C		25°C		15°C		5°C	
Furkan	21.1	BC	21.8	ABC	21.5	ABC	0.0	F
Bilgehan	16.1	D	24.3	A	19.9	C	7.6	E
Özkaynak	22.4	ABC	24.4	A	24.0	AB	0.3	F
Taşkent	21.9	ABC	24.6	A	23.5	AB	0.0	F

LSD (P<0.01): 3.188; CV (%): 14.21

When the interaction between cultivar and temperature was examined as a result of the LSD test, the reactions of cultivars to temperatures to -5°C were realized in different directions. The difference between the germination of Özkaynak variety germinated at 35°C when descending to -5°C and the germination of Bilgehan variety germinated at 35°C showed statistical significance. As a matter of fact, the difference between the reaction of the seeds of the Taşkent variety left to germinate at 25°C and the germination response of the seeds of the Bilgehan variety left to germinate at 15°C when descending to -5 °C was found to be statistically significant (Table 9).

Number of seeds germinated after the last temperature (15°C)

Before data on the number of germinated seeds after the final temperature were obtained, the seeds removed from -5°C were counted after being kept at 0°C for 2 days, at 5°C for 3 days, and at 15°C for 3 days, respectively. In the last germination count, the effect of the production year of the seeds on germination was found to be significant at P<0.05. The difference between the old seeds germinating with a rate of 73.2% and the new seeds germinating with a rate of 67.2% as a result of the LSD test averages is statistically significant. According to the analysis of variance performed in the last germination count, the variety x temperature interaction was effective at P<0.01 importance (Table 10).

Table 10
Variance analysis table of germination in forage peas from -5°C to 15°C

Sources of Variance	DF	SS	MS	F
Variety	3	319.653	106.551	8.3588**
Year	1	76.569	76.569	6.0067*
Variety x Year	3	29.335	9.778	0.7671
Temperature	3	6423.515	2141.172	167.9728**
Variety x Temperature	9	1075.496	119.500	9.3746**
Year x Temperature	3	5.274	1.758	0.1379
Variety x Year x Temperature	9	43.945	4.883	0.3831
Error	96	1223.725	12.747	

*: P<0.05; **: P<0.01

Table 11
Variety x temperature interaction of germination of forage pea seeds from -5°C to 15°C

Variety	Temperature							
	35°C		25°C		15°C		5°C	
Furkan	21.1	A	22.4	A	21.5	A	6.9	B
Bilgehan	7.8	B	24.1	A	20.0	A	7.6	B
Özkaynak	22.6	A	24.3	A	23.6	A	3.5	B
Taşkent	21.9	A	24.5	A	24.0	A	5.1	B

LSD (P<0.01): 4.691; CV (%): 20.34

According to the LSD test, when the averages of germination taking place from -5°C to 15°C are examined, the difference between the number of germinated seeds in the last count of Bilgehan variety germinated at 35°C and the number of germinated seeds of Furkan, Özkaynak and Taşkent varieties germinated at 35°C statistically significant (Table 11).

According to the results obtained from the study, it was observed that the seeds that started to germinate at 5°C kept their vitality and it was determined that they germinated in the last count carried out after 15°C. The difference between varieties in this germination at 5°C was found to be statistically insignificant (Table 11).

Damage status at -5°C ("1-9" scale)

When the damage scale values were examined in the analysis of variance in determining the damage status of all replications at -5°C, it was determined that the interaction of cultivar and temperature was significant at P<0.01 (Table 12). In a similar study carried out in field conditions, it was determined that the effect of the variety factor on the overwintering rate of forage peas at -8°C was found to be effective at P<0.01 significance level to determine the winter hardiness of forage peas (Homer and Groose, 2016).

As a result of the averages obtained from the LSD test, when examining the damage situation at -5°C, the varieties gave different responses with the difference be-

tween germination temperatures. The difference between the damage status of the seeds of the Bilgehan variety germinated at 35°C and the damage status of the Taşkent variety at 25°C was found to be statistically significant. On the other hand, the difference between the damage conditions of Özkaynak and Taşkent cultivars germinated at 15°C and 25°C at -5°C is not statistically significant (Table 12).

Özkaynak variety gave the best results in terms of cold resistance among forage pea lines and varieties, whose winter resistance was examined in Erzurum conditions (Aslan, 2017). In a similar study, the wintering rate was determined as Özkaynak among other forage pea varieties and lines (Kadıoğlu and Tan, 2018).

Table 12
Variance analysis table of damage to forage peas at -5°C

Sources of Variance	DF	SS	MS	F
Variety	3	57.125	19.042	19.8696**
Year	1	1.125	1.125	1.1739
Variety x Year	3	3.625	1.208	1.2609
Temperature	3	101.125	33.708	35.1739**
Variety x Temperature	9	124.625	13.847	14.4493**
Year x Temperature	3	1.625	0.542	0.5652
Variety x Year x Temperature	9	8.625	0.958	1.000
Error	96	92.000	0.958	

**: P<0.01

Table 13
Variety x temperature interaction of damage to forage peas at -5°C

Variety	Temperature							
	35°C		25°C		15°C		5°C	
Furkan	1.3	BC	1.5	BC	1.5	BC	1.0	C
Bilgehan	7.3	A	1.5	BC	1.8	BC	1.0	C
Özkaynak	2.3	B	1.0	C	1.0	C	1.3	BC
Taşkent	2.3	B	1.0	C	1.0	C	1.0	C

LSD (P<0.01): 1.286; CV (%): 56.96

As a result of studies on vetch and pea species, it has been determined that plants with short stature andslow growth and smaller leaf areas are more resistant to cold (Açıkgöz, 1982). It has been determined in the study that the plants germinated in the optimum temperature range are less damaged by cold. It has been determined that the plants germinated above the optimum germination temperature suffer the most damage when kept at -5°C due to the weak tissue structure, development status and chlorophyll status. It has been observed that the seeds left to germinate at 5°C are not damaged at -5°C and maintain their vitality. In a study conducted in field conditions, it was determined that pea plants exposed to cold with less or more leaves than 4-5 leaves were more damaged by cold (Alan and Geren, 2012).

Stem length (cm)

The response of the cultivars to the stem lengths of the germinating forage peas differed with the germination temperatures. This difference was effective at the P<0.01 significance level according to the analysis of variance (Table 14).

In the germination study carried out on vetch species at different temperatures, the highest stem length was determined at 20°C and it was observed that the stem lengths of the plants decreased after 25°C (Çuhadar, 1997).

When the stem lengths of forage peas were examined, the responses of the varieties to the germination temperatures were different as a result of the LSD test. The difference between the stem length of Özkaynak variety germinated at 25°C and the stem length of Furkan variety germinated at 25°C was statistically significant. Although the Taşkent variety germinated at 25°C gave the highest average stem length, the difference between the average height of the Taşkent variety germinated at 25°C and the average stem length of Özkaynak and Bilgehan cultivars germinated at 25°C was found to be insignificant (Table 15).

Table 14
Analysis of variance of forage pea stem length

Sources of Variance	DF	SS	MS	F
Variety	3	49.5817	16.5272	7.8238**
Year	1	5.0880	5.0880	2.4086
Variety x Year	3	0.6802	0.2267	0.1073
Temperature	3	768.7097	256.2365	121.2990**
Variety x Temperature	9	72.2758	8.0306	3.8016**
Year x Temperature	3	5.8122	1.9374	0.9171
Variety x Year x Temperature	9	8.4050	0.9338	0.4421
Error	96	202.7939	2.1124	

**: P<0.01

Table 15
Variety x temperature interaction of forage pea stem length (cm)

Variety	Temperature							
	35°C		25°C		15°C		5°C	
Furkan	3.4	EF	5.2	CDE	4.8	DE	0.2	I
Bilgehan	1.0	GHI	6.8	ABC	2.8	FG	0.3	I
Özkaynak	3.6	EF	7.4	AB	5.2	CDE	0.3	I
Taşkent	2.4	FGH	8.3	A	6.2	BCD	0.5	HI

LSD (P<0.01): 1.910; CV (%): 39.83

Root length (cm)

The response of the cultivars to the root lengths of the germinating forage peas differed with the germination temperatures. According to the analysis of variance, this difference was effective at the significance level of P<0.01 (Table 16).

In a study with peas, the root development of plants was investigated between 12°C and 36°C, and it was found that root growth increased at temperatures up to 30°C and a decrease in root length occurred after 30°C (Leopold and Kriedemann, 1975).

When the root length averages were examined in the LSD test, the response of the cultivars to the germination temperatures occurred in different directions. The difference in root length averages of Taşkent and Özkaynak varieties germinated at 5°C was statistically insignificant (Table 17).

According to the results obtained from seeds germinated at 5, 15, 20, 25 and 30°C, the best germination temperature range was between 18°C and 23°C, but 20°C and 25°C. range was determined as the temperature range with the best root growth (Mosjidis and Zhang, 1995).

Table 16
Variance analysis table of root length of forage pea

Sources of Variance	DF	SS	MS	F
Variety	3	21.1406	7.0468	8.3757**
Year	1	0.0569	0.0569	0.0677
Variety x Year	3	0.9333	0.3111	0.3698
Temperature	3	192.4707	64.1569	76.2550**
Variety x Temperature	9	37.5308	4.1700	4.9565**
Year x Temperature	3	0.7385	0.2461	0.2926
Variety x Year x Temperature	9	10.7949	1.1994	1.4256
Error	96	80.7693	0.8413	

**: P<0.01

Table 17
Variety x temperature interaction of root length of forage pea (cm)

Variety	Temperature							
	35°C		25°C		15°C		5°C	
Furkan	3.2	CDEF	3.6	BCDE	4.3	ABCD	1.6	H
Bilgehan	1.3	H	5.5	A	3.1	DEF	1.6	H
Özkaynak	3.7	BCDE	5.5	A	4.4	ABC	1.8	GH
Taşkent	2.9	EFG	5.3	A	4.8	AB	2.1	FGH

LSD (P<0.01): 1.205; CV (%): 26.86

Total length (cm)

The response of cultivars to total plant length of forage peas differed with germination temperatures. According to the analysis of variance, this difference was effective at the P<0.01 significance level.

When the interaction between the difference in the total heights of the genotypes and the germination temperatures was examined, the difference between the total height averages of the Taşkent variety germinated at 25°C and the total height averages of the Özkaynak variety germinated at 25°C was found to be statistically insignificant. However, the difference between the Tashkent variety germinated at 35°C and the Tashkent variety germinated at 25°C was significant (Table 19).

Table 18
Variance analysis table of total length of forage peas

Sources of Variance	DF	SS	MS	F
Variety	3	133.585	44.528	9.403**
Year	1	4.068	4.068	0.859
Variety x Year	3	1.815	0.605	0.128
Temperature	3	1726.936	575.645	121.555**
Variety x Temperature	9	201.680	22.409	4.732**
Year x Temperature	3	6.137	2.046	0.432
Variety x Year x Temperature	9	34.150	3.794	0.801
Error	96	454.623	4.736	

**: P<0.01

Table 19
Variety x temperature interaction of total length of forage peas (cm)

Variety	Temperature							
	35°C		25°C		15°C		5°C	
Furkan	6.7	EF	8.8	CDE	9.1	CDE	1.9	H
Bilgehan	2.3	H	12.3	AB	5.9	F	1.9	H
Özkaynak	7.2	DEF	12.9	A	9.6	BCD	2.2	H
Taşkent	5.3	FG	13.5	A	11.0	ABC	2.5	GH

LSD (P<0.01): 2.859; CV (%): 30.81

Root length/Stem length

The difference between cultivars in the root/stem ratios of the germinated forage peas was effective at the significance level of P<0.05 (Table 20). The difference between Furkan variety with 4.0% root/stem ratio and Bilgehan variety with 3.2% root/stem ratio as a result of LSD test averages is statistically significant.

Table 20
Variance analysis table of Root length/Stem length ratio of forage pea

Sources of Variance	DF	SS	MS	DF
Variety	3	0.971	0.324	2.8262*
Year	1	0.468	0.468	4.0864*
Variety x Year	3	0.345	0.115	1.0035
Temperature	3	28.527	9.509	83.0245**
Variety x Temperature	9	1.105	0.123	1.0718
Year x Temperature	3	0,809	0.270	2.3559
Variety x Year x Temperature	9	0.844	0.094	0.8181
Error	96	10.995	0.115	

*: P<0.05; **: P<0.01

Table 21
Averages of root length/stem length ratio of forage peas (cm)

Means of Variety							
Furkan		Bilgehan		Özkaynak		Taşkent	
0,97	A	0,76	B	0,90	AB	0,98	A
Means of Temperature							
35°C		25°C		15°C		5°C	
0,80	C	1,50	A	1,10	B	0,90	BC
Means of Year							
Old Seed (2008-2009)				New Seed (2015-2016)			
0,80				0,90			

LSD (Variety: P<0.05) :0.1683; LSD (Temperature: P<0.05): 0.2228; CV (%): 37.32

The difference between the seed production years in the root/stem ratios of the plants was effective at the P<0.05 significance level (Table 20). The difference between the old seeds with 3.2% root/stem ratio and the new seeds with 3.6% root/stem ratio as a result of the LSD test averages was found to be statistically significant (Table 21).

Germination temperatures were effective on the root/stem ratios of the germinating forage pea plants at the significance level of P<0.01 (Table 20). The difference between the root/stem ratio of the plants germinated at 4.4% and 25°C and the root/stem ratio of the plants germinated at 6.0% and 15°C, as a result of the LSD test averages, is statistically significant.

It has been reported that the increase in temperatures after the optimum degree will decrease the root/stem ratio in plants (Akçin, 1981). Based on this, it was determined that although the root/stem ratio of forage peas germinated at relatively 25°C was good, the root/stem ratio decreased at 35°C.

4. Conclusions

As an example of the study supporting this, it was seen that summer sowing gave better results in forage peas planted in winter and summer in Konya conditions. The reason is that due to late winter planting, the plants were exposed to winter cold before reaching sufficient seedling height (Konuk and Tamkoç, 2018).

Since this study was carried out in germination environments, it was carried out with constant temperature applications, and the temperatures were gradually lowered and increased in order to determine the cold damage. However, since environmental conditions are constantly changing, these studies can be supported by conducting them in parallel with field studies. In the results obtained, it can be found that the most suitable germination temperature in winter forage pea varieties, as well as how much the forage pea seedlings growing at the appropriate germination temperature are affected by cold damage. Based on this, for the forage peas to be cultivated commercially, they can spend the winter in a

healthy way only by sowing at the appropriate time in winter sowing and the products can be harvested without losing their yield. Otherwise, failures may occur in winter planting.

5. References

Açıkgöz E (1982). Cold tolarance and its association with seedling marphology and chemical composition in annual forage legumes 1. field peas, *Z.Pflanzenzüchtung* 88: 118-126.

Açıkgöz E (1991). Yem Bitkileri Tarımı. Uludağ Üniversitesi Yayınevi, Bursa.

Akçin A (1981). Genel Tarla Ziraatı. Atatürk Üniversitesi Tarla Bitkileri Bölümü, Ankara.

Alan Ö, Geren H (2012). Bezelye'de (*Pisum sativum* L.) farklı ekim zamanlarının tane verimi ve diğer bazı tarımsal özellikler üzerine etkisi. *Ege Üniversitesi Ziraat Fakültesi Dergisi* 49 (2): 127-134.

Anonymous (1982). Mstat-C statistical program.

Ashby WC, Hellmers H 1(955). Temperature requirements for germination in relation to wild-land seeding. *Journal of Range Management* 8 (2): 80-83.

Aslan B (2017). Erzurum şartlarında kışlık yem bezelyesi geliştirme çalışmaları. Yüksek Lisans Tezi, Atatürk Üniversitesi Fen Bilimleri Enstitüsü, Erzurum.

Çaçan E, Kökten K, Özbay N (2016). Bazı yem bezelyesi hat ve çeşitlerinin farklı sıcaklıklarda çimlenme ve çıkış performanslarının belirlenmesi. *Nevşehir Bilim ve Teknoloji Dergisi TARGİD*, Özel Sayı 62-68.

Çuhadar B (1997). Fiğ tohumlarının çimlenmesi üzerine sıcaklığın etkisi. Yüksek Lisans Tezi, Selçuk Üniversitesi Fen Bilimleri Enstitüsü, Konya.

Düzgüneş O, Kesici T, Kavuncu O, Gürbüz F (1987). Araştırma ve deneme metotları. Ankara Üniversitesi, Ziraat Fakültesi Yayınları, Ankara.

Elçi Ş, Açıkgöz E (1993). Baklagil ve buğdaygil yem bitkileri tanıtma kılavuzu. TİGEM Yayınları, Afşaroğlu Matbaası, Ankara.

Gorecki R, Fordoński G, Bieniaszewski T, Jacuński K (1990). Comparative studies on chilling sensitivity in some legume seeds. *Acta Physiologiae Plantarum* 12 (2): 149-158.

Homer A, Groose RW (2016). Using an "index of merit" to evaluate winterhardy pea lines. *J. Agric. Sci.* 8: 45-53.

Kadıoğlu S, Tan M (2018). Erzurum şartlarında bazı yem bezelyesi hat ve çeşitlerinin tohum verimleri ile bazı özelliklerinin belirlenmesi. *Atatürk Üniversitesi Ziraat Fakültesi Dergisi* 49 (2): 143-149.

Konuk A, Tamkoç A (2018). Yem bezelyesinde kışlık ve yazlık ekimin bazı tarımsal özellikler üzerine etkisi. *Bahri Dağdaş Bitkisel Araştırma Dergisi* 7 (1): 39-50.

Leopold AC, Kriedemann PE (1975). Plant Growth and Development, 2. Edition, Mc Graq-Hill, New York.

Mosjidis J, Zhang X (1995). Seed germination and root growth of several Vicia species at different temperatures. *Seed Science and Technology (Switzerland)* 23 (3): 749-759..

Qin DH (1992). Effect of temperature on seed characters of common vetch [*Vicia sativa*] and hairy vetch [*Vicia villosa*]. *Grassland of China* 5: 18-22.

Tamkoç A (2017). Türkiye'de yem bitkileri ve hayvansal protein üretimi. Hanhana Kalecik Kültür Dergisi, 3.

Selcuk Journal of Agriculture and Food Sciences SJAFS

http://sjafs.selcuk.edu.tr/sjafs/index

(2022) 36 (3), 438-446

e-ISSN: 2458-8377

DOI:10.15316/SJAFS.2022.058

Research Article

Effectiveness of Conventional and Minimized Tillage Practices on Soil Quality Properties and Maize Yield Attributes

Resul MUT[1], Cevdet ŞEKER[1], Hamza NEGİŞ[1,*]

[1]Selçuk University, Faculty of Agriculture, Department of Soil Science and Plant Nutrition, Konya, Türkiye

ARTICLE INFO

Article history:
Received date: 15.08.2022
Accepted date: 14.11.2022

Keywords:
Conventional tillage
Minumum tillage
Direct sowing
Soil water content
Maize yield components

ABSTRACT

The effects of tillage systems on soil properties and crop productivity have been the subject of many studies to ensure sustainable productivity. Aims: In the study, the effects of different tillage methods applied in the intermediate period of wheat-maize rotation on the soil quality properties and corn yield elements were investigated in the pre-planting period (PP) and plant growth period (PGP) of corn. Methods: Conventional tillage (CT), minimum tillage with subsoiling and chisel (MT1), minimum tillage with subsoiling (MT2), minimum tillage with chisel (MT3) and direct sowing (DS) methods were compared. As a result of the tillage practices, at 0-20 cm, the highest bulk density values were measured in the DS and CT methods, and 20-40 cm was determined in the CT method. The penetration resistance values of the soils measured at a depth of 0-80 cm were significantly affected by the applications made in both PP and PGP. The highest saturation value was measured in the MT3, the highest field capacity and plant available water contents were measured in the DS. The effects of the applications on the chemical properties of the soil and the nutrient content of the corn plant were limited. The effects on only grain protein ratio from yield components of corn plant were significant, the highest value was measured in CT. Considering the sustainable management of soils labour requirement and lower costs it was concluded that DS and MT methods are more applicable than CT methods in terms of yield.

1. Introduction

In order to meet the food demand on a global scale, increasing tillage practices causes faster transformations and degradations in the agro-ecosystem. Soil cultivation activities, which have increased progressively in the last fifty years, increased energy costs on the one hand and reducing profitability on the other hand, and cause deterioration in soil properties at the same time (Alskaf et al. 2021; Voorhees and Lindstrom 1984). Soil quality, which can be applied to agricultural and natural ecosystems, has attracted considerable attention worldwide in recent years (Andrews et al. 2002; Doran and Parkin, 1994; Karlen 2004). Sustainable soil management systems often require increased management activities. Instead of meeting these management activities, soil quality tests and accordingly application recommendations can reveal both selection and management functions together (Bünemann et al. 2018). Soil quality is the most practical method for the management practices, interpretation of how the soil and ecosystem are affected, and the continuation of sustainability, as well as the regularity of soil information (Beinat and Nijkamp, 1998). Physical, chemical, and biological quality characteristics of the soils impressed due to the difference in tillage methods and creates differences in plant development, root growth and product yields (Gassel 1982; Gholami et al. 2014). To ensure sustainability in agricultural activities, soil management practices such as reduced tillage or zero tillage can be selected according to the region, climate, and plant. Selection of the most suitable method helps protect soil and water resources, maintain agricultural income and to reduce soil degradation with alternative tillage (Azimzadeh et al. 2008; Blevins et al. 1971; Mujdeci et al. 2017). As a result of the application of conventional tillage methods, the physical and structural properties of the soils deteriorate (Jia et al. 2010; Ren et al. 2018; Shaokun et al 2006). While the soils are compacted due to some tillage tools and field traffic, after processing with conventional tillage tools, the penetration resistance of the top layer of soils generally decreases (Ehlers et al. 1983). However, due to repeated tillage, it is inevitable that a layer limiting the development of plant roots is formed at under the cultivation

* Corresponding author email: *hnegis@selcuk.edu.tr*

depth of soil. Soil porosity tends to decrease with increasing compaction due to intensification of tillage (Şeker 1999; Şeker and Işıldar 2000). Together with the it should be increase reduced soil porosity, it causes the limitation of the use of water and nutrients that plants can benefit from (Hernanz et al. 2000; Topa et al. 2021). Owing to decreasing tillage costs, increasing the amount of water stored in the soil or maintaining its current potential, reduced tillage is considered a long-term recommendation for agro-ecosystems (Li, Liao et al. 2019). Reduced tillage practices provide protection and increase in physical quality by improving soil structure, and accordingly, improve the water holding capacity of soils (Borges et al. 2018; Hellner et al. 2018). There are important interactions between the hydraulic properties and the physical properties of the soil (Li et al. 2019; Perkins et al. 2007; Wang and Shao 2013). Depending on the bulk density and aggregation change of the soils, different tillage techniques affect the hydraulic properties of the soils (Alaboz 2020; Strudley et al. 2008). Since the structure of micro and macro aggregates in soils will differ depending on the differences in tillage, it affects the pore size distribution of the soil and, accordingly, the hydrodynamic properties (Kutilek 2004). Upon evaluation of studies on soil tillage practices, although the medium and long-term effects are better known, the information about the short-term effects is limited. For this reason, in this study, the effects of 5 different tillage methods on the quality characteristics of the soil and the yield components of the corn plant were investigated in pre-planting period (PP) and plant growth period (PGP) at the wheat-corn crop rotation.

2. Materials and Methods

Site location and description

The study was carried out in the province of Aksaray, located in the central south of Turkey, in an area where wheat-corn rotation is practiced (38°24'55.3"N 33°51'01.3"E). The altitude of the study area is 936 m, and the continental climate is dominant in the region. Since the climate data for many years are examined, the temperature was measured as -3.6 °C in the lowest January, the highest in August, 30.6 °C and 12 °C as the average temperature. The region receives most of the precipitation in winter and spring due to its climatic characteristics, and the average precipitation is 361.80 mm (MGM 2020).

The soil at the experimental site area has high clay content (54.40%), medium calcareous (7.47%), low organic matter (1.66%) (Table 1). The soil type was Typic Torrifluvents (Soil Survey Staff 1999).

Experimental design and treatments

The experimental design was laid out in a randomized complete block with three replications, in 15 plots (4x25 m), after wheat harvest. Five tillage treatments and application time were compared as indicated in Table 2. Mean working depth of soil tillage tools used in the study was 20 cm for mould board plough, 60cm for subsoiling (100 cm tillage range), 40 cm for chisel, 30 cm for cultivator, 15 cm for rotatiller, 10 cm for combine rotary and 15 cm for hoeing machine.

Table 1

Basic physico-chemical properties and measurement methods of experimental soil (0-20 cm).

Soil properties		Units	Values	Methods
Texture	Sand	%	33.57	Gee, Bauder, and Klute (1986)
	Silt		12.03	
	Clay		54.40	
Texture class			Clay	
AS		%	49.66	Gugino et al. (2009)
Bulk density		g cm^{-3}	1.23	Blake and Hartge (1986)
Particle density		g cm^{-3}	2.64	
pH		-	8.28	Gugino et al. (2009)
EC		µS cm^{-1}	536	
Lime		%	7.47	McLean (1983)
OM		%	1.66	Wright and Bailey (2001)
AP		mg kg^{-1}	11.48	Olsen and Sommers (1982)
TN		%	0.113	Wright and Bailey (2001)
Fe*		mg kg^{-1}	4.38	Lindsay and Norvell (1969)
Cu*		mg kg^{-1}	1.24	
Mn*		mg kg^{-1}	4.51	
Zn*		mg kg^{-1}	0.95	

AS: Agregatte stability, EC: Electrical conductivity, OM: Organic matter, AP: Available P, TN: Total N, *: DTPA extracted.

Soil measurements and analysis

To determine the soil properties, disturbed soil samples were taken from 0-20 cm depth from different points of the trial area, and it was used in the basic analyses given in Table 1. Soil field measurements and samplings were made in two different periods (pre-plantation (PP) ve plant growth period (PGP)) to determine the effects of tillage practices on the physical quality characteristics of the soil. To determine the effects of applications on other soil quality characteristics, soil samplings were made in a single period (tassel shot time). For this purpose, at every two periods, the bulk density (BD) was determined at 0-20 cm and 20-40 cm soil depths, and the penetration resistance (PR) was measured at 0-80 cm soil depth. Digital Eijkelkamp Penetrologger with code 06.15.SA was used to determine the penetration resistance. In the PGP period, aggregate stability (AS), field capacity (FC), permanent wilting point (PWP), plant available water (PAW) contents, total porosity (TP) and macro porosity (MP) values and soil chemical properties (pH, EC, OM, TN, AP, Fe, Zn, Cu ve Mn) were measured in 0-20 cm soil depth.

Table 2
Field tillage practices planning and treatments dates

Soil tillage/treatments dates	CT	MT1	MT2	MT3	DS
01.10.2019	Moldboard plow	*	*	*	
01.10.2019	*	Subsoiling	Subsoiling	*	
28.10.2019	*	Chisel	*	Chisel	*
28.04.2020	Cultivator	Cultivator	Cultivator	Cultivator	
30.04.2020	Rototiller	Rototiller	Rototiller	Rototiller	
30.04.2020	Combine rotary	Combine rotary	Combine rotary	Combine rotary	
01.05.2020	Sowing	Sowing	Sowing	Sowing	Sowing
19.05.2020	1. Hoeing	1. Hoeing	1. Hoeing	1. Hoeing	1. Hoeing
06.06.2020	2. Hoeing	2. Hoeing	2. Hoeing	2. Hoeing	2. Hoeing

CT; conventional tillage, MT1; minimum tillage with subsoiling and chisel, MT2; minimum tillage with subsoiling, MT3; minimum tillage with chisel and DS; direct seeding

Crop management and measurements

In the experiment, a maize variety, which is in the FAO 700 death group and widely grown in the region, was sown with a pneumatic seeder with 70 cm row spacing and 16 cm plant spacing. Before planting, 13-18-15 fertilizer was applied to all plots as a basal fertilizer at 500 kg ha^{-1}. In the next periods, 270 kg ha^{-1} of urea (46%N) in the first hoe and 600 kg ha-1 of ammonium sulphate (21%N) in the second hoe were applied at the soil surface and was mixed with soil. Herbicide with 2,4-D 2-ethylhexyl ester + florasulam active ingredient in the fight against weeds and insecticides with active ingredients imidacloprid, thiamethoxam and lambda-cyhalothrin were used to combat pests.

In order to determine the effects of the treatments on the growth characteristics and yield of the corn plant; shoot emergence, nutrient content of the leaves (N, P, Fe, Zn, Cu and Mn) (Bayraklı 1987), grain protein ratio (6.25 times the grain nitrogen content) (Wright and Bailey 2001), biomass and grain yield were measured. For seedling emergency, plants at 5 meters from the three rows in the middle were counted during the post-planting shoot period. Biomass and grain yield were measured in 7 m^2 area in each plot.

Statistical Analysis

Analysis of variance was performed to test for significant differences between tillage treatments. Means were compared using the Tukey multiple comparison test at a probability level of 0,05. SPSS statistical software was used in all data analysis.

3. Results and Discussion

Bulk density (BD)

In the PP, the effects of different tillage practices on the BD measured at 0-20 cm and 20-40 cm soil depth was statistically significant ($P<0.05$). Accordingly, at a depth of 0-20 cm, the highest BD was measured in the DS application with 1.16 g cm-3, while the lowest BD value was measured in the MT3 application with 1.08 g cm-3. The difference between the BD values measured in other applications was statistically insignificant and were included in the same group. The highest BD value in the subsurface layer (20-40 cm) was measured in the CT application with 1.23 g cm-3, while the lowest was measured in the MT3 application with 1.13 g cm-3, however, the differences between the MT1, MT2, MT3 and DS methods were statistically insignificant (Figure 1).

In the PGP, the effects of tillage practices on BD measured at 0-20 cm and 20-40 cm soil depth were statistically significant ($P<0.05$). During this period, at a depth of 0-20 cm, the highest BD was measured as 1.22 g cm-3 in the CT application, while the lowest BD value was measured as 1.14 g cm-3 in the MT1 application. The difference between the BD values measured in other applications was statistically insignificant and were included in the same group. While the lowest BD values was in the DS application with 1.16 g cm-3, the BD value measured at 20-40 cm depth was determined with the highest 1.23 g cm-3 in the CT application, however, the differences between the MT1, MT2, MT3 and DS methods were statistically insignificant (Figure 2).

According to these results, DS method at 0-20 cm depth gave higher BD values due to the lack of tillage, while CT method at 20-40 cm depth gave higher results than other methods. The reason for this was evaluated to be caused by the pressure applied to the substrate during cutting and overturning of the plough and tractor traffic formed in the plough track creating a plough layer at 20 cm. The BD values also depend on the structural condition of the soils and are an indicator of soil compaction (Sutherland et al. 2001; Gomez et al. 2002; Karlen 2004; Hall and Raper 2005). The BD values measured in the treatments were below the 1.40 g cm-3 value, which limits plant root development (Lhotský et al. 1984; Badalíková 2010). As a result, although different tillage methods affected the BD values of the soils in different ways, they did not limit plant root growth. However, the effect of soil tillage treatments to be made in the long term will be more decisive (Lal 1993). In addition, it has been observed in the studies that direct sowing does not affect the bulk density much in the short term (Moraes and Benez 1996; So et al. 2009).

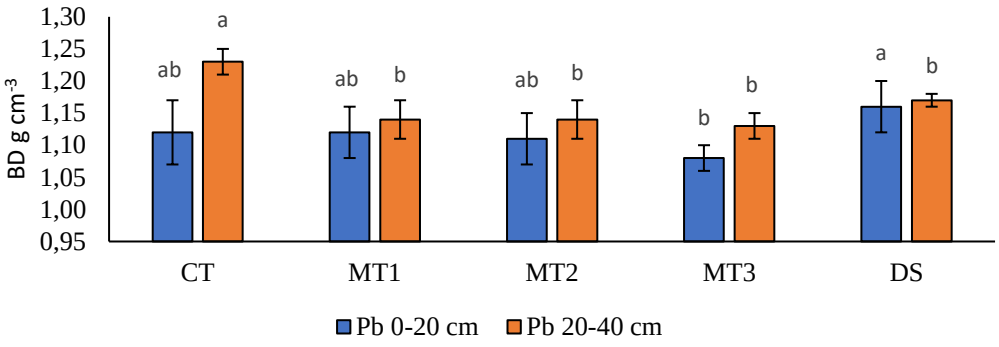


Figure 1
Bulk density (BD) changing in pre-plantation period (CT; conventional tillage, MT1; minimum tillage with subsoiling and chisel, MT2; minimum tillage with subsoiling, MT3; minimum tillage with chisel and DS; direct seeding, means (n = 3) followed by the same letter in a column are not significantly different ($P \leq 0.05$).

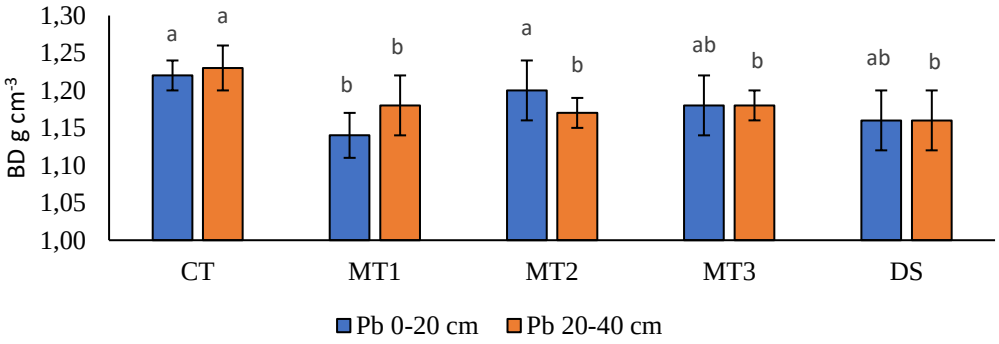


Figure 2
Bulk density (BD) changing in plant growth period (CT; conventional tillage, MT1; minimum tillage with subsoiling and chisel, MT2; minimum tillage with subsoiling, MT3; minimum tillage with chisel and DS; direct seeding, means (n = 3) followed by the same letter in a column are not significantly different ($P \leq 0.05$).

Penetration resistance (PR)

Tillage applications, except for 0-20 cm in PGP, had a statistically significant effect on PD measured at 0-20 cm, 20-40 cm, 40-60 cm, and 60-80 cm depths of soil in PP and PGP ($P < 0.05$).

In the PP, at the soil depth of 0-20 cm, the highest PR value was 1.04 MPa in the MT2 method, while the lowest was 0.55 MPa in the MT1 method. While the difference between the MT2 and DS methods were insignificant, the differences between the CT, MT1 and MT3 methods were also insignificant. This situation shows that subsoiling application (MT2) alone does not create a sufficient level of loosening effect on the soil at the depth of 0-20 cm. It was determined that other applications were at the same level and more effective on loosening 0-20 cm soil (Table 3 and Figure 3). The PR values at the depth of 20-40 cm were measured at the highest 1.66 MPa in the MT2 method, while the lowest 1.29 MPa was measured in the MT1 method. While CT, MT2 and MT3 applications were in the same group with higher PR, MT1 and DS applications were also in the same group with lower PR. This shows that CT, MT2 and MT3 applications have an increasing effect on the soil at 20-40 cm depth (Table 3 and Figure 3). The highest PR value was measured at the depth of 40-60 cm with 2.33 MPa in the CT application and was in the same group with the MT3 application. The lowest was 1.87 MPa in the DS application and was in the same group with the MT1 application. MT2 application was between these two groups with a PR of 2.14 MPa (Table 3). CT and MT3 treatments caused more compaction at 40-60 cm depths, as well as at 20-40 cm depth. This situation is due to both the pressure of the plow base and the trace of the tractor in the conventional tillage method (CT). In addition, it is understood that the subsoil is compaction due to the pressure formed under the cultivation depth in the chisel application (MT3), where soil cultivation is carried out at a depth of 40 cm. Similar effects were also noted by Şeker and Işıldar (2000). Finally, in the PR values at 60-80 cm depth, the CT method gave the highest result (3.24 MPa), while the MT3 method was ranked second with 2.61 MPa and the MT2 method was ranked as the third with 2.37 MPa. With a statistically insignificant difference, the lowest values were found in MT2 and DS methods as 2.09 MPa and 2.02 MPa, respectively (Table 3).

In the PGP, while the highest PR value was measured at the depth of 20-40 cm as 2.85 MPa in the MT3 application. PR value of MT2, DS and CT methods was measured 2.48 MPa, 2.41 MPa and 2.36 MPa, respectively and statistically insignificant differences occurred between them. The lowest value was obtained from the MT1 method as 1.96 MPa (Table 3 and Figure 4). At the depth of 40-60 cm, the highest PR values of 3.13 MPa

was measured in the MT3 application, which is in the same group as CT. These were followed by other applications in different groups, In the MT1, MT3 and DS applications were measured 2.54 MPa, 2.41 MPa and 2.24 MPa, respectively (Table 3). At the depth of 60-80 cm, the highest PR value was measured as 2.80 MPa in the MT3 method, and it was in the same group with MT1 and MT2 applications. DS method was in the same group with MT1 and MT2 methods, and the lowest value was measured as 2.06 MPa in the DS methos (Table 3).

In the PP, at the depth of 0-20 cm, PR values were lower in the MT1 and MT2 methods, while lower values were obtained in the MT1 and DS methods at 20-40 cm depth. While the lowest PR values, at a depth of 40-60 cm like a depth of 20-40 cm, were obtained from the MT1 and DS methods; PR values of the CT, MT2 and MT3 methods exceeded the 2 MPa limit value, which negatively affects plant root growth (McKyes 1985). At the 60-80 cm depth similar to 40-60 cm depth, at the CT, MT2 and MT3 methods measured higher PR values. The CT method also exceeded the limit value of 3 MPa, which stopped plant root growth with 3.24 MPa (Busscher and Sojka 1987; Gugino et al. 2009; Gülser and Candemir 2012). The PR values of the MT1 and DS methods like the MT2 and MT3 methods, also gave results slightly above the threshold value, although they exceeded the 2 MPa threshold value that negatively affected root growth (Gajri et al. 1994; Hall and Raper 2005).

In the PGP, PR values were higher than the pre-sowing period, especially at 0-60 cm depth. The reason for this is that the seed bed preparation, planting process and machine hoeing and pesticide applications during the plant development period create different amounts of pressure in the soil. At the 20-40 cm, except for the MT1 method, at the depth of 20-80 cm, in all tillage methods has been exceeded the 2 MPa limit value that negatively affected the root development of the plant (McKyes 1985). Except for the DS method at planting time, the reason for the compaction at the depth of 20-40 cm is that the cultivator, rototiller and combine rotary applications, creates compaction under the processing depth of the hoe machine during the plant development period. In the MT1 method, both chisel plough and subsoiler were used hence reduction of superficial and deep compression occurred and the PR values remained below the limit value of 2 MPa (McKyes 1985). At the depth of 40-60 cm, PR values in the CT and MT3 methods exceeded the limit value of 3 MPa that hinders plant root growth (Busscher and Sojka, 1987; Gugino et al. 2009; Gülser and Candemir, 2012). In the DS method, the absence of field traffic before sowing as well as the limited field traffic after sowing caused less compaction in the subsoil layers.

Table 3
The effects of different tillage practices on the penetration resistance (MPa) of the soil in two periods

Tillage practices	Pre-plantation				Plant growth period			
	0-20cm	20-40cm	40-60cm	60-80cm	0-20cm	20-40cm	40-60cm	60-80cm
CT	0.68±0.25^{bc**}	1.56±0.25^{a**}	2.33±0.27^{a**}	3.24±0.28^{a**}	1.29±0.45*	2.36±0.39^{b**}	3.04±0.06^{a**}	2.80±0.15^{a**}
MT1	0.55±0.18^{c}	1.29±0.33^{b}	1.96±0.05^{c}	2.09±0.09^{d}	1.56±0.47	1.96±0.26^{c}	2.54±0.10^{b}	2.34±0.08^{b}
MT2	1.04±0.34^{a}	1.66±0.16^{a}	2.14±0.07^{b}	2.37±0.05^{c}	1.67±0.65	2.48±0.10^{b}	2.41±0.05^{c}	2.29±0.12^{b}
MT3	0.60±0.24^{c}	1.50±0.26^{a}	2.29±0.18^{a}	2.61±0.06^{b}	1.68±0.59	2.85±0.26^{a}	3.13±0.13^{a}	2.73±0.14^{a}
DS	0.87±0.14^{ab}	1.33±0.21^{b}	1.87±0.16^{c}	2.02±0.07^{d}	1.71±0.57	2.41±0.05^{b}	2.24±0.16^{d}	2.06±0.06^{c}

CT; conventional tillage, MT1; minimum tillage with subsoiling and chisel, MT2; minimum tillage with subsoiling, MT3; minimum tillage with chisel and DS; direct seeding, *; Not significant, **; means (n = 3) (± standard errors) followed by the same letter in a column are not significantly different ($P \leq 0.05$).

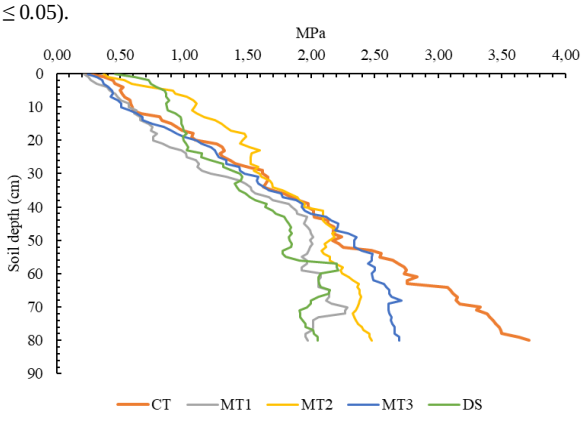


Figure 3
Penetration resistance graphs in the pre-plantation period (0-80 cm soil depth) (CT; conventional tillage, MT1; minimum tillage with subsoiling and chisel, MT2; minimum tillage with subsoiling, MT3; minimum tillage with chisel and DS; direct seeding).

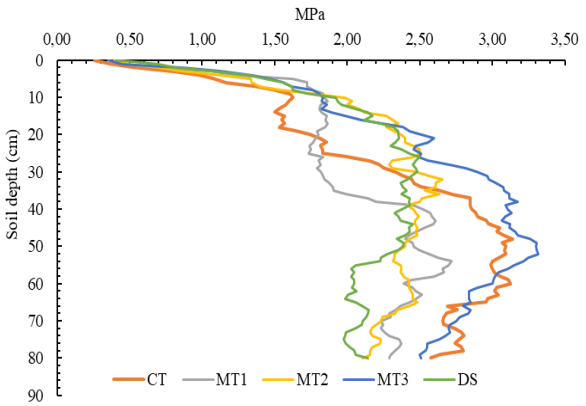


Figure 4
Penetration resistance graphs in the plant growth period (0-80 cm soil depth) (CT; conventional tillage, MT1; minimum tillage with subsoiling and chisel, MT2; minimum tillage with subsoiling, MT3; minimum tillage with chisel and DS; direct seeding).Soil water parameters

Except for permanent wilting point (θPWP), the effects of the treatments on saturation (θS), field capacity (θFC), plant available water (θPAW) and macro porosity (θMP) values were significant by statistically (P<0.05) (Table 4).

While the MT3 method gave a higher θS value compared to other applications, the lowest value (55.65%) was measured in the CT method (Table 4). Differences between CT, MT1, MT2 and DS methods and differences between MT2, MT3 and DS methods were insignificant. The highest θFC value (37.92%) was measured in the DS method, and the lowest (34.97%) in the CT method (Table 4). Only the DS method made a significant difference with the CT method, and the differences between other treatments were insignificant. An increase of approximately 8% was measured in the DS method compared to the CT method. The θFC value in the DS method was higher than others because of the structure of the pores remained intact due to the application of less pressure on the land and the preservation of the existing structure of the soil (Mitchell and Soga 2005; Burgos Hernández et al. 2019). θPWP values, as an indicator of the amount of water that can be retained on the micropores and colloid surface, varied between 24.15-25.91%. (Table 4). The reason for the insignificant change in the θPWP value is that tillage influences the macropores but not the micropore structure. The effect of applications on θPAW values, calculated as the difference of θFC and θPWP values, was significant (P<0.05) (Table 4). The plant available water content measured as 10.81% in the CT method increased by 6.84%, 6.198 and 11.10% in MT1, MT2 and DS methods, compared to CT, respectively. The θMP value of the soil varied between 20.13-23.88%, the highest value in MT3 method and the lowest value in MT3 method was measured. The θMP value, which is an indicator of the aeration level of the soil, was approximately twice of the general limiting value (10%) for plants in all tillage methods (da Silva and Kay 1997; da Silva et al. 1994).

Table 4
The effects of treatments on saturation (θ_S), field capacity (θ_{FC}), permanent wilting point (θ_{PWP}), plant available water (θ_{PAW}) and macro porosity (θ_{MP})

Tillage practices	θ_S	θ_{FC}	θ_{PWP}	θ_{PAW}	θ_{MP}
CT	55.65±2.78^{b**}	34.97±0.71^{b**}	24.16±1.35*	10.81±0.90^{b**}	20.68^{b**}
MT1	56.73±1.75^{b}	35.70±1.61^{ab}	24.15±2.74	11.55±1.85^{ab}	21.03^{ab}
MT2	57.14±0.34^{ab}	36.69±1.94^{ab}	25.21±1.90	11.48±1.91^{ab}	20.45^{b}
MT3	60.18±1.44^{a}	36.30±1.28^{ab}	25.30±1.15	11.00±1.26^{b}	23.88^{a}
DS	58.05±1.60^{ab}	37.92±1.73^{a}	25.91±1.67	12.01±1.71^{a}	20.13^{b}

CT; conventional tillage, MT1; minimum tillage with subsoiling and chisel, MT2; minimum tillage with subsoiling, MT3; minimum tillage with chisel and DS; direct seeding, *; not significant, **; means (n = 3) (± standard errors) followed by the same letter in a column are not significantly different ($P \leq 0.05$).

Chemical properties of the soil

While the effects of different tillage methods on the pH, EC, OM, TN, P and Cu contents of the soil pre-plantation period was statistically insignificant, the effect of Fe, Zn and Mn contents was limitedly significant (Table 5). In the different tillage methos, average pH, EC, OM, TN, P and Cu values of the soils were measured 8.24, 583.74 mS cm-1, 1.65%, 0.12%, 14.71 mg kg-1 and 1.80 mg kg-1 respectively. The highest and the lowest values of Fe, Zn and Mn were measured in CT and DS methods, in MT1 and MT2 methods, DS and MT3 methods, respectively (Table 5).

While the methods based on tillage (CT and MT1) had a limited effect on the Fe and Zn content of the soil, treatments had a variable effect on the Mn content, and a higher value was measured in the DS method, albeit limited. It has been evaluated that this situation may be caused by the change, increase, or decrease in soil aeration due to tillage. At the end of seven years of conventional and reduced tillage practices, soil pH and CaCO3 content were not affected by different tillage methods, while soil organic carbon, total nitrogen and plant-available phosphorus content of the soil surface layer increased partially in reduced tillage (Neugschwandtner et al. 2014). In a five-year study, it was stated that the pH was measured lower because of the mineralization of organic matter in the upper layer of the soil in the no-tillage plots compared to the plots with plough tillage (López-Fando and Pardo 2009). In this study, this effect was not observed in the short term. In the long term, the Fe content of the soil is lower in minimum tillage than in no-till agriculture, in the short-term study, the Fe content beneficial to the plant was found to be lower in the DS method (Obour et al. 2021). This shows that the duration of the tillage application has a different effect on Fe content of the soil.

Table 5
The effects of different tillage practices on the chemical properties of the soil

Tillage practices	pH	EC	OM	TN	P	Fe	Zn	Cu	Mn
		mS cm^{-1}	%	%	mg kg^{-1}	mg kg^{-1}	mg kg^{-1}	mg kg^{-1}	mg kg^{-1}
CT	8.25±0.13*	619.00±68.90*	1.65±0.06*	0.120±0.01*	13.38±0.22*	13.15±0.17^{a**}	1.64±0.02^{d**}	1.69±0.08*	12.41±0.46^{b**}
MT1	8.21±0.08	580.30±31.50	1.66±0.09	0.120±0.01	16.44±0.14	12.69±0.24^{ab}	2.47±0.04^{a}	1.87±0.07	13.60±1.01^{a}
MT2	8.26±0.05	570.03±28.70	1.59±0.03	0.115±0.02	14.74±0.06	12.55±0.19^{ab}	1.38±0.01^{e}	1.81±0.09	11.78±0.06^{c}
MT3	8.25±0.08	581.70±44.70	1.63±0.03	0.118±0.02	12.27±0.47	12.77±0.21^{a}	1.92±0.03^{c}	1.76±0.02	10.05±0.21^{d}
DS	8.23±0.06	567.67±16.44	1.73±0.01	0.126±0.01	16.72±0.43	12.09±0.33^{b}	2.27±0.07^{b}	1.87±0.08	13.34±0.65^{a}

EC: electrical conductivity, OM: organic matter, TN: total nitrogen, P: available phosphorus, Fe: available iron, Zn: available zinc, Cu: available cupper, Mn: available manganese, CT; conventional tillage, MT1; minimum tillage with subsoiling and chisel, MT2; minimum tillage with subsoiling, MT3; minimum tillage with chisel and DS; direct seeding, *; not significant, **; means (n = 3) (± standard errors) followed by the same letter in a column are not significantly different ($P \leq 0.05$).

Yield components of maize

Different tillage practices had a limited and statistically significant (P<0.05) effect only on grain protein proportion, among the effects of maize plant seedling emergence, grain yield, grain protein proportion, total biomass, and dry matter yield (Table 6). The number of seedling emergence, the grain yield, the total biomass amount, and the dry matter yield of the corn plant ranged between 8000-8285 plant da-1, 1252-1369 kg da-1, 8066-8466 kg da-1 and 31.03-33.41 %, respectively (Table 6). The highest protein proportion (5.06%) was measured in the CT application, while the lowest protein proportion (3.81%) was measured in the MT3 application. In terms of protein proportion, the differences between CT, MT2 and DS applications and the differences between MT1 and MT3 applications were insignificant.

Nutrient elements composition of maize leaves

While the effects of the applications on the N, Zn and Mn contents of the corn leaf were significant (P<0.05), the effects on the P, Fe and Cu contents were insignificant (Table 7). The highest N content of corn plant (2.32%) was measured in DS method, while the lowest N content (1.82%) was measured in MT1 method. In terms of N content of the corn plant, the differences between CT, MT1, MT2 and MT3 methods and CT, MT2 and DS methods were insignificant. The highest Zn content (70.10 mg kg-1) was measured in MT2 application, the lowest Zn content (51.60 mg kg-1) was measured in DS method, the differences between CT, MT1 and MT2 methods and MT1, MT3 and DS methods were insignificant. The highest Mn content (77.20 mg kg-1) was measured in MT3 method, the lowest Mn content (50.43 mg kg-1) was measured in CT method, the differences between MT2, MT3 and DS and CT and MT1 methods were insignificant. While the N content of corn cob leaves was below the deficiency limit in all applications, P, Fe, Zn, Cu and Mn contents were measured adequately (Jones Jr 1999).

Table 6
Effects of different tillage practices on yield components of maize

Tillage practices	Seedling emergence (Plants da^{-1})	Grain yield (kg da^{-1})	Protein proportion (%)	Total biomass kg da^{-1}	Dry matter %
CT	8285±285*	1252±39.10*	5.06±0.47^{a**}	8066±464*	33.41±1.32*
MT1	8095±165	1301±88.10	3.91±0.09^{b}	8466±279	31.39±0.86
MT2	8000±285	1347±56.10	4.41±0.40^{ab}	8214±429	30.59±0.13
MT3	8285±285	1280±59.50	3.78±0.37^{b}	8295±780	31.03±1.55
DS	8000±285	1369±44.70	4.43±0.34^{ab}	8338±821	31.51±1.42

CT; conventional tillage, MT1; minimum tillage with subsoiling and chisel, MT2; minimum tillage with subsoiling, MT3; minimum tillage with chisel and DS; direct seeding, *; not significant, **; means (n = 3) (± standard errors) followed by the same letter in a column are not significantly different ($P \leq 0.05$).

Table 7
Effects of different tillage practices on nutrient elements composition of maize leaves

Tillage practices	N	P	Fe	Zn	Cu	Mn
	%			mg kg^{-1}		
CT	2.01±0.23^{ab**}	0.33±1.16*	47.57±10.70*	66.40±6.44^{a**}	17.80±0.76*	50.43±3.26^{c**}
MT1	1.82±0.15^{b}	0.39±0.55	55.70±18.10	59.67±6.10^{ab}	18.56±1.20	58.57±5.84^{bc}
MT2	2.12±0.05^{ab}	0.31±2.19	70.00±4.84	70.10±6.48^{a}	19.25±1.08	68.87±8.81^{ab}
MT3	1.83±0.04^{b}	0.29±0.58	65.30±27.40	52.26±1.44^{b}	18.96±1.66	77.20±4.59^{a}
DS	2.32±0.12^{a}	0.32±0.25	62.37±11.56	51.60±2.52^{b}	17.80±0.76	71.03±2.55^{ab}

CT; conventional tillage, MT1; minimum tillage with subsoiling and chisel, MT2; minimum tillage with subsoiling, MT3; minimum tillage with chisel and DS; direct seeding, *; not significant, **; means (n = 3) (± standard errors) followed by the same letter in a column are not significantly different ($P \leq 0.05$).

4. Conclusions

Soil tillage practices can have significant effects on the efficiency and quality of crop production by affecting the physical and in some cases chemical quality properties of soils. In addition to the effect of reduced tillage practices on soil properties, the cost-reducing effects also make their application widespread. Bulk density and penetration resistance values, which are indicators of soil compaction, are periodically affected by treatments. In the pre-planting period, direct sowing and conventional tillage applications produced higher bulk density values, while the conventional tillage method created higher bulk density values in the plant development period. It has been evaluated that this situation is caused by secondary field traffic such as hoe machine and agricultural spraying. Although tillage practices had negative effects on soil compaction, the amount of macropores did not fall below the limit value, and therefore, no significant changes occurred in the yield parameters of the corn plant. Since the chemical properties of the soil and the yield and yield elements of the corn plant are less affected by the applications, it has come to the fore that soil tillage applications that create less field traffic are preferred.

5. Acknowledgements

This study was taken from a research project supported by Selcuk University (S.U.) BAP Office (Coordinating Office of Scientific Research Projects, Project No: 20201030). The authors would like to thank the S.U.-BAP staffs.

6. References

Alaboz P (2020). Selecting soil properties for assessment of soil aggregation using principal component and clustering analyses, *Soil Research*.

Alskaf K, Mooney S, Sparkes D, Wilson P, Sjögersten S (2021). Short-term impacts of different tillage practices and plant residue retention on soil physical properties and greenhouse gas emissions, *Soil and Tillage Research*, 206: 104803.

Andrews S, Karlen D, Mitchell J (2002). A comparison of soil quality indexing methods for vegetable production systems in Northern California. *Agriculture, ecosystems & environment*, 90: 25-45.

Azimzadeh S, Kouchaki A, Bala M (2008). Study the effects of different plowing management on soil bulk density, porosity, moisture and wheat yield in a rainfed condition, *Journal of agricultural breeding*, 4 (3): 209-224.

Badalíková B 2010. Influence of soil tillage on soil compaction, In: Soil engineering, Eds: Springer, p. 19-30.

Bayraklı F (1987). Toprak ve bitki analizleri, *Ondokuz Mayıs Üniversitesi Yayınları* (17).

Beinat E, Nijkamp P (1998). Land-use management and the path towards sustainability. In: Beinat, E., Nijkamp, P. (eds) Multicriteria Analysis for Land-Use Management. Environment & Management, vol 9. Springer, Dordrecht. https://doi.org/10.1007/978-94-015-9058-7_1

Blake G, Hartge K (1986). Particle density, *Methods of soil analysis: part 1 physical and mineralogical methods*, 5: 377-382.

Blanco-Canqui H, Ruis SJ (2018). No-tillage and soil physical environment, *Geoderma*, 326: 164-200.

Blevins R, Cook D, Phillips S, Phillips R (1971). Influence of No-tillage on Soil Moisture 1, *Agronomy Journal*, 63 (4): 593-596.

Borges JA, Pires LF, Cássaro FA, Roque WL, Heck RJ, Rosa JA, Wolf FG (2018). X-ray microtomography analysis of representative elementary volume (REV) of soil morphological and geometrical properties, *Soil and Tillage Research*, 182: 112-122.

Burgos Hernández TD, Slater BK, Tirado Corbalá R, Shaffer JM (2019). Assessment of long-term tillage practices on physical properties of two Ohio soils, *Soil and Tillage Research*, 186: 270-279.

Bünemann EK, Bongiorno G, Bai Z, Creamer RE, De Deyn G, de Goede R, Fleskens L, Geissen V, Kuyper TW, Mäder P, Pulleman M, Sukkel W, van Groenigen JW, Brussaard L (2018). Soil quality – A critical review. *Soil Biology and Biochemistry*, 120: 105-125.

da Silva AP, Kay BD, Perfect E (1994). Characterization of the Least Limiting Water Range of Soils, *Soil Science Society of America Journal*, 58 (6): 1775-1781.

da Silva AP, Kay BD (1997). Estimating the Least Limiting Water Range of Soils from Properties and Management, *Soil Science Society of America Journal*, 61 (3): 877-883.

Doran JW, Parkin TB (1994). Defining and assessing soil quality. Defining soil quality for a sustainable environment, 1-21.

E. Hall H, L. Raper R (2005). Development and concept evaluation of an on-the-go soil strength measurement system, *Transactions of the ASAE*, 48 (2): 469-477.

Ehlers W, Köpke U, Hesse F, Böhm W (1983). Penetration resistance and root growth of oats in tilled and untilled loess soil, *Soil and Tillage Research*, 3 (3): 261-275.

Gajri PR, Arora VK, Chaudhary MR (1994). Maize growth responses to deep tillage, straw mulching and farmyard manure in coarse textured soils of N.W. India, *Soil Use and Management*, 10 (1): 15-19.

Gassel DK (1982). Tillage Effects on Soil Bulk Density and Mechanical Impedance, In: Predicting tillage effects on soil physical properties and processes, Eds, p. 45-67.

Gee GW, Bauder J, Klute A (1986). Methods of soil analysis, part 1, physical and mineralogical methods, *Soil Science Society of America, American Society of Agronomy*.

Gholami A, Asgari H, Zeinali E (2014). Effect of different tillage systems on soil physical properties and yield of wheat (Case study: Agricultural lands of Hakim Abad village, Chenaran township, Khorasan Razavi province).

Gomez A, Powers RF, Singer MJ, Horwath WR (2002). Soil Compaction Effects on Growth of Young Ponderosa Pine Following Litter Removal in California's Sierra Nevada, *Soil Science Society of America Journal*, 66 (4): 1334-1343.

Gugino BK, Abawi GS, Idowu OJ, Schindelbeck RR, Smith LL, Thies JE, Wolfe DW, Van Es HM (2009). Cornell soil health assessment training manual, Cornell University College of Agriculture and Life Sciences, p.

Gülser C, Candemir F (2012). Changes in penetration resistance of a clay field with organic waste applications, *Eurasian Journal of Soil Science*, 1 (1): 16-21.

Hellner Q, Koestel J, Ulén B, Larsbo M (2018). Effects of tillage and liming on macropore networks derived from X-ray tomography images of a silty clay soil, *Soil Use and Management*, 34 (2): 197-205.

Hernanz J, Peixoto H, Cerisola C, Sánchez-Girón V (2000). An empirical model to predict soil bulk density profiles in field conditions using penetration resistance, moisture content and soil depth, *Journal of Terramechanics*, 37 (4): 167-184.

J. Busscher W, E. Sojka R (1987). Enhancement of Subsoiling Effect on Soil Strength by Conservation Tillage, *Transactions of the ASAE*, 30 (4): 888-0892.

Jia C, Guo H, Yuan K, Yang Q, Sui X, Zhang Y, Fan Q (2010). Effects of different seeding manner on the seedling emergence, over-winter and yield of wheat under maize stalk full returned to the field, *Chin. Agric. Sci. Bull*, 26: 149-154.

Jones Jr JB (1999). Soil and Plant Analysis: Laboratory Registry for the United States and Canada, CRC Press, p.

Karlen DL (2004). Soil quality as an indicator of sustainable tillage practices, *Soil & Tillage Research*, 78 (2): 129-130.

Kutilek M (2004). Soil hydraulic properties as related to soil structure, *Soil and Tillage Research*, 79 (2): 175-184.

Lal R (1993). Tillage effects on soil degradation, soil resilience, soil quality, and sustainability, *Soil and Tillage Research*, 27 (1): 1-8.

Lhotský J, Ehrlich P, Váchal J (1984). Soustava opatření k zúrodňování zhutnělých půd, Ústav vědeckotechnických informací pro zemědělství, p.

Li H, Liao X, Zhu H, Wei X, Shao M (2019a). Soil physical and hydraulic properties under different land uses in the black soil region of Northeast China, *Canadian Journal of Soil Science*, 99 (4): 406-419.

Li Y, Li Z, Cui S, Jagadamma S, Zhang Q (2019b). Residue retention and minimum tillage improve physical environment of the soil in croplands: A global meta-analysis, *Soil and Tillage Research*, 194: 104292.

Lindsay W, Norvell W (1969). Equilibrium relationships of Zn2+, Fe3+, Ca2+, and H+ with EDTA and DTPA in soils, Wiley Online Library.

López-Fando C, Pardo MT (2009). Changes in soil chemical characteristics with different tillage practices in a semi-arid environment, *Soil and Tillage Research*, 104 (2): 278-284.

Martínez E, Fuentes J-P, Silva P, Valle S, Acevedo E (2008). Soil physical properties and wheat root growth as affected by no-tillage and conventional tillage systems in a Mediterranean environment of Chile, *Soil and Tillage Research*, 99 (2): 232-244.

McKyes E (1985). Soil cutting and tillage, Elsevier, p.

McLean E (1983). Soil pH and lime requirement, *Methods of soil analysis: Part 2 Chemical and microbiological properties*, 9: 199-224.

MGM (2020). Müdürlüğü, Meteoroloji Genel Başkanlığı, Araştırma Daire

Klimatoloji Şube Müdürlüğü Kalaba, Ankara, Türkiye.

Mitchell JK, Soga K (2005). Fundamentals of Soil Behavior 3rd ed., JohnWiley &Sons, *Inc. Foundation failure*.

Moraes M, Benez S (1996). Efeitos de diferentes sistemas de preparo do solo em algumas propriedades físicas de uma terra roxa estruturada e na produção de milho para um ano de cultivo, *Engenharia Agrícola*, 16 (2): 31-41.

Mujdeci M, Isildar AA, Uygur V, Alaboz P, Unlu H, Senol H (2017). Cooperative effects of field traffic and organic matter treatments on some compaction-related soil properties, *Solid Earth*, 8 (1): 189-198.

Neugschwandtner R, Liebhard P, Kaul H, Wagentristl H (2014). Soil chemical properties as affected by tillage and crop rotation in a long-term field experiment, *Plant, Soil and Environment*, 60 (2): 57-62.

Obour AK, Holman JD, Simon LM, Schlegel AJ (2021). Strategic Tillage Effects on Crop Yields, Soil Properties, and Weeds in Dryland No-Tillage Systems, *Agronomy*, 11 (4): 662.

Olsen S, Sommers L (1982). Phosphorus. In 'Methods of Soil Analysis'. 2nd edn.(Eds AL Page, RH Miller, DR Keeney) pp. 403–430, *Soil Science Society of America: Madison, WI, USA*.

Perkins D, Haws N, Jawitz J, Das B, Rao P (2007). Soil hydraulic properties as ecological indicators in forested watersheds impacted by mechanized military training, *Ecological Indicators*, 7 (3): 589-597.

Ren B, Li X, Dong S, Liu P, Zhao B, Zhang J (2018). Soil physical properties and maize root growth under different tillage systems in the North China Plain, *The Crop Journal*, 6 (6): 669-676.

Shaokun L, Keru W, Jukai F, Ruizhi X, Shiju G (2006). Factors affecting seeding emergence in sinter wheat under different tillage patterns with maize stalk mulching returned to the field, *Zuo wu xue bao*, 32 (3): 463-465.

So HB, Grabski A, Desborough P (2009). The impact of 14 years of conventional and no-till cultivation on the physical properties and crop yields of a loam soil at Grafton NSW, Australia, *Soil and Tillage Research*, 104 (1): 180-184.

Soil Survey Staff (1999). Keys to soil taxonomy, Pochahontas Press, Inc., Blacksburg, Virginia, USA.

Strudley MW, Green TR, Ascough II JC (2008). Tillage effects on soil hydraulic properties in space and time: State of the science, *Soil and Tillage Research*, 99 (1): 4-48.

Sutherland RA, Bussen JO, Plondke DL, Evans BM, Ziegler AD (2001). Hydrophysical degradation associated with hiking-trail use: a case study of Hawai'iloa Ridge Trail, O'ahu, Hawai'i, *Land Degradation & Development*, 12 (1): 71-86.

Şeker C (1999). Penetrasyon direnci ile bazı toprak özellikleri arasındaki ilişkiler, *Turkish Journal of Agriculture and Forestry*, 23 (supp3): 583-588.

Şeker C, Işıldar A (2000). Tarla trafiğinin toprak profilindeki gözenekliliğe ve sıkışmaya etkisi, *Turkish Journal of Agriculture and Forestry*, 24 (2000): 71-77.

Topa D, Cara IG, Jităreanu G (2021). Long term impact of different tillage systems on carbon pools and stocks, soil bulk density, aggregation and nutrients: A field meta-analysis, *Catena*, 199: 105102.

Voorhees W, Lindstrom M (1984). Long-term effects of tillage method on soil tilth independent of wheel traffic compaction, *Soil Science Society of America Journal*, 48 (1): 152-156.

Wang Y, Shao M (2013). Spatial variability of soil physical properties in a region of the Loess Plateau of PR China subject to wind and water erosion, *Land Degradation & Development*, 24 (3): 296-304.

Wright AF, Bailey JS (2001). Organic carbon, total carbon, and total nitrogen determinations in soils of variable calcium carbonate contents using a Leco CN-2000 dry combustion analyzer, *Communications in Soil Science and Plant Analysis*, 32 (19-20): 3243-3258.

Selcuk Journal of Agriculture and Food Sciences SJAFS

http://sjafs.selcuk.edu.tr/sjafs/index

(2022) 36 (3), 447-457

e-ISSN: 2458-8377

DOI:10.15316/SJAFS.2022.059

Research Article

The Effect of Different Nitrogen Doses on Yield and Quality of Some Winter Canola (*Brassica napus* L.) Cultivars**

Esra YILMAZ[1], Özden ÖZTÜRK[2,*]

[1] Selçuk University, The Graduate School of Natural and Applied Science, Department of Field Crops, Konya, Türkiye
[2] Selçuk University, Faculty of Agriculture, Department of Field Crops, Konya, Türkiye

ARTICLE INFO

Article history:
Received date: 25.10.2022
Accepted date: 14.11.2022

Keywords:
Winter rapeseed
Nitrogen dose
Seed yield
Quality

ABSTRACT

This research was carried out to determine the effect of different nitrogen doses on yield and yield components of winter canola cultivars in Konya conditions during the winter canola growing vegetation period of 2019-2020. Three winter canola cultivars (Linus, PR44W29, Es Neptune) were used as material and six different nitrogen doses (0, 50, 100, 150, 200, 250 kg N ha^{-1}) were applied. The research was established to according to the experimental design of split plots in randomized blocks. Half of the nitrogen fertilizer applications were given in the form of ammonium sulfate (21% N) at planting and the remaining half was given as urea (46% N) at the beginning of flowering. As a result of the study, it was determined that plant height, number of branch on the main stem, number of capsule per plant, capsule length, thousand seed weight seed yield, crude oil yield and crude protein ratio increased, while crude oil ratio decreased in winter canola varieties due to increasing nitrogen doses. As a matter of fact, the plant height values varied between 92.6 and 128.5 cm (respectively, LinusxN_{50} and PR44W29xN_{250}), number of branch (no. main $stem^{-1}$) of 2.4-5.7 (LinusxN_0-PR44W29xN_{250}), number of capsule (no. $plant^{-1}$) of 103.9-218.9 (Es-NeptunexN_0-PR44W29xN_{150}), capsule size of 5.3-6.6 cm (Es-NeptunexN_0-LinusxN_{250}), number of seeds in the capsule of 22.5-30.0 (Es-NeptunexN_{100}-PR44W29xN_{250}), thousand seed weight between 3.2 and 4.7 g (PR44W29xN_0-LinusxN_{200}), seed yield of 1361-5373 kg ha^{-1} (Es-NeptunexN_0-PR44W29xN_{250}), crude oil yield between 547.0 and 2466.0 kg ha^{-1} (Es-NeptunexN_0-PR44W29xN_{150}) and crude protein ratio values ranged of 16.2 23.9% (Es-NeptunexN_0-LinusxN_{200}).It was determined that there were increases in the values of these properties with increasing nitrogen doses in general. In terms of crude oil ratio, the lowest value was determined at PR44W29xN_0 with 39.2%, and the highest value was determined at PR44W29xN_{50} with 46.3%. It has been determined that there are relatively decreases in crude oil ratios with increasing nitrogen doses. The main purpose of cultivation of oil seed crops is to increase the oil yield per unit area. From this point of view, it was concluded that PR444W29 cultivar among the cultivars considered in the study would be more suitable in terms of oil yield with 150 kg N ha^{-1} application for the regions similar to the conditions in which the study was conducted.

1. Introduction

For an adequate and balanced diet, people need to meet the nutritional elements that contain a certain amount of protein, oil, carbohydrates, vitamins and minerals (Öztürk 2000). Among the nutrients, oils have a special importance in nutrition as an energy source. The daily calorie requirement of an adult person is 2800-3000. In a healthy and balanced diet, 30-35% (850-900 calories) of this energy should be taken from oils.1 g of oil is 9.3 calories, an person needs 95 g of oil daily (Kıllı&Beycioğlu 2019). According to FAO, it is stated that a person's annual oil consumption should not be less than 17 kg, otherwise important health problems will occur (Gizlenci et al 2019).

World's total oil production is obtained 87% from plant-based oil seeds and 13% from animal sources. Since the commercial production of animal-derived oils

* Corresponding author email: *ozdenoz@selcuk.edu.tr*
**It is the summary of Esra Yılmaz's Master Thesis

is expensive and these oils contain saturated fatty acids that have a harmful effect on human health, most of the fat needed is met from vegetable oils (Arıoğlu et al 2010). Production of oilseed plants has increased greatly in recent years. While world oilseed production in 2012 was 466.9 million tons, according to FAO 2019 data, world total oilseed production reached 1 billion 101 million tons. Soybean takes the first place in the production of oil crops in the world with 333.8 million tons. This is followed by cotton seed with 82.6 million tons, canola with 70.6 million tons and sunflower with 56.1 million tons (Anonymous 2021).

Vegetable oils constitute an important part of oil production in Turkey. According to FAO datas, a total of 6.390.412 tons of oilseeds were produced. Of this, 2.200.000 tons of cottonseed, 2.100.000 tons of sunflower, 1.525.000 tons of olive, 180.000 tons of canola, 169.328 tons of peanut and 150.000 tons of soybean, and 21.883 tons of safflower and other oilseed plants were obtained. Production of oil crops in our country follows a fluctuating course. While there is an increase in canola, sunflower and cotton, the production amount of oil crops such as safflower and flax decreases. For example; while the flax production in 2012 was 13 tons, there is no 2019 production value. While the amount of safflower production in 2015 was 70.0 thousand tons, there is a serious decrease as 21.3 thousand tons in 2020 (Anonymous 2021).

Although there has been an increase in the production of most oil crops over the years, the population in our country is increasing by almost 1.5% every year. For this reason, almost 70% of the needed oil is supplied by importing crude oil and oilseeds. As a result of this; our country has become foreign-dependent to meet the significant demand for vegetable oil, and billions of dollars of foreign currency are paid for imports. As a matter of fact, the amount of foreign exchange paid for oilseeds and their derivatives was 3.9 billion dollars according to 2020 figures (Kolsarıcı 2021). To meet our need for crude oil and oilseed plants; Planned agricultural policies are needed for the production and processing of oilseed plants (Perk Uçar 2011).

In order to close our vegetable oil deficit and to reduce our foreign dependency, the only solution is to increase the cultivation area, unit area yields and oil production of oil plants. Canola is one of the most produced oilseed crops in the world after soybean and cottonseed. Today, canola can be produced wherever grain is produced in our country. In this case, canola ranks first among the oil crops that can fill our vegetable oil deficit.

World canola production in 2019 is 83.9 million tons on a 40.6 million ha area. The most important producing countries; Canada (18 million tons), China (13 million tons), India (9 million tons), France (3 million tons) and Ukraine (3 million tons). Our country ranks 28th in terms of production. While the world canola yield was 2070 kg/ha in 2019, it was determined as 3430 kg/ha in our country. The provinces with the most common canola production in our country are; Tekirdağ, İstanbul, Konya, Edirne and Çanakkale (Anonymous 2021).

Canola is an important oil plant with winter and summer varieties belonging to the Cruciferae family, *Brassica* genus, with 36-50% oil and 16-24% protein in its seeds (Çorbacı 2011). The climate type, which is similar in our country as well as in countries with a continental climate, has the opportunity to be successfully grown in winter in regions with sufficient spring precipitation despite the low annual total precipitation and in soils with high water holding capacity (Öztürk 2000).

In canola, which has summer and winter varieties, summer varieties reach harvest maturity at the end of July and early August, and winter varieties at the end of June-early July. Most oil crops are not harvested during these periods. Thus, raw materials are provided to oil and feed factories operating at half capacity during these months. Another advantage of having an early maturation period is that it allows second crop farming.

High seed yield (2000-2500 kg ha^{-1}) and oil rate (45-50 %) per unit area are obtained from canola compared to other oil crops. The whole cultivation technique until harvest is suitable for mechanization. Prevent the development of weeds by developing rapidly in spring. Leaves clean soil for the next crop.

In addition to all these, the rise and fluctuations in oil prices have increased the demand for vegetable oils in search of an alternative fuel. Since canola oil has the closest composition to diesel, 80% of biodiesel is produced from canola oil today (Arıoğlu et al 2010).

When all these advantages are evaluated, canola is an important oil plant that will contribute to closing our current vegetable oil deficit. Fertilizer and water are the most important factors in canola cultivation, as in plant production. Fertilizer requirements, seed and oil yield of canola vary according to the amount of precipitation in the region or irrigation and plant cultivar. Determination of fertilizer forms suitable for the cultivar and region, as well as determination of agricultural costs and growing conditions are very important for seed and oil yield in canola.

As in most other plants, the most important plant nutrient in canola is nitrogen. Nitrogen requirement of canola is higher than grains. Nitrogen is a mobile element required at all stages of plant development. It contributes to the positive increase in yield factors such as plant height, number of branches, number of capsules, number of seeds and seed weight, and in parallel with all these, the yield per unit area also increases (Köymen 2018). However, excessive use of nitrogen fertilizers; when combined with soil and climatic factors, it causes high concentrations of nitrate and nitrite to accumulate in drinking, surface and underground waters, bringing human health and the environment to threatening levels. As a result of such unconscious practices, the sustainability of our lands is taking more risks every day and environmental pollution seriously threatens the life of living things (Kılıç&Korkmaz 2012).

The optimum amount of nitrogen needed by canola varies according to varieties (Öztürk&Ada 2009). As a matter of fact, the soil should have 200-250 kg ha^{-1} N content for 2500 kg/ha seed yield (Schjoerring et al 1995). In Argentina, it has been reported that 150 kg/ha of nitrogen application increases the total dry matter and seed yield in a soil with normal soil fertility (Sarandón et al 1996). The amount of nitrogen in fertilization, which is one of the most important elements for a high-yield and quality agriculture, is very important in canola, as it is in most plants. It is necessary to reveal this subject by researching on different regions and varieties (Köymen 2018).

This research was carried out in Konya conditions in order to examine the effects of different nitrogen doses on the yield of some varieties of winter canola, which is a potential oil plant for the Central Anatolia Region, and to determine the most appropriate nitrogen dose for seed yield.

2. Material and Methods

2.1.Material

The trial was established in Konya Province Karatay District Yarma Neighborhood land. Among the registered varieties obtained from Leading Farmer's Association and frequently cultivated in Konya conditions; winter canola cultivars Syngenta-Linus, Pioneer-PR44W29 and United Genetic-Es Neptune were used as material.

2.2.Method

This research, which was carried out to determine the effects of different nitrogen doses on yield and yield components of winter canola varieties in Konya conditions.

The temperature, precipitation and relative humidity values recorded in the growing season of the year (2019-2020) in which the experiment was conducted in Konya/Karatay district and the values of long-term averages are shown in Table 1. Total precipitation was 420.1 mm, average temperature was 6.8 ^{0}C and relative humidity was 71.8 % during the 2019-2020 growing period in which the research was conducted. Especially when the amount of precipitation falling in October of the trial year is 5.5 mm compared to the average of long years, irrigation was done for emergence and germination due to the insufficient rainfall.

Table 1
Some Meteorological Values of the Average of Long Years and October 2019-2020 in the Canola Growing Period (September-June) in Konya Province*

	Mean Temperature (°C)		Total Precipitation (mm)		Mean Relative Humidity (%)	
Months	Long Years	2019-2020	Long Years	2019-2020	Long Years	2019- 2020
September	18.6	15.7	11.3	16.3	46.1	48.5
October	12.4	12.9	29.7	5.5	58.5	60.8
November	5.5	8.2	39.0	30.1	70.1	69.2
December	1.3	1.6	43.9	86.6	76.5	86.3
January	-0.3	-3.1	30.8	53.1	76.0	94.7
February	0.6	-1.1	23.2	34.5	70.3	84.7
March	5.2	3.8	25.5	88.7	62.7	75.0
April	10.9	6.7	35.9	43.1	57.7	65.5
May	15.5	7.7	38.6	40.7	55.4	73.1
June	20.1	16.1	20.5	21.5	47.2	62.3
Total (mm)			298.4	420.1		
Mean	9.0	6.8			62.1	71.8

*Values have been prepared from the records of the Devlet Su İşleri Genel Müdürlüğü.

Soil samples were taken and analyzed before the experiment was set up from a depth of 0-30 cm from the trial area where the research was conducted. The soil of the land has a clay-loam structure, rich in potassium, insufficient in terms of organic matter and phosphorus. The trial land has a slightly alkaline, unsalted and calcareous structure.

This research was established according to the split plots in randomized blocks experimental design with 3 replications. In the study, cultivars (Linus, PR44W29, Es Neptune) were placed on the main plots, and nitrogen doses were randomly placed on the sub-plots (0, 50, 100, 150, 200 and 250 N kg ha^{-1}). Sowing was done by hand on the rows opened with a 1 cm depth at 25 cm row spacing on 20.09.2019. Each sub-plot was arranged as 6 rows of 3 m long.

In the study, phosphorus fertilizer in the form of TSP at 90 kg ha^{-1} P_2O_5 was applied to all plots together with planting. Half of all doses except control (0 kg ha^{-1} N) were planted in all sub-plots in the form of ammonium sulfate (21% N), and the remaining half was in the form of urea (46% N) at the beginning of flowering, to the determined nitrogen amounts (0, 50, 100, 150, 200 and 250 N kg ha^{-1}). Apart from the emergence irrigation, sprinkler irrigation was applied during the flowering and seed setting periods during the trial period.

Before entering the winter, the plants were hand thinning to 5 cm intra row spacing when they had 3-4

leaves. The final thinning and single treatments were made in the early spring at the exit of winter to be 15 cm above the row (Öztürk 2000). Weeds seen in the plots in the spring were cleaned by hand. Hoeing was done by hand and the soil was softened by aerating between rows. In each sub-plot, one row from each side and 50 cm from the plot heads were removed as an edge effect, and the remaining area was harvested manually on 03-07.2020.

In this study; plant height (cm), number of branch (no. main stem^{-1}), number of capsule (no. plant^{-1}), capsule length (cm), capsules on the main stem, number of seeds per capsule (g), seed yield (kg ha^{-1}), crude oil ratio (%), crude protein ratio (%) and crude oil yield (kg ha^{-1}) were investigated and analyzed. Measurements of morphological characteristics were carried out on 10 randomly selected plants from each plot.

The values obtained as a result of the research were subjected to variance analysis in the "MSTAT-C" statistical program according to the split plots in randomized blocks experimental design. The mean values of the transactions, whose differences were determined by performing the F test, were grouped according to the LSD significance test.

3. Results and Discussion

3.1. Plant Height

As could be seen from the examination of Table 2, the differences between cultivars, nitrogen doses and interaction of cultivar x nitrogen dose in terms of plant height were found to be statistically significant. In the study, plant height increased with increasing nitrogen doses in winter canola cultivars. As the average of nitrogen doses; the longest plant height was determined in Es-Neptune (112.6 cm) and PR44W29 (112.4 cm) cultivars, and the shortest in Linus (104.1 cm). Among the nitrogen doses considered as cultivar average, plant height was obtained from the longest 250 kg/ha nitrogen application (118.5 cm) and the shortest control plots (99.4 cm). In the study, the interaction of cultivarxnitrogen dose was statistically significant, and the longest plant height of 128.5 cm was determined in PR44W29, which applied 200 kg/ha of nitrogen. However, the differences between the values obtained in the interactions of PR44W29xN_{250} (126.8 cm), Es-NeptunexN_{200} (123.8 cm) and Es-NeptunexN_{250} (114.6 cm) were statistically insignificant and were included in the same group (a). The shortest plant height was determined as 92.6 cm with Linus in the plots where 50 kg/ha nitrogen was applied and formed the last group (e) (Table 2).

It is also stated in similar studies that increasing nitrogen doses increase plant height in winter canola. In the study conducted by Koç (2000) in Tokat conditions, it was reported that the shortest plant height was 87.5 cm in the control plots, and the longest 159.4 cm was obtained from 210 kg/ha N application. Başalma (1999) determined the plant height in the plots with the lowest nitrogen fertilizer application and the highest 160 kg/ha nitrogen application in the study conducted in Ankara. In other study conducted at different nitrogen doses in winter rapeseed varieties (Gürsoy et al 2019), it was determined that plant heights ranged from 125.40 cm (0 kg/ha N) to 131.02 cm (100 kg/ha N). Sana et al (2003) concluded that the variation in plant height of different varieties may be attributed to their genetic potential. Maestro (1995) and Reddy &Reddy (1998) reported that different brassica varieties differed significantly regarding their plant heights. With this study, it can be said that the differences reported in terms of plant height between the results of the research conducted by different researchers in different locations may have resulted from the climate, especially precipitation, temperature and soil conditions as well as the cultivar characteristics (Üstüner et al 2008; Öztürk 2000).

3.2.Number of Branch on The Main Stem

In terms of the number of branch on the main stem, the differences between cultivars, nitrogen doses and cultivar x nitrogen dose interaction were found to be statistically significant (Table 2). The highest number of branch was determined in PR44W29 with 4.4 per main stem, but the difference between Es-Neptune (4.3) was not statistically significant. Both varieties were included in the same group (a). The lowest number of branch on the main stem was determined in Linus cultivar with 3.6 and formed the last group (b).

As it can be seen Table 2, the highest number of branch in terms of nitrogen doses was obtained in 150 kg and 250 kg nitrogen applications per hectare with 4.9. Statistically, both applications were in the same group (a). The lowest number of branch on the main stem was determined as 3.0 in 0 kg/ha N plots and formed the last group (c).

When the interaction of cultivarxnitrogen dose was examined in terms of the number of branch on the main stem (Table 2), the highest value was determined in PR44W29 cultivar with 250 kg/ha nitrogen applied with 5.7. This value formed the first group (a). The lowest value was obtained in Linus cultivar with 2.4 in control plots and formed the last group (ı).

Table 2
Average values of the morphological characteristics of different nitrogen doses in winter canola cultivars and LSD test groups

Nitrogen Doses (kg/ha)							
Cultivars	N_0	N_{50}	N_{100}	N_{150}	N_{200}	N_{250}	Mean
Plant Height (cm)							
Linus	92.7 e**	92.6 e	108.2 bc	111.4 bc	105.5 bcd	114.2 b	104.1b**
PR44W29	97.8 de	102.0 cd	104.7 bcd	114.3 b	128.5 a	126.8 a	112.4 a
Es-Neptune	107.9 bc	110.4 bc	107.2 bcd	111.6 bc	123.8 a	114.6 a	112.6 a
Mean	99.4 d**	101.7 cd	106.7 c	112.4 b	119.3 a	118.5 a	109.7
	C V (%): 13.61	LSD $_{Cultivar}$: 6.62		LSD $_{Nitrogen\ Dose}$: 5.13		LSD $_{CultivarxN\ Dose}$: 8.89	
Number Of Branch (no. main stem^{-1})							
Linus	2.4 ı**	3.2 h	3.2 h	5.1 abc	3.4 gh	4.3 def	3.6 b**
PR44W29	3.5 fgh	3.6 fgh	4.1 d-g	5.4 ab	4.2 d-g	5.7 a	4.4 a
Es-Neptune	3.0 hı	4.4 cde	3.8 e-h	4.4 cde	4.6 cd	4.7 bcd	4.3 a
Mean	3.0 c**	3.7 b	3.7 b	4.9 a	4.0 b	4.9 a	4.1
	CV (%): 7.62	$LSD_{Cultivar}$: 0.44; $LSD_{Nitrogen\ Dose}$: 0.40; $LSD_{CultivarxNitrogen\ Dose}$: 0.69					
Number Of Capsule (no. plant $^{-1}$)							
Linus	128.4 ef**	108.9 gh	114.8 fgh	151.8 bcd	124.3 efg	145.8 cd	129.0 b**
PR44W29	125.9 ef	205.3 a	144.9 cd	218.9 a	165.5 b	217.8 a	179.0 a
Es-Neptune	103.9 h	108.6 gh	153.8 bc	161.1 bc	146.8 cd	135.8 de	135.0 b
Mean	119.4 d**	140.9 c	137.8 c	177.2 a	145.5 c	166.4 b	147.9**
	CV (%): 4.61	$LSD_{Cultivar}$: 21.48; LSD $_{Nitrogen\ Dose}$: 8.84; $LSD_{CultivarXNitrogen\ Dose}$: 15.31					
Capsule Length (cm)							
Linus	6.4 ab**	6.2 bcd	6.4 ab	6.5 ab	6.5 ab	6.6 a	6.4 a**
PR44W29	5.9 cde	6.1 bcd	5.7 e	6.5 ab	6.2 bc	6.2 bcd	6.1 a
Es-Neptune	5.3 f	5.6 ef	5.6 ef	5.6 ef	5.6 ef	5.8 de	5.6 b
Mean	5.8 c**	5.9 bc	5.8 c	6.2 a	6.1 ab	6.2 a	6.1
	CV (%): 2.63	$LSD_{Cultivar}$: 0.366; LSD $_{Nitrogen\ Dose}$: 0.205; LSD $_{CultivarXNitrogen\ Dose}$: 0.35					
Number Of Seeds In Capsule							
Linus	26.5	26.5	26.6	28.4	27.0	29.4	27.4 a**
PR44W29	27.4	27.6	27.5	31.1	29.0	30.0	28.7 a
Es-Neptune	23.7	23.7	22.5	25.2	24.2	24.7	24.0 b
Mean	25.9 b**	26.0 b	25.4 b	28.5 a	26.7 ab	28.0 a	26.7
		CV (%): 4.90	$LSD_{Cultivar}$: 1.414;	LSD $_{Nitrogen\ Dose}$: 1.696			

** Differences between the means shown with the same letters are not significant at the 1% level.

The high number of branch on the main stem in canola is an important feature that is desired because it increases the number of capsules in the plant and thus affects the seed yield and yield characteristics (Aytaç 2007). Variable number of branches per plant among different varieties, which have been related to be under genetic management control, has also been reported by Labana et al (1987) and Khehra &Singh (1988). As a result of this research; an increase in the number of branches was observed with increasing nitrogen ratios in winter canola. As a matter of fact, it supports our research; in the studies conducted by Başalma (1999), Koç (2000), Köymen (2018), it was reported that the highest number of lateral branches was obtained from the maximum nitrogen fertilizer dose.

3.4.Number Of Capsule Per Plant

In the study, in terms of the number of capsule per plant, the differences between varieties, nitrogen doses and cultivar x nitrogen interaction were found to be statistically significant (Table 2).

As a result of the study, the highest number of capsule in terms of cultivar average was determined in PR44W29 with 179.0 per plant and Linus with the lowest 129.0. However, the difference between Linus and Es-Neptune (135.0 per plant) was not found to be statistically significant and both cultivars were in the last group (b). In terms of nitrogen dose averages, the highest number of capsule per plant was determined at 177.2 with 150 kg/ha N application, and the lowest with 119.4 at 0 kg/ha N application (Table 2).

As it can be seen Table 2, the highest number of capsule in terms of cultivar x nitrogen dose interaction was determined in the PR44W29 cultivar with 150 kg/ha nitrogen applied with 218.9 per plant. This value formed the first group (a). The lowest was detected in Es- Neptune cultivar with 103.9 per plant and 0 kg/ha N applied and formed the last group (h).

In the study conducted by Öztürk&Ada (2009) on examining the relationship between different nitrogen doses and yield and morphological characteristics in summer rapeseed, it was determined that the relationship between seed yield and the number of capsule per

plant and nitrogen dose was positive and important. It has been stated that the number of capsules per plant has a high and positive direct effect on the seed yield.

In the study carried out in Ordu ecological conditions, the number of capsule was determined at the lowest dose of 186.17 per plant and 0 kg/ha N dose, while the highest dose was determined at 242.61 per plant and 200 kg/ha N dose (Köymen 2018). In the study conducted by Çorbacı (2011), in Tekirdağ conditions, the highest number of capsule (206.3 per plant) was determined in the plots where 150 kg/ha N was applied, and the lowest (158.5 per plant) in the control plots. These values are higher than our values and it can be said that the differences may be caused by the climate, soil structure and cultivar of the location where the studies are carried out.

3.5.Capsule Length

In terms of capsule lenght; between cultivars, nitrogen doses and cultivar x nitrogen interaction were found to be statistically significant. As the average of the winter canola cultivars used in the research, the highest capsule length was determined as 6.4 cm in Linus and 6.1 cm in PR44W29 cultivars, which are in the same statistically group (a). The shortest was determined in Es-Neptune with 5.6 cm and formed the other group (b). Among the nitrogen doses used as the average of the cultivars in the study, the longest capsule length was 6.2 cm, and the plots with 150 kg and 250 kg nitrogen per hectare were determined and formed the first group (a). The shortest is 5.8 cm with control and 100 kg /ha nitrogen applications were determined and formed the last group (c) (Table 2).

In terms of cultivar x nitrogen dose interaction, the highest capsule length was determined in Linus cultivar applied with 250 kg nitrogen per hectare with 6.6 cm(a), and the shortest capsule length was determined in Es-Neptune cultivar with 5.3 cm(f) in control plots (Table 2).

Çorbacı (2011) stated that the lowest capsule length varies between 5.31-6.03 cm in the control plots, and the capsule length increases with increasing fertilizer doses. In the study carried out by Başalma (1999) to examine the effect of nitrogen fertilization on yield and yield components of rapeseed in Ankara conditions, the average capsule lengths showed values varying between 5.67-6.69 cm, and it was determined that the average capsule length increased with the increase in nitrogen fertilizer doses. The values obtained as a result of this research regarding the capsule size were similar to the findings of the researchers mentioned above.

3.6. Number of Seeds in Capsule

Differences between cultivars and nitrogen doses in terms of the number of seeds in the capsule were statistically significant however the interaction of cultivar x nitrogen dose was found to be insignificant. In the study, the highest number of seeds in the capsule among the varieties was determined in PR44W29 with 28.7. However, the value was statistically in the same group (a) as Linus (27.4 seed/capsule). The lowest value was determined in Es-Neptune with 24.0 seed/capsule and formed the other group (b) (Table 2).

Among the nitrogen doses, the highest number of seeds in the capsule was determined in the application of 150 kg nitrogen per hectare, with 28.5 seeds. However, the difference between the value obtained from 250 kg/ha nitrogen application (28.0 seed/capsule) was not found to be statistically significant and they were included in the same group (a). The lowest was determined in the application of 100 kg nitrogen per hectare with 25.4 seed/capsule, but the difference between the control (25.9 seed/capsule) and 50 kg/ha nitrogen application (26.0 seed/capsule) was not found to be statistically significant, and they were in the same group (b).

As can be seen from the Table 2, although the interaction of the cultivar x nitrogen dose is insignificant in terms of the number of seeds in the capsule, the highest value was determined in PR44W29xN_{150} with 31.1 and Es-NeptunexN_{100} application with the lowest (22.5).

Koç (2000) reported that the number of seeds in the capsule varied between 18.33-28.33, Köymen (2018) 17.15-20.95, Başalma (1999) 27.8-25.4.The seed number values in the capsule found as a result of this study (between 22.5 and 31.1) are generally similar to the studies on this subject. It can be said that some differences may be due to varieties and climatic characteristics.

3.7.Thousand Seed Weight

As can be seen in Table 3, cultivars, nitrogen doses and cultivar x nitrogen interaction in terms of thousand seed weight were found to be statistically significant. Between the cultivars were determined with the highest 4.1 g in Linus and Es-Neptune, and they were in the same group (a). The lowest value was determined in PR44W29 with 3.7 g and formed the other group (b).

In terms of nitrogen doses, the highest thousand seedweight was determined with 4.2 g at 150 kg/ha and 200 kg/ha nitrogen applications. The difference between the values (4.0 g) obtained from the application of 50 and 250 kg nitrogen doses per hectare was not found to be statistically significant and they were all in the same group (a).

In terms of cultivar x nitrogen dose interaction, the highest thousand seed weight was determined in Linus (a) applied 200 kg nitrogen per hectare, the lowest in PR44W29 planted in control plots (h) with 3.2 g (Table 3).

A high thousand seed weight is an indication that the seeds are large. Therefore, in terms of seed yield and oil rate, it is desirable to have a high seed weight of thousand seed. It is thought that the higher the thousand seed weight, the higher the seed yield and oil content (Mert 2009). In the study carried out by Eryiğit (2005) in Şanlıurfa conditions, the weight of one thousand seeds in canola varied between 2.82-2.96 g. The highest thousand seed weight was 200 kg/ha N, and the lowest was determined in the control plots. It was stated that with

increasing nitrogen dose, thousand seed weight increased and there was a linear relationship between nitrogen doses and thousand seed weight. As a matter of fact, in this study, one thousand seed weight increased with increasing nitrogen doses, and it is thought that the differences between the studies may be due to the differences in the cultivar used, climate and cultural practices. Thus, several investigators (Munir &McNeilly 1992, Hashem et al 1998, Om et al 1998, Sana et al 2003, El-Nakhlawy&Bakhashwai 2009) found significant differences for 1000-seed weight among different brassica varieties.

3.8. Seed Yield

In terms of seed yield, the differences between cultivars, nitrogen doses and cultivar x nitrogen dose interactions were found to be statistically significant. In the study, among the winter canola varieties, the highest seed yield was found in PR44W29 with 3498 kg/ha, and the lowest in Es-Neptune with 2437 kg/ha (Table 3). Among the nitrogen doses, the highest seed yield was determined at 4162 kg/ha in 250 kg nitrogen application per hectare, and the lowest 1630 kg/ha in control plots. As can be seen in Table 3, the highest seed yield in terms of cultivar x nitrogen dose interaction was determined in PR44W29 cultivar with 250 kg/ha nitrogen applied with 5373 kg/ha, while the lowest 1361 kg/ha was determined in the control application of Es-Neptune cultivar.

Nitrogen (N) fertilizer increases yield by influencing a variety of growth parameters such as the number of branches per plant, the number of pods per plant, the total plant weight, the leaf area index. Also, it increases the number and weight of pods, seeds and flowers per plant, and overall crop assimilation, contributing to increased seed yield (Wright et al 1988 and Al-Barrak 2006). Reddy&Reddy (1998) and Khoshnazar et al (2000) found significant differences in seed yield among different varieties of brassica species. The significant and insignificant differences between the studied canola varieties in yield and yield components might be due to the genetic × environment interaction effects. Khoshanazar et al (2000), Kolte et al (2000) and Stringam et al (2000) compared different mustard and rapeseed cultivars and reported that all cultivars differed significantly for seed oil yields (El-Nakhlawy&Bakhashwai 2009). In the study conducted by Köymen (2018), a seed yield of 1410.6 kg per hectare was obtained from the control plots that were not applied nitrogen. It has been reported that the seed yield increased with increasing nitrogen doses and the highest figure was reached at 1977.0 kg per hectare and 200 kg/ha nitrogen application. Similarly, in the study conducted by Başalma (1999), it was stated that the lowest seed yield was obtained from the N_0 dose with 2443.7 kg/ha and the highest N_{120} dose with 2904.9 kg/ha. As a matter of fact, in this study, there was an increase in seed yield with increasing nitrogen doses. The differences between the results of the research in terms of yield values; ecological factors, cultivars used, cultural practices and nitrogen doses are thought to be caused by differences.

Table 3
Average values of thousand seed weight (g) and seed yield (kg/ha) at different nitrogen doses in winter canola cultivars and LSD test groups

	Nitrogen Doses (kg/ha)						
Cultivars	N_0	N_{50}	N_{100}	N_{150}	N_{200}	N_{250}	Mean
	Thousand Seed Weight (g)						
Linus	4.1 bcd**	4.3 abc	3.6 e-h	4.2 bcd	4.7 a	3.9 c-g	4.1a**
PR44W29	3.2 h	3.5 fgh	3.4 gh	4.3 abc	3.6 fgh	4.2 abc	3.7b
Es-Neptune	3.9 c-f	4.2 abc	3.7 ab	4.2 abc	4.5 ab	4.0 cde	4.1a
Mean	3.7b**	4.0a	3.6b	4.2a	4.2a	4.0a	3.9
	CV (%): 4.57 $LSD_{Cultivar}$: 0.097; $LSD_{Nitrogen}$: 0.235; $LSD_{CultivarxNitrpgen\ Dose}$: 0.407						
	Seed Yield (kg/ha)						
Linus	2099 ı**	1522 j	3185 def	2972 efg	3563 cd	4844 b	3031b**
PR44W29	1430 j	2940 efg	2720 fgh	40.6 c	4521 b	5373 a	3498a
Es-Neptune	1361 j	2617 gh	3042 efg	2105 ı	3232 de	2268 hı	2437c
Mean	1630e**	2360d	2982c	3027c	3772b	4162a	2989
	CV (%): 12.77 $LSD_{Çeşit}$: 213.7; $LSD_{Azot\ Dozu}$: 262.2; $LSD_{ÇeşitxAzot\ Dozu}$: 454.2						

** Differences between the means shown with the same letters are not significant at the 1% level.

3.9. Crude Oil Ratio

In terms of crude oil ratio, the differences between cultivars, nitrogen doses and cultivar x nitrogen dose interaction were found to be statistically significant (Table 4). In the study, the highest crude oil ratio was obtained in Linus (43.8%). However, the difference between PR44W29 (43.2%) was found to be statistically insignificant and both varieties were in the same group (a). The lowest value was determined in Es-Neptune with 41.6% and formed group (b). In terms of different nitrogen doses in winter canola cultivars, the average crude oil rate was found to be the highest with 44.3% in the application of 50 kg nitrogen per hectare. The differences between the values obtained from the plots applied 100 kg and 150 kg nitrogen per hectare (44.1% and 43.7%, respectively) were insignificant and were included in the same group (a). The lowest oil rate was determined as 41.1% in the control plots. However, the difference between the crude oil ratio values obtained in N_{200} and N_{250} doses (42.1% and 42.0%, respectively) was found to be statistically insignificant and they were in the same group (b) (Table 4).

Table 4
Average values of crude oil ratio (%, crude oil yield (kg/ha)) and crude protein ratio (%) at different nitrogen doses in winter canola cultivars and LSD test groups

Nitrogen Doses (kg/ha)							
Cultivars	N_0	N_{50}	N_{100}	N_{150}	N_{200}	N_{250}	Mean
Crude Oil Ratio (%)							
Linus	43.8 bc**	45.7 a	43.1 bc	44.4 abc	43.1 bc	42.5 cd	43.8 a**
PR44W29	39.2 e	46.3 a	43.7 bc	43.4 bc	42.5 cd	43.8 bc	43.2 a
Es-Neptune	40.2 e	40.8 de	45.1 ab	43.1 bc	40.8 de	39.5 e	41.6 b
Mean	41.1 b**	44.3 a	44.1 a	43.7 a	42.1 b	42.0 b	42.8
	CV (%): 1.89	$LSD_{Cultivar}$: 1.565	$LSD_{Nitrogen\ Dose}$: 1.051		$LSD_{CultivarxNitrogen\ Dose}$: 1.820		
Crude Oil Yield (kg/ha)							
Linus	918 fg**	696 gh	1373 cd	1322 cde	1536 c	2058 b	1317 b**
PR44W29	561 h	1364 cd	1192 de	2466 a	1923 b	2351 a	1643 a
Es-Neptune	547 h	1071 ef	1372 cd	908 fg	1319 cde	899 fg	1018 c
Mean	675 e**	1043 d	1312 c	1565 b	1593 b	1768 a	1326
	CV (%): 8.06	$LSD_{Cultivar}$: 73.6	$LSD_{Nitrogen\ Dose}$: 138.6		$LSD_{CultivarxNitrogen\ Dose}$: 240.1		
Crude Protein Ratio (%)							
Linus	16.9 gh**	17.3 fg	19.4 e	23.2 ab	23.9 a	22.8 b	20.6 a**
PR44W29	16.4 h	16.8 gh	19.6 e	20.9 cd	21.2 cd	21.6 c	19.4 b
Es-Neptune	16.2 h	16.6 gh	17.7 f	21.5 cd	20.7 d	21.1 cd	19.1 b
Mean	16.5 d**	16.9 c	18.9 b	21.8 a	21.9 a	21.8 a	19.7
	CV (%): 1.73	$LSD_{Cultiva}$:0.4680	$LSD_{Nitrogen\ Dose}$: 0.4396		$LSD_{CultivarxNitrogen\ Dose}$: 0.7614		

In the study, in terms of cultivar x nitrogen dose interaction, the highest crude oil rate was determined with 46.3%, PR44W29 and 50 kg/ha nitrogen application. There was no statistical difference between the value (45.7%) obtained from the 50 kg/ha nitrogen application of Linus and they formed the first group (a). The lowest value was determined with 39.2% in PR44W29 and 0 kg/ha nitrogen application. The difference between this value and the value determined in Es-Neptune cultivar (40.2%) at 0 kg/ha nitrogen application was not statistically significant and formed the last group (e) (Table 4).

Since canola is an oil crop, the main purpose of its cultivation is to obtain crude oil. For this reason, it is an important factor that is desirable that the oil rate in the seed is high.

In order to determine the effects of different nitrogen fertilizer doses applied to rapeseed cultivars on yield and yield components, Gürsoy et al (2019), it was reported that there were decreases in oil ratios with increasing nitrogen doses. The lowest crude oil rate was obtained in the control with 34.10%, and the highest was obtained with a nitrogen dose of 38.09% at 100 kg/ha. As a result of the study carried out by Koç (2000) for two years in Tokat conditions, the lowest oil rate was determined at 41.43% at 0 kg/ha, and the highest at 43.81% with 140 kg nitrogen per hectare plots. In the study conducted by Başalma (1999) in Ankara conditions, it was determined that increasing nitrogen doses caused reductions in oil ratios. In the study, the highest oil rate was determined with 51.17% in the application of 80 kg nitrogen per hectare, and the lowest in the control plots with 37.07%.

As a matter of fact, in this study, it was determined that there were fluctuations in oil ratios depending on the cultivars with increasing nitrogen doses in general. It was observed that the oil ratios decreased with increasing nitrogen doses after N_{150} in Linus and N_{100} in Es-Neptune. In the PR44W29 cultivar, it was determined that the oil ratio followed a fluctuating course after the N_{50} dose. It can be said that the differences seen in oil ratio between this study and other research results may be due to the differences between the cultivars used, the ecologies in which the research was conducted, and the nitrogen doses used. Similarly, Fernandez et al (1986) reported that nitrogen rates of 0-150 kg/ha had no appreciable effect on oil content but rates higher than 200 kg/ha reduced oil content by 8-9%.

3.10. Crude Oil Yield

In terms of crude oil yield, the differences between cultivars, nitrogen doses and cultivar x nitrogen dose interaction were found to be statistically significant. As an average of nitrogen doses, the highest crude oil yield was determined in PR44W29 with 1643 kg/ha in terms of cultivars and formed the first group (a). The lowest was detected in Es-Neptune with 1018 kg/ha and formed the last group (c) (Table 4).

As can be seen from the examination of Table 4, in terms of nitrogen doses, the highest crude oil yield was determined at 1768 kg/ha and 250 kg nitrogen application per hectare, and it was determined in the first group (a), while the lowest 675 kg/ha was determined in the control plots and formed the last group (e). In terms of cultivar x nitrogen dose interaction, the highest crude oil yield was determined in PR44W29 with 2466 kg/ha and 150 kg/ha nitrogen application, and with this value, 250 kg/ha nitrogen application was obtained in PR44W29. There was no statistical difference between 2351 kg/ha, which is the value obtained, and they formed the first group (a).The lowest crude oil yield was determined in

Es-Neptune planted in control plots as 547 kg/ha, and there was no statistical difference between this value and the value determined in PR44W29 (561 kg/ha) in nitrogen application at 0 kg/ha, and both values were combined. they formed the last group (h).

As a matter of fact, similar to the results of this research, in the study conducted by Eryiğit (2005) in Şanlıurfa conditions, it was found that increasing nitrogen doses increased the crude oil yield, the highest oil yield was 1108.6 kg/ha and 200 kg/da N application, the lowest 676.6 kg/ha control. reported from the plots. In the study carried out by Çorbacı (2011) for two years, the highest crude oil yield was found in the fertilizer application plots, and the lowest in the control plots. A similar result was reported in the study conducted by Öztürk (2010) to determine the effects of nitrogen ratios on yield and quality in winter rapeseed varieties.

3.11. Crude Protein Ratio

In terms of crude protein ratio, the differences between cultivars, nitrogen doses and cultivar x nitrogen dose interaction were found to be statistically significant (Table 4). As a result of the research, the highest crude protein ratio was determined in Linus with 20.6% in terms of varieties and formed the first group (a). The lowest value was determined in Es-Neptune with 19.1%, there was no statistical difference between this value and the value determined in PR44W29 (19.4%) and they formed the last group (b). In terms of nitrogen doses, the highest crude protein ratio was determined as 21.9% in 200 kg/ha nitrogen application. According to the LSD test, there was no statistically significant difference between the values determined at 250 kg/ha N and 150 kg/ha N doses (21.8% and 21.8%, respectively), and these three nitrogen doses together formed the first group (a). The lowest crude protein ratio was found in the control plots with 16.5% and formed the last group (d) (Table 4).

In the study, the highest crude protein ratio was found in Linus with 200 kg N application per hectare with 23.9% in terms of cultivar x nitrogen doses and formed the first group (a). The lowest was determined in the Es-NeptunexN_0 interaction with 16.2%, and there was no statistical difference between this value and the value (16.4%) obtained from the PR44W29xN_0 interaction and they formed the last group (h) (Table 4).

Başalma (1999) in Ankara conditions, it was reported that the protein ratio increased with increasing nitrogen ratios and these values ranged between 32.93% and 28.40%. Eryiğit (2005) conducted in Şanlıurfa conditions for two years, reported that with the increase of nitrogen dose, the protein ratio increased in both years, and the highest protein ratio was 21.65% in 200 kg nitrogen application per hectare, and the lowest 18.36% in control plots. As a matter of fact, as a result of this study carried out by us, it was determined that the protein ratio increased as the nitrogen doses increased.

It is a known fact that there is a negative relationship between oil ratio and protein ratio in oil crops (Atakişi 1999). Ilisulu (1973) reported that after oil, the most abundant substance in canola seeds is protein and it covers one-fifth of it overall. In our study, crude protein ratio according to the cultivars and nitrogen doses varied between 16.5% and 21.9%, and these values are in agreement with the values reported by Öztürk (2010) as 20.36-23.0% and by Eryiğit (2005) as 18.31-21.58%. As a matter of fact, although the effect of increasing nitrogen ratios on the crude oil ratio in our study was not very significant, the highest crude oil ratio was determined at the dose of 100 kg/N per hectare, while the highest protein ratio was obtained at the dose of 200 kg/N per hectare. In the studies conducted by Başalma (1999), Öztürk & Ada (2009), and Eryiğit (2005), it was reported that with increasing nitrogen doses, there was a decrease in the fat ratio, while the protein ratio increased. These results confirm the findings obtained as a result of this research conducted by us.

4. Conclusion

For both seed yield and oil yield, nitrogen fertilizer use is frequently preferred in canola as in all field crops. As a matter of fact, canola is an oil crop that increases yield in parallel with the use of fertilizers. Determining the correct nitrogen dose is very important in canola cultivation. It is very important to determine the optimum nitrogen ratio in canola when the high nitrogen application is evaluated in terms of the damage it causes to the country's economy due to the deterioration of the soil and environmental structure and increasing the producer cost.

As a result of the research, it was observed that increasing nitrogen fertilizer doses in winter canola caused significant increases in plant height, number of branches on the main stem, number of capsules per plant, number of seeds per capsule, capsule length and seed yield, but less increase in thousand seed weight. While the crude oil ratio in the cultivars changed inversely with increasing nitrogen doses, the protein ratio increased in parallel with the nitrogen doses.

The highest seed yield was determined in PR44W29xN_{250} application with 5373 kg/ha, crude oil ratio in PR44W29xN_{50} with 46.3%, crude oil yield in PR44W29xN_{150} application with 2466 kg/ha and LinusxN_{200} application with 23.9% crude protein.

According to the results of this research carried out on winter canola; in terms of both seed yield and oil yield; It was determined that PR44W29 was the most suitable cultivar for local conditions. In the study, it was determined that the cultivars showed different responses to increasing nitrogen doses in terms of seed yield and oil yield, and the interaction between cultivars and nitrogen doses was found to be significant.

The main purpose in the cultivation of oil crops is to increase the oil yield per unit area. As a result, it was concluded that for the regions similar to the conditions in which the study was carried out, planting with the PR44W29 cultivar in the form of 150 kg nitrogen application per hectare would be more appropriate in terms

of seed yield. However, this study is only one year, and the necessity of repeating the study in different years and locations should not be overlooked in terms of the reliability of the results.

5. Acknowledgements

A part of this research was presented as oral presentation at the "1. Uluslararası Fen Ve Uygulamalı Bilimler Kongresi" held on June 13-14, 2021 and published as a summary.

The study was supported by Selçuk University Scientific Research Projects Coordinatorship (BAP) allowances numbered as 20201056; and also, we would like to thank the BAP authorities for providing financial support to this research.

6. References

Al- Barrak KM (2006). Irrigation interval and nitrogen level effects on growth and yield of canola (*Brassica napus* L.), *Scientific Journal of King Faisal University* (Basic and Applied Sciences), 7(1): 87-103.

Anonymous (2021). FAOSTAT, www.fao.org, [Visit Date:26.03.2021].

Arıoğlu H, Kolsarıcı Ö, Göksu AT, Güllüoğlu L, Arslan M, Çalışkan S, Söğüt T, Kurt C, Arslanoğlu F (2010). Yağ bitkileri üretiminin artırılması olanakları, *Türkiye Ziraat Mühendisleri Birliği VII. Teknik Kongresi Bildiri Kitabı I, Sayfa*, 361-377.

Atakişi İ (1999). Yağ bitkileri yetiştirme ve ıslahı, *Trakya Üniversitesi Ziraat Fakültesi, Ders Kitabı No:10,* Yayın No:148, Tekirdağ.

Aytaç Z (2007). Bazı kışlık kanola (*Brassica napus ssp. oleifera* L.) çeşitlerinin tarımsal özellikleri ve Eskişehir koşullarına adaptasyonu, Doktora Tezi, *Osmangazi Üniversitesi Fen Bilimleri Enstitüsü* Eskişehir.

Başalma D (1999). Azotlu gübrelemenin kolzanın verim ve verim öğelerine etkisi, *Tarla Bitkileri Merkez Araştırma Enstitüsü Dergisi, Ankara.*, 8 (1-2), 59-67.

Çorbacı S (2011). Tekirdağ koşullarında mikrobiyal ve kimyasal gübre uygulamasının kolza (*Brassica napus ssp. oleifera* L.) bitkisinin verim ve kalite özelliklerine etkisi, Yüksek Lisans Tezi, *Namık Kemal Üniversitesi Fen Bilimleri Enstitüsü,*, Tekirdağ.

El-Nakhlawy FS, Bakhashwain AA (2009). erformance of Canola (*Brassica napus* L.) Seed Yield, Yield Components and Seed Quality under the Effects of Four Genotypes and Nitrogen Fertilizer Rates. *JKAU: Met., Env. & Arid Land Agric. Sci.,* Vol. 20, No. 2, pp: 33-47.

Eryiğit T (2005). Şanlıurfa ili koşullarında bazı kolza çeşitleri için optimum azot dozu ve tohumluk miktarının belirlenmesi, Doktora Tezi, *Yüzüncü Yıl Üniversitesi*, Van.

Fernandez PM, Peyrelongy CA, Contreras FE (1986).The influence of fertilizer application on oil content of rape, *Field Crop Abst*., 39: 798.

Gizlenci Ş, Acar M, Aygün Y (2019). Alternatif bir yakıt biyodizel ve ülkemiz açısından önemi, *Karadeniz Tarımsal Araştırma Enstitüsü,* Samsun.

Gürsoy M, Turan F, Başalma D (2019). Kolza (*Brassica napus* L.) çeşitlerine uygulanan farklı azotlu gübre dozlarının verim ve verim öğelerine etkileri. . *IV. INSAC International Agriculture, Forest and Aquaculture Sciences Congress*, Proceedings Book 11-13.

Hashem A, Majumdar MNA, Hamid A, Hossain MM (1998). Drought stress effects on seed yield, yield attributes, growth, cell membrane stability and gas exchange of synthesized *Brassica napus* L., J. *Agron. Crop Sci.,* 180: 129-136.

İlisulu, K (1973). Yağ bitkileri ve ıslahı, *Çağlayan Kitabevi, İstanbul*, 254-255.

Khoshanazar PR, Ahmadi MR, Ghanndha MR (2000). A study of adaptation and yield capacity of rapeseed (*Brassica napus* L.) cultivars and tines, *Iranian J. Agri. Sci*., 31: 341-352.

Kılıç R, Korkmaz K (2012). Kimyasal gübrelerin tarım topraklarında artık etkileri, *Biyoloji Bilimleri Araştırma Dergisi*, 5 (2), 87-90.

Kıllı F, Beycioğlu, T (2019). Türkiye'de ve Dünyada yağlı tohum ve ham yağ üretim durumu Türkiye yağlı tohum üretimine ilişkin önemli sorunlar, *Uluslararası Anadolu Ziraat Mühendisliği Bilimleri Dergisi*, 1 (5), 17-33.

Khehra MK, Singh P (1988). Sensitivity and performance of some *Brassica napus* genotypes in stress and non-stress environments, *Crop Improvement Ind*., 15: 209-211.

Khoshanazar PR, Ahmadi MR, Ghanndha MR (2000). A study of adaptation and yield capacity of rapeseed (*Brassica napus* L.) cultivars and tines, Iranian J. Agri. Sci., 31: 341-352.

Koç H (2000). Bazı kışlık kolza (*Brassica napus* L. *ssp. oleifera*) çeşitlerinde azotlu gübreleme, *Gaziosmanpaşa Üniveritesi Ziraat Fak. Dergisi*,, 17(11): 83-88.

Kolsarıcı Ö (2021). Dünyada ve Türkiye'de yağlı tohumlu bitkiler yağlı tohum ticareti, Türkiyenin bitkisel yağ açığı ve çözüm önerileri, *Tohum Türked Dergisi*, 26, 6-20.

Kolte SJ, Awasthi RP, Vishwanath R (2000). Divya mustard: a useful source to create Alternaria black spot tolerant dwarf varieties of oilseed Brassica, *Plant Varieties and Seeds* 13: 107-111.

Köymen M (2018). Azotun kışlık kolza çeşitlerinde verim ve verim unsurları üzerine etkisi Yüksek Lisans Tezi, *Ordu Üniversitesi Fen Bilimleri Enstitüsü*, Ordu.

Labana KS, Ahuja KL, Banga SS (1987). Evaluation of some Ethiopian mustard (*Brassica carinata*) genotypes under Indian conditions, *In: 7th International Rapeseed Congress*, Poznan, Poland, p. 115.

Maestro G (1995). Rape, Metapontum area, *Informatore Agrario*, 51: 26-27.

Munir M, McNeilly T (1992). Comparison of variation in yield and yield components in forage and winter oilseed rape, *Pak. J. Agri. Res*., 13: 289-292.

Perk Uçar Y (2011). Kanola (*Brassica napus* L.) çeşitlerinin azot etkinliğinin karşılaştırılması, Yüksek Lisans Tezi, *Ankara Üniversitesi Fen Bilimleri Enstitüsü Toprak Bilimi ve Bitki Besleme,* Ankara.

Om P, Das TK, Singh HB, Singh N (1999). Performance of three *Brassica* species as affected by time of sowing and nitrogen, I. Yield attributes and yield, *Annals Agri. Res*., 20: 448-454.

Öztürk, Ö (2000). Bazı kışlık kolza çeşitlerinde farklı ekim zamanı ve sıra arası uygulamalarının verim, verim unsurları ve kalite üzerine etkileri, Doktora Tezi, *Selçuk Ünivesitesi Fen Blimleri Enstitüsü*, Konya.

Öztürk Ö, Ada R (2009). Yazlık kolzada farklı azot dozlarının verim ile morfolojik özellikler arasındaki ilişkilerin incelenmesi, *Selçuk Tarım ve Gıda Bilimleri Dergisi*, 23 (49), 24-29.

Öztürk Ö (2010). Effects of source and rate of nitrogen fertilizer on yield, yield components and quality of winter rapeseed (*Brassica napus* L.), *Chilean journal of agricultural research*, 70 (1), 132-141.

Reddy CS, Reddy PR (1998). Performance of mustard varieties on alfisols of Rayataseema Region of Andhra Pradesh, *J. Oilseeds Res*., 15: 379-380.

Sana MA, Ali M, Malik A, Saleem MF, Rafiq M. (2003). Comparative yield potential and oil contents of different canola cultivars (*Brassica napus* L.), *Pakistan Journal of Agronomy*, 2(1):1-7.

Sarandón SJ, Chamorro AM, Tamagno LN, Bezus R (1996). Respuesta de la colza-canola (*Brassica napus* L. *sp. oleifera forma annua*) a la fertilización con N a la siembra: Efecto sobre la acumulación y partición de la materia seca, el rendimiento y sus componentes, *Revista de la Facultad de Agronomía*, 101.

Schjoerring J, Bock J, Gammelvind L, Jensen C, Mogensen V (1995). Nitrogen incorporation and remobilization in different shoot components of field-grown winter oilseed rape (*Brassica napus* L.) as affected by rate of nitrogen application and irrigation, *Plant and Soil*, 177 (2), 255-264.

Stringam GR, Degenhardt DF, Thalgarajah MR, Bansal VK, Hawkins GP (2002). Kelsey summer rape, *Can. J. Plant Sci.*, 82: 559-560.

Üstüner N, Kolsarıcı Ö, Kaya M (2008). Farklı gelişme dönemlerinde uygulanan azotlu gübre formlarının kışlık kolza (*Brassica napus ssp*. *oleifera* L.)'nın verim ve verim öğelerine etkileri, *Tarla Bitkileri Merkez Araştırma Enstitüsü Dergisi*, 17 (1-2).

Wright GC, Smith CJ, Woodroffe MR (1988). The effect of irrigation and nitrogen fertilizer on rapeseed (*Brassica napus*) production in South- Eastern Australia, I. Growth and seed yield, *Irrig. Sci*.,9:1-13.

Selcuk Journal of Agriculture and Food Sciences

http://sjafs.selcuk.edu.tr/sjafs/index

Research Article

SJAFS
(2022) 36 (3), 458-465
e-ISSN: 2458-8377
DOI:10.15316/SJAFS.2022.060

Profitability Analysis of Catfish (*Clarias gariepinus*) Production in Edo State, Nigeria

Osagie Stephen IMADE[1*], Joseph AHMADU[1]

1Benin University, Department of Agricultural Economics and Extension Services, Benin City, Nigeria

ARTICLE INFO

Article history:
Received date: 02.05.2022
Accepted date: 17.11.2022

Keywords:
Cost
Revenue
Constraints
Farmers
Catfish
Nigeria

ABSTRACT

The continuous importation of fish portends a colossal loss of foreign exchange reserved to Nigeria which requires urgent attention to boost fish production. It is against this backdrop that this study analyzed the profitability analysis of catfish production in Edo State, Nigeria. It specifically describe the production characteristics of the catfish farmers, estimate the input and output quantities of catfish, determine the profitability and identify the constraints associated with catfish production in the study area. Multi-stage sampling procedure was employed to select a total of 468 catfish farmers from the study area. Data collection was achieved through the administration of structured questionnaire. Data analysis was done using descriptive statistics, budgetary techniques and 4-Point Likert-type scale. The results revealed that the farmers in the study area used more of personal savings (58.12%), family land (59.40%) and local feed (82.48%). Stocked more at juvenile stage (70.30%) and produced average output of 4859.51kg per production cycle. The results also showed the average total cost incurred and revenue realized were ₦ 543.67 and ₦ 752.56 per kg fish respectively. The catfish production was profitable with average gross margin, net profit and return per naira invested of ₦224.35, ₦208.90 and ₦1.38 per production cycle respectively. The major constraints faced by catfish farmers were high cost of food (3.96), lack of capital (3.65) and lack of inadequate power supply (3.51) are very serious constraints among others. Since the catfish production was profitable, the farmers should be encouraged to combat these constraints and expand their holdings to boost production.

1. Introduction

There has been growing concerns in the recent years at the low level of animal protein intake which is essentially higher in quality than that of plant as it contains all essential amino acids for growth (Ahmadu, Boheje Odum and Osariemen, 2021). Fish happens to be the cheapest source of protein available to man (Imade and Ogieva 2022) and fish production is often viewed as one of the means of increasing food production in food deficient countries like Nigeria. According to Odioko & Becer, (2022), "the Nigerian Minister of Agriculture and Rural Development, stated that Nigeria's total fish production is estimated at 1.123 million metric tonnes (Vanguard, 2021), to which marine catches contributed 36 per cent, inland waters catch contributed 33 per cent and aquaculture 31 per cent (FAO, 2021). Akinsorotan et al., (2019) reported that the yearly fish demand of Nigeria is about 2.1 million metric tons with Nigeria only able to meet up just about 38.1% of its fish needs and depends on imports to cover the shortfall of about 61.9% of its population need yearly. In 2021, the Nigerian Ministry of Agriculture and Rural Development put the fish demand of the country at 3.6 million metric tonnes of which the country only meet up about 31.19% and depends on importation to meet up the huge gap of about 68.80% (Vanguard, 2021). Fish remains an important dietary element for Nigeria, especially in the southern part of the country where fish is highly valued and one of the cheapest sources of animal protein available to many Nigerians (FAO, 2021). The fishery sector is 1.09% of the national GDP in 2020 and 0.97% in the Q3 of 2021 (NBS, 2021)".

Many species of fish are cultivated all over the world, but catfish is taking the lead due to its uniqueness (Ahmadu and Egbodion 2017). The favoured catfish cultured in Nigeria is *Clarias gariepinus*. *Clarias gariepinus* is regarded as an excellent aquaculture species because it grows fast and feeds on a variety of agricultural by-products; it is hardy and can tolerate extreme temperature, easy to produce in captivity with high annual production, good feed conversion rate and

* Corresponding author email:*osagieimade@yahoo.com*

healthy for human consumption. Most consumers prefer catfish to other fish species because of its low calorie value, low carbohydrate content, high protein, low fat, low bone content and fine flavor (Ahmadu and Egbodion 2017). In addition, it is quick and easy to prepare and above all, it has great taste. These qualities coupled with its high growth rate, its ability to feed on virtually anything and the fact that its market value is higher than those of other fish species such as tilapia makes the catfish the pride of most fish farmers in Nigeria (Imade and Ogieva 2022).

Catfish has the potential to contribute to sustainable development and poverty reduction in Nigeria by generating income and employment. The production of Catfish as an economic resource is undertaken by a large number of people especially the small-scale farmers in Nigeria (Alawode and Ajagbe 2020). The Nigerian vast aquatic medium comprising numerous water bodies like rivers, streams, lake reservoirs, flood plains, irrigation canals, and coastal swamps offer great potentials for catfish production in Nigeria. The United Nation (UN) noted in its 2016 State of World Fisheries and Aquaculture report that nearly a third of wild stocks are overfished (FAO 2016). Thus, there is need to annexed the potentials of catfish production to fill this gap. This can only be possible if adequate information on the profitability of the production is known. It is in this view that this study seeks to analyze the profitability of catfish production in Edo State of Nigeria.

The specific objectives of the study are to: describe the production characteristics of the catfish farmers; estimate the input and output quantities of catfish production; determine the profitability of catfish production; and identify the constraints associated with catfish production in Edo States, Nigeria.

2. Methodology

Study Area

The study was carried out in Edo State of Nigeria. Edo State is located in the south-south region of Nigeria with Benin City as capital. The State is found in the forest zone of the country. Edo State was created on August 27, 1991 from former Bendel State. The popular language groups include Edo (Binis), Esan, Afemai (Owan/Etsako), Ora, Akoko Edo and Ibibio. The state lies between latitudes 05^{o} 44'N and 07^{o} 34'N of the Equator and longitudes 06^{o} 04'E and 06^{o} 43'E of the Greenwich Meridian. It covers land area of 17,802km^{2} with a projected population in 2018 of 4,592,935 people (Imade and Ahmadu. 2022). The tropical region is characterized by two distinct seasons: the wet season with a mean temperature of 25^{o}C, which starts by April and ends by October and the dry season with a mean temperature of 28^{o}C, which is from November to March. Relative humidity and rainfall are high giving rise to thick vegetation cover with an average rainfall in the range of 1500 - 2500mm (Ebomwonyi, Omorogie, Noutcha, Abajue and Okiwelu, 2019).

The major occupation of the inhabitants of the State is agriculture. Agricultural practices carried out in the area include arable and tree crops production, fishing, snailry, aquaculture, poultry and livestock rearing. Edo State is divided into three Agricultural zones according to Agricultural Development Programme (ADP) delineation as follows: Edo Central, Edo North and Edo South zones. Edo Central is divided into five Local Government Areas (LGAs) as follows: Esan Central, Esan West, Esan North-East, Esan South-East and Igueben LGAs. Edo North comprises six LGAs, namely, Owan West, Akoko Edo, Etsako West, Etsako East, Owan East and Etsako central LGAs. Edo South consists of seven LGAs, namely, Oredo, Ovia South West, Ovia North East, Ikpoba-Okha, Egor, Uhunmwode and Orhionmwon LGAs. In all, there are a total of Eighteen Local Government Areas (LGAs) in Edo State. The State is blessed with freshwater swamp and others coastine areas that is sultable for fish farmers (Ahmadu, Boheje Odum and Osariemen, 2021)

Experimental Design and method of data analysis

The data for the study were drawn from a population of 570 catfish farmers using structured questionnaire in Edo State. Multistage sampling procedure was adopted in selecting farmers for the study. The first stage involved a purposive random sampling of two Local Government Areas (LGAs) each from the various Agricultural Zones where catfish production is dominant in the state. Oredo and Ikpoba-Okha LGAs were from Edo South, Esan North-East and Esan South-East LGAs from Edo Central, and Owan West and Owan East LGAs from Edo North Agricultural Zone. A total of Six LGAs were sampled from State.

Snowballing sampling technique was adopted in the second stage where catfish farmers in the various LGAs were identified in addition to the list obtained from Agricultural Development Programme (ADP) zone in state for this study.

The formula for determining the sample size according to Ryan (2013), was used to select the respondents in the third stage,. This formula is expressed as

$$n = N/(1 + Ne\hat{}2) \quad (1)$$

Where n = Corrected Sample Size, N = Population Size, e = Margin of Error (MoE), e = 0.05 or 5%.

The Catfish Farmers drawn from the LGAs using the sample size formula gave a total sample size of 487 Catfish Farmers used for this study. However, a total of 468 respondents provided useful information for data analysis. The data covered the quantities of inputs and outputs of catfish production and their respective prices. The constraints faced by the farmers were also sourced. Data collected were analyzed using descriptive statistics, budgetary analysis and Likert scale.

Descriptive statistics used include means, frequency count, percentages and standard deviation in tables was used to describe the production characteristics of the catfish farmers and examine the input and output

quantities of catfish production. Budgetary analysis was employed to determine the profitability analysis of catfish production per production cycle.

Model specification

The budgetary tools used are gross margin, net profit and return on investment as adapted from Ahmadu and Egbodion (2017), it is specified as follows:

$$GM = TR - TVC \quad (2)$$

$$TC = TVC + TFC \quad (3)$$

$$Gross\ Ratio = TC/TR \quad (4)$$

$$TR = P*Q \quad (5)$$

$$NFI = \Sigma P_{Yi}Y_i - \Sigma P_{Xj}X_j - \Sigma Fk \quad (6)$$

Where,

GM = Gross Margin (NGN)

TR = Total Revenue (NGN)

TVC = Total Variable Cost (NGN)

TFC = Total Fixed Cost (NGN)

TC = Total Cost (NGN)

P = Market Price (NGN)

Q = Total Quantity (KG)

NFI = Net farm Income (NGN)/production cycle;

P_{Yi} = Unit price of the output of catfish (NGN)

Y_i = Total output of catfish (KG);

P_{Xj} = Unit price of variable inputs (NGN)

X_j = Quantity of variable inputs (where j = 1,2,3……n)

Fk = Cost of fixed inputs (NGN) (where k = 1,2,3….n)

Σ = Summation sign.

Estimates of the operating ratio reveal the level of liquidity in the catfish production business. It is estimated as the percentage of the ratio of total variable costs to the total revenue derived from the business. The Benefit-Cost (B/C) ratio measures the relationship between the costs borne to the benefits obtained from a business venture.

$$ROS = \pi/TR \quad (7)$$

$$OR = TVC/TR \quad (8)$$

$$B/C = TR/TC \quad (9)$$

$$ROI = \pi/TC \quad (10)$$

$$\%ROI = 100*ROI \quad (11)$$

Where,

ROS = Return on Sales (NGN)

OR = Operation Ratio

B/C = Benefit-Cost Ratio

ROI = Return on investment (NGN)

%ROI = Percentage Basic of ROI

The analyses of constraints associated with catfish production in the study area were identified using 4-point Likert-type scale (Likert 1932). The response to the various constraints were served in such a way that the response indicating the most serious constraints were given the highest scale (that is 4) as a 4-point scale, the responses were grouped into 4, that is:

Very Serious (VS) = 4

Serious (S) = 3

Not Serious (NS) = and

Not a Problem (NP) = 1

for a given constraint. A mean score (greater than) > 2.5 were regarded as serious constraints.

3. Results and Discussion

3.1. Socio-economic Characteristics of Catfish Farmers

The results of the study presented in Table 1 indicate that males dominated the catfish production in the study area and this is in agreement with the finding of Ajiboye, Adekunmi, Osundare, Oluwatusin, Toluwase and Amao (2020). This may be connected to the rigorous nature of catfish production activities which requires physical strength and men are known to be more physically endowed. Numa, Obayelu, Sanusi and Bada (2017) also support the males dominating the business in Delta South agricultural zone.

The age distribution of the respondents showed that most of the catfish farmers were relatively young; hence can contribute to improvement in their catfish business. According to Ahmadu, Boheje Odum and Osariemen (2021), catfish farmers in this age category have the propensity for improved agricultural production. Majority (76.71%) of the respondents were married with high household sizes. This emphasizes the advantage of higher family labour to single-member household catfish farmers as corroborates by Olajide and Omonona (2019). Table 1 also showed that most of the farmers were literate with 96.58% of them having at least secondary education. This result confirms the findings of Ahmadu, Boheje Odum and Osariemen (2021) who reported that most catfish farmers in their study area were literate. The result compares favorably with Olajide and Omonona (2019) who reported that the years of education enhance the farmers' capacity to interprets and apply information on improved farm practices. Majority of catfish farmers (66.03%) operate on part time basis. This implies, the farmers had other supporting occupation and catfish production were secondary occupation for them as means of increasing their household income. This is contrary to the findings of Yaqoob and Fasakin (2021) that farming is a major occupation for self-reliance and income generation. As regards production experience, about 52% of the respondents had over 8 years of experience. According to the submission of Olajide and Omonona (2019), the ability to manage catfish ponds efficiently depends on the years of experience, which they found was directly related to the total productivity of the farm.

Table 1
Socio-Economic Characteristics of Catfish Farmers in Edo State.

Variables	Edo State							
	Edo South		Edo Central		Edo North		Total	
	Freq (186)	% (100)	Freq (138)	% (100)	Freq (144)	% (100)	Freq (468)	% (100)
Sex								
Male	138	74.19	100	72.46	72	50.00	310	66.24
Female	48	25.81	38	27.54	72	50.00	158	33.76
Age (years)								
24 – 35	45	24.19	34	24.64	60	41.67	139	29.70
36 – 47	95	51.08	84	60.87	54	37.50	233	49.79
48 – 59	40	21.51	18	13.04	30	20.83	88	18.80
60 – 71	6	3.23	2	1.45	-	-	8	1.71
Mean	41.13		39.29		39.14		39.98	
Std. Dev.	8.48		7.40		7.63		7.95	
Marital status								
Single	27	14.52	22	15.94	22	15.28	71	15.17
Married	153	82.26	114	82.61	92	63.89	359	76.71
Separated	3	1.61	2	1.45	10	6.94	15	3.21
Widow	-	-	-	-	4	2.78	4	0.85
Divorce	3	1.61	-	-	16	11.11	19	4.06
Labour Type								
Family	45	24.19	24	17.39	14	9.72	83	17.73
Hired	33	17.74	24	17.39	46	31.95	103	22.01
Both	108	58.07	90	65.22	84	58.33	282	60.26
Household Size								
1 – 4	100	53.76	116	84.06	72	50.00	288	61.54
5 – 8	80	43.01	20	14.49	50	34.72	150	32.05
9 – 12	6	3.23	2	1.45	22	15.28	30	6.41
Mean	4		3		5		4	
Level of Education								
non-formal	-	-	-	-	2	1.39	2	0.43
Primary	4	2.15	-	-	10	6.94	14	2.99
Secondary	37	19.89	16	11.59	48	33.33	101	21.58
Tertiary	145	77.96	122	88.41	84	58.33	351	75.00
Nature of Catfish Production								
part-time	117	62.90	90	65.22	102	70.83	309	66.03
full-time	69	37.10	48	34.78	42	29.17	159	33.97
Production Experience								
1 – 7	112	60.22	97	68.12	20	13.89	226	48.29
8 – 14	67	36.02	42	30.43	102	70.83	211	45.09
15 –21	7	3.76	2	1.45	22	15.28	31	6.62
Mean	6.31		6.28		9.42		7.25	
Std. Dev.	3.32		2.46		2.60		3.21	

Source: Computed from Field Survey, 2021

3.2. Production Characteristics of Catfish Farmers

Table 2 showed the production characteristic of catfish farmers who participated in catfish production. The results revealed that the percentage of the farmers who used personal savings was 59.14%, 63.77%, 51.39% and 58.12% in Edo-South, Edo-Central, Edo-North and Edo State respectively. This implies that majority of the catfish farmers in the study area fund their production with their personal savings. This agrees with Michael and Duru (2020) where they reported that 80.80% of the respondents financed their fish farm by themselves. In fact, the need to increase farmers' access to credit is necessary in order to ensure agricultural development (Imade and Ogieva 2022). The results also revealed that the majority of the catfish farmers used family land (59.40%) in Edo State then, followed by the leased/rented land (26.28%). This majority of family lands indicate more access to cheap and low cost farming land used for catfish business.

Various types of pond culture systems were used for catfish production in the study area. 40.60% of the catfish farmers used concrete pond, followed by plastic/tarpaulin pond (36.75%) and earthen pond (22.65%). Thus, the concrete pond types were the most common used in the study area. This finding agrees with Imade and Ahmadu. (2022) who stated that majority of fish farmers in Edo State used concrete ponds for fish production. The use of concrete pond might be due to the convenience offered, in being easy to clean and manage, and the ease of harvesting and draining. 70.30% of the farmers stocked at juvenile stage. The majority that stocked at juvenile stage might be so because the farmers could well identify good culture stock even from the juvenile stage so as to reduce mortality of the catfish and as well aid the growth and maturity rate. Catfish farmers used more of local feed (79.57%, 88.41%, 80.56% and 82.48% in Edo South, Edo Central, Edo North and Edo State respectively). This agrees with Ekanem et al. (2012) who compared the growth performance and food utilization of catfish feed on local, midst and import feed and concluded that locally formulated feeds are good alternative in catfish production. More of family and hired labour (60.26%) was also used in the area. This agrees with the finding of Ahmadu and Egbodion (2017) also confirm that both

family and hired labour are used more in catfish production in Lagos Metropolis, Nigeria. The results (Table 2) further revealed that borehole was the major water source (75.00%) in the area. This implies that the respondents depended heavily on borehole water that directly increases the cost of production.

Table 2
Production Characteristics of Catfish Farmers

Variables	Edo State							
	Edo South		Edo Central		Edo North		Total	
	Freq (N=186)	% (100)	Freq (N=138)	% (100)	Freq (N=144)	% (100)	Freq (N=468)	% (100)
Source of finance								
Personal savings	110	59.14	88	63.77	74	51.39	272	58.12
Credit/loan	46	24.73	44	31.88	56	38.89	146	31.20
Government agency	30	16.13	6	4.35	14	9.72	50	10.68
Farm Land Acquisition								
Gift	3	1.61	-	-	4	2.78	7	1.50
Communal	12	6.45	14	10.14	6	4.17	32	6.84
Family	92	49.46	90	65.22	96	66.67	278	59.40
Lease/Rent	55	29.57	30	21.74	38	26.38	123	26.28
Purchase	24	12.90	4	2.90	-	-	28	5.98
Pond Type								
Earthen	48	25.81	24	17.39	34	23.61	106	22.65
Concrete	66	35.48	60	43.48	64	44.45	190	40.60
Plastic/Tarpaulin	72	38.71	54	39.13	46	31.94	172	36.75
Stocking Stage								
Fingerlings	13	6.99	-	-	80	55.56	93	19.87
Juvenile	131	70.43	138	100.00	60	41.67	329	70.30
Post Juvenile	42	22.58	-	-	4	2.77	46	9.83
Feed Type								
Imported	6	3.23	-	-	4	2.78	10	2.14
Local	148	79.57	122	88.41	116	80.56	386	82.48
Midst	32	17.20	16	11.59	24	16.66	72	15.38
Labour Type								
Family	45	24.19	24	17.39	14	9.72	83	17.73
Hired	33	17.74	24	17.39	46	31.95	103	22.01
Both	108	58.07	90	65.22	84	58.33	282	60.26
Water Source								
River	51	27.42	24	17.39	42	29.17	117	25.00
Borehole	135	72.58	114	82.61	102	70.83	351	75.00

Source: Computed from Field Survey, 2021

3.3. Average quantites of Inputs and Output of catfish production

The average quantities of inputs and output of catfish production is presented in Table 3.

The average feed quantity of catfish production in the study area was 0.78kg per cycle for a kg of catfish produced. Edo-South fertilizer quantity (0.28kg per cycle per kg fish) was the highest while Edo-North had the least fertilizer quantity (0.18kg per cycle per kg fish). The results in Table 3, also recorded that the average number of catfish produced was 4153.43, the weight of a catfish was 1.17kg and the output quantity produced was 4859.51kg per cycle per average of 584.17m^2 pond sizes of the catfish farmers in the study area. The prices per kg of mature catfish in Edo-South, Edo-Central and Edo-North were respectively NGN 760.22, NGN 760.87 and NGN 734.72, and the study area price per kg was NGN 752.56. This was supported by Ebukiba and Anthony (2019), who reported that higher output is due to efficiency utilization of resources used in catfish production.

Table 3
Distribution of Catfish Farmers by Input-Output Quantities per cycle

Variable	Edo State			
	Edo South (186) Mean	Edo Central (138) Mean	Edo North (144) Mean	Total (468) Mean
Inputs				
Feed Quantity /kg of catfish	0.71	0.75	0.90	0.78
Labour Used /kg of catfish	0.002	0.002	0.002	0.002
Fertilizer Quantity/kg of catfish	0.28	0.20	0.18	0.22
Outputs				
Average number of catfish /farmer	3412.36	4261.81	5168.46	4153.43
Average Weight/ Catfish (kg)	1.24	1.20	1.05	1.17
Pond Size(M^2)/ farmer	576.61	566.96	610.42	584.17
Output (kg)	4231.33	5114.17	5426.88	4859.51
Price/kg of Mature Catfish	760.22	760.87	734.72	752.56

Source: Computed from Field Survey, 2021

3.4. Profitability (costs and returns) analysis of catfish production

The results in Table 4, revealed that the average total cost incurred in catfish production across the state were NGN 500.04, NGN 547.54, NGN 596.32 and NGN 543.67, and revenue of NGN 760.22, NGN 760.87, NGN 734.72 and NGN 752.56 per kg fish was realized respectively in Edo-South, Edo-Central, Edo-North and Edo State as a pool. On average in the study area as a whole, a kg of catfish produced gave an average gross margin of NGN 224.35 and net profit of NGN 208.90 per production cycle. The return on sales was 0.28 indicating that for every one Naira of catfish sold, 28k profit was made. The gross ratio 0.72 implies that from every N1.00 return to the farm enterprise, 72k expenses were incurred in the production. The operating ratio of 0.70 means 70% of the revenue was used to offset the variable costs. The B/C ratio (1.38) and the rate of return on investment of 0.38 imply that for every one Naira invested in catfish production by each catfish farmer, returns (NGN 1.38) and profit of NGN 0.38 was obtained. The Profitability analyses show that, the catfish production was profitable in the Study area. This result is consistent with the findings of Alawode and Ajagbe.(2020) on the profitability of small-scale catfish production in Southwest Nigeria. Also, it is similar to the findings by Ebukiba and Anthony (2019) which concluded that catfish production is a profitable business in Nassarawa State as well as Onyekuru., Ihemezie and Chima (2019) which also concluded that catfish production in Enugu State is profitable.

Table 4
Profitability analysis of Catfish production per cycle per kg catfish

Variables	Edo State			
	Edo South (186) Mean	Edo Central (138) Mean	Edo North (144) Mean	Total (468) Mean
Returns (NGN)				
Total Revenue per kg fish	760.22	760.87	734.72	752.56
Variable Costs (NGN)				
Unit cost of stocking	28.28	35.90	25.62	29.69
Cost of fertilizer	44.35	31.87	28.42	35.77
Cost of lime	0.01	0.01	0.01	0.01
Cost of feeding	404.56	453.03	525.15	455.96
Cost of medication	2.25	2.53	2.17	2.31
Cost of transportation	1.20	0.96	1.18	1.12
Cost of energy	3.01	2.92	1.88	2.64
Cost of labour	0.92	0.67	0.55	0.73
Total Variable Cost (NGN)	484.51	527.89	584.99	528.21
Fixed Costs (NGN)				
Rent on Land	3.85	3.37	2.81	3.39
Cost of Pond construction	2.39	2.25	1.73	2.155
Harvesting nets Cost	0.39	0.36	0.31	0.36
Cost of scale	0.00	0.00	0.00	0.00
Cost of Sorting Filter	0.15	0.06	0.09	0.10
Plastic container Cost	1.19	0.92	0.96	1.04
Cost of generator	3.56	3.30	2.20	3.06
Cost of Shovel	0.09	0.10	0.10	0.10
Cost of Borehole	3.91	9.28	3.15	5.26
Total Fixed Cost (NGN)	15.53	19.65	11.34	15.46
Total Cost per kg fish (NGN)	500.04	547.54	596.32	543.67
Profitability Index				
Gross margin (NGN)	275.71	232.99	149.74	224.35
Net Profit (NGN)	260.18	213.33	138.40	208.90
Return on Sales	0.34	0.28	0.19	0.28
Gross Ratio	0.66	0.72	0.81	0.72
Operation Ratio	0.64	0.69	0.80	0.70
Benefit-Cost Ratio	1.52	1.39	1.23	1.38
Return on Investment	0.52	0.39	0.23	0.38
Percentage Basic of ROI	52.00	39.00	23.20	38.40

Source: Computed from Field Survey, 2021

3.5. Constraint faced by Cafish Farmers

The production constraints faced by catfish farmers in the study area are presented in Table 5. Out of the 16 constraints under consideration, 15 of them are rated serious with their mean scores greater than the bench mark of 2.5. They influence catfish productivity negatively. Top among the constraints are high cost of food, lack of capital and credit at affordable rates, and inadequate power supply respectively. Except for high spread of pest and disease that shows not serious, other constraints presented are greater than the bench mark of 2.5, meaning serious constraints. These results agreed with the findings of Saagulo *et al.* (2017) and Ahmadu, Boheje Odum and Osariemen (2021) that also reported these constraints are serious constraints in their respective study areas.

Table 5
Constraints faced by Catfish Farmers

Variable	Edo State			
	Edo South (186) Mean	Edo Central (138) Mean	Edo North (144) Mean	Total (468) Mean
Feed high cost	3.95	4	3.92	3.96
Lack of capital	3.63	3.71	3.61	3.65
Lack of credit at affordable rates	3.68	3.77	3.56	3.36
Lack of skilled manpower	3.24	3.28	3.25	3.25
Scarcity of fingerlings/ juveniles	3.37	3.39	3.28	3.35
Lack of modern technologies	3.26	3.30	3.22	3.26
High cost of transportation	3.36	3.29	3.19	3.29
High cost of labour	3.40	3.49	3.39	3.43
Inadequate access to farm land	3.31	3.35	3.28	3.31
Inadequate water supply	3.48	3.48	3.42	3.46
Inadequate power supply	3.45	3.55	3.56	3.51
Poor storage facilities	3.24	3.32	3.33	3.29
Ineffective extension services	3.21	3.30	3.31	3.27
Lack of organized market	2.79	2.97	2.83	2.86
Inadequate data collection	2.74	2.91	2.83	2.82
High spread of pest and disease	2.07	2.32	2.19	2.18

Source: Computed from Field Survey, 2021
Mean score $\geq$ 2.5 is a serious constraint.

4. Conclusıon and Recommendatıons

The study analyzed the profitability of catfish production in Edo State. It revealed that the cost of feeds was the major cost component in catfish production but it was found to profitable with more profit in Edo South and least profit in Edo North agricultural zone. The major constraints identified to confront catfish production were high cost of food, lack of capital and credit at affordable rates respectively. In boosting catfish production effort should be geared towards reduction in the cost of feeds and making credit facilities accessible to the farmers to finance their production. Government, private and non-governmental organizations as well as financial institutions should be encouraged to provide accessible financial support to the catfish farmers at affordable rate so as to increase their productivity. Catfish farmers should come together to form co-operative societies and/or join Catfish Farmers Association to ease their accessibility to farm inputs, through cooperate effort reduce cost of feed and organized marketing channels to enable sales of marketable catfish.

5. References

Ahmadu J, Boheje Odum EE, Osariemen FO (2021). Profitability and Profit Efficiency of Catfish Fingerlings Production in Edo South, Nigeria. Zagadnienia Ekonomiki Rolnej Problems of Agricultural Economics 4(369): 5-20

Ahmadu J, Egbodion J (2017). Analysis of Costs and Returns in Catfish Production in Lagos Metropolis, Nigeria. *Biological and Environmental Sciences Journal for the Tropics* 14(1): 16 – 21

Ajiboye A., Adekunmi AO, Osundare FO, Oluwatusin FM, Toluwase SOW, Amao SA (2020). Analysis of Technical Efficiency of Catfish Production in Ekiti State of Nigeria. INTERNATIONAL JOURNAL OF INNOVATIVE RESEARCH & DEVELOPMENT, 9(5): 137-142.

Akinsorotan AM, Akinsorotan OA, Jimohb JO, Adene IC & Akiwowo UA (2019). Offshore aquaculture practice; a potential for meeting Nigeria fish demand – a review. Journal of Physics: Conf. Series 1299,012111. DOI: 10.1088/17426596/1299/1/012111

Alawode, OO and Ajagbe SO (2020). "Profitability of Small-Scale Catfish Production In Southwest Nigeria: the challenges" Scientific Papers-Series Management Economic Engineering In Agriculture And Rural Development, 20 (3):77-88.

Awotide BA, Abdoulaye T, Alene A, Manyong VM (2015). Impact of Access to Credit on Agricultural Productivity: Evidence from Smallholder Cassava Farmers in Nigeria. *A Contributed paper Prepared for Oral Presentation at the International Conference of Agricultural Economists (ICAE) Milan, Italy*. August 9-14, pp34

Ebomwonyi A, Omorogie AO, Noutcha MAE, Abajue MC, Okiwelu SN (2019). Further Analyses of Trends in Malaria Diagnoses and Incidence Rates (2014-2018) in Edo State, Nigeria. International Journal of TROPICAL DISEASE & Health, 39(4), pp. 1-12.

Ebukiba SE. Anthony L (2019). "Economic Analysis of CatFish (Clarias gariepenus) Production in Karu Local Government Area, Nassarawa State, Nigeria". Journal of Agriculture and Veterinary Science. 12(3):41-48.

Ekanem AP, Eyo VS, Obiekezie AI, Enin UI, Udo PJ (2012). A comparative study of the growth performance and food utilization of the African catfish (*Clarias gariepinus*) feed. *Journal of Marine Biology and Oceanography* 1(2): 1-6

Food and Agriculture Organization FAO (2016). Food and Agriculture Organization of the United Nations. *The state of World Fisheries and Aquaculture* 43

Food and Agriculture Organization (FAO) (2021). The State of World Fisheries and Aquaculture 2020. http://www.fao.org/state-of-fisheries-aquaculture (18.08.2021)

Imade OS. Ahmadu, J (2022). Technical Efficiency of Catfish (Clarias gariepenus) Farmers in Edo State, Nigeria. Agricultural Economics and Extension Research Studies (AGEERS) Vol 10 (1): 18-26

Imade OS. Ogieva OF (2022). Economic Efficiency of Catfish Production in Edo State, Nigeria. Agricultural Economics and Extension Research Studies (AGEERS) Vol 10 (1): 35-46

Likert R (1932). A Technique for the Measurement of Attitudes. *Archives of Psychology* 140: 1–55

Michael CO, Duhu CR (2020). Sources of Information of Fish Farmers in Enugu State. *International journal of Agriculture and Rural Development (IJARD)* 23(1):4744 -4751

National Bureau of Statistics, Nigeria (NBS) (2021). Nigeria's Fish Production: 2010-2015. https://www.nigerianstat.gov.ng

Numa VD, Obayelu AE, Sanusi RA, Bada BA (2017). Effects of Oil Spillage on Poverty Status of Artisanal Fishing Households in River State, Nigeria. Conference Proceedings of the 18th Annual National Conference of the Nigerian Association of Agricultural Economics Held at Federal University of Agriculture, Abeokuta, Nigeria. 16th – 19th Oct, 2017. Pp 527-532

Odioko E, Becer ZA (2022). The Economic Analysis of The Nigerian Fisheries Sector: A Review. Journal of Anatolian Environmental and Animal Sciences 7(2), 216-226.

Olajide OO, Omonon BT (2019). Productivity of Catfish Production in Osun State, Nigeria. International Journal of Agricultural Research, Sustainability, and Food Sufficiency (IJARSFS) Vol. 6(04): Pp. 409-420

Onyekuru NA, Ihemezie EJ and Chima CC (2019). "Socioeconomic And Profitability Analysis of Catfish Productıon: A Case Study of Nsukka Local Government Area of Enugu State, Nigeria. Agro-Science. 18(2):51-58.

Ryan T (2013). Sample Size Determination and Power. John Wiley and Sons, 8(4): 207-232

Saagulo MN, Alhassan EH, Amikuzuno J (2017). Determinants of Choice of fishing activity along the Volta lake in Yeyi, Ghana. *Ghana Journal of Development Studies* 14(2): 6

Vanguard (2021). Nigeria has 2.5m metric tonnes of fish deficit- Minister. Vanguardngr.com. https://www.google.com/amp/s/www.vanguardngr.com/2021/03/nigeria-has-2.5m-metric-tonnes-of-fish-deficit-minister/amp/

Yaqoob AM, Fasakin JI (2021). Determinants of Profit Efficiency Among Catfish Farmers in South Western Nigeria: Stochastic Profit Frontier Approach. IOSR Journal of Agriculture and Veterinary Science (IOSR-JAVS) Volume 14, Issue 10 Ser. II, PP 01-09.

Selcuk Journal of Agriculture and Food Sciences
SJAFS
http://sjafs.selcuk.edu.tr/sjafs/index
(2022) 36 (3), 466-475
Research Article
e-ISSN: 2458-8377
DOI:10.15316/SJAFS.2022.061

Evaluation of Agro-Industries According To Management Functions: Case of Konya Province

Gürhan ÖZAYDIN[1*], Mithat DİREK[1]
[1]Selcuk University, Faculty of Agriculture, Department of Agricultural Economics, Konya, Türkiye

ARTICLE INFO

Article history:
Received date: 05.10.2022
Accepted date: 28.11.2022

Keywords:
Agro Industries
Agro-based industries
Agro-linked industries
Management functions

ABSTRACT

The study is to investigate the current situation in agricultural industry enterprises according to business functions. The data used in the research were made voluntarily through face-to-face interviews. The sample volume of the study was determined as 199 according to the quota (stratified purposive sampling) sampling method in the agro-industries of Konya, using TOBB data. According to the research results, it has been determined that the management functions in agro-industries are mostly carried out by business owners, business managers are generally male individuals, and their education level is generally at the undergraduate level. In agro-industries, raw materials are mostly (43.22%) brought from the province of Istanbul. It has been determined that the majority of agro-industries operate in national and international (71 enterprises) markets. The majority of the exports (15.08%) in the analyzed agro-industries are allocated to Iraq. It is stated that the average annual turnover of agro-industries is approximately 61 million TL. In agro-industries, income is one of the important factors used in determining the level of enterprises. When we look at the sector, it can be said that the existing industry consists of workshop-type family businesses rather than institutionalized companies and they have just started to institutionalize.

1. Introduction

While some of the products obtained as a result of agricultural production reach the consumer without being involved in any process, the other part can be consumed only after processing. Agro-industries are examined in two groups, agro-based, and agro-linked industries. In order to increase the efficiency in agricultural production, fertilization, and agricultural struggle, agricultural mechanization technologies are carried out by agro-linked industries. Agro-industrial enterprises consist of both the industry branch that provides input to agriculture and the sector that transforms agricultural raw materials into processed and semi-finished products. In order to increase efficiency in agricultural production, fertilization, agricultural struggle, and agricultural mechanization technologies are carried out by agro-linked industries. The branch of industry that uses agricultural products as raw materials, processes agricultural products, and makes them usable and transportable over long distances is agro-based industries.

In agro-industries, businesses engage in many activities to survive and achieve their goals. There is a systematic path to be followed for all activities that need to be achieved. Institutions and organizations consisting of certain elements and operating in line with specific goals have a functional system within themselves (Genç, 2017). Business activities that are grouped according to their similarities are called “business functions” (Yüksel, 2008). This study aims to examine the structures of agro-industrial enterprises according to their business functions.

2. Materials and Methods

The main material of the study consists of primary data obtained from agro-industry business managers operating in Konya. Agro-industries were examined according to the "International Standard Industrial Classification (ISIC)" classification method established by the UN. According to this classification, agro-industries:

A: Agro-based industries 1) Food industry 2) Beverage industry 3) Tobacco and products Industry 4) Textile and clothing industry 5) Leather and products industry 6) Forest products industry 7) Paper industry

B: Agro-linked industries 1) Agricultural equipment and machinery industry 2) Fertilizer industry 3) Agro-pharmaceutical industry 4) Seed industry

* Corresponding author email: *gurhan@selcuk.edu.tr*

Within the scope of the study, the agro-industry enterprises located in the province of Konya in the union of chambers and commodity exchanges of Turkey (TOBB) records constitute the total population. The current situation of the agricultural industry enterprises in the research area is given in Table 1 and it is a total of 899 enterprises. Since the criterion of capacity as a criterion in determining samples from the total population is not homogeneous based on enterprises, quota sampling was made by considering the number of personnel of the enterprises. Quota sampling (stratified purposive sampling) is the grouping of the researcher from the population according to certain characteristics and sampling until a certain number of them is reached (Moser & Stuart, 1953; Yağar & Dökme, 2018). Accordingly, the number of enterprises in the sample was determined not less than 20% of the enterprises operating in the same field (Table 1). TOBB has classified enterprises as 1-9, 10-49, and 50-249, enterprises with 250 or more employees (Mecek, 2020). To analyze the data obtained because of the study in a homogeneous way, the number of personnel of the enterprises was divided into strata, considering the classification of TOBB.

Table 1
Distribution of agro-industrial enterprises in Konya according to sub-sectors and determination of sample enterprises

Agro-industry sectors		1-9 *	Eg. **	10-49 *	Eg. **	50-249 *	Eg. **	250-+ *	Eg. **	Sum Pop. ***	Sum Eg. ****	Eg. **
Agro-based industries	Food industry	175	35	185	37	64	16	10	2	434	90	20,74
	Beverage industry	2	1	7	3	2	1	-	-	11	5	45,45
	Tobacco and products Industry	-	-	-	-	-	-	-	-	-	-	-,--
	Textile and clothing industry	23	5	25	5	16	4	5	1	69	15	21,74
	Leather and products industry	33	7	25	5	4	1	-	-	62	13	20,97
	Forest products industry	23	4	15	4	4	1	-	-	42	9	21,43
	Paper industry	14	4	11	5	6	2	-	-	31	11	35,48
Agro-based industries		270	56	268	59	96	25	15	3	649	143	22,50
Agro-linked industries	Fertilizer industry	20	4	10	3	2	1	-	-	32	8	25,00
	Agri. equipment and machinery industry	114	23	70	15	19	4	-	-	203	42	20,69
	Agro-pharmaceutical industry	8	3	3	2	4	1	-	-	15	6	40,00
	Seed industry	-	-	-	-	-	-	-	-	-	-	-,--
Agro-linked industries		142	30	83	20	25	6	-	-	250	56	22,40
Agro-industries		412	84	351	79	121	33	15	3	899	199	22,14

Note: *Total population according to the number of personnel, **Number of businesses to be interviewed, ***Total population in industry sub-branches, ****Number of sample businesses to be drawn from the total population, ***** What percentage of the total population will be interviewed according to sampling

Data related to businesses in agro-industries are classified based on business functions. Although there are various classifications for business functions, the generally accepted classification is developed by Fayol (1916) in his book "General and Industrial Management".

a. General administration (Management) function

b. Production management function

c. Marketing management function

d. Finance and accounting management function

e. Human resource management function

f. R&D management function

The research data is presented in a table by analyzing frequency and percentage calculations.

The attitudes, behaviors, and perceptions of the business managers in the agro-industries in Konya were analyzed using a 5-point Likert-type scale. A Likert-type scale is the most practical method for measuring attitudes. Therefore, this method is used similar data (Powers & Xie, 2008; Ünlüer & Güneş, 2013; Christoforou et all. 2018;)

3. Findings and Discussion

In terms of business functions of the agro-industries examined within the scope of the study, general administration function (agricultural industry sub-sectors, legal structure, establishment date, etc.), production function (input supply method, where they obtain input from, etc.), marketing function (market network, export status, etc.), R&D management function (R&D status, number of patents, status of following innovations, etc.), finance and accounting function (annual turnover), and human resources function (number of personnel, personnel qualifications, etc.) were examined.

3.1. Management structures according to business functions in agro-industries

The management statuses according to the management functions in the examined agro-industries are shown in Table 2. In the general administrative management of agro-industries in Konya, 67.34% were managed by the business owners, 22.61% by the family of the business, and 10.05% by a professional manager. While the professional manager is 11.89% in agro-based industries, this rate is 5.36% in agro-linked industries. In the study conducted by Mazgal&Bozoglu (2006) in Samsun province, it was determined that only 1.70% of the agriculture-based industrial enterprises were managed by professional managers. General administrative management in agro-industrial enterprises is usually carried out by business owners. The current management and business entrepreneurship qualities of the business owner or business family are the important factors that make up the success of businesses. This situation can bring various advantages to the company, as well as lead to negative consequences. Being flexible and fast in the decision-making process creates a positive effect on the business in businesses managed by the owner or his family. Despite this, the decisions made by the owner of the business due to the lack of management, knowledge, and experience can lead to negative consequences for the business.

In the production management of agro-industries, 56.78% were managed by the business owners and 20.60% are managed by the business family. In agro-industries, professional managers were found in general administration management with a ratio of 10.05%, while in production management with a share of 22.61%. While business owners hold the general administration into their own hands, they can also prefer professional managers in the field of production. While the professional manager was 22.38% in agro-based industries, this rate was 23.21% in agro-linked industries.

In the marketing function of the enterprises, 48.74% were managed by the business owners and 17.59% by the business family. The fact that there are more professional managers in the marketing department compared to other business departments shows that companies give more importance to marketing. While the professional manager was 30.77% in agriculture-based industries, this rate was 41.07% in agriculture-related industries.

In agro-industrial enterprises, 22.11% of the R&D management was managed by the business owners and 10.05% by the business family. Enterprises that do not have an R&D department in agro-industries and that do not have a business manager had a large share of 40.70%. This shows that R&D was not given enough importance in agro-industries. While the professional manager was 25.87% in agriculture-based industries, this rate was 30.36% in agro-linked industries.

Table 2
Management structures of the agro-industries studied

Agro-industry sectors		Business owner		Business family		Professional Manager		Do not have Department		Total	
		Count	%	Count	%	Count	%	Count	%	Count	%
General administration	Agro-based	99	69,23	27	18,88	17	11,89	-	-	143	100,00
	Agro-linked	35	62,50	18	32,14	3	5,36	-	-	56	100,00
	Agro-industries	134	67,34	45	22,61	20	10,05	-	-	199	100,00
Production	Agro-based	83	58,04	28	19,58	32	22,38	-	-	143	100,00
	Agro-linked	30	53,57	13	23,21	13	23,21	-	-	56	100,00
	Agro-industries	113	56,78	41	20,60	45	22,61	-	-	199	100,00
Marketing	Agro-based	74	51,75	25	17,48	44	30,77	-	-	143	100,00
	Agro-linked	23	41,07	10	17,86	23	41,07	-	-	56	100,00
	Agro-industries	97	48,74	35	17,59	67	33,67	-	-	199	100,00
Finance and accounting	Agro-based	39	27,27	22	15,38	82	57,34	-	-	143	100,00
	Agro-linked	19	33,93	8	14,29	29	51,79	-	-	56	100,00
	Agro-industries	58	29,15	30	15,08	111	55,78	-	-	199	100,00
R&D	Agro-based	30	20,98	12	8,39	37	25,87	64	44,76	143	100,00
	Agro-linked	14	25,00	8	14,29	17	30,36	17	30,36	56	100,00
	Agro-industries	44	22,11	20	10,05	54	27,14	81	40,70	199	100,00
Human resource	Agro-based	51	35,66	16	11,19	28	19,58	48	33,57	143	100,00
	Agro-linked	18	32,14	8	14,29	15	26,79	15	26,79	56	100,00
	Agro-industries	69	34,67	24	12,06	43	21,61	63	31,66	199	100,00

3.2. Structure of the general administration (Management) function in agro-industries

The gender and education status of business owners and professional managers in the examined agro-industries is shown in Table 3. It was determined that 95.67% of the business owners were male and 4.33% are female founders or partners. In the study conducted by Keskin (2014) on the situation of women entrepreneurs in Turkey, it was stated that the rate of employers and self-employed women was 12.10% in 2012 and 14.90% of total entrepreneurs were women entrepreneurs. It was determined that 82.10% of the agro-industries consisted of male professional managers. There is a similar situation in the study by Üçler and Karaçor (2015). In the study on the industry in Konya, it was stated that male business managers managed 76.40% of them. In addition, in a study by Ulas and Çakır (2004) in the agro-based industry in Van determined that 97.70% of business managers were men.

While only 4.28% of agro-industry business owners were women, the rate of professional managers they employ was 17.90%. While the rate of female professional managers in agro-based industrial enterprises was 16.03%, this rate was determined as 23.08% in agro-linked industries.

It was determined that the majority of the business owners of agricultural industrial enterprises (62.50%) were primary or high school graduates. Insufficient education of business owners or partners is one of the major causes of business failure. Due to the lack of education, the developments in the growth process of the enterprise may not be properly controlled and problems related to business management may occur. However, especially considering the country's scale, it is seen that there are many entrepreneurs who set an example of success despite their low education levels. The common features of these examples are; Despite their low level of education, it is shown that they have the opportunity to work with people who have experience or expertise in the master-apprentice relationship.

Table 3
General administrative management gender and education status in the analyzed Agro-industries

Agro-industry sectors			Business owner			Pro. Manager		
			Agro-based	Agro-linked	Agro-industries	Agro-based	Agro-linked	Agro-industries
Education	Primary edu.	Count	0,62	0,43	0,56	0,08	0,09	0,08
		%	28,12	23,30	26,92	4,23	5,88	4,64
	High school	Count	0,85	0,48	0,74	0,21	0,32	0,24
		%	38,66	26,21	35,58	11,54	21,18	13,91
	Associate Deg.	Count	0,04	0,11	0,06	0,25	0,20	0,24
		%	1,92	5,83	2,88	13,85	12,94	13,62
	Bachelor	Count	0,62	0,77	0,66	1,25	0,88	1,15
		%	28,43	41,75	31,73	68,85	57,65	66,09
	Master Deg.	Count	0,06	0,05	0,06	0,03	0,04	0,03
		%	2,88	2,91	2,88	1,54	2,35	1,74
	Total	Count	2,19	1,84	2,09	1,82	1,52	1,73
		%	100,00	100,00	100,00	100,00	100,00	100,00
Gender	Female	Count	0,10	0,05	0,09	0,22	0,30	0,25
		%	4,79	2,91	4,33	12,31	20,00	14,20
	Male	Count	2,08	1,79	2,00	1,59	1,21	1,49
		%	95,21	97,09	95,67	87,69	80,00	85,80
	Total	Count	2,19	1,84	2,09	1,82	1,52	1,73
		%	100,00	100,00	100,00	100,00	100,00	100,00

The legal statuses of the examined agro-industries are shown in Table 4. 72.86% of agro-industries were limited liability companies, 16.58% were joint stock companies and 10.55% were individual property. While there were 70.63% limited liability companies in agro-based industries, this rate was 78.57% in agro-linked industries. In the study of Herdem (2014) on software and R&D companies in Konya, it was determined that most enterprises (63.60%) were limited liability companies. Ulaş & Çakır (2006); In the research conducted in the agro-industries in Van, it was determined that 38.88% of the companies were joint stock companies. Contrary to this, in the study by Çelikkaya (2010) on SMEs in Gebze determined that the joint stock company rate was 86.70% and the limited company rate was 13.30%. The legal structure of businesses depends on the cities they are in, the structure of the business, the situation of the entrepreneurs, the sector, etc. appears to vary accordingly.

Table 4
Legal status of examined agro-industries

Agro-industry sectors	Joint stock		Individual property		Limited liability		Total	
	Count	%	Count	%	Count	%	Count	%
Agro-based industries	24	16,78	18	12,59	101	70,63	143	100
Agro-linked industries	9	16,07	3	5,36	44	78,57	56	100
Agro-industries	33	16,58	21	10,55	145	72,86	199	100

3.3. Structure of the product management function in agro-industries

Production: It is expressed as increasing the use of goods or services used to meet human needs. The production unit is defined as the provision of raw materials or inputs and the realization of production to carry out the production activities of the enterprise (Kobu, 2017).

The establishment dates of the examined agro-industries are given in Table 5. The establishment dates of enterprises in agro-industries vary. It is seen that 42.71% of agro-industries were established between 1991 and 2010 and continued their activities. Businesses that have been continuing since 2011 have a rate of 27.64%. In a study conducted by Üçler & Karaçor (2015) on industrial enterprises in the province of Konya, it was stated that 31.00% of the establishment dates of the enterprises were between 2001-2009 and 32.50% between 1990-2000. In addition, in the study conducted by Mazgal & Bozoğlu (2006) on agriculture-based industrial enterprises in Samsun, it was stated that the majority of the establishment dates were started after 1991.

In the study by Doğan (2013) on the industrialization process of Turkey, it was reported that since 1950, decisions have been taken to accelerate development in Turkey and the world and to support private investors. With the Second Five-Year Development Plan, it was stated that policies were followed in order to make and support investments in the field of industry in other provinces apart from metropolitan cities. In the sixth five-year development plan implemented between 1990 and 1994, it was stated that the importance of industry increased, and industrialization was the main source of development. This information confirms the establishment dates of the agricultural industries established in the research area.

Table 5
Distribution of the examined agro-industries by date of establishment

Agro-industry sectors	1950-1970		1971-1990		1991-2010		2011-+		Toplam	
	Count	%	Count	%	Count	%	Count	%	Count	%
Agro-based industries	12	8,39	30	20,98	63	44,06	38	26,57	143	100
Agro-linked industries	5	8,93	12	21,43	22	39,29	17	30,36	56	100
Agro-industries	17	8,54	42	21,11	85	42,71	55	27,64	199	100

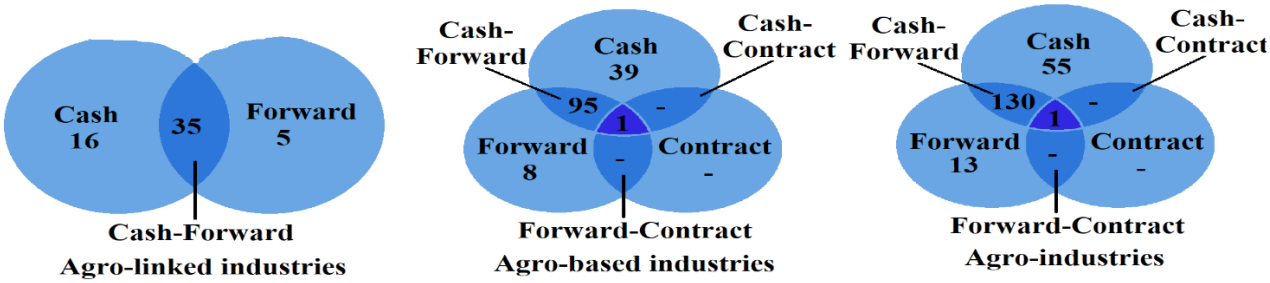
The raw material supply method in the examined agro-industries is shown in Table 6. 61.00% of the agro-industries purchased the input in cash. The rate of agro-industry enterprises that purchase input on a forward basis was 38.75%. While the agro-based industries purchased the raw material in cash at 61.61%, the agro-linked industries remained at the rate of 59.46%. In the study conducted by Yalcin & Esengün (2008) on food industry enterprises in the province of Tokat, it was determined that the enterprises did not engage in contracted farming, and received the inputs on a forward basis or in cash.

Table 6
The raw material supply method in the examined agro-industries

Agro-industry sectors	Cash Purchase	Forward Purchase	Contract farming	Total
Agro-based industries	61,61	38,05	0,34	100
Agro-linked industries	59,46	40,54	-	100
Agro-industries	61,00	38,75	0,25	100

The raw material supply limits of the examined agro-industries are shown in Figure 1. It has been determined that the majority of agro-industrial enterprises (130 enterprises) use both cash and forward basis purchases. While the number of businesses that purchase raw materials only in cash is 55, the number of those that purchase only in time is 13. It has been determined that the majority of the agro-based industries (95 enterprises) examined use both cash and forward basis purchases. While the number of businesses that purchase raw materials only in cash is 39, the number of those that purchase only in time is 8. It was observed that only 1 enterprise used the contract farming method. It has been determined that the majority of the agro-linked industries (35 enterprises) examined use both cash and forward basis purchases. While the number of businesses that purchase raw materials only in cash is 16, the number of only time deposits is determined as 5. The contracted (agricultural) production model is a form of production and marketing because the companies receive the product to be obtained under certain conditions, even though the product at the planting-planting time or the farmer assumes the responsibility of realizing a certain cultivation area and production (Hekimoğlu & Altındeger, 2012). Since there are enterprises that provide input to agriculture in agro-linked industries, contract farming cannot be done in these enterprises.

Figure 1
The cluster of raw material purchase methods (number of enterprises) in the agro-industries studied



The distribution of raw materials used in the examined agro-industries is given in Table 8. It has been determined that 44.82% of the agro-industries allocate the input from within the province. The ratio of enterprises allocating input from other provinces was determined as 42.83%. It has been determined that while the industries based on agriculture allocate the raw materials mostly (46.91%) from other provinces, the industries related to agriculture mostly (50.45%) from within the province.

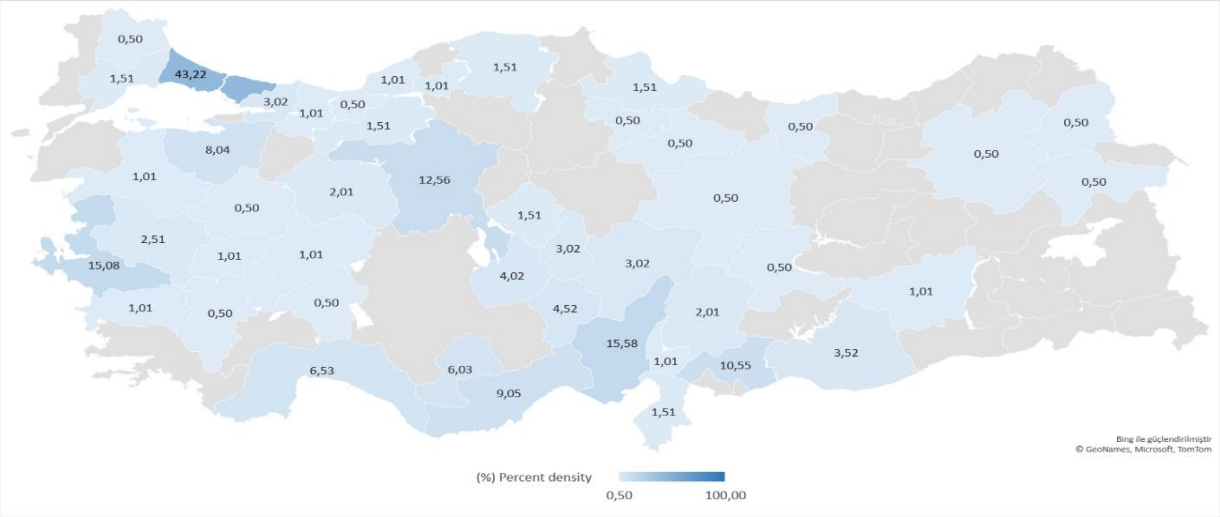
Table 7
Distribution of raw material supply locations used in the examined agro-industries (%)

Agro-industry sectors	Within the province	Other provinces	Own production	From abroad	Toplam
Agro-based industries	42,62	46,91	3,39	7,08	100
Agro-linked industries	50,45	32,41	4,29	12,86	100
Agro-industries	44,82	42,83	3,64	8,70	100

Figure 2 shows where the raw materials used in the examined agro-industries were obtained from outside the province of Konya. While creating the raw material supply map, the places where the enterprises are supplied with raw materials are determined by frequency and percentage calculations. It has been determined that agro-industries supply raw materials mostly (43.22% of enterprises) from Istanbul on the basis of provinces. Istanbul was followed by Adana (15.58%), Izmir (15.08%), Ankara (12.56%), Gaziantep (10.50%), Mersin (9.05%), Bursa (8.04%) respectively, Antalya (6.53%), Karaman (6.03%), and Aksaray (4.02%). Except for these provinces, raw material supply remained at the rate of 2.38% and Şanlıurfa, Kocaeli, Kayseri, Manisa, etc. provinces have been identified. It has been observed that the examined agro-based industries mostly supply inputs from the province of Istanbul (24.62%). It is stated that Adana is in second place in the supply of

inputs with 10,77% and Gaziantep in third place (7.69%). It is seen that the input supply in the industries related to agriculture was mostly met in the province of Istanbul (29.73%). However, it was determined that while the input supply in agro-linked industries is in second place in İzmir (17.57%), it was in fifth place in agro-based industries. Agro-linked industries; It was determined that while obtaining input supply from provinces such as Aksaray, Manisa, Nevşehir, Şanlıurfa, and Karabük, agro-industries did not bring raw materials from these provinces. It was determined that while agro-based industries use provinces such as Gaziantep, Antalya, Karaman, and Niğde, agro-linked industries didn't establish a relationship with these provinces in terms of input supply.

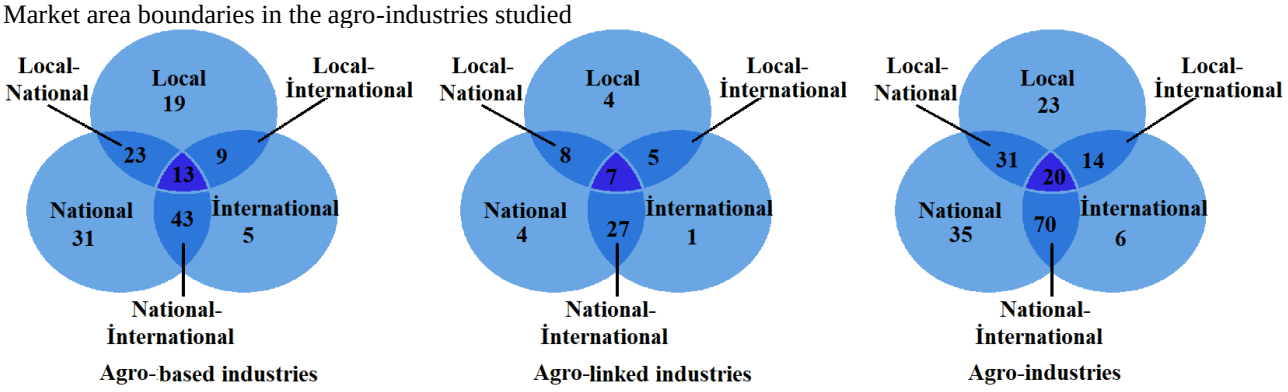
Figure 2
Provinces from which the examined agro-industries supply raw materials (%)



3.4. Structure of the marketing management function in agro-industries

In agro-industries, it is of great importance for enterprises to operate on an international and national scale and for the development of national and international trade. The scope of activity of the examined agro-industries is given in Figure 3 based on enterprises. It was determined that the majority of agro-industries operated in national and international (70 enterprises) markets. However, there were 35 enterprises operating only in the national market, while 31 enterprises operated in both regional and national areas. In a study conducted by Dinçer (2010) on SMEs in Eskişehir, it was stated that 38.60% of the enterprises operate regionally, 52.90% nationally, and 39.80% internationally. It was determined that the majority of the examined agriculture-based industries consist of companies that use the national-international (43 enterprises) field of activity. While the number of enterprises with a market network in the national area was 111, 31 of these enterprises only created a market network in the national area and did not form a regional or international market network. In the study Bakkaloglu (2018) on Turkish food industry enterprises, it was determined that while the majority of the enterprises constituted the national-international field of activity, only the enterprises with a regional market network drew attention when their small enterprises were taken into account. A result similar to the situation in the agro-based industries occurs in the examined agro-linked industries.

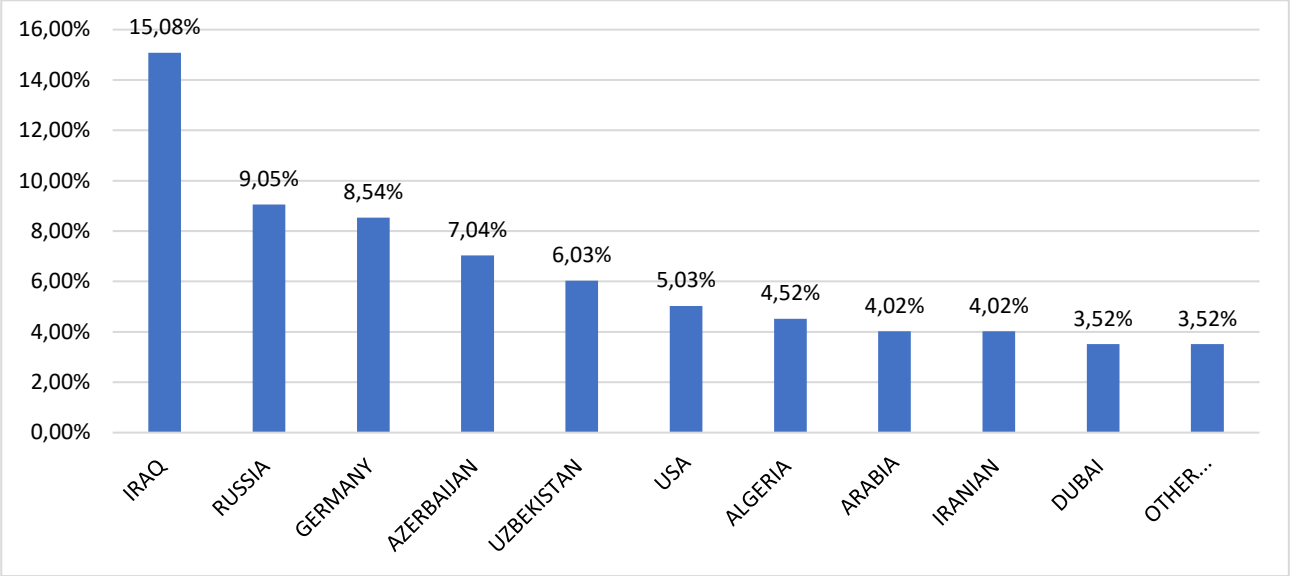
Figure 3
Market area boundaries in the agro-industries studied



The exported countries in the examined agro-industries are shown in Figure 4. The majority of exports (15.08%) in agro-industries were allocated to Iraq. Iraq is followed by Russia (9.05%), Germany (8.54%), Azerbaijan (7.04%), Uzbekistan (6.03%), etc., respectively. countries followed. Apart from these top ten countries, agro-industries include Palestine, France, Qatar, England, Italy, Libya, Egypt, Romania, Jordan, Belgium, etc. countries were also exported. In the study conducted by KSO (2022) on Konya's economic data, it was stated that the export of Konya province in 2021 was in the eleventh rank in Turkey. In addition, most of the exports are from Iraq, USA, Germany, Russia, etc. countries have been identified.

When the non-exporters in agro-industry enterprises are investigated, insufficient infrastructure, insufficient production, insufficient financing power, excessive raw material prices, inability to deliver the product to the customer in the country, insufficient operating capacity, small company, lack of personnel, insufficient technology, product exportability, unsuitable, etc. causes have been identified.

Figure 4
Countries to which exports are made in the analyzed agro-industries



3.5. Structure of the finance and accounting management function in agro-industries

Annual sales turnovers of the examined agro-industries are shown in Table 8. It was determined that 46.73% of agro-industrial enterprises had an income between 0 and 550 thousand $. It was stated that the average annual turnover of agro-industries was approximately 3.352 thousand $. While 23.08% of the agro-based industries earn more than 2.201 thousand $, this rate was seen as 14.29% in the agro-linked industries. While the average annual turnover in agro-based industries was 1.594 thousand $, it was 3.132 thousand $ in agro-linked industries. In the study conducted by Bakkaloglu (2018) on the Turkish food industries, the majority of the enterprises had an income of more than 190 thousand $, in the study conducted by Dinçer (2010) on SMEs in Eskişehir, only 5.00% of the companies were 660 thousand $ reported having more income. The country's economic conditions also change the enterprises' income over the years.

Table 8
Annual sales turnover in the examined agro-industries (thousand $)

Agro-industry sectors	0-550		551-1.100		1.101-1.650		1.651-2.200		2201-+		Total	
	Count	%	Count	%	Count	%	Count	%	Count	%	Count	%
Agro-based industries	68	47,55	22	15,38	13	9,09	7	4,90	33	23,08	143	100,00
Agro-linked industries	25	44,64	14	25,00	5	8,93	4	7,14	8	14,29	56	100,00
Agro-industries	93	46,73	36	18,09	18	9,05	11	5,53	41	20,60	199	100,00

Note: (1$=18,20 TL in 2020), (1$=5,29 in 2018), (1$=1,53 TL in 2010)

3.6. Structure of the R&D management function in agro-industries

The concept of research and development is generally understood as the research and development methods used in producing and developing new products. However, developing methods that will increase the company's profitability in marketing, finance, accounting, and human resources management also has an important function in achieving company goals (Kahveci $ Baş, 2015).

The status of R&D activities in the examined agro-industries is given in Table 9. It was determined that 43.72% of the agro-industries enterprices actively carried out R&D studies, 33.67% did not carry out R&D studies, and 22.61% did R&D studies, even if they were

not laboratories, even partially. While the enterprises that did not perform R&D in agro-based industries constituted 28.67%, this rate was 46.43% in agro-linked industries. Tan et al. (2017) stated that 75.16% of the enterprises in the province of Çanakkale on agro-based industries, and in the study conducted by Mazgal and Bozoğlu (2006) in the province of Samsun, 80.70% of the enterprises did not carry out R&D activities.

Table 9
The status of doing R&D activities in the examined agro-industries

Agro-industry sectors	Enterprises doing R&D		Enterprises partially engaged in R&D		Enterprises that do not do R&D		Total	
	Count	%	Count	%	Count	%	Count	%
Agro-based industries	63	44,06	39	27,27	41	28,67	143	100,00
Agro-linked industries	24	42,86	6	10,71	26	46,43	56	100,00
Agro-industries	87	43,72	45	22,61	67	33,67	199	100,00

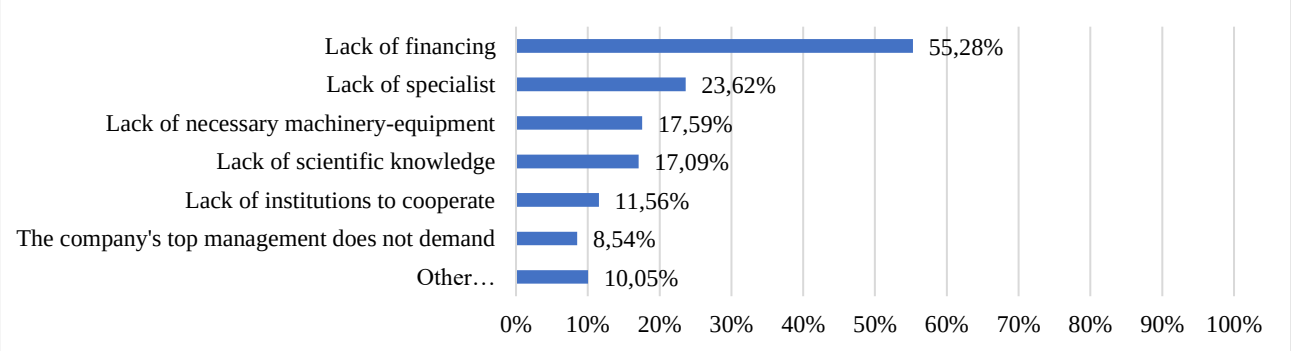
In the examined agro-industries, the number of patents applied by the enterprises during the period and received since the establishment date is given in Table 10. It was determined that agro-industrial enterprises had an average of 5,93 patents. It was stated that 16.44% of these patent numbers were applied during this period and 83.56% were previously obtained. According to TÜRKPATENT (2022) data, the patent applications of agro-industry enterprises in 2020 were determined as 2.251. While the number of patent applications and patented products was 5,72 in agro-based industries, this rate was 6,46 in agro-linked industries.

Table 10
Number of patents applied or received in the agro-industries examined (Average)

Agro-industry sectors	Applied to		Received		Total	
	Count	%	Count	%	Count	%
Agro-based industries	0,76	13,33	4,96	86,67	5,72	100,00
Agro-linked industries	1,52	23,48	4,95	76,52	6,46	100,00
Agro-industries	0,97	16,44	4,95	83,56	5,93	100,00

The difficulties encountered while developing technology in the agro-industries studied are given in Figure 5. Problems faced by agro-industrial enterprises while developing technology; It was determined that 55.28% of them had insufficient financing, 23.62% of them could not find enough experts in the market, 17.59% of them could not reach the necessary machinery and equipment, and 17.09% of them had a lack of scientific knowledge. In addition, 10.05% of the official procedures (Bureaucracy), the inadequacy of state support, lack of qualified personnel, the location of the establishment, etc., it was stated that they had difficulties in developing technology due to situations.

Figure 5
Challenges encountered while developing technology in the agro-industries examined



It is shown in Table 11 whether the enterprises in the examined agro-industries cooperate with universities. It was determined that 76.88% of enterprises in agro-industries did not cooperate with universities. Even in the study conducted by Gül (2012) in the technology region of İzmir, it was determined that 40.50% of the companies were not in cooperation with any university, and 59.50% were in cooperation with universities. The study of Yardımcı and Müftüoğlu (2015) on industrial enterprises determined that 76.69% of the enterprises do not cooperate with the university-industry.

Table 11
Collaboration status with universities in the examined agro-industries

Agro-industry sectors	Yes		No		Total	
	Count	%	Count	%	Count	%
Agro-based industries	21	14,69	122	85,31	143	100,00
Agro-linked industries	25	44,64	31	55,36	56	100,00
Agro-industries	46	23,12	153	76,88	199	100,00

4. Results and Discussion

According to the results of the research, mostly general administration (89.95%), production (77.39%), and marketing (66.33%) management within the business departments were carried out by business owners or family members. In addition, it was observed that there was no manager in R&D (32.16%) and human resources (46.73%) management. It is known that one of the solutions to the problems of family businesses in Turkey is possible with the institutionalization of businesses. Global competition conditions require institutionalization and professional management team in enterprises. However, if family companies have started to hire professional or assistant managers, adaptation and transition processes will be easier. A "management system" should be established by the general operating rules in enterprises and care should be taken to employ professional managers.

It was determined that the education level of business owners in agro-industries was generally high school (35.58%) and bachelor's (31.73%), but business owners who had completed their graduate education (2.09%) were not sufficient. While the undergraduate level provides general competence in a particular field, it provides specialization in a postgraduate field. Considering the ratio of enterprises that did not cooperate with universities (76,88%), it is seen that the ties between agro-industries and universities were weak. In agro-industrial enterprises, especially business owners and managers should focus on undergraduate and graduate education and get to know universities more closely.

It was determined that the legal structure of the agro-industries examined, 72.86% of them were limited liability companies, and the establishment dates were concentrated between 1991 and 2010 (42.71%). This situation was the result of the policies created for the industry in the sixth Five-year development plan.

The vast majority of agro-industrial enterprises provided raw materials in cash (61.00) and from within the province (44.82%). In addition, raw materials were procured (42.83%) from outside the province of Konya for the examined agro-industries. Except for Konya province, raw materials were mostly (25.60%) procured from Istanbul. While Adana (10.77%) and Gaziantep (7.69) stand out in agriculture-based industries, İzmir (17.57%) and Ankara (8.11%) were in agro-linked industries. Also, Germany, China, Italy, Russia, etc. Raw materials are also imported from other countries. Procuring raw materials from other provinces (especially distant provinces) or abroad increases the input costs (transportation fees, etc.) of the enterprises. While allocating the necessary raw materials for production in the enterprises, they should be procured at an affordable price. The department responsible for raw material supply should work efficiently to carry out its activities and functions rationally and economically.

It was determined that most of the enterprises in the examined agro-industries operate in the national and international (71 enterprises) areas. In addition, 35 enterprises operating only in the national area drew attention. Only 6 enterprises were exporting. The development of exports is an important issue for businesses as well as for countries. Especially the integration and liberalization brought about by increasing competition and globalization increase the importance of exports. It seems possible with the development and export of high technology machinery equipment with high added value in the industry sector.

It was determined that 49.75% of the enterprises have 1-3 quality certificates in the quality certificates of agro-industrial enterprises. It was stated that 40.59% of the enterprises received OHSAS (Occupational health and safety) and 31.19% of them ISO 9001 (quality management) certificate. Although each country has its quality standards, quality certificates such as ISO, which is the reason why it is preferred by businesses, will help companies make a good start on the environment. Especially international enterprises should understand the standards of each country and produce accordingly.

In agro-industries, income is one of the important factors used in determining the status of enterprises. The employment structures of enterprises are also one of the factors that can be used to determine the characteristics of enterprises. One of the most important problems in Turkey is the high unemployment rate. In addition, the need for qualified labor in agro-industries is extremely important. For this reason, unemployment rates can be reduced by providing training activities and training qualified personnel. To meet the qualified labordemand of agro-industrial companies in the study area, more effective cooperation should be established between industrial organizations and technical high schools in the region. In addition, the necessary training should be given to the technical personnel present in the enterprise through the said institutions. In this context, industry, universities, government institutions, etc. cooperation between stakeholders is needed.

It was determined that 43.72% of the enterprises in agro-industries carried out R&D activities, and 33.67% did not carry out any R&D activities. It was stated that they generally used domestic fairs (71.86%) and internet-media tools to follow innovations. It was shown that the difficulties faced by enterprises while developing technology were due to the lack of finance 55.28% and the lack of specialists or qualified personnel 23.62%. Industrial enterprises need to initiate systematic plans to improve their R&D activities and employ qualified personnel. Due to the financial problems of the enterprises, it is necessary to provide sufficient resources from the institutions and organizations that support R&D and to benefit more from universities. Successful R&D studies should be carried out to raise financial performance to a positive level. Especially small or medium-sized industrial companies KOSGEB, TUBITAK, etc. institutions should be supported.

It was determined that 76.88% of the agro-industries didn't cooperate with universities. University-industry cooperation can benefit the industry, universities, and the welfare of society separately. The most important problem between industrialists and universities is the lack of communication and it needs to be solved urgently. The information obtained from the university should be transferred to the industry as soon as possible, and the technology in the sector to the university. University curricula should be oriented toward the educational needs of the sector, and university students should spend certain days of the week in the industry, transferring what they learned from education to the business, and the knowledge they gained from the business to education.

5. References

Bakkaloglu Z (2018). Research and Development (R&D) Status of Food Industry in Turkey and Suggestions for Improvement. Academic Food Journal, 16, 183-191.

Christoforou A, Dachner N, Mendelson R, Tarasuk V (2018). Front of Package Nutrition References are Positively Associated with Food Processing. Public health nutrition, 21(1), 58-67.

Çelikkaya S (2010). Kobi'lerde Ar-Ge ve İhracat Desteği Gebze Organize Sanayi Bölgesi Üzerine Bir Araştirma. Department of Economics. Suleyman Demirel University. Isparta-Turkey

Dincer A (2010). İşletmelerin İnovasyon Gücünün Ölçümlenmesi ve Eskişehir İlinde Faaliyette Bulunan Kobilerde Bir Uygulama. In Department of Business Administration. Dumlupinar University Institute of Social Sciences. Kütahya-Turkey

Doğan M (2013). Türkiye Sanayileşme Sürecine Genel Bir Bakış. Marmara Geographical Review, 28, 211-231. 2013.

Genç N (2017). Yönetim ve Organizasyon. 5th Edition Seçkin yayıncılık. Ankara 1-424.

Gül TG (2012). İzmir Teknoloji Geliştirme Bölgesinde, Teknoloji Geliştirme Faaliyetlerinin Firma Performansi Üzerine Etkilerinin Ölçülmesi. Social Sciences Institute. Ege University. İzmir-Turkey.

Hekimoğlu B, Altındeğer B (2012). Tarımda Sözleşmeli Üretim Modeli ve Samsun İli Yaklaşımı. TC Samsun Governorship Provincial Directorate of Food, Agriculture and Livestock. Samsun.

Herdem P (2014). Rekabet Avantajı Sağlamada Bir Araç Olarak Kümelenme: Konya Yazılım ve Ar-Ge Firmaları Üzerine Bir Alan Araştırması. In Industrial Engineering Department. Selcuk University. Konya -Turkey

Kahveci M (2015). Baş F. Üretim işletmelerinde ürün mamul geliştirmenin Ar-Ge departmanında teknik aşamaları ve bilgi teknolojilerinin kullanımı. Accounting and Auditing Review, 15: 75-88.

Keskin S (2014). Türkiye'de kadın girişimcilerin durumu. Journal of Entrepreneurship and Developmen, 9: 71-94.

Kobu B (2017). Üretim Yönetimi. Beta basım yayım. İstanbul.

KSO (2022). Konya Ekonomik Verileri. Konya: Konya Sanayi Odası (KSO).

Mazgal B, Bozoğlu B (2006). Samsun ilinde tarıma dayalı sanayinin geliştirilebilmesi üzerine bir araştırma. Department of Agricultural Economics. Ondokuz Mayiz University. Samsun 1-114.

Mecek G (2020). Küçük ve Orta Büyüklükteki İşletmelerin (KOBİ) Uluslararası Tanımlama Ölçütleri ve Kavramlaştırılması, Ekonomi İşletme Siyaset ve Uluslararası İlişkiler Dergisi, 6 (1), 29-55.

Moser CA, Stuart A (1953). An experimental study of quota sampling. Journal of the Royal Statistical Society. Series A (General), 116(4), 349-405.

Powers D, Xie Y (2008). Statistical methods for categorical data analysis (Vol. 2). Bingley: Emerald Group Publishing.

Tan S, Tepeli C, Everest B (2017). Investigation of R&D Potential of the Agricultural Industry Firms: Çanakkale Province Case. In International Conference On Eurasian Economies. Beykent University. İstanbul 379-385.

TÜRKPATENT (2022). Patent ve Faydalı Model Başvurularının NACE Sınıflandırmasına Göre Sektörel Dağılımı. https://www.turkpatent.gov.tr/patent-ve-faydali-model. web (Türk Patent ve Marka Kurumu).

Ulas F, Çakır C (2004). The Fields of Occupation, Socio-Economic Structures and Capacity Use of Agribusiness Firms in Van Province. Graduate school of natural sciences. Yuzuncu Yıl University. Van.

Unluer M, Günes E (2013). An Analysis of Factors Affecting Repayment of Agricultural Credits. Journal of Agricultural Faculty of Gaziosmanpasa University, 30(2), 86-93.

Üçler YT, Karaçor Z (2015). Bölgesel Kalkınmada Üniversite-Sanayi İşbirliği Yöntemleri: Konya Üniversitelerinde ve Sanayisinde Bir Araştırma. Journal of Selcuk University Vocational School of Social Sciences. 18. 89-116.

Yağar F, Dökme S (2018). Niteliksel Araştırmaların Planlanması: Araştırma Soruları, Örneklem Seçimi, Geçerlik ve Güvenirlik. Gazi Journal of health sciences. 3, 1-9.

Yalcin H, Esengün K (2008). Food Regulation in EU-Turkey and Food Industry Farms Structure in Tokat. Journal of the Agricultural Faculty of Gaziosmanpaşa University, 25, 37-49.

Yardımcı A, Müftüoğlu EB (2015). Üniversite Sanayi İşbirliğine Sanayi kesiminin Bakışı. Ankara University SBF Journal, 70, 815-838.

Yüksel O (2008). Uluslararası finansal raporlama standardı III kapsamında işletme birleşmeleri ve muhasebeleştirilmesi. Phd. Marmara University. İstanbul-Turkey.

Selcuk Journal of Agriculture and Food Sciences
SJAFS
http://sjafs.selcuk.edu.tr/sjafs/index
(2022) 36 (3), 476-481
Research Article
e-ISSN: 2458-8377
DOI:10.15316/SJAFS.2022.062

Determination of the ADF and IVOMD Content of Sugarcane Using Near Infrared Spectroscopy Coupled with Chemometrics

Ozcan CATALTAS[1,*], Kemal TUTUNCU[2]
[1,2]Selçuk University, Faculty of Technology, Department of Electrical Electronic Engineering, Konya, Türkiye

ARTICLE INFO

Article history:
Received date: 31.10.2022
Accepted date: 28.11.2022

Keywords:
Near-infrared spectroscopy
Chemometrics
Partial least squares regression
Food analysis

ABSTRACT

Sugarcane is a plant whose quality parameters are required to be determined both for being one of the substances used in sugar production and for being used as animal feed. Near-infrared spectroscopy is a technique that has already been used for predicting the parameters of various plants and has gained popularity in recent years. This study proposes a near-infrared spectroscopy-based model for the rapid and effortless analysis of acid detergent fiber fraction and vitro organic matter digestibility parameters of the sugarcane plant. Partial least squares regression was combined with common preprocessing methods for modeling. This model yielded an R^2_{CV} value of 0.935 and 0.953 for the acid detergent fiber fraction and vitro organic matter digestibility parameters, respectively. Then, the spectra from three handheld spectrometers were combined using a proposed combination method to generate new spectra with higher spectral resolution. New models were built using these generated spectra and compared to the previous result. Obtained results showed that combining spectra from different spectrometers can improve model performance.

1. Introduction

Near-Infrared Spectroscopy (NIRS) has become a popular method in recent years as it allows non-destructive and fast analysis. NIRS, which is based on measuring vibrational frequencies in the near-infrared (NIR) region (780-2500 nm) in the electromagnetic spectrum and is characterized by combination vibrations with molecular overtones, stands out as an advantageous method. Chemical bonds with high vibrational frequencies, such as C-H, O-H, and N-H form overtone and combination bands in the NIR region and their intensity can be measured in this region (Ciurczak et al. 2021). Thus, it can be easily used in qualitative and quantitative analysis in many areas, such as agriculture, food, and pharmacy. Although it has been used in many different areas, the most intensive studies have been carried out in the food sector. Several studies have been conducted to determine food quality (Mohamed et al. 2021; Teye et al. 2020; Yang et al. 2022), detect food adulteration (De Girolamo et al. 2020; Genis et al. 2021; Laborde et al. 2021), and classify food types (Fu et al. 2022).

Sugarcane is one of the primary materials for sugar production. Its production is demanding as it requires special growing conditions (Guo et al. 2014). For this reason, it is important to determine the quality of the products and to apply the necessary fertilization or insecticide spraying. For this purpose, several studies have been carried out to determine the mineral content (Steidle Neto et al. 2017), to predict the crude protein and sugar content (Ryckewaert et al. 2022), to determine the lignin content of sugarcane (Assis et al. 2017), to screen sugarcane breeding (Guo et al. 2014), and to determine soil organic carbon in sugarcane fields (Zhao et al. 2022). Although sugarcane is considered inferior as an animal feed, its high dry matter and organic matter digestibility means it can be a good feed, particularly for ruminants (So et al. 2020).

Spectra acquisition in NIRS has traditionally been performed with expensive laboratory-type instruments (Schuler et al. 2009). In recent years, however, mini/micro spectrometers have continued to be developed that work in a narrow spectrum or with low spectral resolution. The increase in the variety of these spectrometers, their widespread use, and the increase in resolution will facilitate more comprehensive studies in the NIRS.

The spectra obtained from spectrometers cannot be used directly as they may contain several distortions. Although various preprocessing methods have been developed to suppress these distortions, there is no global method. On the other hand, various methods are being developed to extract meaningful data from the

* Corresponding author email: *ozcancataltas@selcuk.edu.tr*

preprocessed spectrum. Application examples of artificial neural networks (Pérez-Marín et al. 2007), deep learning (Cui & Fearn 2018), or ensemble methods (Kim et al. 2023; Mishra et al. 2020) have been encountered frequently in recent years. However, partial least square regression (PLSR), a cult method for qualitative analysis, is still the most popular method.

The objective of our work is twofold: to develop a model for the prediction of acid detergent fiber fraction (ADF) and in vitro organic matter digestibility (IVOMD) content of sugarcane samples using the spectra of different handheld spectrometers and to investigate whether combining the spectra of different devices would be beneficial.

The paper is organized as follows: Section two provides brief information on preprocessing techniques and partial least squares regression. The properties of the used dataset and spectra combining procedure are also given in section two. Performance metrics and results are presented in section three. Section four contains the main conclusion of our study.

2. Materials and Methods

2.1. Dataset description

This study used the sugarcane dataset to build and evaluate a reliable model. This dataset contains spectra of 60 sugarcane samples which measured different regions of sugarcane. These spectra were obtained from eight different spectroscopy instruments; each has different ranges and resolutions. Of these, NIRScan Nano (Texas Instrument), MicroNIR1700 (Viavi), and TellSpec were included in this study. The main reason for our choice is that all three devices operate in a spectral range close to each other. Detailed information about the chosen instruments is shown in Table 1. The dataset also included four reference values: Total sugar content (TS), crude protein (CP), ADF, and IVOMD. Because (Ryckewaert et al. 2022) used TS and CP in their study, only ADF and IVOMD parameters were included. Statistical information of these parameters is given in Table 2. Absorbance spectra and their relation to the ADF and IVOMD parameters are shown in Figures 1 and 2. The dataset is available at (Zgouz et al. 2020).

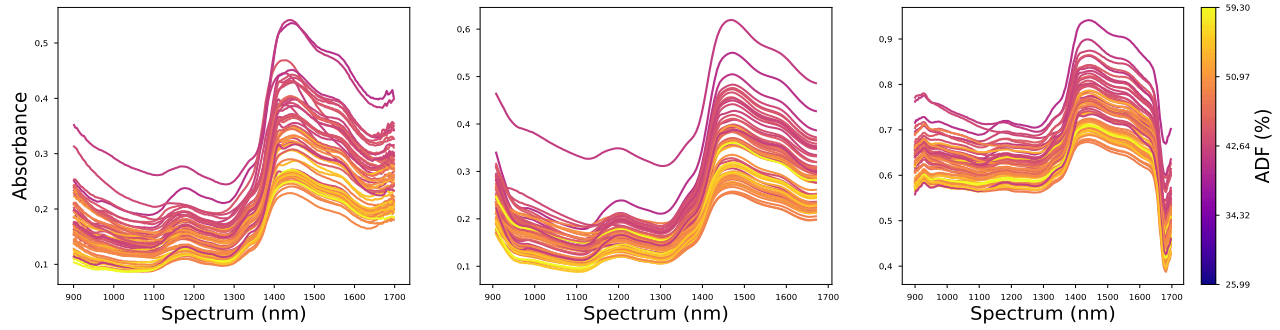
Table 1
Spectral range and resolution of the instruments used in the study

Instrument	Spectral Range (nm)	Resolution (nm)	Number of Features
NIRScan Nano	901-1701	3.9	228
MicroNIR1700	908-1676	6.1	125
TellSpec	900-1700	3.8	256

Table 2
Statistical information on ADF and IVOMD parameters

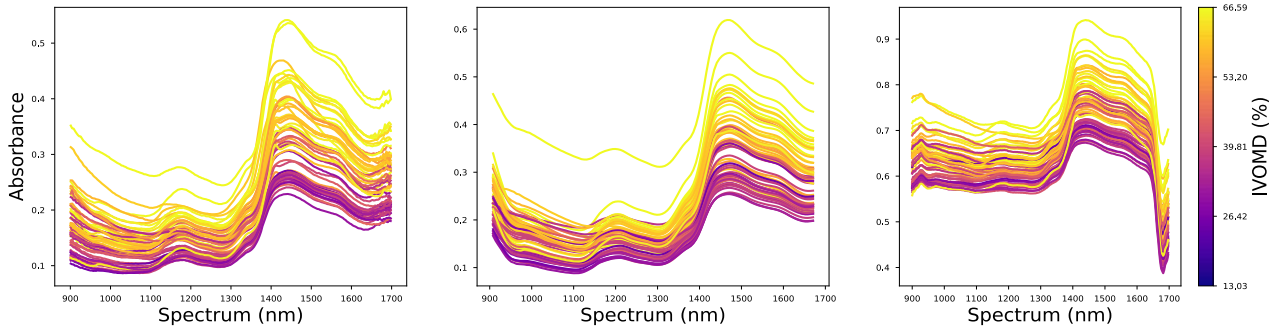
Parameter	Min	Max	Mean	Std
ADF (% DM)	25.99	59.30	39.17	8.61
IVOMD (% DM)	13.03	66.59	41.03	14.97

DM: Dry Matter



a) b) c)

Figure 1
The relation of used spectra and ADF content. These spectra belong to a) NIRScan Nano, b) MicroNIR1700, c) TellSpec



a) b) c)

Figure 2
The relation of used spectra and IVOMD content. These spectra belong to a) NIRScan Nano, b) MicroNIR1700, c) TellSpec

2.2. Preprocessing the spectra

Preprocessing the spectra is one of the crucial steps in NIRS that directly affects the performance of models. The sugarcane spectra were preprocessed before applying regression analysis using a combination of common preprocessing methods. Standard Normal Variate (SNV) (Barnes et al. 1989), Multiplicative Scatter Correction (MSC) (Geladi et al. 1985), Savitzky-Golay (SG) (Savitzky & Golay 1964), and a combination of these methods were used as preprocessing method.

2.3. Partial least squares regression

The main goal of a regression model is to find a relationship between independent variables and dependent variable(s). However, in some cases, as in spectroscopy data, many independent variables correlate with others. This situation is named as multicollinearity problem (Frank & Friedman 1993). To deal with this problem, (Wold et al. 1983) proposed partial least squares regression. PLSR proposes a solution by reducing the dimensionality of correlated variables.

2.4. Spectra combination procedure

In this study, a spectrum combining technique was proposed that combines spectra from three instruments. Since the spectra of different instruments contain baseline differences, each spectrum in the dataset was first normalized to a range between 0 and 1. After, these spectra were combined column by column and sorted by wavelength number. In this new spectrum, peaks were seen across the spectrum. A five-point moving average filter was used to eliminate these peaks. The combined dataset contains 609 features. The combination procedure is illustrated in Figure 3.

3. Results and Discussion

In this paper, preprocessing methods and regression models were implemented in Python (version 3.7.13) using scikit-learn library (version 1.0.2) (Lemaitre 2021). Two popular metrics were chosen to evaluate model performance. One of them, the coefficient of determination (R^2), is a measure that corresponds to the proportion of variation for a dependent variable that is explained by the independent variables. R^2 values close to 1 are preferable. The second metric, root mean squared error (RMSE), represents the square root of the average squared difference between the target value and the predicted value. Since RMSE is based on error, values closer to 0 indicate that the model performs better. The formulas of R^2 and RMSE are given in Equations 1 and 2.

$$R^2 = 1 - \frac{\sum_{i=1}^{N}(y_i - \hat{y}_i)^2}{\sum_{i=1}^{N}(y_i - \bar{y}_i)^2} \quad (1)$$

$$RMSE = \sqrt{\frac{1}{N}\sum_{i=1}^{N}(y_i - \hat{y}_i)^2} \quad (2)$$

In Equations 1 and 2, N is the number of samples, y_i, $\hat{y}_i$, and $\bar{y}_i$ are real, predicted, and mean values of the target, respectively.

This study proposes a chemometrics-based method for determining ADF and IVOMD parameters of sugarcane. The first experiment was carried out using classical PLSR. Different preprocessing methods were applied to the spectra taken from NIRScan, MicroNIR1700, and TellSpec instruments. At this point, popular preprocessing methods such as SNV, MSC, SG, and their binary combinations were preferred. In addition, a search space was created to determine the optimal values of window length, the order of the polynomial, and the order of derivative for SG. This search space was 5-29 for window length and 0-2 for the order of polynomial and derivative. Models were built to determine ADF and IVOMD parameters with preprocessed spectra and PLSR. At this stage, the dataset was randomly divided for calibration and testing at a rate of 66% to 33%. That is, 40 samples are used for the calibration of the model and 20 samples are reserved for the testing. Then, the optimal latent variables (LV) value for PLSR was determined using 5-fold cross-validation (CV) with the calibration set. In determining the LV, 1-15 values were selected as the search space. Table 3 and Table 4 show the performance results of the created CV model for the most appropriate LV value. Then, a new model was created using the calibration data set for the most appropriate LV value. The performance of this model was evaluated using the test set and is given in Table 3 and Table 4.

Table 3
Obtained results on prediction of the ADF parameter.

Instrument	Preprocessing	LV	R^2_{test}	R^2_{CV}	$RMSE_{test}$	$RMSE_{CV}$
Combined	SG(1,2,9)	7	0.885	0.953	3.281	1.743
NIRScan Nano	SG(1,2,13) + MSC	8	0.823	0.935	4.074	2.044
NIRScan Nano	MSC + SG(0,1,21)	13	0.912	0.899	2.868	2.553
MicroNIR1700	SG(2,2,5) + SNV	4	0.926	0.849	2.639	3.118
Combined	MSC + SG(2,2,19)	9	0.939	0.797	2.385	3.628
MicroNIR1700	SG(2,2,9) + SNV	3	0.951	0.797	2.138	3.624
TellSpec	SNV + SG(1,2,19)	5	0.845	0.671	3.813	4.610
TellSpec	SNV + SG(2,2,25)	4	0.945	0.606	2.276	5.047

The results are sorted based on R^2_{CV} in descending order. LV: Latent variables, SG: Savitzky-Golay (window length, the order of the polynomial, the order of the derivative)

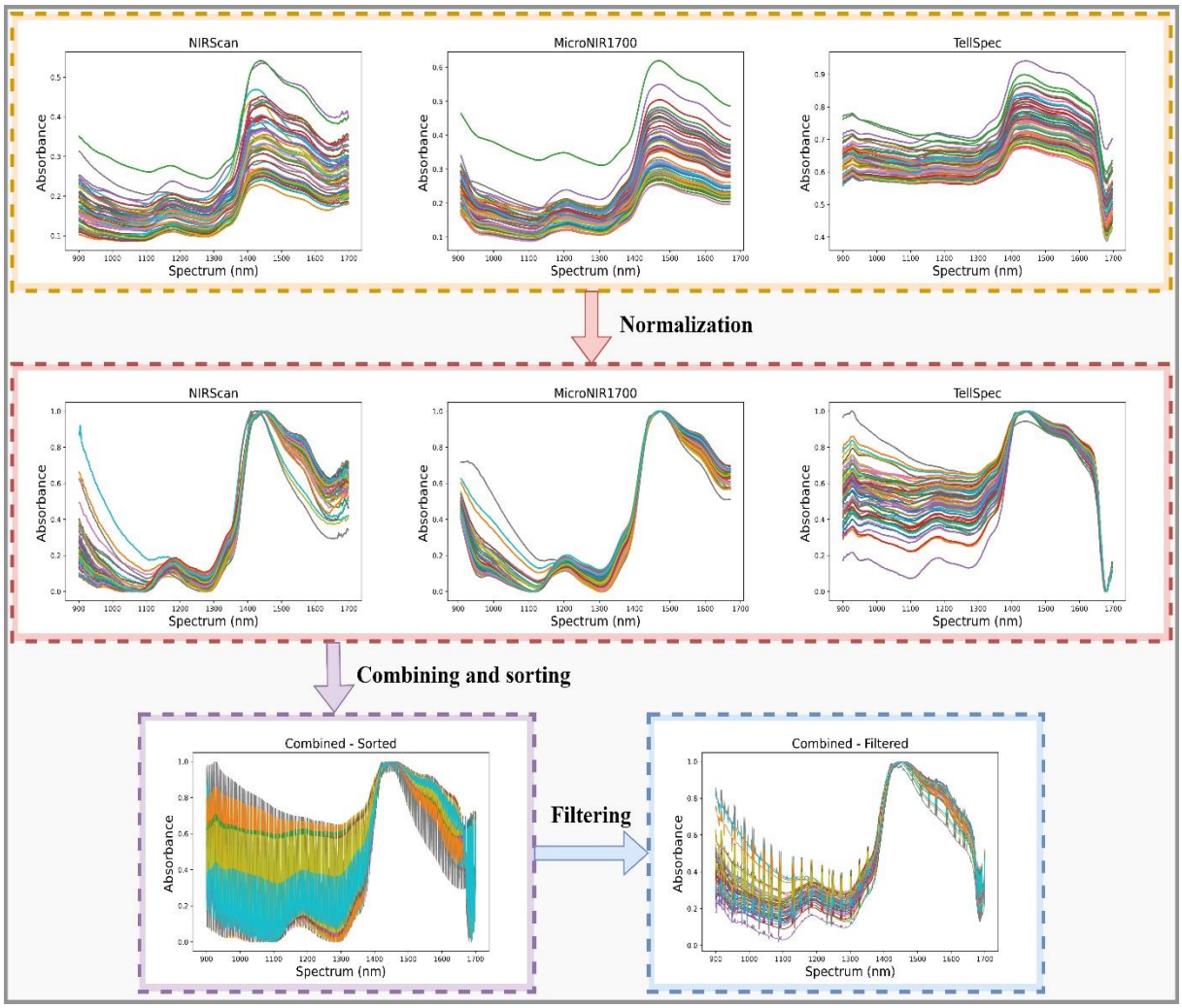


Figure 3
Application steps of the proposed spectrum combining algorithm

In the second experiment, the spectra of three instruments were combined, as explained in section 2.4. After the combination, the obtained spectra had a mean resolution value of 1.318 ± 0.899 nm. Similar preprocessing and regression process was done for the new spectra. Obtained results are shown in Table 3 and Table 4.

When we examine Table 3, the best performance according to R^2_{CV} and $RMSE_{CV}$ metrics belongs to the combined spectra. The higher R^2_{test} value was obtained with MicroNIR1700 spectra. For predicting the IVOMD parameter, the spectra of NIRScan Nano showed higher R^2_{CV} and lower $RMSE_{CV}$ values, while the combined spectra showed higher R^2_{test} and lower $RMSE_{test}$ values.

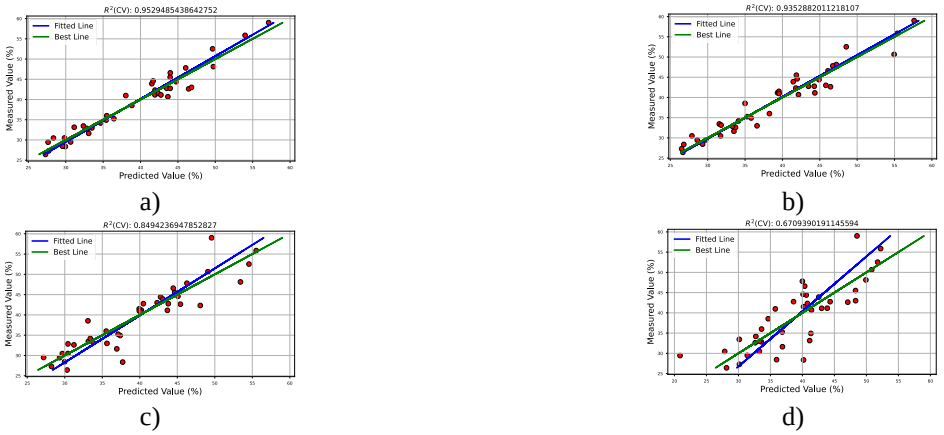


Figure 4
Predicted vs. Measured ADF values. Best values are given according to R^2_{CV} for instruments; a) Combined, b)NIRScan Nano, c) MicroNIR1700, d) TellSpec

Table 4
Obtained results on prediction of the IVOMD parameter.

Instrument	Preprocessing	LV	R^2_{test}	R^2_{CV}	$RMSE_{test}$	$RMSE_{CV}$
NIRScan Nano	SG(1,2,19) + MSC	12	0.746	0.953	8.064	3.128
Combined	SG(1,2,17)	10	0.858	0.945	6.037	3.375
NIRScan Nano	SG(0,0,5) + SNV	13	0.899	0.879	5.089	5.023
MicroNIR1700	SNV + SG(2,2,5)	4	0.893	0.867	5.228	5.257
Combined	SG(2,2,21)	5	0.930	0.861	4.244	5.373
MicroNIR1700	SG(2,2,13)	4	0.866	0.778	5.863	6.797
TellSpec	SG(1,2,21) + SNV	4	0.789	0.673	7.363	8.241
TellSpec	SNV + SG(2,2,13)	3	0.909	0.541	4.840	9.770

The results are sorted based on R^2_{CV} in descending order. LV: Latent variables, SG: Savitzky-Golay (window length, the order of the polynomial, the order of the derivative)

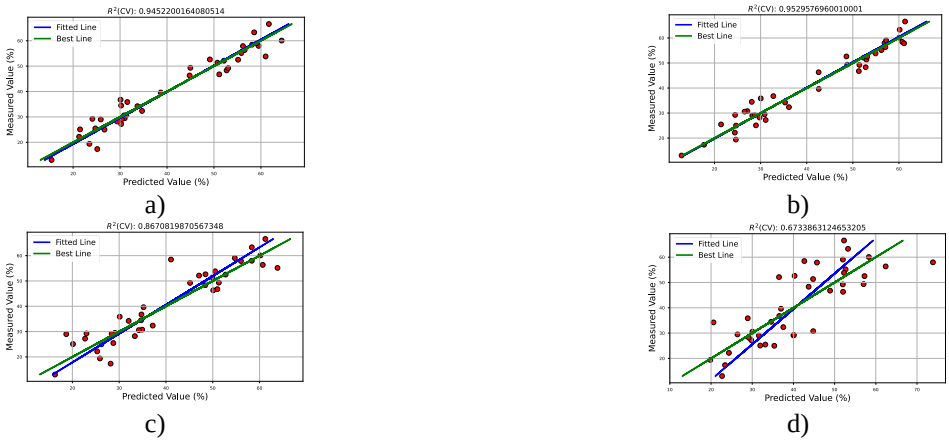


Figure 5
Predicted vs. Measured IVOMD values. Best values are given according to R^2_{CV} for instruments; a) Combined, b) NIRScan Nano, c) MicroNIR1700, d) TellSpec

Table 5
The mean value of obtained results for every preprocessing combination

Instrument	ADF		IVOMD	
	R^2_{test}	R^2_{CV}	R^2_{test}	R^2_{CV}
Combined	0.876	0.830	0.843	0.849
NIRScan Nano	0.848	0.869	0.808	0.879
MicroNIR1700	0.832	0.685	0.819	0.734
TellSpec	0.817	0.602	0.767	0.598

Each value in this table was calculated by taking the average of 390 combinations.

Figure 4 and Figure 5 show the predicted and measured output for each spectra. As expected, more successful results were obtained with the spectra from the NIRscan Nano, since the NIRscan Nano has a higher spectral resolution than the MicroNIR1700 device. On the contrary, the worst performance for both parameters was obtained with the spectra of TellSpec, although it has the highest spectral resolution among the three instruments. This may be due to the spectral sensitivity of the device.

One of the conclusions from this study is that the preprocessing method to be used varies according to the spectrum. All of the high-success models were obtained with different combinations of preprocessing methods or different parameters. This is the weakest point of classical chemometrics. Another conclusion is that combining the spectra of different instruments can yield better results. Table 5 shows the mean values of obtained results. It is essential to take the mean of all combinations to eliminate the parameter selection effect of preprocessing methods. According to Table 5, combined spectra give the highest scores according to the R^2_{test} metric for both targets and the second highest scores according to the R^2_{CV} metric.

4. Conclusion

In this study, the ADF and IVOMD parameters of sugarcane were determined noninvasively using near-infrared spectroscopy. PLSR, coupled with common preprocessing methods, was utilized for the building prediction model. The best results were obtained with NIRScan Nano according to R^2_{CV} for both parameters (0.928 for ADF and 0.947 for IVOMD). The spectra from three instruments were combined to increase spectral resolution. New PLSR models were developed using these spectra. Obtained results have shown that combining spectra from different spectrometers helps to improve model performance. Future studies can focus on developing machine learning-based spectra combination algorithms.

5. Acknowledgements

This study was partially supported Selcuk University Academic Staff Training Program (2018-OYP-041).

6. References

Assis C, Ramos RS, Silva LA, Kist V, Barbosa MHP, and Teófilo RF (2017). Prediction of Lignin Content in Different Parts of Sugarcane Using Near-Infrared Spectroscopy (NIR), Ordered Predictors Selection (OPS), and Partial Least Squares (PLS). *Applied Spectroscopy* 71:2001-2012.

Barnes RJ, Dhanoa MS, and Lister SJ (1989). Standard Normal Variate Transformation and De-Trending of Near-Infrared Diffuse Reflectance Spectra. *Applied Spectroscopy* 43:772-777. 10.1366/0003702894202201

Ciurczak EW, Igne B, Workman Jr J, and Burns DA. 2021. *Handbook of near-infrared analysis*: CRC press.

Cui C and Fearn T (2018). Modern practical convolutional neural networks for multivariate regression: Applications to NIR calibration. *Chemometrics and Intelligent Laboratory Systems* 182:9-20. https://doi.org/10.1016/j.chemolab.2018.07.008

De Girolamo A, Arroyo MC, Cervellieri S, Cortese M, Pascale M, Logrieco AF, and Lippolis V (2020). Detection of durum wheat pasta adulteration with common wheat by infrared spectroscopy and chemometrics: A case study. *LWT* 127:109368. https://doi.org/10.1016/j.lwt.2020.109368

Frank IE, and Friedman JH (1993). A Statistical View of Some Chemometrics Regression Tools. *Technometrics* 35:109-135. 10.2307/1269656

Fu L, Sun J, Wang S, Xu M, Yao K, Cao Y, and Tang N (2022). Identification of maize seed varieties based on stacked sparse autoencoder and near-infrared hyperspectral imaging technology. *Journal of Food Process Engineering* 45. 10.1111/jfpe.14120

Geladi P, MacDougall D, and Martens H (1985). Linearization and Scatter-Correction for Near-Infrared Reflectance Spectra of Meat. *Applied Spectroscopy* 39:491-500. 10.1366/0003702854248656

Genis HE, Durna S, and Boyaci IH (2021). Determination of green pea and spinach adulteration in pistachio nuts using NIR spectroscopy. *LWT* 136:110008. https://doi.org/10.1016/j.lwt.2020.110008

Guo H, Chen J, Pan T, Wang J, and Cao G (2014). Vis-NIR wavelength selection for non-destructive discriminant analysis of breed screening of transgenic sugarcane. *Analytical Methods* 6:8810-8816. 10.1039/C4AY01833H

Kim S-Y, Hong S-J, Kim E, Lee C-H, and Kim G (2023). Application of ensemble neural-network method to integrated sugar content prediction model for citrus fruit using Vis/NIR spectroscopy. *Journal of Food Engineering* 338:111254. https://doi.org/10.1016/j.jfoodeng.2022.111254

Laborde A, Puig-Castellví F, Jouan-Rimbaud Bouveresse D, Eveleigh L, Cordella C, and Jaillais B (2021). Detection of chocolate powder adulteration with peanut using near-infrared hyperspectral imaging and Multivariate Curve Resolution. *Food Control* 119:107454. https://doi.org/10.1016/j.foodcont.2020.107454

Lemaitre G (2021). scikit-learn. *Available at https://github.com/scikit-learn/scikit-learn*.

Mishra P, Biancolillo A, Roger JM, Marini F, and Rutledge DN (2020). New data preprocessing trends based on ensemble of multiple preprocessing techniques. *TrAC Trends in Analytical Chemistry* 132:116045. https://doi.org/10.1016/j.trac.2020.116045

Mohamed H, Nagy P, Agbaba J, and Kamal-Eldin A (2021). Use of near and mid infra-red spectroscopy for analysis of protein, fat, lactose and total solids in raw cow and camel milk. *Food Chemistry* 334:127436. https://doi.org/10.1016/j.foodchem.2020.127436

Pérez-Marín D, Garrido-Varo A, and Guerrero JE (2007). Non-linear regression methods in NIRS quantitative analysis. *Talanta* 72:28-42. https://doi.org/10.1016/j.talanta.2006.10.036

Ryckewaert M, Chaix G, Héran D, Zgouz A, and Bendoula R (2022). Evaluation of a combination of NIR micro-spectrometers to predict chemical properties of sugarcane forage using a multi-block approach. *Biosystems Engineering* 217:18-25. https://doi.org/10.1016/j.biosystemseng.2022.02.019

Savitzky A, and Golay MJE (1964). Smoothing and Differentiation of Data by Simplified Least Squares Procedures. *Analytical Chemistry* 36:1627-1639. 10.1021/ac60214a047

Schuler LP, Milne JS, Dell JM, and Faraone L (2009). MEMS-based microspectrometer technologies for NIR and MIR wavelengths. *Journal of Physics D: Applied Physics* 42:133001. 10.1088/0022-3727/42/13/133001

So S, Cherdthong A, and Wanapat M (2020). Improving sugarcane bagasse quality as ruminant feed with Lactobacillus, cellulase, and molasses. *Journal of Animal Science and Technology* 62:648.

Steidle Neto AJ, Toledo JV, Zolnier S, Lopes DdC, Pires CV, and Silva TGFd (2017). Prediction of mineral contents in sugarcane cultivated under saline conditions based on stalk scanning by Vis/NIR spectral reflectance. *Biosystems Engineering* 156:17-26. https://doi.org/10.1016/j.biosystemseng.2017.01.003

Teye E, Anyidoho E, Agbemafle R, Sam-Amoah LK, and Elliott C (2020). Cocoa bean and cocoa bean products quality evaluation by NIR spectroscopy and chemometrics: A review. *Infrared Physics & Technology* 104:103127. https://doi.org/10.1016/j.infrared.2019.103127

Wold S, Martens H, and Wold H (1983). The multivariate calibration problem in chemistry solved by the PLS method. In: Kågström B, and Ruhe A, editors. Matrix Pencils. Berlin, Heidelberg: Springer Berlin Heidelberg. p 286-293.

Yang J, Luo X, Zhang X, Passos D, Xie L, Rao X, Xu H, Ting KC, Lin T, and Ying Y (2022). A deep learning approach to improving spectral analysis of fruit quality under interseason variation. *Food Control* 140:109108. https://doi.org/10.1016/j.foodcont.2022.109108

Zgouz A, Héran D, Barthès B, Bastianelli D, Bonnal L, Baeten V, Lurol S, Bonin M, Roger J-M, Bendoula R, and Chaix G (2020). Dataset of visible-near infrared handheld and micro-spectrometers – comparison of the prediction accuracy of sugarcane properties. *Data in Brief* 31:106013. https://doi.org/10.1016/j.dib.2020.106013

Zhao X, Zhao D, Wang J, and Triantafilis J (2022). Soil organic carbon (SOC) prediction in Australian sugarcane fields using Vis–NIR spectroscopy with different model setting approaches. *Geoderma Regional* 30:e00566. https://doi.org/10.1016/j.geodrs.2022.e00566

Selcuk Journal of Agriculture and Food Sciences
SJAFS
http://sjafs.selcuk.edu.tr/sjafs/index
(2022) 36 (3), 482-492
Research Article
e-ISSN: 2458-8377
DOI:10.15316/SJAFS.2022.063

The effects of Ortho Silicon (Optysil) and *Ascophyllum nodosum* Based Seaweed Extract (KelpGreen) Applications on the Quality of Table Grape cvs. Gök Üzüm and Müşküle

Zeki KARA[1,*], Burcu Nur UĞUR[2], Osman DOĞAN[1]
[1]Selçuk University, Faculty of Agriculture, Department of Horticulture, Konya, Türkiye
[2]Selçuk University, Institute of Science, Department of Horticulture, Konya, Türkiye

ARTICLE INFO

Article history:
Received date: 20.04.2022
Accepted date: 05.12.2022

Keywords:
Maturity index
Plant activators
Post harvest
Quality retention
Weight loss

ABSTRACT

Table grapes are one of the most consumed non-climacteric fruits globally, and practices for their quality are socioeconomically important. In this study, the effects of the combined and separate applications of two plant activators Ortho Silicon (Si, Optysil, 0.5 mL L^{-1}) and *Ascophyllum nodosum*-based seaweed extract (ANE, KelpGreen 2.5 ml L^{-1}) were tested by two applications after fruit set 15 days intervals to table grapes cvs. Gök Üzüm and Müşküle in a producer vineyard at Hadim town of Konya province in middle Anatolia 37°2'15"N 32°34'53" E, 1060 m above sea level. The effects of the applications on ripening, cluster and berry characteristics and post harvest shelf life during 10 days of storage at room conditions were analyzed. Both plant activators provided an increase in cluster size and an improvement in ripening. Si+ANE was more effective on the maturation and quality retention during the post-harvest shelf-life period. All applications provided reduction in weight loss (WL), decay rate and berry dullness, and it reduced berry separation force due to the drying of the peduncle and decreased the maturity index (MI) increase in the post harvest period. Thus, it contributed to the formation and retention of fruit quality. According to the data obtained from this study, improvements in the sustainable preservation of table grape quality can be achieved by applying Ortho Silicon and *A. nodosum* based seaweed extract separately and together between after fruit set and before veraison.

1. Introduction

Table grapes are one of the most consumed non-climacteric fruits worldwide. Table grape is a fruit with a relatively low physiological activity rate, which does not ripen further after harvest. Its quality depends on different attributes related to appearance, colour, texture, flavour, and aroma. “Veraison” begins with maturation, accumulation of sugars, softening of berries, synthesis of anthocyanin, metabolism of organic acids and accumulation of aroma compounds. Soluble solids content (°Brix) and sugar/acid ratios are primary indicators of table grape quality and are minimum requirements for each variety. Table grape flavour is a complex and important quality characteristic as it is a mixture of hundreds of different volatile compounds synthesized during ripening. After harvest, table grapes deteriorate rather quickly, as they are exposed to significant WL because of the drying of the stem and the peduncle, causing browning, WL, and berry softening. In addition, rot by *Botrytis cinerea* also causes large losses, limiting conservation (Palou et al., 2010). Many internal factors such as the structure and consistency of the kernel and fruit flesh, and the ripening rate, as well as external factors such as temperature and relative humidity, are effective in maintaining quality.

Consumers' high preference for table grapes is due to their excellent organoleptic and nutritional qualities, and consumption has increased significantly in recent years. According to 2019 data from the International Vine and Wine Organization (OIV), about 36% of total grape production is for fresh consumption, with China being the largest consumer, followed by India and the European Union (EU). Table grape production has doubled in the last two decades. According to USDA data, world production for 2019/20 is estimated to be around 23.4 million tons (Romero et al., 2020). Turkey supplies 51% of approximately 3.7 million tons of grape production to the local and global markets as table grapes (TÜİK, 2022).

* Corresponding author email: *zkara@selcuk.edu.tr*

Silicon is a beneficial element for higher plants, as its effects are often linked to morphological, physiological, and molecular aspects of plants (Ma et al., 2004; Lobato et al., 2009). Silicon acts as a semi-essential element for plants because its deficiency can cause various abnormalities in the growth, reproduction, and general development of plants (Lobato et al., 2009). Silicon feeding to plants improves the plant protection mechanism against diseases and insects (Dallagnol et al., 2011; Guntzer et al., 2012; Liu et al., 2014). Exogenous silicon application protected against UV-B stress by stimulating the antioxidant defense system of soybean, wheat, and corn seedlings (Tripathi et al., 2017), accelerated plant ripening (Matichenkov, 1990), increased growth in citrus fruits by 30 to 80%, increased fruit ripening. It accelerated by 2-4 weeks and increased yield (Taranovskaia, 1939) and °Brix (Matichenkov et al., 2001). Silica nanoparticles provided protection by reducing oxidative stress in pea seedlings, and the activities of enzymes such as superoxide dismutase and ascorbate peroxidase increased significantly with silica nanoparticles (Tripathi et al., 2015). It has been determined that silicon can replace phosphate in DNA and RNA molecules and increase the stability of these molecules (Snyder et al., 2016). Silicon increased the chlorophyll density in the leaf (Adatia and Besford, 1986), thus allowing the plant to use light more efficiently and to tolerate low or high light levels. Higher soluble silicon causes higher concentrations of the enzyme ribulose biphosphate carboxylase to be produced in leaf tissue (Adatia and Besford 1986). By regulating CO_2 metabolism, this enzyme supports plants to use CO_2 more efficiently.

Seaweeds are macroalgae that fit in the class Phaeophyceae and are best known as brown algae. They are mainly composed of polysaccharides such as laminarin, fucoidan and alginate. Many seaweed-based products are known to be beneficial for humans and plants. Seaweed extracts contain various bioactive compounds. Such bioactive compounds induce resistance in plants to different biotic and abiotic stresses. Seaweed extracts may also contain numerous plant bioactive inorganic and organic compounds such as mannitol, polysaccharides, oligosaccharides, phytohormones (auxins, cytokinins, gibberellins, betaine), antioxidants and vitamins. It also contains a low concentration of minerals (calcium, boron, zinc, potassium, phosphorus, magnesium, and some other trace elements). Seaweed extract can promote plant growth and increase the rate of photosynthesis. Seaweed extracts increased seed germination rates, crop growth, yield, and product shelf life. It can reduce the effect of diseases due to fungal, viral, and bacterial pathogens (Singh et al., 2021).

A bio stimulant can be defined as any beneficial microorganism or any organic substance that can increase plant growth, increase nutrient uptake, increase tolerance to abiotic and biotic stress, and increase crop yield. Bio stimulants have been obtained from seaweeds, bacteria, higher plants, fungi, humic acid, and other industrially processed materials according to their source. Phaeophycean seaweed, also known as brown seaweed, is the largest group with 2000 species. Their maximum biomass is found on the rocky coasts of the temperate zone of different countries. Products based on the brown seaweed *Ascophyllum nodosum* (L.) are mostly used in agriculture (Blunden and Gordon, 1986). Increased resistance to stress, improved crop yield, early germination of seeds, etc. There are numerous reports on the role of seaweed-based bio stimulants for crop protection and crop production (Beckett and Van Staden, 1989; Hankins and Hockey, 1990; Norrie and Keathley, 2005). Polysaccharide-rich extracts of seaweed have been shown to have an enhancing effect on plant growth (Hernández Herrera et al., 2016). Such activity of the extracts suggested the role of oligosaccharides as signalling molecules for the regulation of phytohormone-related genes in treated plants. However, polysaccharide-rich extracts that promote root growth in mung bean plants also demonstrated the presence of synthetic hormones in the core. A recent report suggested the role of *Ascophyllum nodosum* extract (ANE) in reducing mycotoxin production in wheat plants infected with Fusarium head blight (Gunupuru et al., 2019).

Emerging formulations based on ANE can improve plant growth as well as increase tolerance to abiotic stresses including heat, drought, and salinity. Several plant metabolic pathways are targeted by seaweed extracts to improve plant growth and tolerance to abiotic stresses (Craigie, 2011). Currently, more than 50 companies around the world produce seaweed extracts to stimulate plant growth, and these seaweed extracts are based on different types of seaweed found in the sea. ANE are the products that attract the most attention (Sharma et al., 2014). Several plant species have shown growth promotion under application of seaweed extract, but the mechanism behind such activity has not been very well studied (Battacharyya et al., 2015). Application of seaweed extracts increased seedling growth of lettuce (*Lactuca sativa* L.) (Moller and Smith, 1998). ANE formulation has been reported to increase growth and K^+ accumulation in almond plants (Prunus dulcis). Commercial products Grozyme and Megafol showed a similar effect on foliar application and stimulated plant growth (Saa et al., 2015).

In this study, the effects of pure and combined applications of Ortho Silicon and Seaweed extract (ANE) on product quality shelf life of table grapes cvs. Gök Üzüm and Müşküle were investigated.

2. Materials and Methods

In this study, table grape cvs. Gök Üzüm and Müşküle (*Vitis vinifera* L.) were used in producer vineyards in Hadim town of Konya province in middle Anatolia 37°2'15"N 32°34'53" E, 1060 m above sea level. After fruit set 15 days intervals Ortho Silicone (0.5 ml L^{-1}) and Seaweed extract (2.5 ml L^{-1}) were pulverized (Uwakiem, 2015) on vines. The samples were harvested in late (23rd) September as the normal commercial harvest time. The alternations of some quality parameters such as MI (°Brix % / Titratable acidity g L^{-1}), WL (%),

Skin rupture force (N), Berry detachment force (N), Berry brightness (Hue, h°, by using a cR- 400 chromometer, Konica Minolta, Japan), and Decay rate (%) were monitored 3 days interval for 10 days.

The study was designed as completely randomized blocks, Ortho Silicone and Seaweed extract effects were compared in SPSS 17.0 statistical program (SPSS Inc, Chicago, IL, USA) Duncan multiple comparison test, the applications JMP 7 statistical programs with Student's t test at $p< 0.05$ significance level.

3. Results and Discussion

The effects of the applications were evaluated as the effects on the product quality determined at the harvest and the effects on the shelf life monitored by keeping it in room conditions for 10 days after harvest.

3.1. Quality Parameters

The effects of seaweed, silicon, and seaweed+silicone applications on cvs. Gök Üzüm and Müşküle on cluster characteristics were significant. In both cultivars, the lowest cluster length and width were determined in the control, while the applications increased the cluster length and width. The effects of individual applications of silicon and seaweed were in the same statical group in both grape cultivars. The longest and widest clusters were obtained from seaweed+silicone treatment (Table 1). Berry length and width were also affected by the treatments like the cluster. The lowest values were recorded in the control in both cultivars, while the highest values were recorded from seaweed+silicone combined applications. On the other hand, the effects of applications on berry density were insignificant (Table 1).

Table 1
Cluster and berry characteristics

Cultivars	Application	Cluster length	Cluster width	Berry length	Berry width	Berry density
Gök Üzüm	Control	20.96±0.62 b	13.38±0.70 b	14.57±0.14 b	16.16±0.10 c	1.05±0.04
	Seaweed	21.25±0.43 b	13.79±0.65 b	15.69±0.50 a	17.20±0.68 b	1.06±0.03
	Silicone	21.67±0.31 b	14.46±0.59 ab	15.57±0.47 a	17.11±0.46 bc	1.05±0.04
	Seaweed+Silicone	22.79±0.44 a	15.38±0.82 a	16.02±0.43 a	18.19±0.63 a	1.06±0.02
	LSD≤.05	1.04	1.48	0.78	0.85	ns
Müşküle	Control	18.50±0.50 c	9.29±0.64 b	19.95±0.12 b	21.47±0.52 b	1.03±0.02
	Seaweed	19.88±0.45 b	9.92±0.26 b	20.21±0.10 b	21.97±0.33 b	1.05±0.06
	Silicone	19.83±0.51 b	9.96±0.58 b	20.30±0.41 ab	22.15±0.10 b	1.05±0.01
	Seaweed+Silicone	20.83±0.19 a	10.88±0.25 a	20.81±0.34 a	22.98±0.30 a	1.05±0.02
	LSD≤.05	0.97	0.95	0.46	0.58	ns

The effects of the applications of Ortho Silicone cvs. Gök Üzüm and Müşküle on the juice yield were significant. While the lowest must yield was detected in seaweed application to cv. Gök Üzüm, the highest yield was in control (Table 2). In cv. Müşküle, the lowest juice yield was recorded in seaweed+silicone application, while the highest yield was recorded in silicon application (Table 2). Differential effects of cultivars on seaweed, silicon, and seaweed+silicone treatments were attributed to genotypic response difference.

Table 2
Effects on berry composition

Cultivars	Application	Must yield	Maturity index	°Brix	Ph
Gök Üzüm	Control	74.43±3.59 a	7.05±0.35 b	19.93±0.55 c	3.56±0.04 c
	Seaweed	57.20±7.98 b	7.54±0.96 ab	20.97±0.46 b	3.76±0.01 ab
	Silicone	71.50±7.48 a	8.01±0.25 ab	22.00±0.20 a	3.73±0.03 b
	Seaweed+Silicone	62.47±8.61 ab	8.19±0.39 a	22.30±0.17 a	3.79±0.01 a
	LSD≤.05	12.24	1.27	0.51	0.03
Müşküle	Control	70.48±2.36 b	8.36±0.44 b	20.50±0.30	3.97±0.04 ab
	Seaweed	64.37±5.77 bc	8.89±0.53 ab	21.07±0.40	3.97±0.02 ab
	Silicone	79.80±2.28 a	9.82±0.82 a	21.20±0.56	3.88±0.01 b
	Seaweed+Silicone	60.94±5.06 c	10.08±0.67 a	21.37±0.55	4.02±0.12 a
	LSD≤.05	8.37	1.42	ns	0.13

MI was also significantly affected by the treatments in both cultivars. The greatest effect, in other words, the acceleration of ripening was recorded in seaweed+silicone application in both cultivars. The effects on °Brix were insignificant in cv. Müşküle, but significant in cv. Gök Üzüm. The highest must pH value was recorded in Si+ANE application in both grape varieties.

3.2. Weight Loss

As a result of the silicon, seaweed, and Si+ANE treatments applied to the cv. Gök Üzüm, the lowest weight losses on the 3rd, 7th and 10th days were determined in the Si+ANE application (2.52±0.11%, 3.79±0.17% and 6.31±0.28%). The highest WL were found in the control (3.60±0.82%, 7.74±0.28%, and 12.35±1.19%) (Figure 1a). WL increased as the waiting time increased after harvest. The WL of cv. Müşküle was less than that of cv. Gök Üzüm in all treatments. The lowest WL was Si+ANE (0.46±0.43%, 1.83±0.69% and 4.17±0.20%) in the three analysis periods, 3rd, 7th, and 10th determined in the application. The highest WL in this period was observed in the control (2.98±0.71%,

5.34±0.28% and 9.47±0.86%) (Figure 1b). All the applications made contributed to the extension of the postharvest life by reducing the WL in both grape cultivars. The effect of the applications made on the cv. Müşküle on WL was positive compared to the cv. Gök Üzüm and less WL was observed.

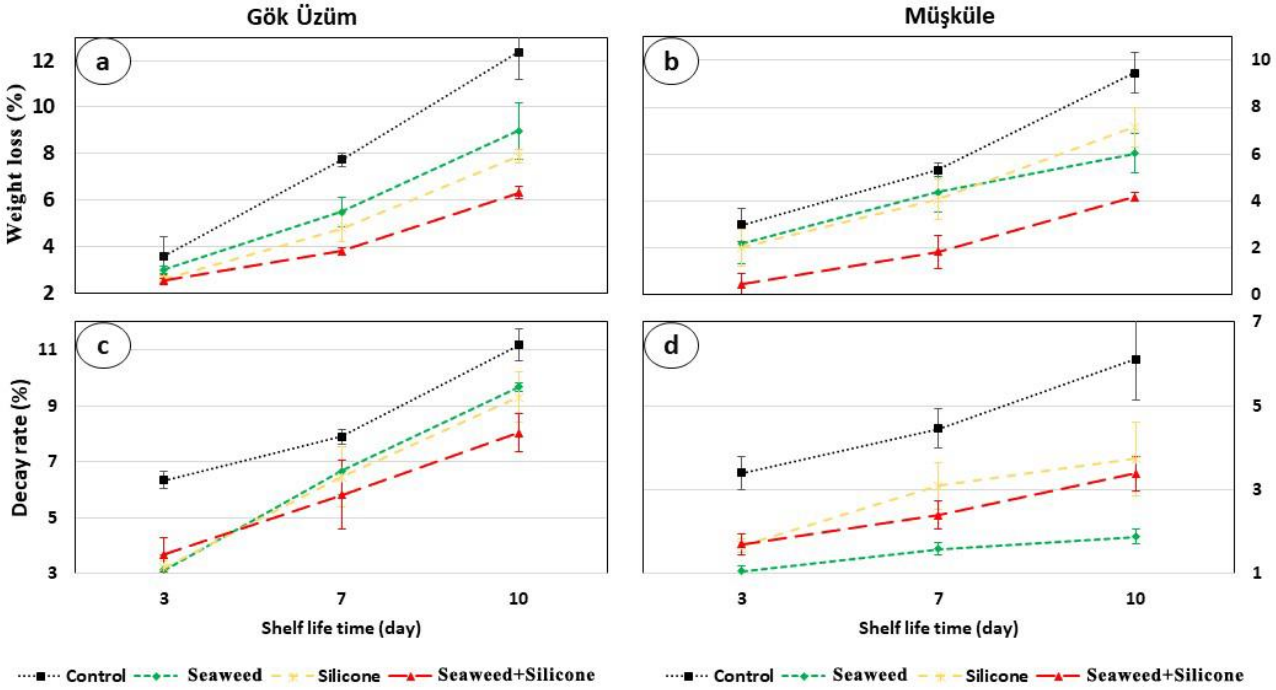


Figure 1
Effects of WL (a and b) and decay rates (c and d)

WL occurred at varying rates in the postharvest process of table grapes, depending on the various factors mentioned (Serrano et al., 2005). Different applications and new developments that have the effect of reducing WL results have shed light on today's modern preservation techniques. Like the results of some previous studies (Serrano et al., 2005; Valero et al., 2006), our different practices in this study were effective in reducing WL. ANE application increased the isopropanol fraction in vines (*Vitis vinifera* L.) and water potential and stomatal conductivity in K^+ and Ca_2^+ flows under drought stress (Mancuso et al., 2006). With the accumulation of K+, ionic and osmotic stresses can be overcome. Application of ANE increased water use efficiency under drought stress in the orange tree *Citrus sinensis* (Spann and Little, 2011). The use of ANE in vineyards with irregular irrigation may be a useful application by increasing water use efficiency in drought stress.

3.3. Decay Rate

The effect of the applications on the decay rate in cv. Gök Üzüm was highest in the control (6.34±0.30%, 7.90±0.26% and 11.16±0.57%) at the end of the 3rd, 7th, and 10th days, while the lowest decay was in the seaweed on the 3rd day (3.14±0.53%), and on the 7th and 10th days, Si+ANE (5.81±1.23% and 8.04±0.67%) was applied (Figure 1c). In cv. Müşküle, the highest decay rates were determined in the control (3.40±0.39%, 4.46±0.46% and 6.11±0.97%) during the post-harvest period, while the lowest decay was obtained by seaweed (1.07±0.12%, 1.58±0.15% and 1.89±0.17%), (Figure 1d). All applications were more effective in the cv. Müşküle and reduced rot in both cultivars.

Plant diseases pose a great threat to agricultural production and cause serious crop yield and quality loss worldwide (Etesami and Alikhani, 2017). The use of mineral nutrition to increase disease resistance in plants may be a practical alternative (Marschner, 1995). Silicon stands out among the minerals due to its effectiveness in reducing the severity of various plant diseases (Epstein, 1999). As a result of the emergence of serious physiological diseases and the reduction of quality and storage problems, the use of silicone has increasingly attracted people's attention. The use of silicon is known as one of the most environmentally friendly and sustainable ways to combat plant diseases and pests. It has been determined that silicon increases plant cell wall properties (Lux et al., 2002) and plant resistance against diseases (Fauteux et al., 2005).

Silicic acid polymerization within the apoplast creates an amorphous silica barrier and helps deter pathogen infection (Guerriero et al., 2016). Improved overall mechanical strength and an added outer layer of protection for plants explain many of the reported benefits in crop quality and yield after silicon fertilization. Successful infection enters the host plant by overcoming physical barriers of plant pathogens such as the waxy layer, cuticle, and cell walls. Physical barrier formation is a mechanism to control plant diseases (Kim et al., 2002), and make plant cells more vulnerable to fungal pathogen invasion and subsequent enzymatic degradation (Fau-

teux et al., 2005). Silicon supports plant growth by forming an outer protective layer and increasing the mechanical strength of plants. Silicon is typically cross-linked to hemicellulose, which improves the mechanical properties and regeneration of cell walls (Guerriero et al 2016). Silicon plays a role in cell wall stiffness and reinforcement and helps increase elasticity during elongation (Marschner, 2012). In primary cell walls, silicon interacts with cell wall components such as polyphenols and pectins, contributing to increased flexibility during longitudinal growth (Fauteux et al 2005; Liang et al 2015a). Well fed plants are known to be more resistant to diseases. Another mechanism by which silicon increases resistance to plant diseases is that it affects plant mineral nutrition. It contributes to the uptake of silicon substances, can increase the concentration of essential nutrients in plants. The silicate anion competes for adsorption sites in the soil, increasing the availability of sulfate, nitrate and phosphate in the soil and the ability of plants to retain these anions (Pozza et al., 2015). Marschner (1988) reported that macronutrient and micronutrient imbalances affect the growth power of plants, defense responses and affect the host's susceptibility to diseases. They can work directly on secondary metabolic pathways where cell wall defense system expansion and formation of fungistatic phenolic compounds occur by increasing mid lamella resistance or by creating physical and chemical barriers as in calcium conditions.

Climate changes and unscientific agricultural practices directly affect agricultural products negatively by weakening the defence system in plants and causing diseases (Anderson et al., 2004; Ayliffe and Lagudah., 2004). Plant diseases directly damage productivity. To prevent disease infection, plants have developed inducible defence processes (Conrath et al., 2002; Wiesel et al., 2014). Two types of disease defence mechanisms have been identified, namely induced systemic resistance (ISR) and systemic acquired resistance (SAR). To protect plants from a variety of pathogens, ISR mediates defence responses to jasmonate (JA) and ethylene (ET), while salicylic acid (SA) is important for PR (pathogenesis-related) gene activation for SAR mechanisms (Gaffney et al., 1993; Van Loon et al., 1998). In plants, elicitor molecules from pathogens are responsible for stimulating the defence system (Conrath et al., 2002; Wiesel et al., 2014). Not only chitin, lipopolysaccharides, and flagella of microbes, but also some chemically synthesized components such as 2,6-dichloro isonicotinic acid, b-aminobutyric acid, chitosan, benzothiadiazole and methyl jasmonate can induce SAR/ISR mechanisms against a wide variety of pathogens (Dixon, 2001; Mercier et al, 2001; Bektas and Eulgem, 2015; Iriti and Varoni, 2015). Different seaweeds have equipped themselves with important defence mechanisms to protect themselves from their pathogens (Potin et al., 1999; Shukla et al., 2016). In seaweeds, they show resistance to a wide variety of pathogens, with some important bioactive components such as carrageenans, fucans, ulvans, and fucose-containing polymers (or laminarins) (Klarzynski et al., 2003; Sangha et al., 2010). These bioactive components of seaweed work as elicitor molecules and play a role in stimulating defence mechanisms against pathogens (Khan et al., 2009; Sharma et al., 2014; Shukla et al 2016). These elicitors can act as pathogen-associated molecular models (PAMPs) (Sharma et al., 2014). PAMPs bind to host pattern recognition receptors (PRRs), which are transmembrane proteins, and protect plants by inducing the defence mechanisms ISR and SAR through a systemic signal (Eckardt, 2008; Zipfel, 2009). Plants treated with *A. nodosum* extract (ANE) showed a higher defence response than control plants during pathogen infections. ANE bioactive components induce defensive responses against different pathogens (Patier et al., 1995; Sharma et al., 2014). ANE induced defence against *Phytophthora melonis* in cucumber plants (Abkhoo and Sabbagh, 2016). ANE induced the activation of some important disease resistance enzymes such as polyphenol oxidase (PPO), peroxidase (PO), phenylalanine ammonia-lyase (PAL), lipoxygenase and β 1,3-glucanase (Abkhoo and Sabbagh 2016). Panjehkeh and Abkhoo, (2016), determined that ANE can induce resistance (ISR) against *Phytophthora capsici* in tomato. In cucumber, ANE has been shown to reduce the ability of fungal pathogens to cause disease by inducing certain defence genes and enzymes (Jayaraman et al., 2011), JANE induces plant immunity by increasing the concentration of ROS through hydrogen peroxide synthesis (Cook et al., 2018). *A. thaliana* plants treated with 1 g/L ANE showed resistance to the necrotic fungal pathogen *Sclerotinia sclerotiorum* (Subramanian et al., 2011). In carrots, ANE induced defence-related enzymes (Mukherjee and Patel, 2020) and prevented disease development of *A. radicina* and *B. cinerea* (Jayaraj et al., 2008).

In previous studies, the effectiveness of silicon and ANE applications with different mechanisms has been reported. In this study, the application of the two products separately and together positively affected the quality and post-harvest shelf life of the harvested table grapes.

3.4. Berry Detachment Force

Applications to cv. Gök Üzüm increased the berry detachment force of the stem. The lowest stem breaking resistances were determined in the control (2.25±0.12, 1.79±0.03, 1.26±0.14 and 1.12±0.10) at harvest (0^{th} day), 3^{rd}, 7^{th}, and 10^{th} days. The highest values in seaweed application (3.94±0.03) at harvest (0^{th} day), silicon application (2.75±0.24 and 1.86±0.13) on 3^{rd} and 7^{th} days, and Si+ANE application (1.54±0.16) on 10^{th} day determined (Figure 2a). In the applications made to cv. Müşküle, the lowest stem rupture resistance was found in the control (5.03±0.34, 4.15±0.30, 3.08±0.16, 2.92±0.11), while the highest values were found in the Si+ANE application on the 0^{th}, and 7^{th} days (6.82±0.36, 4.63±0.37) and Si+ANE (5.07±0.14, 3.65±0.19) applications on the 3^{rd}, and 10^{th} days. Applications of both cultivars increased stem breaking strength (Figure 2b). As the waiting time increased after harvest, the stem

breaking resistance decreased in all applications and varieties.

3.5. Skin Rupture Force

All applications increased the skin rupture resistance of cv. Gök Üzüm, while the increases were more limited in cv. Müşküle. The lowest skin rupture strength was found in the control (1.72±0.09, 1.38±0.15, 1.35±0.06, 1.16±0.10) in all analyses, and the highest rupture strength was found in the silicone application (2.09±0.18, 2.03±0.11, 1.78±0.18, 1.59±0.10) (Fig. 2c). In cv. Müşküle, the lowest skin rupture resistance was recorded in the control (2.55±0.21, 2.04±0.24, 1.52±0.08) on the 0^{th}, 3^{rd}, and 10^{th} days, and in the silicone application (1.72±0.06) on the 7^{th} day. The highest skin rupture resistance was determined in the 0^{th}, and 3^{rd} days of Si+ANE application (2.79±0.13, 2.27±0.16) and the 7^{th} and 10^{th} days of the seaweed application (2.11±0.06, 1.84±0.11) (Figure 2d). As the waiting time after harvest increased, the skin rupture resistance of the samples taken from all applications decreased.

The berry skin rupture varies according to cultivars. Skin thickness, berry skin rupture and extracted anthocyanin rate have a significant regression (Segade et al., 2008). Previous studies the hardness of fruit at the end of storage period dropped slightly in all applications (Letaief et al 2008) and this decline over time pectin polymers have been reported to be associated with fragmentation (Pretel et al., 2006).

Previous studies have shown that since silicon improves photosynthesis by promoting leaf chlorophyll content, preventing decay and premature aging, promoting growth and development, regulating the absorption of potassium, nitrogen, phosphorus, and micronutrients, effectively preventing cracking, premature defoliation, and other physiological diseases, in leaves and fruits. It has been found to prevent moisture loss, increase fruit firmness, and improve storage and transportation (Jiangyu and Xuelong, 2005).

Optimal pre-harvest and post-harvest management practices ensure the preservation of fruit quality over longer storage periods and increase consumer confidence (Tesfay et al., 2011). Silicon can increase the amount of free phenol. In other words, silicon acts as the main elicitor in increasing free polyphenol levels. Silicon applications in avocado increased fruit quality by increasing free phenol accumulation in mesocarp. Silicon may be an important factor in improving postharvest fruit quality by increasing the free phenols released from the membrane bound form and consequently increasing the antioxidant pool in fruit (Tesfay et al 2011). Fruits treated with silicone lose less weight than control. Silicon probably contributes to retaining moisture in the fruit. Silicone applications reduce fruit respiration and WL by wrapping the fruit stomata with a silicone layer (Hammash and Assi 2007). Reduced activity levels of polyphenol oxidase limit mesocarp blackening of cut avocados (Bower and Dennison 2005). Decreased polyphenol oxidase activity causes leaching of membrane-bound phenols and acts as an antioxidant without oxidant interference to reduce browning. Silicon can reduce oxidant accumulation as it functions to bind cellular oxygen. Silicon is oxidized to form solid silicon dioxide, where a lattice is formed with a silicon atom surrounded by four oxygen atoms (Bekker et al 2007).

Hanumanthaiah et al. (2015), reported that foliar application of silicon at 15-day intervals it effectively improved quality parameters such as (26.67 °Brix), shelf life (6.33 days), skin/fruit ratio (7.44), acidity (0.26%), reducing sugar (19.93%) and non-reducing sugar (2.24). In addition, calcium silicate applications increased chlorophyll a and b and total chlorophyll levels in bananas (Putra et al., 2010).

Costa et al., (2015) identified an increase in diameter of mango trees treated with 1600 kg ha^{-1} of agrosilicone. More et al. (2015), stabilized silicic acid spray improved yield and quality in the early stages of fruit growth (before flowering and 15, 30, 45 and 60 days after flowering) in Alphonso mango (*Mangifera indica* L.). Silicon application improves endogenous levels of indole-3-acetic acid (IAA), gibberellins (GA) and cytokinins (CK) in mango trees, while abscisic acid (ABA), peroxidase (POX), catalase (CAT) and superoxide dismutase (SOD) enzymes levels (Helaly et al., 2017).

Silicon fertilization increased fruit size and quality in apples (Cai and Qian, 1995). Silicone applications can significantly increase the silicon content in apple leaves, fruit skin, flesh, and whole fruit. This situation favorably affects apple fruits, since silicon is the main component of the cell wall, it accumulates in the plant cell wall and root cortical cells, forming a layer of silica with host cells that acts as a barrier against pathogen invasion. Therefore, silicon plays an effective role in improving flesh firmness and pest resistance (Wang et al., 2016). Polygalacturonase (PG) is the predominant factor of cell wall and pectin degradation and is a key enzyme of various fruit softening processes. PG is an enzyme that can break down pectin; however, pectin plays a very important role in maintaining the indoor and outdoor environments and maintaining the firmness of fruit skins. In the apple storage process, a lower PG content better preserves apple quality and produces a longer storage period in apple trees treated with silicon fertilization (100 g/tree). The yield of trees treated with silicon decreased 19.7%. Silicon is the main component that maintains fruit firmness by tightening the cell wall structure. Therefore, a high amount of silicon fertilization can increase the cell wall's ability to defend against malondialdehyde (MDA) and PG activity, and subsequently prolong the storage time of the harvested apple. Gao et al., (2006), studied the effects of silicon applications on apple inner black necrosis (IBN) caused by high manganese levels, simultaneous administration of both silicon and manganese at 400 mg/kg effectively inhibited IBN development in Fuji apples. Su et al., (2011) showed that silicon can significantly reduce the titratable acid content of apple fruits grown in acidic soils, increase the contents of °Brix and vitamin C, but not affect the hardness.

In our study, skin rupture force decreased with the loss of water towards the end of the postharvest period. Due to the drying of pedicels, WL increased, and the skin rupture force decreased. In both varieties, our treatments gave better values than control. It shows that seaweed and silicate fertilization can effectively improve fruit quality and contribute to increasing yield and maintaining quality after harvest.

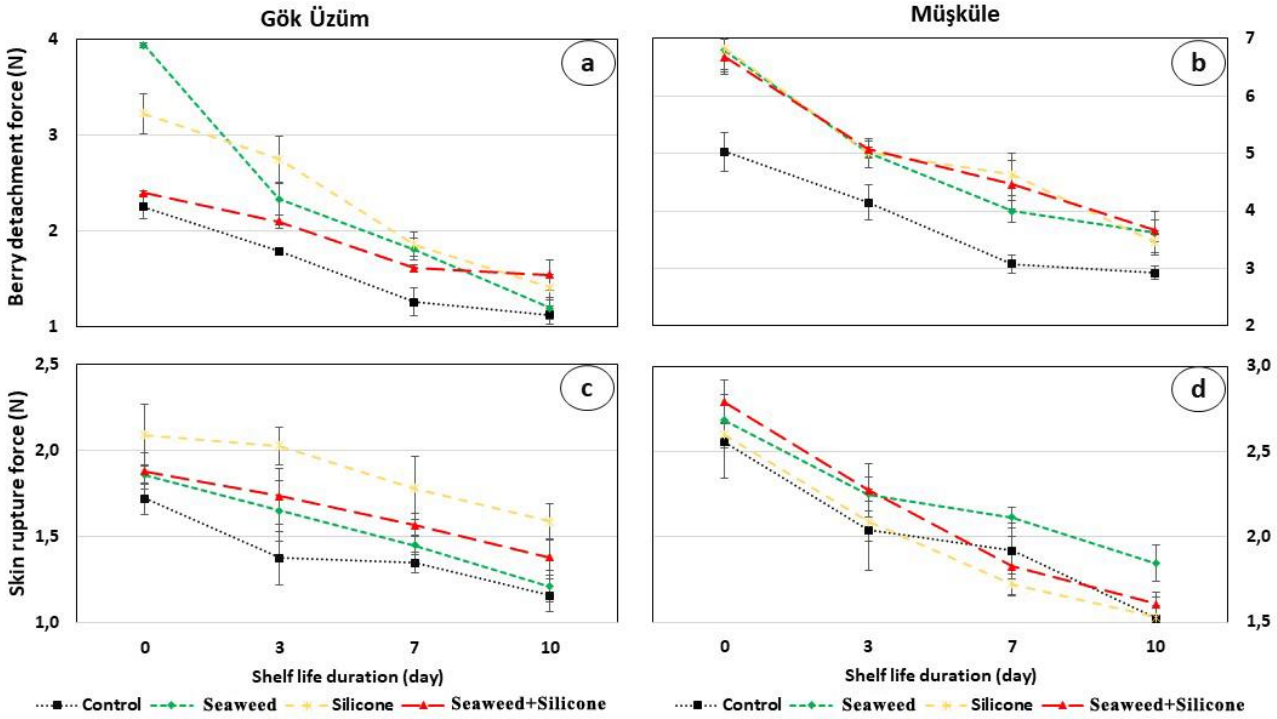


Figure 2
Effects on stem break (a and b) and shell tear resistance (c and d)

3.6. Berry Brightness (*Hue Value, h°)*

The lowest h° values in the applications made to the cv. Gök Üzüm were silicon application on the 0th day (124.39±7.23), the Si+ANE application on the 3rd day (121.87±2.01), and the seaweed application on the 7th and 10th days (110.68±6.31 and 08.66±4.33) was determined. The highest h° values were recorded in the control (133.07±3.29, 131.55±4.52, 120.59±4.88) on days 0th, 3rd, and 7th, and in Si+ANE application (116.68±7.47) on day 10th (Figure 3a). The lowest h° values in the cv. Müşküle were determined in seaweed (114.02±2.58, 105.05±7.57) days 0th, and 7th, control (108.40±0.45) on the 3rd day and silicon application (101.73±7.98) on the 10th day. The highest h° values were recorded in the Si+ANE application in the post-harvest period. In general, our applications slowed down the decrease in the h° value in both varieties, in other words, the dulling of the berry colours.

The quality of grapes largely depends on skin colour. Skin colour often varies depending on anthocyanin content and composition (Carreño et al., 1995). Anthocyanin composition is determined primarily by anthocyanin accumulation due to genetic factors and various agro-ecological factors (diversity, climate, and cultural practices) (Cacho et al., 1992; Pomar et al., 2005; Segade et al., 2008). According to the findings of this study, the brightness value decreased as the post-harvest period was prolonged. As reported in previous studies, towards the end of the post-harvest period, the brightness value gradually decreases and the fruits become opaque (Artés-Hernández et al., 2004; Pretel et al., 2006).

3.7. Maturity Index

Seaweed, silicon, and Si+ANE applications to the cvs. Gök Üzüm and Müşküle increased the MI in the post-harvest period. The lowest MI values were 14.09±0.69, 15.09±1.92, 16.01±0.49 and 16.38±0.78 in the control group, respectively, while the highest values were 17.82±0.99, 20.85±0.67, 20.60±0.62, and 24.56±1.53 in Si+ANE application (Figure 3c). Cv. Müşküle was affected similarly to MI cv. Gök Üzüm. The lowest MI was found to be 16.71±0.89, 17.78±1.05, 19.63±1.63 and 20.16±1.34 in the control group, respectively, while the highest values were 22.49±1.23, 27.73±0.89, 26.42±2.11 and 30.02±1.89 in Si+ANE application according to the analysis dates.

Crisosto and Mitchell (2002) and Sabir A et al. (2006), are also reported at the end of the storage period °Brix and acidity changes depending on the MI values can be seen in the increase. In this study, which was maintained in the form of cluster of 'AK' MI values in applications other than grape varieties have been detected only slight increases during storage period.

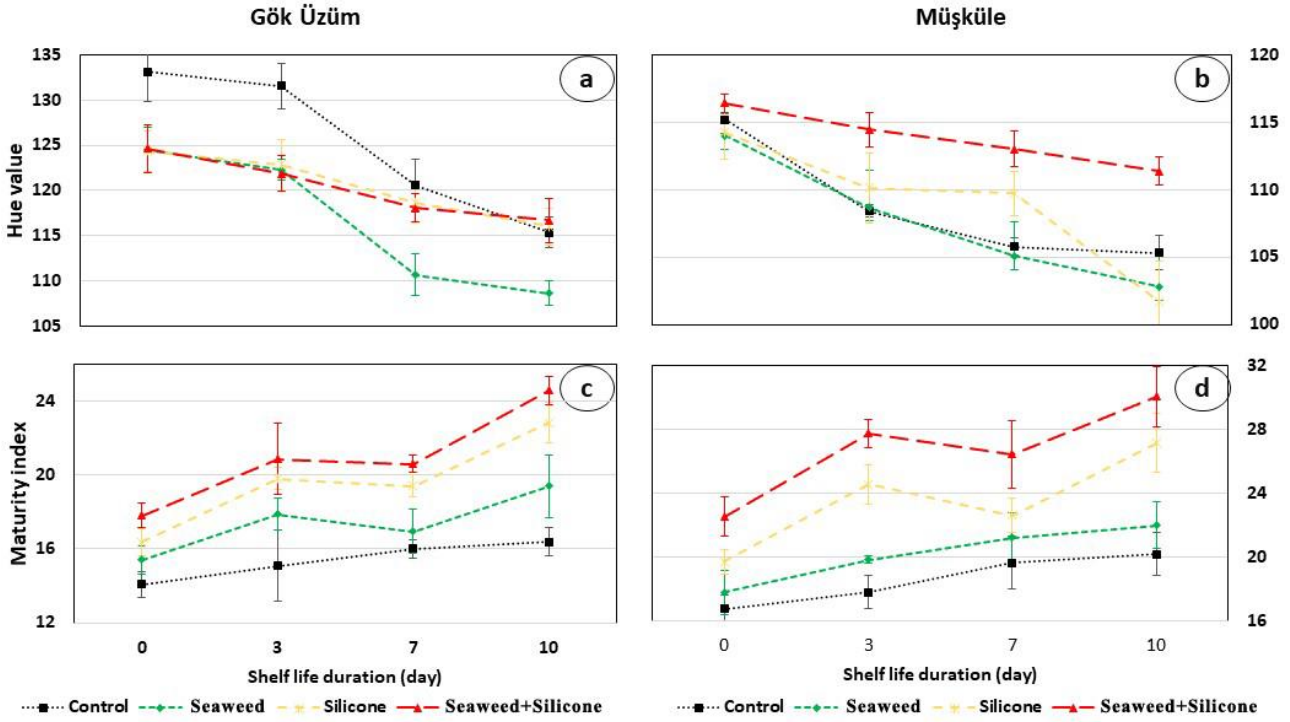


Figure 3
Effects on hue value (a and b) and MI (c and d)

4. Conclusion

Application of Ortho silicon (0.5 mL L^{-1}) and seaweed extract (2.5 mL L^{-1}) to cvs. Müşküle and Gök Üzüm after fruit set 15 days intervals resulted in improvements in the peduncle and in accelerating of ripening. The improvement in the studied properties increased with the co-administration of these two plant activators. In the 4-repeat analyzes carried out during the 10-day storage period at room conditions after harvest, the preservation of quality characteristics such as WL, berry detachment force, skin rupture force, MI, brightness (hue h°) and decay rate were positively affected by the applications. In this process, the combined application of Si+ANE was found to be more effective than the individual applications of these activators. According to the data obtained from this study, improvements in the sustainable preservation of quality can be achieved by applying Ortho Silicon and seaweed extract separately and together after fruit setting and before veraison to preserve the quality of table grape production and postharvest.

5. Acknowledgements

This study was supported by the Scientific Research Projects Coordinatorship of Selçuk University as the thesis project numbered 19201084.

6. References

Abkhoo J, Sabbagh S (2016). Control of Phytophthora melonis damping-off, induction of defense responses, and gene expression of cucumber treated with commercial extract from *Ascophyllum nodosum*. *Journal of Applied Phycology*, 28 (2): 1333-1342.

Adatia M, Besford R (1986). The effects of silicon on cucumber plants grown in recirculating nutrient solution. *Annals of botany*, 58 (3): 343-351.

Anderson PK, Cunningham AA, Patel NG, Morales FJ, Epstein PR, Daszak P (2004). Emerging infectious diseases of plants: pathogen pollution, climate change and agrotechnology drivers. *Trends in ecology & evolution*, 19 (10): 535-544.

Artés-Hernández F, Aguayo E, Artés F (2004). Alternative atmosphere treatments for keeping quality of 'Autumn seedless' table grapes during long-term cold storage. *Postharvest biology and Technology*, 31 (1): 59-67.

Ayliffe MA, Lagudah ES (2004). Molecular genetics of disease resistance in cereals. *Annals of botany*, 94 (6): 765-773.

Battacharyya D, Babgohari M, Rathor P, Prithiviraj B (2015). Seaweed extracts as biostimulants in horticulture. *Scientia horticulturae*, 196: 39-48.

Beckett R, Van Staden J (1989). The effect of seaweed concentrates on the growth and yield of potassium stressed wheat. *Plant and Soil*, 116 (1): 29-36.

Bekker T, Aveling T, Kaiser C, Labuschagne N, Regnier T (2007). Accumulation of total phenolics due to silicon application in roots of avocado trees infected with *Phytophthora cinnamomi*, *Actas VI Congreso Mundial del Aguacate*.

Bektas Y, Eulgem T (2015). Synthetic plant defense elicitors. *Frontiers in Plant Science*, 5: 804.

Blunden G, Gordon S (1986). Betaines and their sulphonio analogues in marine algae. *Progress in phycological research* 4: 39-80.

Bower J, Dennison M (2005). A process to prevent browning of frozen avocado halves and chunks. *South African Avocado Growers' Association Yearbook*, 28: 40-41.

Cacho J, Fernández P, Ferreira V, Castells JE (1992). Evolution of five anthocyanidin-3-glucosides in the skin of the Tempranillo, Moristel, and Garnacha grape varieties and influence of climatological variables. *American journal of enology and viticulture* ,43 (3): 244-248.

Cai DL, Qian FJ (1995). Effect of Si fertilization on yield and quality of apples. J. Reg. Res. Dev., 14: 64–66.

Carreño J, Martínez A, Almela L, Fernández-López J (1995). Proposal of an index for the objective evaluation of the colour of red table grapes. *Food Research International*, 28 (4): 373-377.

Conrath U, Pieterse CM, Mauch-Mani B (2002). Priming in plant–pathogen interactions. *Trends in plant science*, 7 (5): 210-216.

Cook J, Zhang J, Norrie J, Blal B, Cheng Z (2018). Seaweed extract (Stella Maris®) activates innate immune responses in *Arabidopsis thaliana* and protects host against bacterial pathogens. *Marine drugs*, 16 (7): 221.

Costa I, Pereira M, Mizobutsi G, Maia V, Silva J, Oliveira J, Oliveira M, Souza V, Nietsche S, Santos E (2015). Influence of silicon fertilization on 'Palmer'mango tree cultivation. *Acta horticulturae*, 1075: 229-234.

Craigie J (2011). Seaweed extract stimuli in plant science and agriculture. *Journal of Applied Phycology*, 23 (3): 371-393.

Crisosto CH, Mitchell GF (2002). Postharvest handling systems: small fruits, 357-374 pp., Postharvest Technology of Horticultural Crops, Kader, A.A. (Ed.), University of California Agricultural and Natural Resources, Publication 3311, USA.

Dallagnol LJ, Rodrigues FA, DaMatta FM, Mielli MV, Pereira SC (2011). Deficiency in silicon uptake affects cytological, physiological, and biochemical events in the rice–Bipolaris oryzae interaction. *Phytopathology*, 101 (1): 92-104.

Dixon RA (2001). Natural products and plant disease resistance. *Nature*, 411 (6839): 843-847.

Eckardt NA (2008). Chitin signaling in plants: insights into the perception of fungal pathogens and rhizobacterial symbionts, American Society of Plant Biologists.

Epstein E (1999). Silicon. *Annual review of plant biology*, 50 (1): 641-664.

Etesami H, Alikhani HA (2017). Evaluation of gram-positive rhizosphere and endophytic bacteria for biological control of fungal rice (*Oryzia sativa* L.) pathogens. *European Journal of Plant Pathology*, 147 (1): 7-14.

Fauteux F, Rémus-Borel W, Menzies JG, Bélanger RR (2005). Silicon and plant disease resistance against pathogenic fungi. *FEMS Microbiology letters*, 249 (1): 1-6.

Gaffney T, Friedrich L, Vernooij B, Negrotto D, Nye G, Uknes S, Ward E, Kessmann H, Ryals J (1993). Requirement of salicylic acid for the induction of systemic acquired resistance. *Science*, 261 (5122): 754-756.

Gao YM, Xu J, Gao SQ (2006). Effects of silicon application on apple internal bark necrosis induced by high content of manganese. *Plant Nutr. Fertil. Sci.*, 12: 571–577.

Guerriero G, Hausman J-F, Legay S (2016). Silicon and the plant extracellular matrix. *Frontiers in Plant Science* 7: 463.

Guntzer F, Keller C, Meunier J-D (2012). Benefits of plant silicon for crops: a review. *Agronomy for sustainable development*, 32 (1): 201-213.

Gunupuru L, Patel J, Sumarah M, Renaud J, Mantin E, Prithiviraj B (2019). A plant biostimulant made from the marine brown algae *Ascophyllum nodosum* and chitosan reduce Fusarium head blight and mycotoxin contamination in wheat. *PLoS One*, 14 (9): e0220562.

Hammash F, Assi N (2007). influence of pre-storage waxing and wrapping on quality attributes of stored'Shamouti'oranges. *Acta horticulturae*.

Hankins S, Hockey H (1990). The effect of a liquid seaweed extract from *Ascophyllum nodosum* (*Fucales, Phaeophyta*) on the two-spotted red spider mite *Tetranychus urticae*, *Thirteenth International Seaweed Symposium*, pp. 555-559.

Hanumanthaiah M, Kulapatihipparagi R, Renuka D, Kiran Kumar K, Santhosha K (2015). Effect of soil and foliar application of silicon on fruit quality parameters of banana cv. Neypoovan under hill zone. *Plant Archives*, 15 (1): 221-224.

Helaly MN, El-Hoseiny H, El-Sheery NI, Rastogi A, Kalaji HM (2017). Regulation and physiological role of silicon in alleviating drought stress of mango. *Plant physiology and biochemistry*, 118: 31-44.

Hernández Herrera R, Santacruz Ruvalcaba F, Zañudo Hernández J, Hernández Carmona G (2016). Activity of seaweed extracts and polysaccharide-enriched extracts from *Ulva lactuca* and *Padina gymnospora* as growth promoters of tomato and mung bean plants. *Journal of Applied Phycology*, 28 (4): 2549-2560.

Iriti M, Varoni EM (2015). Chitosan-induced antiviral activity and innate immunity in plants. *Environmental Science and Pollution Research*, 22 (4): 2935-2944.

Jayaraj J, Wan A, Rahman M, Punja Z (2008). Seaweed extract reduces foliar fungal diseases on carrot. *Crop Protection*, 27 (10): 1360-1366.

Jayaraman J, Norrie J, Punja ZK (2011). Commercial extract from the brown seaweed *Ascophyllum nodosum* reduces fungal diseases in greenhouse cucumber. *Journal of Applied Phycology*, 23 (3): 353-361.

Jiangyu F, Xuelong M (2005). Progress of silicon improving plant resistance to stress. *Zhongguo Nong xue Tong bao= Chinese Agricultural Science Bulletin*, 21 (11): 304-306.

Khan W, Rayirath UP, Subramanian S, Jithesh MN, Rayorath P, Hodges DM, Critchley AT, Craigie JS, Norrie J, Prithiviraj B (2009). Seaweed extracts as biostimulants

of plant growth and development. *Journal of Plant Growth Regulation*, 28 (4): 386-399.

Kim SG, Kim KW, Park EW, Choi D (2002). Silicon-induced cell wall fortification of rice leaves: a possible cellular mechanism of enhanced host resistance to blast. *Phytopathology*, 92 (10): 1095-1103.

Klarzynski O, Descamps V, Plesse B, Yvin J-C, Kloareg B, Fritig B (2003). Sulfated fucan oligosaccharides elicit defense responses in tobacco and local and systemic resistance against tobacco mosaic virus. *Molecular Plant-Microbe Interactions*, 16 (2): 115-122.

Letaief H, Rolle L, Zeppa G, Gerbi V (2008). Assessment of grape skin hardness by a puncture test. *Journal of the Science of Food and Agriculture*, 88 (9): 1567-1575.

Liang Y, Nikolic M, Bélanger R, Gong H, Song A (2015a). Effect of silicon on crop growth, yield and quality, In: Silicon in Agriculture, Eds: Springer, pp. 209-223.

Liu H, Li JM, Zheng G, Du QG, Pan TH, Chang YB (2014). Effects of silicon on plant growth and Fruit quality of cucumber. *Acta. Agric. Boreali-Occidentalis Sinica*. 23: 117–210.

Lobato A, Coimbra G, Neto M, Costa R, Santos F, Oliveira N, Luz L, Barreto A, Pereira B, Alves G (2009). Protective action of silicon on water relations and photosynthetic pigments in pepper plants induced to water deficit. *Research Journal of Biological Sciences*, 4 (5): 617-623.

Lux A, Luxová M, Hattori T, Inanaga S, Sugimoto Y (2002). Silicification in sorghum (*Sorghum bicolor*) cultivars with different drought tolerance. *Physiologia plantarum*, 115 (1): 87-92.

Ma JF, Mitani N, Nagao S, Konishi S, Tamai K, Iwashita T, Yano M (2004). Characterization of the silicon uptake system and molecular mapping of the silicon transporter gene in rice. *Plant physiology*, 136 (2): 3284-3289.

Mancuso S, Briand X, Mugnai S, Azzarello E (2006). Marine bioactive substances (IPA Extract) improve foliar ion uptake and water stress tolerance in potted" *Vitis vinifera*" plants. *Marine Bioactive Substances (IPA Extract) Improve Foliar Ion Uptake and Water Stress Tolerance in Potted" Vitis vinifera" Plants*: 1000-1006.

Marschner H (1988). Mechanisms of manganese acquisition by roots from soils, In: Manganese in soils and plants, Eds: Springer, pp. 191-204.

Marschner H (1995). Mineral Nutrition of Higher Plants Second Edition Academic Press Edition London.

Marschner P (2012). Marschner's mineral nutrition of higher plants 3rd edition, Elsevier Publishers, Oxford, United Kingdom.

Matichenkov V (1990). Amorphous oxide of silicon in soddy podzolic soil and its influence on plants. *Autoref. Diss. Cand., Moscow State University, Moscow*.

Matichenkov V, Bocharnikova E, Calvert D (2001). Response of citrus to silicon soil amendments, *Proc. Fla. State Hort. Soc*, pp. 94-97.

Mercier L, Lafitte C, Borderies G, Briand X, Esquerré-Tugayé MT, Fournier J (2001). The algal polysaccharide carrageenans can act as an elicitor of plant defence. *New phytologist*, 149 (1): 43-51.

Moller M, Smith M (1998). The significance of the mineral component of seaweed suspensions on lettuce (*Lactuca sativa* L.) seedling growth. *Journal of Plant Physiology*, 153 (5-6): 658-663.

More S, Gokhale N, Shinde S, Korake G (2015). Effect of different sources of silica on nutrient content of leaves and fruit at different stages of Alphonso mango (*Mangifera indica* L.) in lateritic soil. *Journal of Progressive Agriculture*, 6 (2): 84-88.

Mukherjee A, Patel J (2020). Seaweed extract: biostimulator of plant defense and plant productivity. *International Journal of Environmental Science and Technology*, 17 (1): 553-558.

Norrie J, Keathley J (2005). Benefits Of Ascophyllum Nodosum Marine-Plant Extract Applications To Â´ Thompson Seedlessâ´ Grape Production, *X International Symposium on Plant Bioregulators in Fruit Production, 727*, pp. 243-248.

Palou L, Serrano M, Martínez-Romero D, Valero D (2010). New approaches for postharvest quality retention of table grapes. *Fresh Produce*, 4 (1): 103-110.

Panjehkeh N, Abkhoo J (2016). Influence of marine brown alga extract (Dalgin) on damping-off tolerance of tomato. *J. Mater. Environ. Sci.*, 7: 2369-2374.

Patier P, Potin P, Rochas C, Kloareg B, Yvin J-C, Liénart Y (1995). Free or silica-bound oligokappa-carrageenans elicit laminarinase activity in Rubus cells and protoplasts. *Plant Science*, 110 (1): 27-35.

Pomar F, Novo M, Masa A (2005). Varietal differences among the anthocyanin profiles of 50 red table grape cultivars studied by high performance liquid chromatography. *Journal of Chromatographya*, 1094 (1-2): 34-41.

Potin P, Bouarab K, Küpper F, Kloareg B (1999). Oligosaccharide recognition signals and defence reactions in marine plant-microbe interactions. *Current opinion in microbiology*, 2 (3): 276-283.

Pozza EA, Pozza AAA, Botelho DMdS (2015). Silicon in plant disease control. *Revista Ceres*, 62: 323-331.

Pretel M, Martinez-Madrid M, Martinez J, Carreno J, Romojaro F (2006). Prolonged storage of 'Aledo'table grapes in a slightly CO_2 enriched atmosphere in combination with generators of SO2. *LWT-Food Science and Technology*, 39 (10): 1109-1116.

Putra E, Zakaria W, Abdullah N, Saleh G (2010). Weak neck of Musa sp. cv. Rastali: A review on it's genetic, crop nutrition and post harvest. *J. Agron*, 9: 45-51.

Romero I, Vazquez-Hernandez M, Maestro-Gaitan I, Escribano MI, Merodio C, Sanchez-Ballesta MT (2020). Table Grapes during Postharvest Storage: A Review of the Mechanisms Implicated in the Beneficial Effects of Treatments Applied for Quality Retention. *International journal of molecular sciences*, 21 (23): 9320.

Saa S, Rio O, Castro S, Brown P (2015). Foliar application of microbial and plant based biostimulants increases growth and potassium uptake in almond (*Prunus dulcis* [Mill.] DA Webb). *Frontiers in Plant Science*, 6: 87.

Sabir A, Sabir FK, Tangolar S, Agar IT (2006). Farklı konsantrasyonlarda etanol uygulamasına sahip SO 2 jeneratörlerinin üzüm cv'nin soğuk muhafazası üzerindeki etkilerinin karşılaştırılması. Alphonse Lavallée. *J Çukurova Üni Tarım Fak.*, 21: 45–50.

Sangha JS, Ravichandran S, Prithiviraj K, Critchley AT, Prithiviraj B (2010). Sulfated macroalgal polysaccharides λ-carrageenan and ι-carrageenan differentially alter *Arabidopsis thaliana* resistance to Sclerotinia sclerotiorum. *Physiological and molecular plant pathology*, 75 (1-2): 38-45.

Segade SR, Rolle L, Gerbi V, Orriols I (2008). Phenolic ripeness assessment of grape skin by texture analysis. *Journal of Food Composition and Analysis*, 21 (8): 644-649.

Serrano M, Martinez-Romero D, Castillo S, Guillén F, Valero D (2005). The use of natural antifungal compounds improves the beneficial effect of MAP in sweet cherry storage. *Innovative food science & emerging technologies*, 6 (1): 115-123.

Sharma H, Fleming C, Selby C, Rao J, Martin T (2014). Plant biostimulants: a review on the processing of macroalgae and use of extracts for crop management to reduce abiotic and biotic stresses. *Journal of Applied Phycology*, 26 (1): 465-490.

Shukla PS, Borza T, Critchley AT, Prithiviraj B (2016). Carrageenans from red sea weeds as promoters of growth and elicitors of defense response in plants. *Frontiers in Marine Science*, 3: 81.

Singh KP, Jahagirdar S, Sarma BK (2021). Emerging Trends in Plant Pathology, Springer, pp.

Snyder GH, Matichenkov VV, Datnoff LE (2016). Silicon, In: Handbook of plant nutrition, Eds: CRC Press, pp. 567-584.

Spann TM, Little HA (2011). Applications of a commercial extract of the brown seaweed *Ascophyllum nodosum* increases drought tolerance in container-grown 'Hamlin'sweet orange nursery trees. *HortScience*, 46 (4): 577-582.

Su XW, Wei SC, Jiang YM, Huang Y (2011). Effects of silicon on quality of apple fruit and Mn content in plants on acid soils. *Shandong Agric. Sci.*, 6.

Subramanian S, Sangha JS, Gray BA, Singh RP, Hiltz D, Critchley AT, Prithiviraj B (2011). Extracts of the marine brown macroalga, *Ascophyllum nodosum*, induce jasmonic acid dependent systemic resistance in *Arabidopsis thaliana* against *Pseudomonas syringae* pv. tomato DC3000 and *Sclerotinia sclerotiorum*. *European Journal of Plant Pathology*, 131 (2): 237-248.

Taranovskaia V (1939). The silicication of subtropic greenhouse and plantations. *Soviet subtropics* 7: 32-37.

Tesfay S, Bertling I, Bower J (2011). Effects of postharvest potassium silicate application on phenolics and other antioxidant systems aligned to avocado fruit quality. *Postharvest biology and Technology*, 60 (2): 92-99.

Tripathi DK, Singh VP, Prasad SM, Chauhan DK, Dubey NK (2015). Silicon nanoparticles (SiNp) alleviate chromium (VI) phytotoxicity in *Pisum sativum* (L.) seedlings. *Plant physiology and biochemistry*, 96: 189-198.

Tripathi DK, Singh S, Singh VP, Prasad SM, Dubey NK, Chauhan DK (2017). Silicon nanoparticles more effectively alleviated UV-B stress than silicon in wheat (*Triticum aestivum*) seedlings. *Plant physiology and biochemistry* 110: 70-81.

TÜİK (2022). https://biruni.tuik.gov.tr/medas/?kn=92&locale=tr. [20.04.2022].

Uwakiem MK (2015). Effect of spraying silicon, selenium, and humic acid on fruiting of Early Sweet grapevines, *The 2 nd Inter. Conf. on Hort. Crops. 15-18 March., Egypt. J. Hort*, pp. 333-343.

Valero D, Valverde J, Martínez-Romero D, Guillén F, Castillo S, Serrano M (2006). The combination of modified atmosphere packaging with eugenol or thymol to maintain quality, safety and functional properties of table grapes. *Postharvest biology and Technology*, 41 (3): 317-327.

Van Loon L, Bakker P, Pieterse C (1998). Systemic resistance induced by rhizosphere bacteria. *Annual review of phytopathology*, 36 (1): 453-483.

Wang M, Wang X, Wang JJ (2016). Effect of silicon application on silicon contents in "Fuji" Apple in *Loess Plateau*. *Communications in soil science and plant analysis*, 47 (20): 2325-2333.

Wiesel L, Newton AC, Elliott I, Booty D, Gilroy EM, Birch PR, Hein I (2014). Molecular effects of resistance elicitors from biological origin and their potential for crop protection. *Frontiers in Plant Science*, 5: 655.

Zipfel C (2009). Early molecular events in PAMP-triggered immunity. *Current opinion in plant biology*, 12 (4): 414-420.

Selcuk Journal of Agriculture and Food Sciences SJAFS

http://sjafs.selcuk.edu.tr/sjafs/index

(2022) 36 (3), 493-500

e-ISSN: 2458-8377

DOI:10.15316/SJAFS.2022.064

Research Article

Determination of Densities and Frequencies of Problematic Weed Species in Onion Planting Areas of Ankara and Çorum Provinces

Nilgün ARIKAN[1], İzzet KADIOĞLU[2]

[1]General Directorate of Agricultural Research and Policies, Çankaya, Ankara, Türkiye
[2] Retired Faculty Member Anamur, Mersin, Türkiye

ARTICLE INFO

Article history:
Received date: 13.10.2022
Accepted date: 08.12.2022

Keywords:
Onion
Weed
Survey
Density
Frequency of Occurrence

ABSTRACT

This study was conducted in 2019 in order to determine the weed species, their densities and frequency of occurrences in the onion production areas of Ankara and Çorum Provinces. The surveys were carried out in 78 fields, 55 in Ankara and 23 in Çorum. As a result of the surveys, 75 weed species, two of which are parasite, belonging to 28 families and 64 genera, were determined in the onion fields of Ankara Province. The families with the most species were listed as Asteraceae (11), Poaceae (9), Fabaceae (6), Chenopodiaceae (5) and Apiaceae (5). Other families were determined between 1-3 species. Of these weeds, *Convolvulus arvensis* L. (1.2 plants/m^2), *Xantium strumarium* L. (0.50 plants/m^2), *Amaranthus retroflexus* L. (0.40 plants/m^2), *Cuscuta* sp. (0.37 parazited onion/m^2), *A. blitoides* S. Watson (0.36 plants/m^2) were the 5 most intense species. According to the frequency of occurrence, the first five species are determined as; *C. arvensis* L. (90.90%), *X. strumarium* L. (65.45%), *A. retroflexus* L. (54.54), *Chenopodium album* L. (52.72%), *A. albus* L. (47.27%). In the onion cultivation areas of Çorum Province, 61 weed species one of which is parasite, belonging to 24 families and 51 genera were determined. The families with the most species were listed as Asteraceae (9), Poaceae,(8) Apiacea (5), Polygonaceae (4), Fabaceae (4); as for other families, between 1-3 species were determined. Considering the densities per m^2 of these weeds, the 5 most common weed species are determined as *C. arvensis* L. (0.80 plants/m^2), *Anethum graveolen* L. (0.56 plants/m^2), *A. retroflexus* L. (0.41 plants/m^2), *Chrozophora tinctoria* (L.) Raf. (0.32 plants/m^2), *X. spinosum* L. (0.31 plants/m^2), the first five species in terms of frequency of incidence are; *C. arvensis* L. (86.95), *A. refroflexus* L. (56.52%), *X. strumarium* L. (56.52%), *Cirsium arvense* (L.) Scop. (52.17%), *C. album* L. (52.17%).

1. Introduction

Onion is a vegetable that can be grown in different parts of the world and consumed in many different ways, and it has been farmed for over 4000 years. The homeland of the onion starts from the Mediterranean basin and extends to Iran and Afghanistan. The most widely known and farmed type of onion, which belongs to the Alliaceae family, is *Allium cepa* L. (Albayrak and Elmacı, 2017).

Although onion is not a type used directly in cooking, it ranks third in the world vegetable production after legumes and tomatoes (Bayram, 2021). Onion, which has an important place in human nutrition, is consumed in two ways as dry and fresh. In addition to containing many important vitamins and minerals, it is a medicinal plant known to be used since the beginning of human history (Yılmaz et al., 2006).

Temperature and day length are two important factors in onion cultivation. Although onion is a heat tolerant vegetable, it is more productive in cooler climates. During this period, the average temperature demand is 12-13 ^{0}C. The onion needs higher temperature after it starts to tie the head. The optimum temperature demanded by the onion, which is 18-20 ^{0}C during this period, rises to 23-27 ^{0}C during the ripening phase of the heads (Anonymous, 2022a). Onions are grouped into short day, medium day and long days depending on day length requirements. During the head formation stage, short-day varieties require 8-10 hours, medium-day varieties require 10-12 hours, and long-day varieties require 13-15 hours of day length (Beşirli et al.2021).

* Corresponding author email: *nilgun_arikan@yahoo.com*

While India, China and Nigeria are the countries with the largest onion cultivation areas in the world, China, India and the USA are the first three ones in production (Bayram, 2021). In 2020, 105 million tons of onion production was realized in approximately 5.5 million hectares of land in 138 countries in the world. According to the Food and Agriculture Organization (FAO) 2020 data, Turkey ranks 12th in the world onion cultivation area and 5th in production. While the world onion yield is 1.908 kg/da, Turkey's yield is 3.244 kg/da, which is higher than the world average. It has been reported that the amount of dry onion in world foreign trade was 16.6 million tons in 2020, with a monetary value of $ 7.1 billion, and the average ton price in 2020 was $233. World onion exports amounted to 8.5 million tons, with a value of 3.6 billion dollars in 2020, and imports amounted to 8.1 million tons and 3.5 billion dollars. Considering the amount of production, Türkiye is a self-sufficient and exporting country in dry onions, and ranks 10th with an export of 221 thousand tons (Anonymous, 2022b). In our country, dry onion proficiency level was determined as 114.2% and per capita consumption was determined as 21.4 kg. Although onion cultivation is carried out in every region of our country with different climatic conditions, it is seen that the production is concentrated in the Central Northern part of Central Anatolia, the Central Black Sea and the Mediterranean Region for early types (Anonymous, 2022c). On the basis of provinces; It is produced intensively in Ankara, Amasya, Hatay, Çorum, Tokat, Adana, Eskişehir, Bursa, Konya, Balıkesir, Tekirdağ, Karaman, Aksaray, Gaziantep, Antalya, Yozgat, Afyon, Kahramanmaraş provinces. The province with the largest onion cultivation area is Ankara with 165 thousand decares. Ankara is followed by Çorum with 105 thousand decares, Amasya with 70 thousand decares, and Hatay, Tokat, Eskişehir and Adana, respectively.

Ankara is in the first place with 835 thousand tons of dry onion production, as it is in the cultivation area. Ankara is followed by Çorum with a production of 295 thousand tons, Amasya with a production of 286 thousand tons, and Hatay, Eskişehir and Adana, respectively (TÜİK, 2022).

Turkey had an onion planting area of 722.319 decares in 2012, and 1.735.857 tons of onions were produced. In the years two thousand and twenty-one, onion production areas in Turkey decreased by approximately 3.9% compared to 2012 and were realized on a total area of 698.972 decares. While the cultivation areas decreased, the amount of product obtained increased by 44% and reached 2.5 million tons of dry onion production (TUIK, 2022).

In the provinces of Ankara and Çorum, where the study was conducted, the cultivation area was 88.507 and 46.463 decares, respectively, in 2012; production amount was stated as 268.224 and 123.886 tons. In the years two thousand and twenty-one, the total planting and production amounts are 165.767 decares and 835.269 tons for Ankara; for Çorum, it was determined as 105.739 decares and 295.503 tons.

In the provinces of Ankara and Çorum, where the study was conducted, the increase in planting and product amounts from 2012 to 2021 was 87% and 211% for Ankara, respectively; for Çorum, it was 127% and 138%. The reason for this is thought to have successful results in the selection of the right variety in production, the use of healthy seeds, the improvement of cultivation techniques, the intensive use of agricultural technology, and the methods for combating harmful organisms that cause problems for onions.

Commercial onion production is made in three ways; 1. Production by direct seed sowing, 2. Shallot production, 3. Seedling production, (Beşirli et al., 2021). Many factors affect the yield in onion production areas. Of these factors; disease agents (fungal, bacterial, viral), harmful insects and weeds take the lead. These harmful organisms not only reduce the yield, but also reduce the market value of the product. As it is known, weeds are compete with cultivated plants in terms of growth factors that are water, nutrients, light and place, live as semi-or full parasites in cultivated plants, negatively inhibit the development of cultivated plants by showing allelopathic effects, harm human and animal health with some toxic chemicals they contain, cause significant yield and quality losses. This causes more cost, especially in the early period (Özer et al., 1997; Işık et al., 2015). In addition to causing direct damage to cultivated plants, weeds cause many disease factors and host or intermediately host pests, which are problematic in cultivated plants, causing them to survive in the environment and to pass on to cultivated plants (Özer et al., 1997; Kitiş, 2011).

Onion is a plant that has little competition with weeds because it is a slow-growing, short, shallow rooted plant with a weak canopy. Additionally, it has been reported that the cylindrical shaped, upright growing leaves cannot suppress weed growth because they cannot shade in the soil (Ghosheh, 2004).

In the studies conducted abroad, it has been revealed that the onion is affected by weeds during the whole development period depending on the weed species and density, but the most competition is in the first 1.5-2 months when annual weeds are dominant (Anonymous, 2008; Güncan and Karaca, 2018). It is stated that especially in onion production from seeds, product losses due to competition are very high, and the highest competition is at the beginning of germination. It is estimated that losses from weeds are much higher than losses from pests and diseases (Tripathy et al., 2013). Additionally, a 15% weed density in the first 6 weeks reduces the yield by 86%, and a 50% weed density reduces the yield by 91% (Klingman and Ashton 1982; Torun, 2017), In another study, it was reported that weed competition reduced the average onion yield by 62% compared to the weed-free control (Qasem, 2006), and season-long crop-weed competition reduced the onion yield by 81.2% compared to the weed-free condition (Prakash et al., 2000). In another study conducted in Pakistan, it was stated that weed competition caused a 71% and 76%

decrease in onion yield in the first and second years, respectively (Khokhar et al., 2006).

In the studies conducted in our country, it has been revealed that the product losses in the fields where weed control is not done or done at late stages are between 20-100%, and it also affects the quality negatively creating small-headed onions (Özer et al., 1997; Anonymous, 2008).

In another study, it was determined that the competition between onion and weed started with the emergence of the onion, and there was a 55% decrease in yield and a 32% decrease in onion diameter with the prolongation of the competition between onions and weeds. It was determined that weeds, which were determined to cause great decreases in onion yield and quality, should be controlled, especially in the first 4-5 weeks (Kızılkaya et al., 2001).

For this reason, in order to benefit more from the existing agricultural areas, as the world population is increasing rapidly, it is necessary to minimize the weed problems in the onion production areas and to increase the amount and quality of the product taken from the unit area.

For this, besides breeding methods, it becomes necessary to fight against weeds; chemical, mechanical and cultural methods are applied in the control. However, hoeing and manual plucking is not economical in places where worker wages are high (Özer et al., 1997). For an effective and correct control method, it is of great importance to know the weed species, their density and biology, which are the problem.

This study was carried out in order to determine the weed species, densities and incidence frequencies in the onion cultivation areas of Ankara and Çorum provinces, where both the cultivation area and production are high in our country.

2. Materials and Methods

The main material of the study consists of weeds in Ankara and Çorum provinces and districts where onions are produced. The survey studies required to determine the weed species that are problematic in terms of frequency and density in onion planting areas were carried out in a total of 78 fields, 55 for Ankara and 23 for Çorum, in areas where onion production is intense in May-August 2019 (Tablo 1).

Survey studies have been carried out in at least 1% of the onion planting areas. The distribution of onion production areas in Ankara and Çorum provinces is unequal and concentrated in a certain regions (Figure 1).

Figure 1
Sampling areas

Table 1
Onion planting areas (da) in the provinces where the survey was conducted and the number of sampling fields

Names of the provinces	Onion production areas (da)	Number of fields to be sampled
Çorum	41.054	23
Ankara	97.085	55
Total	138.139	78

(TUİK, 2019)

Care was taken to ensure that the fields to be surveyed were at least 3 km away from each other. Additionally, attention was paid to the fact that it was in different directions. The counts to be made in the field in the cultivation areas to be sampled were started from within 10 m inside in the direction of the diagonals, avoiding the field edge effect. 1 m^2 frame is used at each sampling point, 4 for 1-5 decares, 6 for 5-10 decares, 8 for 10-20 decares, 12 for 20-60 decares, and 16 points for larger fields are taken with a 1 m^2 frame. Plants were counted according to species, and the number of weeds detected in the counts were recorded together with the estimated cultivation areas (Kadıoğlu et al., 1993). Each of the narrow-leaved weeds of the wheatgrass group was counted as a sister plant. As for the parasitic plant *Cuscuta* sp., the number of parasitized onions per m^2 was taken into account. The herbariums of the species that could not be identified under field conditions were tried to be diagnosed under laboratory conditions, and the diagnoses were performed by utilising Davis PH (1965-1985), Uluğ et al. (1993), Özer et al. (1999), Özgür (2013).

The formulas given below were used to determine the weed species, their densities (number/m2) and the frequency of occurrence (%) (Odum, 1971).

$Y=b/m$

Y: densities (number of plant /m^2)

m: number of surveys

b: Number of plants in total m^2 in surveys made at the counting point

Incidence Frequency: It is the value that shows the % of a weed species encountered in the surveyed regions.

Incidence Frequency (%) = (n/m) x 100

n = Total number of fields with a species (units)

m = Total number of fields observed (units)

3. Results and Discussion

As a result of the surveys carried out at 55 sampling points in 2019 in order to determine the weed types, densities and frequency of occurrence in the onion cultivation areas of Ankara Province, 75 weed species, two of which are parasites (*Cuscuta* sp. and *Orobanche* sp.), belonging to 28 families and 64 genera, were determined. Follow-up studies are continuing to determine whether onion or weed is the host plant of *Orobanche* sp. or not (Figure 2). As a result of the study, when it is determined that it is the host of the onion, it will be the first record in our country. When the determined weed species are considered on the basis of family, Asteraceae

(11), Poaceae (9), Fabaceae (6), Chenopodiaceae (5) and Apiaceae (5) families are in the first place. Other families were found between 1-3 species. Considering the densities per m^2 of these weeds, the 5 most common weed species are *C. arvensis* L. (1.2 plants/m^2), *X. strumarium* L. (0.50 plants/m^2), *A. retroflexus* L. (0.40 plants/m^2). *Cuscuta* sp. (0.37 parasitic onions/m^2), *A. blitoides* S.Watson (0.36 plants/m^2). When evaluated according to the frequency of occurrence, the first five species are; *C. arvensis* L. (90.90%), *X. strumarium* L. (65.45%), *A. retroflexus* L.(54.54%), *C. album* L. (52.72%) and *A. albus* L. (47.27%) determined (Table 2).

In the surveys carried out at 23 sampling points in the onion cultivation areas of Çorum, 61 weed species belonging to 24 families and 51 genera were determined. The families with the most species were listed as Asteraceae (9), Poaceae (8) Apiaceae (5), Polygonaceae (4), Fabaceae (4), while other families were determined between 1-3 species. Considering the densities per m2 of these weeds, the 5 most common weed species are *C. arvensis* L. (0.80 plants/m^2), *A. graveolen* L. (0.56 plants/m^2), *A. retroflexus* L. (0.41 plants/m^2), *C. tinctoria* (L.) Raf. (0.32 plants/m^2), *X. spinosum* L. (0.31 plants/m^2), the first five species according to the frequency of occurrence; *C. arvensis* L. (86.95%), *A. refroflexus* L. (56.52%), *X. strumarium* L. (56.52%), *C. arvense* (L.) Scop (52.17%), *C. album* L. (52.17%) (Table 2).

Figure 2
Orobanche sp.

Table 2
Weed Species, Densities and Incidence Frequency in Onion Production Areas in Ankara and Çorum Province

		Ankara		Çorum	
Family	Weeds	Density (plant/m^2)	Frequency of incidence (%)	Density (plant/m^2)	Frequency of incidence (%)
Amaranthaceae	*Amaranthus albus* L.	0.17	47.27	0.16	34.78
	Amaranthus blitoides S.Watson	0.36	43.63	0.05	8.69
	Amaranthus retroflexus L.	0.40	54.54	0.41	56.52
Apiaceae	*Anethum graveolens* L.	0.11	20.00	0.56	52.00
	Bifora radians Bieb.	0.005	1.81	0.02	4.34
	Daucus carota L.	0.004	3.63	0.01	4.34
	Echinophora tenuifolia L.	0.07	18.18	0.10	17.39
	Turgenia latifolia L.Hoffm.	0.008	5.45	0.01	4.34
Aristolochiaceae	*Aristolochia maurorum* L.	0.03	7.27	0.05	13.04
Asclepiadaceae	*Cynanchum acutum* L.	0.01	7.27	0	0
Asteraceae	*Acroptilon repens* (L.) DC	0.10	23.63	0.22	17.00
	Centaure solstitialis L	0	0	0.01	4.34
	Chondrilla juncea L.	0.002	1.81	0	0
	Cichorium intybus L.	0.005	1.81	0	0
	Cirsium arvense (L.) Scop	0.11	21.81	0.24	52.17
	Helianthus annus L.	0.002	1.81	0.01	4.34
	Lactuca serriola L.	0.002	1.81	0	0
	Matricaria chamomilla L.	0	0	0.02	8.69
	Senecio vulgaris L.	0.01	3.63	0.005	4.34
	Silybum marianum L. Gaertn.	0.002	3.63	0	0
	Sonchus oleraceus L.	0.01	10.90	0.02	8.69
	Xanthium spinosum L.	0.27	52.72	0.31	47.82
	Xanthium strumarium L.	0.50	65.45	0.30	56.52
Boraginaceae	*Echium vulgare* L.	0.001	1.81	0.01	8.69
	Heliotropium europaeum L.	0.12	21.81	0.20	21.73
Brassicaceae	*Boreava orientalis* Jaub and Spach	0	0	0.05	13.04
	Sinapis arvensis L.	0.09	34.54	0.17	47.82
	Siymsbrium officinale (L.) Scop.	0.01	12.72	0.05	17.39
Caryophyllaceae	*Agrostemma githago* L.	0.002	1.81	0	0
Chenopodiaceae	*Chenopodium album* L.	0.30	52.72	0.18	52.17
	Chenopodium opulifolium Schrad.	0.06	14.54	0.10	17.39
	Chenopodium urbicum L.	0.02	7.27	0	0
	Chenopodium vulvaria L.	0.14	30.90	0.24	43.47
	Salsola kali L.	0.16	40.00	0	0

Table 2 (Continuation)
Weed Species, Densities and Incidence Frequency in Onion Production Areas in Ankara and Çorum Province

Convolvulaceae	*Convolvulus arvensis* L.	1.20	90.90	0.80	86.95
	Convolvulus galacticus Roston. ex Choisy.	0.01	1.81	0.13	13.04
Cuscutaceae	*Cuscuta* sp.	0.37 parasited onion nummer/m^2	23.63	0.09 parasited onion number/m^2	4.16
Euphorbiaceae	*Chrozophora tinctoria (*L.) Rafin	0.08	29.09	0.32	30.43
	Euphorbia prostrata Aiton	0.17	43.63	0.28	47.82
	Euphorbia sp.	0.008	7.2	0.08	43.47
Equi*setaceae*	*Equisetum arvense L.*	0.007	5.45	0.06	13.04
Fabaceae	*Alhagi pseudalhagi (Bieb)* Resv	0.20	27.27	0.05	8.69
	Cicer arietinum L.	0.001	1.81	0	0
	Glycyrrhiza sp.	0.001	1.81	0	0
	Medicago sativa L.	0.05	7.27	0.01	4.34
	Melilotus officinalis (L.) Desr.	0.01	9.09	0.08	30.43
	Vicia sativa L.	0.002	1.81	0.005	4.34
Juglandaceae	*Juglans regia*	0.001	1.81	0	0
Lamiaceae	*Molucella leavis* L.	0.03	1.81	0	0
Malvaceae	*Abutilon theophrasti* Medik.	0.005	3.63	0	0
	Hibiscus trionum L.	0.25	12.72	0.14	34.78
	Malva neglecta Wallr.	0.03	16.36	0.04	13.04
Orobanchaceae	*Orobanche* sp.	0.002	3.63	0	0
*P*lantaginaceae	*Plantago majör* L.	0.01	5.45	0.02	8.69
Poaceae	*Avena fatua* L.	0.14	25.45	0.29	34.78
	Avena sterilis L.	0	0	0.08	13.04
	Cynodon dactylon (L.) Pers.	0.33	29.09	0.28	30.43
	Echinochloa crus-galli (L.) P. B	0.14	21.81	0.15	17.39
	Hordeum vulgare L.	0.004	1.81	0	0
	Lolium perenne L.	0.02	3.63	0.03	4.34
	Phragmites australis (Cav) Trin. ex. Steudel	0.07	12.72	0.13	17.39
	Setaria verticillata (L.)P.B.	0.15	14.54	0.12	13.04
	Sorghum halepense (L.) Pers	0.10	18.18	0.22	21.73
	Triticum aestivum L.	0.005	1.81	0	0
Polygonaceae	*Polygonum aviculare* L.	0.10	29.09	0.21	30.43
	Polygonum convolvulus L.	0.004	3.63	0.02	8.69
	Polygonum cognatum Meissn.	0.045	1.81	0.02	8.69
	Rumex crispus L.	0.007	1.81	0	0
	Rumex sp.	0	0	0.01	13.04
Portulacaceae	*Portulaca oleracea* L.	0.12	27.27	0.05	21.73
Primulaceae	*Anagallis arvensis* L.	0.002	1.81	0.03	8.69
Ranunculaceae	*Consolida orientalis* (Gay)Schröd.	0.005	3.63	0.005	4.34
Resedaceae	*Reseda lutea* L.	0.03	14.5	0.12	8.69
Rubiaceae	*Galium aparine* L.	0.04	3.63	0.02	4.34
	Galium tricornutum Dandy.	0.005	1.81	0	0
	Rubia tinctorum L.	0.01	1.81	0	0
Scrophulariaceae	*Kickxia spuria* L. Dumort	0	0	0.03	13.04
Solanaceae	*Datura stramonium* L.	0.16	34.54	0.28	21.73
	Solanum nigrum L.	0.12	30.90	0.11	39.13
Zygophyllaceae	*Peganum harmala* L.	0.005	1.81	0	0
	Tribulus terrestris L.	0.29	36.36	0.10	13.04

For Ankara and Çorum provinces, the number of species with a frequency of over 10% and above was determined as 36 and 39 species, respectively. The number of species with a density of more than 0.10 plant/m^2 was determined as 25 for Ankara and 26 for Çorum.

Centaure solstitialis L, *Matricaria chamomilla* L., *Avena sterilis* L., *Boreava orientalis* Jaub and Spach, *Kickxia spuria* L. Dumor. weed species were not found in the survey areas carried out in the onion fields in Ankara, while *Cynanchum acutum L., Chondrilla juncea* L., *Cichorium intybus* L, *Lactuca serriola* L., *Silybum marianum* L. Gaertn., *Agrostemma githago* L., *Chenopodium urbicum* L., *Salsola kali* L., *Cicer arietinum* L., *Glycyrrhiza* sp, *Juglans regia, Molucella leavis* L., *Abutilon theophrasti* Medik., *Orobanche* sp., *Hordeum vulgare* L., *Triticum aestivum* L., *Galium tricornutum* Dandy., *Rubia tinctorum* L. *Peganum harmala* L. species were not found in onion fields of Çorum provivce.

Studies on the determination of weeds in onion cultivation areas in our country are at a limited level, and it has been observed that there are no studies on weeds in the onion cultivation areas of Ankara and Çorum Provinces in the literature research. In this context, when the study is evaluated, it has the feature of being the first.

In this context, when the study is evaluated, the weed species encountered in the mentioned provinces have been determined, and the frequency and densities of these species have been determined for the first time throughout this study.

When we look at similar studies carried out on weeds in onion production, survey studies were carried out by

Alsan (1986) to determine the weeds in the onion fields of the Eastern Anatolia Region. The surveys were carried out in 14 fields, in onion fields in the provinces of Tunceli (Mazigirt and Pertek districts), Erzincan (Central district) and Sivas (Suşehri district). In the results of working; It was determined that weed species found in onion fields belong to 26 genera in Tunceli, 22 genera in Erzincan and 17 genera in Sivas. In Tunceli Province, *C. arvensis* L. *M. officinalis* Lam. Thuill, *Chenopodium* spp., especially *C. album* L., *P. aviculare* L. *Sorghum halepense* (L.) Pers., *Setaria viridis* (L.) P. Beauv., *Cynodon dactylon* (L.) Pers., *Echinochloa crus-galli* (L.) P. Beauv.), *Cuscuta* sp.; in Erzincan *A. retroflexus* L, *C. albüm* L., *C. arvensis* L, *C. arvense* (L.) Scop., *Solanum nigrum* L., *Hibiscus trionum* L., *Sinapis arvensis* L., *Setaria viridis* (L.) P. Beauv., *E. crus-galli* (L.) P. Beauv., *Cuscuta* sp.; in Sivas *A. retroflexus* L. *C. albüm* L., *C. arvense* (L.) Scop. *C. arvensis* L., *P. convolvulus* L, *Euphorbia glyptosperma* Engilm., *Rapistrum rugosum* (L.) Al., *S. viridis* (L.) P. Beauv., *Cuscuta* sp. were determined to be species found in onion fields.

In a study conducted by Zengin (1997), he determined the weeds, their densities, prevalence rates and community formation status in onion fields in Erzurum region. It was found that 41 weed species belonging to 18 families in the research regions have an average density of 95.87 units/m^2. *A. retroflexus, S. viridis L., C. album L.* and *C. arvensis* are very dense in onion cultivation areas, respectively, *C. album* L., *C. arvensis* L., *A. retroflexus* L. and *S. viridis* (L) were determined as the most common species.

In the survey studies conducted in Kazova and Kelkit valleys of Tokat province, 73 weed species belonging to 27 families in Kazova and a total of 83 weed species belonging to 31 families in Kelkit Valley were determined. The species with the highest density in Kazova was determined as *C. arvensis* L. with 13.81%, and *A. retroflexus* L. with 17.48% in Kelkit Valley (Kızılkaya, 2003)

Mennan and Işık (2003) investigated the change in weed flora of onion production areas by comparing the results of the survey conducted in Amasya Province in 1976 and the years 1999-2000. While 23 weed species were recorded in the first survey, it was reported that 87 weed species were detected in the second survey. According to the density, the most common species in the first survey were *C. album, A. retroflexus, C. arvensis, Heliotropium europaeum* and *S. nigrum*; In the study conducted between 1999-2000, the first 5 species with the highest incidence are respectively; *C. arvensis* L., *X. strumarium* L., *A. retroflexus* L., *Galium aparine, S. arvensis*. In the second survey, it was determined that X. *strumarium, C. arvense, S. arvensis, G. aparine* and *Bifora radians* gained more importance in the 25-year period.

In the study conducted by Gürbüz (2007), 105 weed species belonging to 30 plant families were determined. According to the number of weed species, the largest 3 families were reported to be Asteraceae (17), Poaceae (14) and Fabaceae (9). While the incidence of 57 of the weed species determined in the studies was over 10%, *Medicago polymorpha* L., *C. arvensis* L., *Avena sterilis* L., *C. album* L. and *S. arvensis* L. were found to be in the first 5 places.

In a study carried out in the onion cultivation areas of Tekirdağ province, 39 weed species belonging to 21 families were determined. *Convolvulus* spp., *S. arvensis* L., *Avena spp*., *C. album* L., *Euphorbia* spp., *Adonis flammea* Jacq., *S. nigrum* L., *Cirsium* sp., *X. strumarium* L., *P. aviculare* L. species reported to be the most dense species on the basis of (Yaşar, 2012).

In a study conducted in Hatay, 82 genera and 93 weed species belonging to 29 families, 2 of which are monocotyledonous, 26 of which are dicotyledonous and 1 of which are parasitic, were determined. *A. sterilis* L., *S. arvensis* L., *C. arvensis* L were found to be the most common weed species, *A. sterilis* L., *S. arvensis* L. and *A. retroflexus* L. were determined to be the most intense weeds. (Kaya and Üremiş, 2019).

When the previous survey studies in onion fields were compared with the study we conducted in Ankara, it was found that it was similar to the weed species detected in the survey studies conducted in Amasya by Mennan and Işık (2003).

When compared on the basis of families, in the study conducted by Gürbüz (2007) in the onion production areas covering Adana, Hatay and Mersin provinces, it was seen that the largest family according to the number of weed species overlapped as Asteraceae, Poaceae and Fabaceae. When the survey studies carried out in onion cultivation areas grown both in summer and winter are examined, it is seen that *C. arvensis* L, and *A. retroflexus* are common weed species.

The differences in the weed types and densities detected in the survey results are thought to vary depending on the different climate and soil characteristics of the regions, production technique, topographic factors, height, crop pattern, weed control methods and time, and cultivation techniques.

As a result, weeds, which are one of the most important plant protection problems in onions, as in other crop plants, compete with the onion in terms of nutrients, water, light and place, affect its development, cause a large amount of yield and quality losses.by hosting diseases and pests.

For this reason, the determination of weed species, density and incidence is important in order to be able to fight economically, effectively and correctly in onions that have weak competition.

In the interviews with the producers encountered in the field during the survey, it was stated that they mainly used chemical control and then hoeing at least once in weed control, and it was reported that this increased production costs. It is accepted that the competitive power of *C. arvensis* L, *A retroflexus* L. and *X. strumarium* L. weed species that are common in surveys in Ankara and Çorum are high. Since weed seeds are carried to long distances by wind, birds, animals, irrigation water, tools,

equipment or people, and spread over large areas thanks to their seed forming abilities and high adaptability, the producers are provided with the Integrated Chemical and Mechanical Control, which includes cultural measures to prevent contamination, especially by rotation. Training activities should be increased on the need to implement Integrated Weed Management (IWM)

Finally, it is thought that this study will contribure to ensure healthy, adequate and balanced nutrition of the increasing population in our country and in the world, increasing the yield and quality in onion production, ensuring the necessary agricultural food production, and to the researches to be carried out within the framework of "Integrated Weed Management" against these weeds detected in order to increase the useful life of existing herbicides.

4. References

Alsan C (1986). Survey studies on weeds in onion (*Allium cepa* L.) fields in eastern anatolian region, *Plant Protection Bulletin*, 26 (1), p.1-11.

Albayrak B, Elmacı ÖL (2017). The determınatıon of nıtrogen and sulphur applıcatıons on yıeld and some qualıty crıterıa of onıon (*Allium cepa* L.). *Bahçe* 46 (1): 21–30

Anonymous (2008). Zirai Mücadele Teknik Talimatları, Soğan Tarlalarında Yabancı Otlar. T.C. Gıda Tarım ve Hayvancılık Bakanlığı, Tarımsal Araştırmalar ve Politikalar Genel Müdürlüğü, Bitki Sağlığı Araştırmaları Daire Başkanlığı, Cilt 6, p: 286, Ankara.

Anonymous (2022a). Soğan Yetiştiriciliği. https://istanbul.tarimorman.gov.tr/Belgeler/Kutu-Menu/Brosurler/Sebzecilik/sogan.pdf, (Date of Access 8.10.2022)

Anonymous (2022b). Ürün Masaları Soğan Bülteni, Ocak 2022. T.C. Tarım ve Orman Bakanlığı, Bitkisel Üretim ve Genel Müdürlüğü, Tarla ve Bahçe Bitkileri Daire Başkanlığı, Ankara Sayı:18

https://www.tarimorman.gov.tr/BUGEM/Belgeler/B%C3%BClteniler/OCAK%202022/Kuru%20So%C4%9Fan%20Ocak%20B%C3%BClteni.pdf (Date of Access 21.05.2022)

Anonymous (2022c). Ürün Masaları Soğan Bülteni. Mayıs 2022. T. C. Tarım ve Orman Bakanlığı, Bitkisel Üretim Genel Müdürlüğü, Tarla ve Bahçe Bitkileri Daire Başkanlığı, Ankara, Sayı:19.

https://www.istib.org.tr/resim/siteici/files/118_Kuru%20So%C4%9Fan%20B%C3%BCltени%20-%20%C3%9Cr%C3%BCn%20Masalar%C4%B1%20(May%C4%B1s%202022).pdf, (Date of Access 15.09.2022)

Bayram U (2021). Ürün Raporu Kuru Soğan, 2021. T.C. Tarım ve Orman Bakanlığı, Tarımsal Ekonomi ve Politika Geliştirme Enstitüsü, Yayın no:336, Ankara.

Beşirli G, Sönmez İ, Albayrak B, Polat Z (2021). Organik Soğan Yetiştiriciliği, T.C. Tarım ve Orman Bakanlığı, Tarımsal Araştırmalar ve Politikalar Genel Müdürlüğü, Atatürk Bahçe Kültürleri Merkez Araştırma Enstitüsü Müdürlüğü No: 10: s 53, Yalova. https://arastirma.tarimorman.gov.tr/yalovabahce/Belgeler/Yayinlar/110-organik.sogan.pdf (Date of Access 18.09.2022)

Davis PH (1965-1985). Flora of Turkey and the East Eagean Islands, vol. 1- 9, Edinburgh University Press, Edinburgh.

FAO (2020). Food and Agriculture Organization of the United Nations https://www.fao.org/faostat/en/#data/QCL

Ghosheh HZ (2004). Single herbicide treatments for control of broadleaved weeds in onion (*Allium cepa*), *Crop Protection* 23 No:6 p.539–542.

Güncan A, Karaca M (2018). Yabancı Ot Mücadelesi. Güncellenmiş ve İlaveli Dördüncü Baskı. Selçuk Üniversitesi Basımevi. s:334. ISBN: 975 448 178 4.

Gürbüz R (2007). Çukurova soğan üretim alanlarında görülen yabancı otların öneminin ve bazı herbisitlerin yabancı otlanma ile soğan verimine olan etkilerinin araştırılması, Yüksek Lisans Tezi, Çukurova Üniversitesi Fen Bilimleri Enstitüsü, Adana.

Işık D, Dok M, Altop Kaya E, Mennan H (2015). Applicability of early tillage and glyphosate together with pre and post emergence weed control in corn production, *Journal of Agricultural Sciences*. 21 (2015): p.596-605.

Kadıoğlu İ, Uluğ E, Üremiş İ (1993). Akdeniz bölgesi pamuk ekim alanlarında görülen yabancı otlar üzerinde araştırmalar. Türkiye I. Herboloji Kongresi, 3-5 Şubat, Adana, s.151-156.

3-5 Şubat, Adana, s. 151-156.

Kaya H, Üremiş İ (2019). Determination of weed species, their frequencies and densities in onion fields in Hatay province. *Mustafa Kemal University Journal of Agricultural Sciences* 24 (1):21-30

Khokhar KM, Mahmood T, Shakeel M, Chaudhry MF (2006). Evaluation of integrated weed management practices for onion in Pakistan. *Crop Protection* 25 (2006): 968–972

Kitiş YE (2011). Yabancı ot mücadelesinde malç ve solarizasyon uygulamaları GAP VI. Tarım Kongresi, 09–12 Mayıs, Şanlıurfa, p.463-468.

Kızılkaya A, Önen H, Özer Z (2001). Researches on the effects of weed competition on onion yield. *Turkish Journal of Weed Science*, 4(2): p.58-65.

Kızılkaya A (2003). Tokat ili (Kazova ve Kelkit Vadisi)'nde baş soğan (*Allium Cepa* L.) yetiştirilen alanlarda sorun olan yabancı otlar ve farklı dönemlerde yabancı otların yok edilmesi ile ekonomik eşiğin belirlenmesi üzerinde araştırmalar. Yüksek Lisans Tezi, Gaziosmanpaşa Üniversitesi Fen Bilimleri Enstitüsü, Tokat

Klingman GC, Ashton FM (1982). Weed science, principles and practices (second edition). Wiley Interscience Pub., New York, USA.

Mennan H, Işık D (2003). Invasıve weed specıes ın onıon productıon systems durıng the last 25 years ın amasya, Turkey. *Pakistan Journal of Botany* 35(2): p:155-160.

Qasem JR (2006). Chemical weed control in seedbed sown onion (*Allium cepa* L.). *Crop Protect* 25 (6): p.618– 622.

Odum EP (1971). Fundamentals of ecology. W. B. Saunders Company, Philadeiphia, London, Toronto, 574 p.

Özer Z, Kadıoğlu İ, Önen H, Tursun N (1997). Herboloji (Yabancı Ot Bilimi), Gaziosmanpaşa Üniversitesi, Ziraat Fakültesi Yayınları, No:20, Kitaplar serisi No:10, p.388. Tokat

Özer Z, Önen H, Tursun N, Uygur FN (1999). Türkiye'nin bazı önemli yabancı otları (tanımları ve kimyasal savaşımları), Gaziosmanpaşa Üniversitesi, Ziraat Fakültesi Yayınları, No:38, Kitap Seri No:16, p. 438. Tokat.

Özgür OE (2013). Şeker Pancarı Tarla Çiçekleri, I ve II. (Weeds of Sugar Beet), Ankara, s.410. ISBN 9786056099847(1.c), ISBN 9786056099854(2.c)

Prakash V, Pandey AK, Singh RD, Mani VP (2000). Integrated weed management in winter onion (*Allium cepa* L.) under mid-hills conditions of North-Western Himalayas. *Indian Journal of Agronomy* 45 (4): p.816-821.

Torun H (2017). Determination of weed species, frequencies and densities in leaf edible vegetable minor crops in Eastern Mediterranean Region, *Plant Protection Bulletin* 57(3): 279 – 291.

Tripathy P, Sahoo BB, Patel D, Dash DK (2013). Weed management studies in onion (*Allium cepa* L.), *Journal of Crop and Weed* 9(2):210-212.

Tüik (2019). http://www.tuik.gov.tr/PreTablo.do?alt_id=1001, (Date of Access 19.02.2019)

Tüik (2022). https://biruni.tuik.gov.tr/medas/?kn=104&locale=tr, (Date of Access 21.05.2022)

Uluğ E, Kadıoğlu İ, Üremiş İ (1993). Türkiye'nin yabancı otları ve bazı özellikleri, Türkiye Cumhuriyeti Tarım ve Köy İşleri Bakanlığı, Zirai Mücadele Araştırma Enstitüsü Müdürlüğü, Yayın No:78, Adana, 513.

Yaşar S (2012). Tekirdağ ili soğan ekim alanlarında görülen önemli yabancı ot türleri, yoğunlukları ve rastlanma sıklıklarının belirlenmesi. Yüksek Lisans Tezi, Namık Kemal Üniversitesi, Fen Bilimleri Enstitüsü, Tekirdağ.

Yılmaz H, Demircan V, Erel G (2006). Profitability ındicators and production costs of dry onions in mainly producing provinces. *Çukurova University Faculty Journal of Agriculture* 21(2), p.39-48.

Zengin H (1997). Erzurum yöresi soğan tarlalarındaki yabancı otlar, yoğunlukları, yaygınlıkları ve topluluk oluşturma durumları üzerinde araştırmalar, *Atatürk Üniversitesi Ziraaat Fakültesi Dergisi* 28 (3), 433-440.

Selcuk Journal of Agriculture and Food Sciences SJAFS

http://sjafs.selcuk.edu.tr/sjafs/index

(2022) 36 (3), 501-506

e-ISSN: 2458-8377

DOI:10.15316/SJAFS.2022.065

Research Article

Determination of Chemical Fertilizer and Various Organic Fertilizers on Some Agricultural Characteristics of Green Bean (*Phaseolus vulgaris* L.)

Muhammet KÜÇÜK, Ercan CEYHAN

[1] Selçuk University, The Graduate School of Natural and Applied Science, Department of Field Crops, Konya, Türkiye
[2] Selçuk University, Faculty of Agriculture, Department of Field Crops, Konya, Türkiye

ARTICLE INFO

Article history:
Received date: 09.08.2022
Accepted date: 12.12.2022

Keywords:
Chemical Fertilizer
Fresh Bean
Organic Fertilizers
Seed Yield

ABSTRACT

This research was conducted to determine the effects of chemical fertilizers, some organic fertilizers, and control applications on seed yield and some agricultural characteristics of fresh beans in 2021. The trial was carried out in the Faculty of Agriculture of the University of Selçuk more precisely in the research and application station of Prof. Dr. Abdülkadir AKÇİN. In the study, pure bean lines PV2001, Riberia, PV2002, and PV2003 developed by hybridization were used as materials in the experiment. Then the seed yield, some agricultural characteristics, and the protein rate were determined. In this study, it was determined that the effects of chemical fertilizers and some organic fertilizer applications on all other traits examined were found to be statistically significant. As a result of the research, it was determined that the height of the bean plant ranged between 39.81 cm (control) and 51.19 cm (cow manure), the number of pods per plant changed between 14.36 (control) and 27.92 per plant (chemical fertilizer), the number of seeds per plant was 53.76 (control) and 127.52 (chemical fertilizer), the seed yield changed between 116.81 kg da^{-1} (control) and 239.48 kg da^{-1} (fertilizer chemical), the weight of 100 g of the seeds ranged between 36.632 g (control) and 40.91 g (sheep manure) and the protein content varies between 26.48 % (cow manure) and 27.49 % (control). The reactions of the bean lines used in the study to chemical fertilizers and some organic fertilizer applications showed differences. As a result, when the results obtained in this one-year study were reviewed, it was revealed that cow and sheep manure could be used in the organic cultivation of beans.

1. Introduction

Green bean is a vegetable that has an important place in human nutrition and health and is consumed fondly as fresh, dry, and canned. It is generally grown easily in every region of our country and is known to have an important place in covering the needs of the people for fresh vegetables. Bean, which is a type of vegetable that originates from Central America, has a very wide distribution area in the world. Its production in 2020 was 23.276.716 tons in the world. Asian and European countries have a significant share in this production. The production of China, which is the largest producer of green beans in the world, was 17.964.222 tons in 2020. Turkey, on the other hand, ranks 4th with a production of 547.349 tons (FAO, 2022).

Bean plant has important roles in human nutrition as well as in sustainable agriculture and animal nutrition. With the *Rhizobium phaseoli* bacteria in the nodules in its roots, it fixes the free nitrogen in the air, increases the nitrogen amount of the soil, and leaves a nitrogen-rich soil for the plants to be planted after it (Şehirali, 1998).

Throughout history, humanity has done many studies to increase productivity in this respect but has ignored the damage to natural resources and the environment. The desired yield increase was achieved with pesticides and fertilizers used in large quantities, but the natural balance began to deteriorate. Also, there have been consequences over time such as loss of quality in the product, deterioration of the soil structure, end of microorganism activities, decrease in the amount of organic matter in the soil, an increase of diseases and harmful factors, and soil erosion, which affect plant production negatively (Akgün, 2018).

Fertilizers support the plant at the stage when the macro and microelements it takes from the soil are insufficient. The right dosage and form of fertilizers contribute to plant growth and increase the amount of product in a unit area. Fertilizers can be applied in chemical and organic forms (Kaya and Erdönmez, 2020).

* Corresponding author email: *eceyhan@selcuk.edu.tr*

Today, it is a matter that must be taken as a basis to make the most use of it without damaging the air, water, and soil. Producers become conscious of organic agriculture, good agricultural practices, and sustainable agriculture, which is one of the new agricultural practices. In this way, the purpose is to restore the natural balance, develop environmentally friendly production systems, and increase production by providing organic and green fertilization instead of synthetic chemical drugs and fertilizers (Başdemir et al., 2020).

In line with the purpose and target of the present study, the possibilities of reducing the dependence on chemical fertilizers and re-gain of the organic materials that are gradually depleted with intense agriculture were investigated. The differences between traditional fertilization and different organic fertilizer sources in terms of their effects on yield and some yield elements in beans were also uncovered in this study.

2. Materials and Methods

The Riberia genotype and three fresh bean pure lines, which are the candidates for registration, of Dr. Ercan CEYHAN (PV2001, PV2002, and PV2003 pure lines), were used as the study material in the study.

Some characteristics of sheep manure used as the study material were; moisture 10.35%, organic matter 74.1%, pH 7.53, nitrogen 1.63%, water-soluble P_2O_5 0.17%, water-soluble K_2O 3.26%, water-soluble Zn 1.60 ppm, and water-soluble Fe 4.80 ppm. Some characteristics of the cow manure were; moisture 2.96%, organic matter 76.67%, pH 7.38, nitrogen 1.55%, water-soluble P_2O_5 0.16%, water-soluble K_2O 3.00, water-soluble Zn 2.40 ppm, and water-soluble Fe 29.60 ppm.

According to the 17-year meteorological observations made in the city of Konya during the vegetation period in the year, the experiments were established, the 17-year average temperature, total precipitation, and average relative humidity were 22.5ºC, 80.1 mm, and 40.8%, respectively. In 2021, when the experiment was established, it was 21.8ºC, 105.2 mm, and 42.2%.

The previous plant was wheat in the study area. The soil was plowed deeply in October after the wheat plant was harvested, allowing it to spend the winter in this way. In March, the trial area was first plowed and parceled out, and then organic fertilizers (cow and sheep manures) were applied to the main plots. Control (without manure) was randomly placed on the main plots with 5 kg/da of nitrogen as pure substance from chemical fertilizer (urea), 3000 kg/da of bovine (cow) animal manure, and 2000 kg/da of ovine (sheep) animal manure. According to the trial design, control (without manure), 0.72 kg/da of urea manure, 195 kg of cow manure, and 130 kg of sheep manure were applied to the main plots. Organic fertilizers and chemical fertilizers, which were weighed for each main parcel, were distributed evenly to the parcels with the help of a shovel and rake and mixed with the soil with a hand tiller rotavator.

The trial was performed on the annealed soil with 3 replications according to the "Divided Plots Trial in Random Blocks" pattern on May 2, 2021, in Selçuk University, Faculty of Agriculture, Prof. Dr. Abdülkadir AKÇİN Research and Application Station. In the study, fertilizers were placed on the main plots and the varieties were placed randomly on the sub-plots, which were 5.0 m x 2.5 m = 12.5 m^2. The seeds were sown manually at a depth of 4 cm in rows opened with a marker, with 50 cm between rows and 8 cm between rows. Five rows were planted in each plot.

To clean the trial plots from weeds, to break the cream layer formed after irrigation, and to ensure the deterioration of capillarity, 2 hoeings and 5 times irrigation were applied depending on the water needs of the bean plant depending on the climatic conditions during the plant development period. The first irrigation was carried out during the period when the plants were 10-15 cm after emergence because of the lack of precipitation, and the second irrigation was carried out just before the pod planting, at the time of flowering, and the other irrigations were performed during the pod binding period according to the climatic conditions. Harvesting was carried out by hand between 20 and 31 August 2021 when 90% of the plants were ripe in each genotype.

Each agricultural characteristic examined in the trial was subjected to statistical analysis separately, and the differences in the averages calculated for each treatment were grouped according to the "LSD Test" as 1% in some and 5% in others (Yurtsever, 1984). Statistical analyzes were made using the JUMP package program in the study.

3. Results and Discussion

3.1. Plant Height

According to the analysis of variance results, it was found that the effects of the fertilizer types on plant height were statistically significant at the rate of 5% in four different bean genotypes that received organic and chemical fertilizers (Table 1). According to Table 2, it was determined that the fertilizer application with the highest average plant height was cow manure at 51.19 cm, and the control application with a minimum of 39.81 cm. According to the LSD test, cow manure, sheep manure, and chemical manure were in the first group (group a), and the control group was in the second group (group b) (Table 2). When other study results were examined, it was found that there were similar results. Aldemir (2019) reported that in his study that was conducted on the chickpea plant, he obtained the longest plant height of 43.18 cm from the plots on which barnyard manure was applied, and the shortest plant height from the control plot of 40.11 cm. When Göksu (2012) examined the two-year results of the study on pea plants, he reported that organic and chemical fertilizers increased plant height significantly when compared to control and microbial fertilizers. Bulut (2013), on the other hand, reported that he obtained the highest plant height from the

plots where chicken manure was used and the lowest from the control plots in his study that was conducted on bean plants.

Table 1
Variance analysis of the plant height of the bean genotypes that received organic and chemical fertilizers

Variance Sources	DF	Plant Height	Number of Pod per Plant	Number of Seed per Plant
Replication	2	25,860	0,378	4,286
Fertilizer (F)	3	318,256*	407,003**	11447,100**
Error 1	6	40,436	2,114	93,434
Genotype (G)	3	52,320	3,663	40,395
F X G İntrac.	9	40,956	2,061	102,820
Error 2	24	27,739	3,647	130,050
Variance Sources	DF	Seed Yield	Weight of Hundred Seed	Protein Ratio
Replication	2	212,979	0,052	1,082
Fertilizer (F)	3	30940,100**	49,750**	2,157
Error 1	6	153,094	1,271	1,327
Genotype (G)	3	1434,810**	61,457**	4,714**
F X G İntrac.	9	212,083	5,107*	2,105
Error 2	24	152,360	1,288	0,927

* : $p < 0.05$

Table 2
Variance analysis of the plant height and pod per plant of the bean genotypes that received organic and chemical fertilizers

Genotype	Chemical and Organic Fertilizers								Mean
	Control		Chemical Fertilizer		Cow Manure		Sheep Manure		
Plant Height (cm)									
PV2001	34,89		50,00		50,55		48,44		45,97
Riberia	39,56		51,11		47,78		43,89		45,58
PV2002	40,67		51,78		53,22		43,33		47,25
PV2003	44,11		47,56		53,22		55,89		50,19
Mean	39,81	b	50,11	a	51,19	a	47,89	a	47,25
Pod per Plant (number)									
PV2001	14,89		26,89		19,22		17,63		19,66
Riberia	15,67		28,11		18,92		17,81		20,13
PV2002	13,56		27,79		17,89		16,44		18,92
PV2003	13,33		28,89		20,27		17,70		20,05
Mean	14,36	c	27,92	a	19,08	b	17,40	b	19,69

*: The differences between the means denoted by the same letter were not statistically significant.
Plant height $LSD_{Fertilizer}$: 6.35; Pod per plant: $LSD_{Fertilizer}$: 2.22

The variation of the seed yields of the genotypes used in the trial according to different fertilizer applications was statistically insignificant (Table 1). As an average of the fertilizer applications, the highest plant height was measured at 50.19 cm from the PV2003 genotype. This was followed by plant heights of PV2002 (47.25 cm) and PV2001 (45.97) genotypes, in decreasing order. The lowest plant height was measured at 45.58 cm from the Riberia genotype (Table 2). Other researchers working on this subject obtained similar results (Ceyhan, 2004; Ülker and Ceyhan, 2008a; Varankaya and Ceyhan, 2012; Kepildek and Ceyhan, 2021).

3.2. Number of Pod per Plant

According to the analysis of variance results, it was determined that the effects of the organic and chemical fertilizers applied to four different bean genotypes on the number of pods were statistically significant at the 1% level (Table 1). According to Table 3, it was determined that the average number of pods was a maximum of 27.92 number per plant of chemical fertilizers, and the minimum average of 14.36 number per plant was the control application. According to the LSD test, it was determined that chemical fertilizer was in the first group (group a), cow and sheep manure was in the second group (group b), and control application was in the third group (group c) (Table 3). Erdönmez (2020) found the effects of different doses of fertilizers applied to soybean on the number of pods to be significant and reported that the highest number of pods was obtained in the HG2 fertilizer application at 172.80 number per plant, and the lowest in SG3 fertilizer application at 132.03 80 number per plant. Bulut (2013) reported that organic fertilizer applications in bean - Bacteria and no-Bacteria conditions affected the number of pods per plant, and the mean number of pods was found to be between 6.13 and 8.23 80 number of pods per plant, and the highest was obtained from chicken manure application.

Table 3
Variance analysis of the seed per plant and seed yield of the bean genotypes that received organic and chemical fertilizers

Genotype	Chemical and Organic Fertilizers								Mean	
	Control		Chemical Fertilizers		Cow Manure		Sheep Manure			
Seed per Plant (number)										
PV2001	53,78		130,29		91,23		65,00		85,07	
Riberia	57,93		127,00		83,42		80,43		87,20	
PV2002	44,93		126,62		90,20		77,09		84,71	
PV2003	58,40		126,19		87,26		82,55		88,60	
Mean	53,76	c	127,52	a	88,03	b	76,27	b	86,39	
Seed Yield (kg da^{-1})										
PV2001	122,35		240,79		173,13		157,86		173,53	a
Riberia	96,38		225,03		153,52		146,64		155,39	b
PV2002	119,56		246,86		170,96		187,16		181,14	a
PV2003	128,93		245,23		156,65		163,04		173,46	a
Mean	116,81	c	239,48	a	163,57	b	163,68	b	170,88	

*: The differences between the means denoted by the same letter were not statistically significant.
Seed per Plant: $LSD_{Fertilizer}$: 14.63; Seed yield: $LSD_{Fertilizer}$: 18.73, $LSD_{Genotype}$: 14.09

The variation of the pods per plant of the genotypes used in the trial according to different fertilizer applications was statistically insignificant (Table 1). As an average of the fertilizer applications, the highest the pods per plant was obtained at 20.13 number from the Riberia genotype. This was followed by seeds per plant of PV2003 (20.05 number) and PV2001 (19.66 number) genotypes, in decreasing order. The lowest pods per plant was obtained at 18.92 number from the PV2002 genotype (Table 2). Other researchers working on this subject obtained similar results (Ceyhan, 2004; Ülker and Ceyhan, 2008a; Varankaya and Ceyhan, 2012; Kepildek and Ceyhan, 2021).

3.3. Number of Seeds per Plant

According to the analysis of variance, the effects of organic and chemical fertilizers applied to bean genotypes on the number of seed per plant were found to be statistically significant at the level of 1% level (Table 1). According to Table 3, the maximum number of seeds per plant was obtained from chemical fertilizer with 127.52 number, and at least 53.76 number from control application. According to the results of the LSD test, chemical fertilizer was found to be in the first group (group a), cow and sheep manure in the second group (group b), and control application in the third group (group c) (Table 3). Bulut (2013) reported that the effects of grafting and fertilizer applications on the number of seeds per plant were important, and the mean value of the fertilizers varied between 25.08 - 34.76 mumber per plant. Aldemir (2019), on the other hand, reported that he obtained the least seed yield from the control plot, as in our study.

According to the results of the analysis of variance, the effects of four different bean varieties that received organic and chemical fertilizers on the number of seed per plant were statistically insignificant at the 1% level (Table 3). According to Table 3, it was determined that the highest number of seeds per plant was in the PV2003 genotype with 88.60, and the least average number of seeds was in the PV2002 variety with 84.71 (Table 3). Aldemir (2019) reported that the fertilizers that received different chickpea cultivars had a significant effect on the number of seed per plant and that 44.83 in the Gökçe variety, 39.24 in the Azkan variety, and 31.83 in Aydın 92 variety. According to the results of the present study, this result was different.

3.4. Seeed Yield

According to the results of the analysis of variance, the difference between the seed yield of organic and chemical fertilizers that were applied to four different bean genotypes was found to be statistically significant at the 1% level (Table 1). According to Table 3, the maximum seed yield was in chemical fertilizer with 239.48 kg da^{-1}, and the control application with the least 116.81 kg da^{-1}. According to the results of the LSD test, it was determined that chemical fertilizer was in the first group (group a), sheep and cow manure in the second group (group b), and control application was in the third group (group c) (Table 3). Gül (2018) reported that they obtained the highest seed yield in soybean plants with chicken manure and Bulut (2013) reported that they obtained the highest seed yield in bean plants with chicken manure with 141.33 kg da^{-1}.

According to the analysis of variance because of the study, it was found that the difference in seed yield between four different bean varieties, which received organic and chemical fertilizers, was significant at the level of 1% level (Table 1). In the study, it was found that the maximum seed yield was 181.14 kg da^{-1} in the PV2002 genotype, and the least seed yield was 155.39 kg da^{-1} in the Riberia genotype (Table 3). According to the LSD test results, PV2001, PV2002, and PV2003 genotypes were in the first group (group a), and the Riberia genotype was in the second group (group b) (Table 3). Aldemir (2019) reported that different fertilizer applications in the chickpea plant affected the seed yield in three different chickpea cultivars significantly and obtained a similar result to our analysis results. Göksu

(2012), because of his study on peas, reported that the fertilizers did not have effects on the cultivars and contradicted our result.

3.5. Weight of Hundred Seed

According to the results of the analysis of variance, the difference between the hundred-seed weight of organic and chemical fertilizers applied to four different bean genotypes was found to be statistically significant at the rate of 1% level (Table 1). When Table 4 is examined, the mean 100-seed weight in the parcels with sheep manure was the highest at 40.91 g. The lowest was obtained from the control plots with 36.32 g (Table 4). According to the results of the LSD test, sheep manure was in the first group (group a), cow manure was in the second group (group ab), chemical fertilizer was in the third group (group b), and control application was in the fourth group (group c) (Table 4). Göksu (2012) obtained the highest values in terms of hundred seed weight and yield in pea plants with manure application, Bulut (2013), on the other hand, reported that the mean values of fertilizer application in bean plants were between 27.43-28.73 g and the highest weight was 28.73 g from chicken manure application.

Table 4
Variance analysis of the weight of hundred seed and protein ratio of the bean genotypes that received organic and chemical fertilizers

Genotype	Chemical and Organic Fertilizers								Mean	
	Control		Chemical Fertilizers		Cow Manure		Sheep Manure			
Weight of Hundred Seed (g)										
PV2001	32,43	b	36,71	f	36,53	f	37,68	ef	35,84	c
Riberia	37,53	ef	38,72	de	39,19	de	40,49	cd	38,98	b
PV2002	38,90	de	38,95	de	41,60	bc	41,36	c	40,20	ab
PV2003	36,44	f	39,91	cd	43,45	ab	44,12	a	40,98	b
Mean	36,32	c	38,57	b	40,20	ab	40,91	a	39,00	
Protein Ratio (%)										
PV2001	27,36		27,88		26,65		28,49		27,60	a
Riberia	28,72		27,14		25,97		27,22		27,26	ab
PV2002	26,79		26,42		25,92		25,95		26,27	b
PV2003	27,07		25,55		27,39		25,95		26,49	b
Mean	27,49		26,75		26,48		26,91		26,91	

*: The differences between the means denoted by the same letter were not statistically significant.
Weight of hundred seed: $LSD_{Fertilizer}$: 1.71, $LSD_{Genotype}$: 1.30, $LSD_{Genotype\ x\ Fertilizer}$: 1.91; Protein ratio: $LSD_{Genotype}$: 2.22

According to the results of the trial, the difference between genotypes in terms of the hundred-seed weight of bean genotypes applied with organic and chemical fertilizers was found to be significant at the 5% level (Table 1). According to Table 4, it was found that the PV2003 genotype had the highest seed yield at 40.98 g, and the PV2001 genotype had the least at 35.84 g. According to the LSD test, the PV2002 genotype was in the first group (group ab), Riberia and PV2003 were in the second group (group b), and PV2001 was in the third group (group c) (Table 4). Aldemir (2019) reported that the hundred-seed weight of Gökçe and Azkan cultivars were higher than that of the Aydın 92 cultivar when the chickpea cultivars were compared.

According to the results of the analysis of variance, the difference between the hundred-seed weight of the cultivar x fertilizer interaction was statistically significant at the 5% level (Table 4). When Table 4 was examined, it was found that the highest hundred-seed weight was 44.12 g from sheep manure application with the PV2003 genotype. It was determined that the parcels that had the least weight were 36.53 g of chemical fertilizer of the PV2001 genotype (Table 4). In their study, Elsiddig et al. (1998) reported that organic matter applications applied to chickpea plants increased the hundred-seed weight.

3.6. Protein Ratio: According to the test results, it was found that organic and chemical fertilizers were statistically insignificant in terms of protein ratios (Table 1). As seen in Table 4, it was found that the highest protein rate was in the control application at 27.49%, and the lowest was in the cow manure at 26.48% (Table 4). Göksu (2012) reported that as a result of the applications in peas, the values varied between 21.15% and 24.07%, the highest value was obtained from 1 NP application, and the lowest values were obtained from control and bacteria applications. Bulut (2013) reported that the protein ratio in seed applications is between 18.28% and 21.05% as a result of fertilizer applications, the highest protein ratio is obtained in the chicken application and the lowest in the control application. Gül (2018), on the other hand, reported that the highest crude protein ratio was obtained from leonardite (45.89%) application in soybean organic fertilizer applications and the lowest from chicken manure application (45.5%).

According to the results of the analysis of variance, the effects of organic and chemical fertilizers applied to four different bean genotypes on the protein ratio were found to be statistically significant at the 1% level (Table 1). Among the four different bean genotypes used in the trial, PV2001 had the highest protein rate at 27.66% and the lowest protein rate was PV2002 with 26.27% (Table 4). According to the results of the LSD test, the

PV2001 genotype was found in the first group (group a), the Riberia genotype in the second group (group ab), and the PV2002 and PV2003 genotypes in the third group (group b) (Table 4). Gül (2018) reported that the crude protein ratio in the soybean plant was 45.71% in the Arısoy variety and 45.63% in the Nova variety and that the Arısoy variety had a higher value. Aldemir (2019) reported the effect of chickpea varieties on protein ratio as significant. Other researchers working on this subject obtained similar results (Ceyhan, 2004; Ülker and Ceyhan, 2008b; Varankaya and Ceyhan, 2012; Kepildek and Ceyhan, 2021).

4. Conclusions

The reactions of the bean lines used in the study to chemical fertilizer and some organic fertilizer applications showed differences. As a result, when the results obtained in this one-year study were examined, it was found that cow and sheep manure could be used in organic bean cultivation.

5. Acknowledgements

This paper was summarized from Master Thesis of the first author (Muhammet KÜÇÜK).

6. References

Akgün H (2018). Leonardit ve Organik Deniz Yosunu Gübrelerinin Fasulye Bitkisinin Gelişimi ile Toprağın Fiziko-kimyasal Özelliklerine Etkisi, Giresun Üniversitesi Fen Bilimleri Enstitüsü, , Giresun, 82.

Aldemir B (2019). Nohut (*Cicer arietinum L.*) Tarımında Gül Posası, Ahır Gübresi ve Bakteri Aşılamanın Verim ve Bazı Verim Öğelerine Etkisi, Isparta Uygulamalı Bilimler Üniversitesi Lisansüstü Eğitim Enstitüsü, Isparta, 51.

Başdemir F, Türk Z, İpekeşen S, Tunç M, Eliş S ve Bicer BT (2020). Bazı Bakla (*Vicia faba L.*) Çeşitlerinde Gübre Uygulamalarının Verim ve Verim Unsurlarına Etkisi, Türk Tarım ve Doğa Bilimleri Dergisi, 7 (3), 749-456.

Bulut N (2013). Aşılı Aşısız Koşullarda Fasulyede *(Phaseolus vulgaris L.*) Organik Gübrelerin Verim ve Verim Öğeleri Üzerine Etkisi, Yüzüncü Yıl Üniversitesi Fen Bilimleri Enstitüsü, Van, 47.

Ceyhan E (2004). Effect of Sowing Dates on Some Yield Components and Yield of Dry Bean *(Phaseolus vulgaris L.*) Cultivars, Turkish Journal of Field Crops, 9 (2), 87-95.

Elsiddig A, Lsheikh E, Ekhlas M ve Mohamedzein M (1998). Effect of Biologial, Organic and Chemical Fertilizers on Yield, Hydration Coefficient, Cookability, Mineral Composition of Groundnut Seeds, Food Chemistry, 63, 253-257.

Erdönmez HK (2020). Kahramanmaraş Şartlarında Farklı Gübre Formlarının Soya Fasulyesinin (*Glycine mx. (L.) Merrill*) Verim ve Kalite Unsuları Üzerine Etkisi, Kahramanmaraş Sütçü İmam Üniversitesi Fen Bilimleri Enstitüsü, 50.

FAO (2022). Crops and Livestock Products, https://www.fao.org/faostat/en/#data/QCL/visualize, [26/07/2022].

Göksu E (2012). Bezelye (*Pisum sativum L.)*'de Kimyasal, Organik ve Mikrobiyal Gübrelemenin Verim ve Verim Özelliklerine Etkileri, Uludağ Üniversitesi Fen Bilimleri Enstitüsü, Bursa, 124.

Gül S (2018). II. Ürün Olarak Yeetiştirilen Soyada (*Glycine max. (L.) Merr.*) Organik Gübre Uygulamalarının Verim Unsurları ve Bazı Kalite Kriterleri Üzerine Etkisi, Ondokuz Mayıs Üniversitesi Fen Bilimleri Enstitüsü, Samsun, 62.

Kaya AR ve Erdönmez HK (2020). Farklı Kökenli Gübre Form ve Dozlarının Soya Fasulyesinin (*Glycine max. (L.) Merrill*) Tohum Çimlenmesi ve Fide Gelişimi Üzerine Etkisi, Türk Doğa ve Fen Dergisi, 9 (Özel Sayı), 73-79.

Şehirali S (1998). Yemeklik Tane Baklagiller, Ankara, Ankara Üniversitesi Tarla Bitkileri Yayınları, p. 21-25.

Ülker M ve Ceyhan E (2008a). Orta Anadolu Ekolojik Şartlarında Yetiştirilen Fasulye (*Phaseolus vulgaris L.*) Genotiplerinin Protein ve Bazı Mineral Oranlarının Belirlenmesi, S.Ü. Ziraat Fakültesi Dergisi, 22 (46), 90-97.

Ülker M ve Ceyhan E (2008b). Orta Anadolu Ekolojik Şartlarında Yetiştirilen Fasulye (*Phaseolus vulgaris L.*) Genotiplerinin Bazı Tarımsal Özelliklerinin Belirlenmesi, S.Ü. Ziraat Fakültesi Dergisi, 22 (46), 77-89.

Varankaya S ve Ceyhan E (2012). Yozgat Ekolojik Şartlarında Yetiştirilen Fasulye *(Phaseolus vulgaris L.)* Genotiplerinin Bazı Tarımsal Özelliklerinin Belirlenmesi, Selçuk Tarım ve Gıda Bilimleri Dergisi, 26 (1), 27-33.

Yurtsever N (1984). Deneysel İstatistik Metotları, Ankara, Tarım Orman ve Köyişleri Bakanlığı Köy Hizmetleri Genel Müdürlüğü, p.

Selcuk Journal of Agriculture and Food Sciences SJAFS

http://sjafs.selcuk.edu.tr/sjafs/index

(2022) 36 (3), 507-514

e-ISSN: 2458-8377

DOI:10.15316/SJAFS.2022.066

Research Article

Drying of Melons in a Solar Tunnel Dryer: The Effect of Ascorbic Acid Solution on Drying Kinetics and Color Parameters

Sevil KARAASLAN[1,*], Barbaros Salih KUMBUL[1], Kamil EKİNCİ[1]

[1] Isparta University of Applied Sciences, Faculty of Agriculture, Department of Agricultural Machinery and Technologies Engineering, Isparta, Turkey

ARTICLE INFO

Article history:
Received date: 30.10.2022
Accepted date: 14.12.2022

Keywords:
Melon
Drying
Modelling
Colour

ABSTRACT

This study aims to present the performance of the solar tunnel dryer, which is required for the drying of two different types of melon with and without pretreatment. The solar tunnel dryer used in the study was built at Isparta University of Applied Sciences, Department of Agricultural Machinery and Technologies Engineering. The dryer consists of a flat plate solar collector, drying tunnel, solar cell module and a small axial fan. During the drying process, solar radiation, drying air temperature, relative humidity and air velocity were measured continuously in different parts of the dryer. At the same time, the mass loss of the melon slices was measured every two hours during the drying period. On the other hand, colour measurements were made on untreated and pre-treated melon slices, fresh and after drying, before and after the trials. Fresh melon samples were washed with tap water, and cut in half with a knife, the core in the middle was removed and sliced by hand in a half-moon shape without peeling before pre-treatment. 1% ascorbic acid was applied to the fruits as a pretreatment. After the preparations, the solar tunnel drying behaviour of untreated and pretreated sliced melon samples was investigated. Drying characteristic curves were evaluated according to five different mathematical models. In this study, it was determined that the L*, a* and b* values of melon varieties were preserved without pretreatment. The lowest total color change was observed in the bleached samples in both types of melon samples.

1. Introduction

Melon (*Cucumis melo*), which is from the cucurbitaceae family, is fragrant, aromatic, and pleasantly delicious, usually, It is an oval or round shaped, yellow, greenish-yellow or pinkish-orange fleshy, juicy large fruit. The origin of the melon is defined as Asia Minor (Anatolia) and Iran, and it is stated that it was cultivated in these regions 5000 years ago. According to FAOSTAT data, the production amount of melon produced in many regions of Turkey, especially with local varieties, was 1,724,856 tons in 2020.

Melon production is most intense in the Mediterranean, Western Anatolia and the Aegean. Turkey has taken its place among the dec melon producing countries in the world with its melon production. China (39%), the largest melon producer in the world, is followed by Turkey (9%). The majority of melon production in Turkey (85%) is made with Kırkağaç, Hasanbey, Yuva and Sarı Kışlık melon varieties, and the remaining part is with Cantalupensis (C. melo L. var cantalupensis) group melon varieties such as Ananas and Galia.

Solar drying has been used since ancient times to dry plants, seeds, fruits, meat, fish, wood and other agricultural and forest products. In recent years, various attempts have been made to benefit from solar energy, which is a free and renewable energy source, to protect agricultural and forest products, and to improve drying with solar energy. However, the limitations of open-field drying for large-scale production are well known. These include high labour costs, large space requirements, lack of ability to control the drying process, possible spoilage due to biochemical or microbiological reactions, insect infestation, etc. countable. Forced convection sun drying of agricultural products in closed structures is an important way to reduce post-harvest losses and poor-quality dried products compared to traditional open sun drying methods. In most developing countries and rural areas, grid-tied electricity and other non-renewable energy sources are difficult, unreliable and very expensive to supply. In such cases, solar dryers

* Corresponding author email: *sevilkaraaslan@isparta.edu.tr*

are becoming increasingly attractive in commercial spaces (Xingxing et al. 2012).

Fruits, vegetables, and dried agricultural products are good sources of energy, minerals, and vitamins. However, during the drying process, changes occur in the nutrient contents of the product (Sablani 2006). Product (colour, texture) and nutritional quality are becoming more and more important for dried fruits and vegetables. The improvement of these qualities can be achieved by pretreatments before drying. Pre-treatments suitable for agricultural products can improve the drying process by reducing drying time, providing higher quality products and energy savings.

When the previous studies are examined, it is seen that the studies on the drying characteristics of melon are insufficient, and the drying times of the melon are quite long in these studies. In this study, drying characteristics of two different melon varieties were experimentally investigated by using parameters such as different drying air temperatures, melon slice thickness and drying air velocity in a tunnel type solar dryer located in the field environment. In addition, the mathematical thin layer drying model was determined by comparing five different model equations that best describe it based on experimental data.

The main purpose of this study is to evaluate the solar tunnel dryer for thin layer drying of Galia and Kırkagac melons. In addition, the effects of pretreatment with 1% ascorbic acid solution and 3 different slice thicknesses on drying properties were determined. The best explanatory mathematical thin layer drying model to the experimental data is determined by comparing 5 different models. Finally, one of the aims is to determine the colour differences between dried and fresh fruit samples.

2. Materials and Methods

Sample Preparation

Two types of melons, Galia and Kırkağaç, were obtained from a local market in Isparta, Turkey.Before starting the experiments, the melons were peeled and sliced into 2 mm, 4 mm and 6 mm thick half moons using a cutting machine.The initial moisture content of the melon slices was determined using the oven method at 105°C for 24 hours.

Table 1
The characteristics of melon varieties

The characteristics of melon varieties		
	Galia	Kırkagac
Fruit color	light green	cream color
Fruit peel color	yellow	orange, with black dots
Weight of fruit	2-2,5 kg	3-4 kg
Shape of the fruit	round	oval
Shelf life of the fruit	long	long

Triple samples were used to determine the moisture content of melon varieties and average values were reported as 83,7927 kg water/kg dry matter (w.b.) for Galia Melon variety and 88,1213 kg water/kg dry matter (w.b.) for Kırkağaç melon variety. Melon slices were pretreated with solution of ascorbic acid (0.1%) for 2 min. The other lot was untreated (control).The characteristics of melon varieties are given in Table 1.

Solar Tunnel Drying of Melons

The researchers used a tunnel-type solar dryer that was designed and built in the Department of Agricultural Machinery and Technologies Engineering at Isparta University of Applied Sciences. The dryer used in the study consisted of a drying tunnel, a flat plate solar collector, a small axial fan, and a solar cell module. All parts are located on a frame made of metal. The solar collector has hexagonal channels. Its base is painted black and is directly connected to the drying tunnel. The solar collector is covered with a transparent polycarbonate sheet. A 150-W solar cell module is installed in the dryer to move the air by means of fan support. The collector has a surface area that is 2 m long and 1.9 m wide. The area of the drying tunnel is exactly twice the area of the collector. The solar tunnel dryer faces south in the east-west direction and does not have shade between 9:00 am and 5:00 pm.

Figure 1
The experimental solar tunnel dryer

Moisture Content

The content of moisture was calculated by drying 50 grams of melon fruit in a hot air oven at 105°C for 24 h. The measurement was performed three times for each experiment and averaged.

Moisture Ratio

The moisture ratio (MR) of the melon samples is determined (1) using the Equation (1) below.

$$MR = \frac{M_t - M_e}{M_0 - M_e} \quad (1)$$

where MR is moisture ratio (dimensionless), M_t is the moisture content of the sample at any t time ($kg_{water}/kg_{dry\ solids}$), M_e is equilibrium moisture content ($kg_{water}/kg_{\ dry\ solids}$), and Mo is the initial moisture ($kg_{water}/kg_{dry\ solids}$). M_e is relatively small for a long drying time compared to M_t or M_0. Therefore, Me is accepted as numerically zero in this study. Thus, *MR* can be simplified as $MR=M_t/M_0$. (Tunckal Doymaz 2020).

Drying Rate

The drying rate was a highly significant parameter when it comes to drying kinetics. In order to reveal the association of the drying duration of melon and the drying rate, the drying rate of melon slices was determined as follows:

$$DR = \frac{M_{t+\Delta t} - M_t}{\Delta t} \quad (2)$$

where *DR* is the drying rate ($kg_{water}/kg_{dry\ solids}$ min), M_t and M_{t+dt} are the moisture contents at t and $t+dt$ respectively, and *t* is the drying time (min) (Beige 2016).

Mathematical Modeling

Continuous data on moisture content that were obtained according to different size characteristics were turned into moisture content. Table 2 shows five different mathematical models that are used for the moisture ratio of melon samples in the literature.

Table 2
Models employed for fitting of experimental data

No	Model name	Model Equation	References
1	Page	MR=exp(-kt^n)	Diamente and Munro (1993)
2	Henderson and Pabis	MR=a exp(-kt)	Yagcioglu et al. (1999)
3	Logarithmic	MR=a exp(-kt)+c	Rahman et al. 1998
4	Midilli *et al.*	MR=a exp(-kt^n)+bt	Midilli et al. 2002
5	Two-term	MR=a exp(-k_0t)+b exp(k_1t)	Rahman and Perera (1996)

a, b, c, g, k, k0 , k1 , m, n – empirical constants and coefficients in drying models

The non-linear regression analysis of equations was carried out with Sigma Plot 12.0 statistical software and the drying parameters and coefficients (a, b, c, k, k_1, k_2, n) of these equations were calculated. The determination of the best mathematical model was based upon three statistical values of the correlation coefficient R^2 obtained by the non-linear analysis under different drying conditions, the chi-square value of χ^2, and the root mean square error (RMSE) (Doymaz et al. 2015). The lower the $\chi 2$ and RMSE values and the higher the R^2 value, the better the goodness of fit (Falade & Ogunwolu 2014). They were chosen as the criteria for goodness of fit.

The equations of R^2, SEE, RMSE and χ^2 were given in (3), (4), (5) and (6), respectively:

$$R^2 = 1 - \frac{\sum_{i=1}^{N}\left(MR_{exp,i} - MR_{pre,i}\right)^2}{\sum_{i=1}^{N}\left(\overline{MR}_{exp} - MR_{pre,i}\right)^2} \quad (3)$$

$$SEE = \frac{\sum_{i-1}^{n}\left(MR_{exp,i} - MR_{pre,i}\right)^2}{d_f} \quad (4)$$

$$RMSE = \sqrt{\frac{\sum_{i=1}^{N}\left(MR_{pre,i} - MR_{exp,i}\right)^2}{N}} \quad (5)$$

$$\chi^2 = \frac{\sum_{i=1}^{N}\left(MR_{exp,i} - MR_{pre,i}\right)^2}{N - z} \quad (6)$$

where MR_{exp} and MR_{pre} are experimental and predicted values of moisture ratio, respectively. The value z is the number of constants in the model and N is the total number of observations and d_f is the number of degrees of freedom of regression model.

Colour Parameters

Colour measurements of fresh and dried melon samples prepared with and without pretreatment were performed by reading L* (brightness/darkness), a* (redness/greenness) and b* (yellowness/blueness) values using Minolta Chroma CR-100 colour measuring device (Konica, Minolta, Tokyo, Japonya). The measurements were repeated 5 times and evaluated by taking the average of the obtained values. After the color measurement process; Chroma (C), hue angle (alpha) and ΔE values were also calculated from L*, a*, b* values. The equations used in the calculation of C, alpha and ΔE are given below (Vega-Galvez et al. 2009) (Eq.7-Eq.9).

$$C = \sqrt{(a^2 + b^2)} \quad (7)$$

$$\alpha^{\circ} = \tan^{-1}\left(\frac{b}{a}\right) \quad (8)$$

$$\Delta E = \sqrt{(L^* - {L_0}^*)^2 + (a^* - {a_0}^*)^2 + (b^* - {b_0}^*)^2} \quad (9)$$

Statistical Analysis

Sigma Plot (Scientific Graph System, version 12.00) software was used to perform the statistical analysis. Non-linear regression analysis was carried out by means of Sigma-Plot (version 12.00) in order to calculate equation parameters. The results of the regression analysis include of melon samples under solar tunnel drying; R^2, coefficient of determination; χ^2, chi-square value, and RMSE, the root mean square error.

All the samples were carried out in triplicate and all the data of the experiments were analysed by statistical software SPSS (Version 21.0, SPSS Inc., Chicago, IL, USA). The results of the experiments were expressed as mean ± standard deviation. Analysis of variance (ANOVA) was used to find the significant terms of each response in the model and values of $p \leq 0.05$ were considered statistically significant.

3. Results and Discussion

Figure 2 shows the panel inlet and outlet temperature as a function of drying time. The panel inlet temperature varied between 34.4°C and 47.1°C while the panel outlet temperature ranged from 39.3°C to 52.7°C. Figure 3 presents air velocity change as a function of time and days. According to Figure 3, the velocity values obtained from the fans reached a peak approximately in the middle of the day. The solar cell provided the fan with power; therefore, the velocity of air flow varied as a function of solar irradiation. Solar irradiance levels were low in the morning and afternoon as solar angles varied throughout the day.

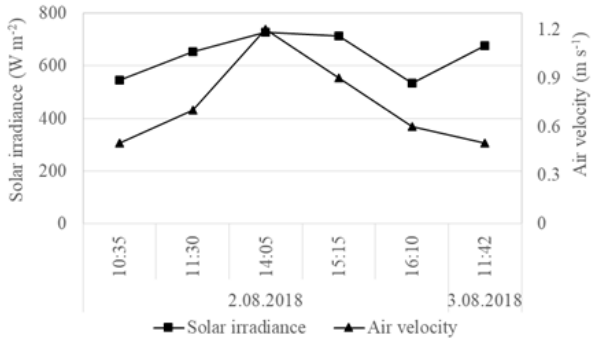


Figure 2
Change of solar irradiance and air velocity of drying air at the outlet of drying tunnel as a function of time

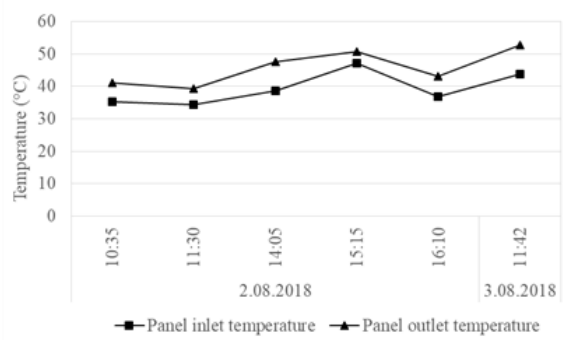


Figure 3
Change of panel inlet and outlet temperature as a function of time

In Figure 4 and 5, the effect of slice thickness of without pretreated and pretreated samples on the moisture ratio of two different melon varieties, Galia and Kırkagac, is given. As seen in Figure 4, the drying time of the melon increased significantly with the increase in slice thickness in both melon varieties. In both types of melons, the effect of pretreatment was seen at the end of drying. The pretreated samples were dried in shorter drying times than the without pretreated samples in both varieties. Samples with low slice thickness and pretreated were dried in the shortest time. Samples with a large slice thickness and without pretreatment were dried for the longest time. The drying time required for the drying of the Galia variety melon was found to be 3360, 4560 and 4800 minutes for 2mm, 4mm and 6mm slice thicknesses without pretreatment, respectively. However, the drying time required for drying the Galia variety melon was determined as 1680, 1680 and 3120 minutes for pretreated 2mm, 4mm and 6mm slice thicknesses, respectively. In another melon variety, Kırkağaç, the required drying time was found to be 2880, 3360 and 4560 minutes for slice thicknesses of 2mm, 4mm and 6mm without pretreatment, respectively. In the pretreated application, the drying times were determined as 1920, 2880 and 3360 minutes for 2mm, 4mm and 6mm slice thicknesses, respectively. The following study can be given as an example to show the effectiveness of preprocessing. According to a study on sliced persimmon, the samples were dried in a hot air dryer at three different temperatures (50 °C, 60 °C and 70 °C), respectively. It was stated that at the end of the drying processes, the pre-treated slices dried faster than the others (Doymaz 2012).

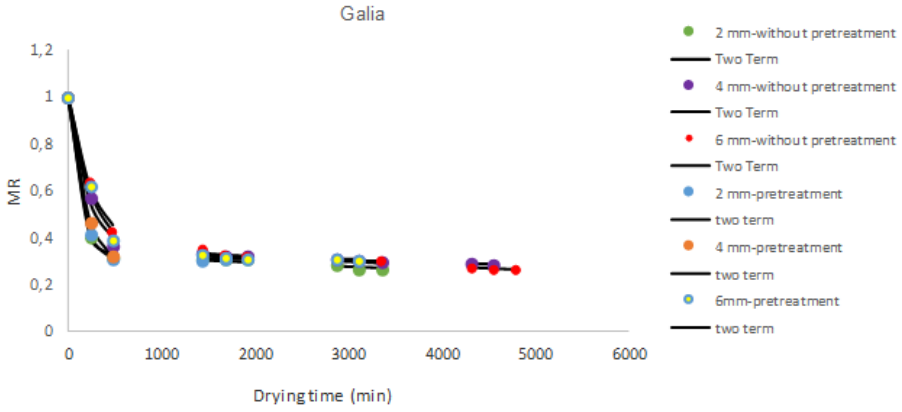


Figure 4
Drying curves of "Galia melon" dried with and without pretreatment at different slice thicknesses

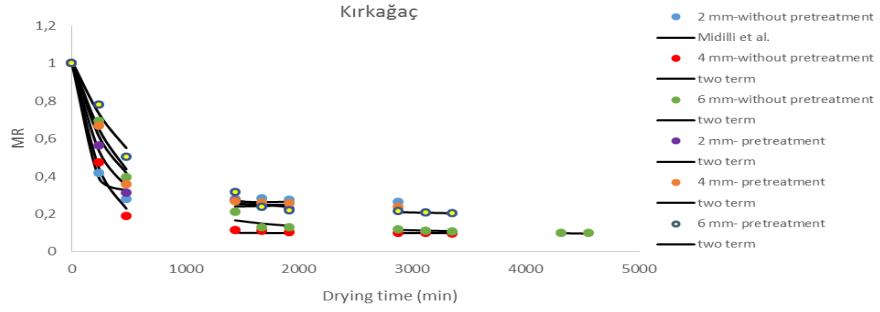


Figure 5
Drying curves of "Kırkağaç melon" dried with and without pretreatment at different slice thicknesses

Figure 6 and 7 show the variation of drying time and drying rate of Galia and Kırkagac melon samples dried with and without pre-treatment at different slice thicknesses in a solar tunnel drying system. Accordingly, the total drying rates were determined as 0.210333 kg [H_2O] kg-1dry solids.min for 2 mm sample thickness, 0,151 kg [H_2O] kg-1dry solids.min for 4 mm, and 0,12925 kg [H_2O] kg-1dry solids.min for 6 mm for the without pre-treatment of Galia melon varieties, respectively.

Additionally the total drying rates were determined as 0.203 kg [H_2O] kg-1dry solids.min for 2 mm sample thickness, 0.184583 kg [H_2O] kg-1dry solids.min for 4 mm, and 0,135 kg [H_2O] kg-1dry solids.min for 6 mm for the pre-treatment of Galia melon varieties, respectively.

In the Kırkağaç melon varieties, the drying rates differed as 0.211792 kg [H_2O] kg-1dry solids.min for 2mm slice thickness, 0,19575 kg [H_2O] kg-1dry solids.min for 4 mm slice thickness, 0.119167 kg [H_2O] kg-1dry solids.min for 6 mm slice thickness and 0,161375 kg [H_2O] kg-1dry solids.min for 2 mm, 0,124167 kg [H_2O] kg-1dry solids.min for 4 mm, 0,105583 kg [H_2O] kg-1dry solids.min for 6 mm respectively, according to without pretreated and pretreated applications.

Results showed that drying rate was between 0,001875 and 0.00033 kg [H_2O] kg-1dry solids.min both without pretreatment and pretreatment for Galia melon samples at the final stage of drying. In addition, drying rates for dried Kırkagac melon, for untreated and pretreated samples varied between 0.001375 and 0.00075 kg [H_2O] kg-1dry solids.min.

Results showed that drying rate sharply increased within four hours and then decreased. In this study conducted with Galia and Kırkagac melon varieties, all drying methods were carried out in the period of falling drying rate. Also, there is no constant rate drying period in the drying rate curves. Parallel results were obtained in the study conducted with grape fruit by Yaldız et al. (2001).

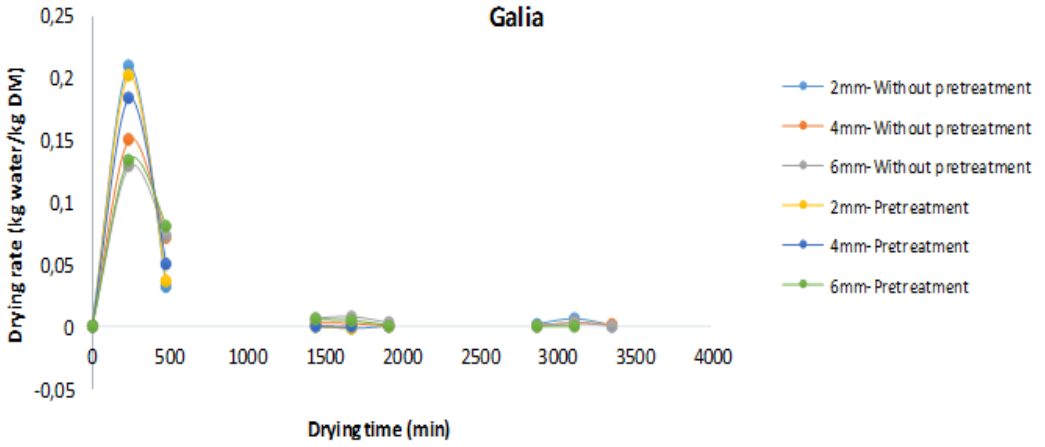


Figure 6
Drying rate curves of "Galia melon" dried with and without pretreatment at different slice thicknesses

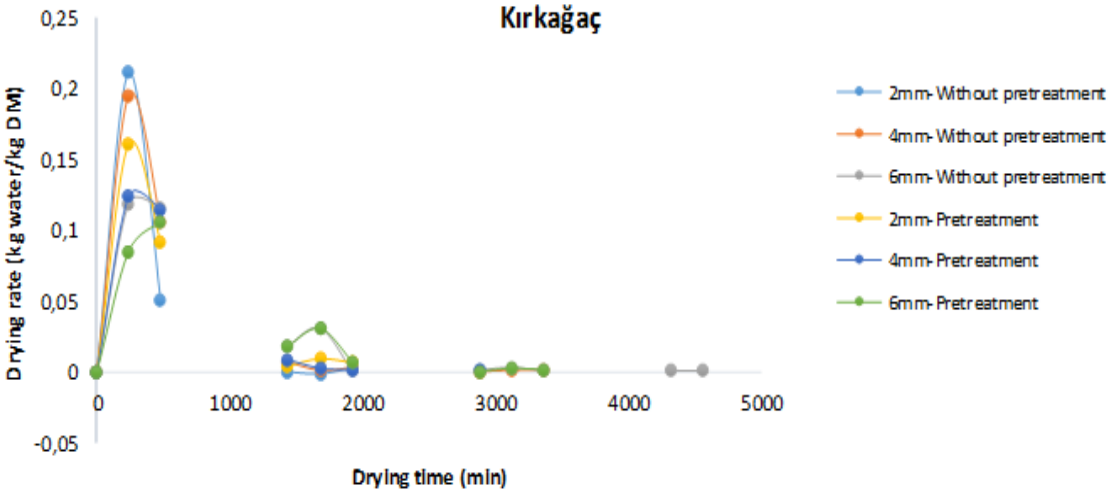


Figure 7
Drying rate curves of "Kırkağaç melon" dried with and without pretreatment at different slice thicknesses

Table 3 shows the model constants and statistical analysis data including R2, SEE, RMSE and $\chi 2$ values for all thin layer drying models suitable for moisture content data.

For the Galia melon variety, statistical parameter estimates in both conditions showed that the values of R^2, SEE, RMSE and $\chi 2$ ranged from 0,4743 to 1,0000, 0,0006 to 0,2148, 0,00027 to 0,17768 and 0,00002 to

0,092203, respectively. When a comparison was made between the 5 models and the statistical values, the Two Term model showed the greater R^2 and the lesser RMSE and χ 2 values with respect to other drying models at without pretreatment and pretreatment conditions.

On the other hand, statistical parameter estimates in both conditions showed that the values of R^2, SEE, RMSE and $\chi 2$ ranged from 0,5227 to 0,9961, 0,0221 to 0,2045, 0,01801 to 0,1728 and 0,00134 to 0,36968, respectively for the kırkagac varieties.

The drying characteristic curves were estimated against five mathematical models, and all other data showed that the Two Term model equation was the best descriptive model, except for the experiments of untreated thin-layered Kırkağaç melon samples with a slice thickness of 2 mm. On the other hand, in the untreated Kırkagac melon sample with a slice thickness of 2 mm, Midilli et al. model equation was found to be the best descriptive model.

Colour Measurement

Colour values of fresh and dried Galia and Kırkagac melon samples are shown in Tables 4 and 5. The brightness values (L*) of the melon samples (Pineapple and Kırkağaç) dried in the solar tunnel dryer increased compared to the fresh samples. While the L* values were 61.98 before drying in the Galia variety melon samples, it was determined that this value changed between 62.84 and 73.15 after drying. On the other hand, the Galia melon samples treated with ascorbic acid were compared with the untreated samples and it was determined that the brightness values (L*) after the pretreatment applications were the closest to fresh. Likewise, the same is true for dried Kırkagac melon samples. While the L* values before drying were 61.38 in Kırkagac variety melon samples, it was determined that this value changed between 66.9 and 80.71 after drying. In other words, it was observed that the brightness values of the samples dried with the pre-treatment drying method were closer to the brightness values of the fresh product (L*). In general, the brightness values of both melon cultivars dried by the application of ascorbic acid solution decreased and the values closest to fresh were obtained. Color changes often take place in dried foods due to non-enzymatic browning reactions (Sroy et al. 2022).

The a* value on the color scale means red or green. An increase in a* means a redder chroma, known as an indicator of the browning reaction. Also, the b* value on the color scale indicates yellowness or blueness. An increasing b* value ($P < 0.05$) indicates the more yellow sample. While no significant change was observed in b* values of Galia melon samples, an increase was detected in pretreated applications of a* values. The a* (redness) value was affected by the applied pretreatment. It was determined that higher a* values ($P < 0.05$) were obtained in the pretreated samples compared to the untreated samples.

Another criterion used to compare the color qualities of the samples is their b* values. It is desired that this value be as low as possible in quality dried melon varieties. When the samples were compared in terms of b* values, the lowest value was determined in the products dried without pretreatment. When the Kırkağaç melon samples are examined in terms of b* (yellowness) values, the situation is slightly different compared to a* values. b* values decreased in applications without pretreatment. However, in preprocessed applications, results closer to the fresh value were obtained. On the other hand, in a* (redness) values, the closest results to the fresh value were obtained in applications without pretreatment. As in the Galia melon samples, an increase in the a* values obtained as a result of the drying process applied with the pretreatment was observed in the Kırkagac melon samples.

The chroma value is one of the most effective factors in the appearance of the products and is effective in product preference. The chroma value shows the tone of the color in the products, and the values are low in pale colors and high in bright colors (Çetin, 2019). C values, b* (yellowness) values of both dried melon samples closely followed. However, the highest C values for both melon varieties were determined as 33.89 for Galia variety with 2mm thickness pre-treated and 28.46 for Kırkagac variety with 2mm thickness pre-treated, respectively. This showed us that the 2mm thick pretreated samples in both varieties were significantly ($P<0.05$) more vivid in color than any of the other dried and fresh samples.

Kırkagac melon without preprocessing samples, the hue angle decreased while the total color difference increased. On the contrary, in the pretreated samples, the hue angle increased while the total color difference decreased. On the other hand, no significant difference was observed in the total color difference values in Galia melon samples in both applications.

In this study, it was determined that better results were obtained when the L*, a* and b* values of melon varieties were dried without pretreatment.

Table 3
Statistical results obtained from the selected drying models

		Melon varieties											
		Galia						Kırkağaç					
		Without pretreatment			Pretreatment			Without pretreatment			Pretreatment		
		2mm	4mm	6mm	2mm	4mm	6mm	2mm	4mm	6mm	2mm	4mm	6mm
Page	R^2	0,9940	0,9712	0,9746	0,9918	0,9842	0,9625	0,9864	0,9732	0,9732	0,9718	0,9520	0,9672
	SEE	0,0195	0,0385	0,0363	0,0316	0,0432	0,0520	0,0345	0,0535	0,0518	0,0568	0,0699	0,0569
	RMSE	0,017215	0,034874	0,033101	0,024477	0,033494	0,045045	0,029144	0,04719	0,046839	0,046358	0,059086	0,050138
	χ^2	0,000874	0,002788	0,002411	0,001681	0,00292	0,004699	0,002466 4	0,011921	0,015767	0,005125	0,008337	0,007742
Henderson and Pabis	R^2	0,4743	0,5548	0,6626	0,5674	0,6099	0,6739	0,5227	0,9195	0,9266	0,7765	0,7983	0,9053
	SEE	0,1824	0,1515	0,1322	0,2294	0,2148	0,1534	0,2045	0,0927	0,0858	0,1597	0,1433	0,0966
	RMSE	0,160889	0,13701	0,120655	0,177685	0,166393	0,132879	0,172834	0,08178	0,07756	0,130388	0,121083	0,085172
	χ^2	0,050281	0,039565	0,032246	0,092203	0,073338	0,041401	0,074784	0,23778	0,369689	0,047849	0,055081	0,036018
Logarithmic	R^2	0,9943	0,8605	0,7741	0,9996	0,9985	0,9950	0,9607	0,9961	0,9922	0,9927	0,9855	0,9898
	SEE	0,0206	0,0899	0,1140	0,0085	0,0164	0,0208	0,0656	0,0221	0,0297	0,0333	0,0429	0,0343
	RMS	0,016823	0,076707	0,098725	0,00539	0,01041	0,016471	0,049602	0,018019	0,025347	0,023593	0,032452	0,02798
	χ^2	0,000981	0,017439	0,028768	0,00009	0,000314	0,000599	0,008218	0,001343	0,002676	0,001632	0,002219	0,001876
Two-term	R^2	0,9981	0,9958	0,9968	1,0000	0,9998	0,9950	0,9790	0,9961	0,9930	0,9929	0,9855	0,9899
	SEE	0,0131	0,0167	0,0143	0,0006	0,0081	0,0203	0,0553	0,0242	0,0299	0,0403	0,0495	0,0374
	RMSE	0,009769	0,013349	0,011708	0,00027	0,003295	0,016467	0,036188	0,018033	0,023884	0,023238	0,032426	0,027866
	χ^2	0,000315	0,000429	0,000309	0,00002	0,00004	0,00060	0,003904	0,001365	0,002015	0,001692	0,002107	0,00183
Midilli *et al.*	R^2	0,9940	0,9802	0,9840	0,9995	0,9988	0,9832	0,9909	0,9896	0,9894	0,9888	0,9734	0,9878
	SEE	0,0231	0,0362	0,0321	0,0132	0,0208	0,0427	0,0365	0,0394	0,0369	0,0505	0,0672	0,0409
	RMSE	0,017221	0,028887	0,026246	0,005896	0,009308	0,030169	0,023887	0,029355	0,02943	0,029129	0,043948	0,030524
	χ^2	0,000873	0,00191	0,00153	0,000115	0,000282	0,002001	0,001769	0,012353	0,002855	0,003421	0,004657	0,002016

Table 4
Colour measurements of "Galia"melon fruit in different slice thicknesses (Statistical analysis conducted by one-way ANOVA)

Galia	L*	a*	b*	C*	α*	ΔE
Fresh	$61,98^{b}$ (±5,29)	$1,991^{b}$(±1,113)	$26,23^{ab}$(±6,36)	$26,32^{b}$(±6,4)	$85,744^{a}$(±2,049)	-
2 mm-Without pretreatment	$73,15^{a}$(±2,42)	$1,872^{b}$(±1,191)	$27,5^{ab}$(±2,93)	$27,58^{ab}$(±2,99)	$86,27^{a}$(±2,159)	11,61(2,31)
4 mm-Without pretreatment	$69,36^{ab}$(±5,89)	$2,008^{b}$(±1,508)	$27,144^{ab}$(±1,739)	$27,248^{ab}$(±1,813)	$85,85^{a}$(±2,94)	8,77(3,6)
6 mm-Without pretreatment	$69,27^{ab}$(±5,76)	$1,52^{b}$(±0,87)	$23,832^{b}$(±1,164)	$23,893^{b}$(±1,176)	$86,379^{a}$(±2,069)	8,78(3,58)
2 mm-Pretreatment	$65,22^{ab}$(±8,46)	$5,86^{a}$(±3,03)	$33,24^{a}$(±3,05)	$33,89^{a}$(±2,54)	$79,68^{b}$(±5,87)	11,64(3,72)
4 mm-Pretreatment	$62,84^{ab}$(±4,22)	$6,07^{a}$(±1,232)	$29,92^{ab}$(±2,78)	$30,55^{ab}$(±2,87)	$78,564^{b}$(±1,896)	6,81(2,79)
6 mm-Pretreatment	$63,04^{ab}$(±4,89)	$5,27^{a}$(±1,322)	$28,47^{ab}$(±4,32)	$28,96^{ab}$(±4,46)	$79,617^{b}$(±1,401)	7,08(1,63)

The statistics of each color parameter column were applied separately, and the differences between the means with different letters in the same column were significant ($p<0.05$).

Table 5
Color measurements of "Kırkagac"melon fruit in different slice thicknesses (Statistical analysis conducted by one-way ANOVA)

Kırkagac	L*	a*	b*	C*	α*	ΔE
Fresh	$61,38^{d}$(±5,82)	$-1,255^{b}$(±1,981)	$24,4^{ab}$(±4,23)	$24,51^{ab}$(±4,18)	$-41,7^{b}$(±4,6)	-
2 mm-Without pretreatment	$80,718^{a}$(±1,546)	$-1,244^{b}$(±0,23)	$15,148^{c}$(±0,549)	$15,201^{c}$(±0,528)	$-85,277^{b}$(±1,044)	$21,45^{a}$(1,31)
4 mm-Without pretreatment	$76,16^{ab}$(±2,91)	$-0,89^{b}$(±0,501)	$14,596^{c}$(±2,011)	$14,634^{c}$(±1,976)	$-86,26^{b}$(±2,49)	$17,78^{ab}$(3,32)
6 mm-Without pretreatment	$73,96^{abc}$(±5,8)	$-0,7^{b}$(±0,289)	$15,12^{c}$(±2,49)	$15,14^{c}$(±2,48)	$-87,186^{b}$(±1,411)	$15,77^{abc}$(5,94)
2 mm-Pretreatment	$66,9^{cd}$(±4,6)	$2,79^{a}$(±2,24)	$28,28^{a}$(±4,6)	$28,46^{a}$(±4,81)	$84,78^{a}$(±3,34)	$9,46^{cd}$(3,59)
4 mm-Pretreatment	$68,05^{bcd}$(±4,05)	$0,826^{ab}$(±0,856)	$21,224^{abc}$(±1,861)	$21,254^{abc}$(±1,865)	$51,8^{a}$(±1,902)	$8,44^{d}$(2,29)
6 mm-Pretreatment	$71,72^{bc}$(±3,09)	$-0,084^{ab}$(±0,513)	$20,128^{bc}$(±1,8)	$20,134^{bc}$(±1,796)	$-17,7^{b}$(±1,291)	$11,40^{bcd}$(2,99)

The statistics of each color parameter column were applied separately, and the differences between the means with different letters in the same column were significant ($p<0.05$).

4. Conclusions

The solar tunnel dryer produced in our department can be used for drying melon fruit as well as drying different agricultural products under the climatic conditions of Isparta. In this study, a constant drying rate period was not observed, on the contrary, all drying processes took place in a decreasing rate period. However, the moisture content values were determined as

83.7927 kg water/kg dry matter (w.b.) for the Galia variety and 88.1213 kg water/kg dry matter (w.b.) for the Kırkagac variety, 10.78% and 12.52%, respectively. The trials lasted only two days, depending on the weather conditions, in the solar tunnel dryer. At the

same time, the melon samples dried in the solar tunnel dryer were completely protected from birds, insects, dust and bad weather conditions due to the closed system. Dried melon variety samples were brighter and more yellow when dried with without pretreatment compared to drying pretreatment. No significant effect of the slice thickness applications carried out within the study directly on the final product was detected.

5. References

Beigi M (2016). Hot air drying of apple slices: dehydration characteristics and quality assessment. *Heat and Mass Transfer* 52(8): 1435-1442. http://dx.doi.org/10.1007/s00231-015-1646-8.

Çetin N (2019). Effect of Drying Conditions on Color Properties of Apples and Oranges. *European Journal of Science and Technology* 17: 463-470.

Diamente LM, Munro PA (1993). Mathematical modeling of the thin layer solar drying of sweet potato slices. *Solar Energy* 51(4): 271–276.

Doymaz I (2012). Evaluation of some thin-layer drying-models of persimmon slices (Diospyros kaki L.). *Energy Conversion and Management* 56: 199-205.

Doymaz I, Kipcak AS, Piskin S (2015). Microwave drying of green bean slices: drying kinetics and physical quality. *Czech J. Food Sci*. 33: 367–376.

Falade K, Ogunwolu OS (2014). Modeling of drying patterns of fresh and osmotically pretreated cooking banana and plantain slices. *Journal of Food Processing and Preservation* 38: 373–388.

Midilli A, Kucuk H, Yapar Z (2002). A new model for single-layer drying. *Drying Technology* 20(7): 1503–1513.

Rahman MS, Perera CO (1996). Effect of pre-treatment on airdrying rate and thin layer drying characteristics of fresh cheery. *In Proceedings of the 10th international drying symposium* Krakow Poland.

Rahman MS, Perera CO, Thebaud C (1998). Desorption isotherm and heat pump drying kinetics of peas. *Food Research International* 30(7): 485–491.

Sablani SS (2006). Drying of Fruits and Vegetables Retention of Nutritional/ Functional Quality. *Drying Technol*ogy 24:428–432.

Sroy S, Miller FA, Fundo JF, Silva CLM, Brandão TRS (2022). Freeze-Drying Processes Applied to Melon Peel: Assessment of Physicochemical Attributes and Intrinsic Microflora Survival during Storage. *Foods* 11: 1499.

Tunckal C, Doymaz I (2020). Performance analysis and mathematical modelling of banana slices in a heat pump drying system. *Renewable Energy* 150: 918-923.

Xingxing Z, Xudong Z, Stefan S, Jihuan X, Xiaotong Y (2012). Review of R&D progress and practical application of the solar photovoltaic/thermal (PV/T) Technologies. *Renewable and Sustainable Energy Reviews* 16: 599-617.

Yagcioglu A, Degirmencioglu A, Cagatay F (1999). Drying characteristics of laurel leaves under different drying conditions. *In Proceedings of the 7th international congress on agricultural mechanization and energy*, Adana, Turkey.

Yaldiz O, Ertekin C, Uzun HI (2001). Mathematical modelling of thin layer solar drying of Sultana grapes. *Energy* 26(5): 457- 465.

Selcuk Journal of Agriculture and Food Sciences SJAFS

http://sjafs.selcuk.edu.tr/sjafs/index

(2022) 36 (3), 515-520

e-ISSN: 2458-8377

DOI:10.15316/SJAFS.2022.067

Research Article

Classification and Analysis of Tomato Species with Convolutional Neural Networks

Yavuz Selim TASPINAR[1,*]

[1]Selcuk University, Doganhisar Vocational School, Department of Transport and Traffic Services, Konya, Türkiye

ARTICLE INFO

Article history:
Received date: 16.11.2022
Accepted date: 19.12.2022

Keywords:
Classification
Computer Vision
Food Quality
Nutrients
Tomato Varieties

ABSTRACT

Tomatoes are one of the most used vegetables. There are varieties that can grow in different climates. The taste, usage area and commercial value of each are different from each other. For this reason, identifying and sorting tomato species after the production stage is a problem. In addition, since tomato is a sensitive vegetable, it is extremely important to separate it from a distance. For this purpose, the classification of tomato images belonging to 9 different tomato species was carried out in the study. In total, a dataset containing 6810 tomato images in 9 classes was used. Three different pre-trained Convolutional Neural Network (CNN) models were used with the transfer learning method to classify the images. AlexNet, InceptionV3 and VGG16 models were used for classification. As a result of the classifications made, the highest classification belongs to the AlexNet model with 100%. Evaluation of the performances of the models was also made with precision, recall, F1 Score and specificity performance metrics. It is foreseen that the proposed methods can be used for the separation of tomatoes.

1. Introduction

The main food sources of humans are protein, carbohydrates and fats. They can get these nutrients directly or indirectly from various fruits and vegetables. From past to present, vegetable and fruit cultivation has developed rapidly. As a result, different species emerged. The genetics of the seeds have been changed to meet the demands of people and increase the yield. As a result of these changes, different species emerged. Many different types have emerged in tomato from vegetable varieties. There are differences in color, texture, odor and flavor according to the species. For this reason, the differentiation of tomato species has therefore become important. In the literature, there are studies on the identification of tomato species and other vegetable and fruit species with image processing methods.

Jhaawar has differentiated oranges according to their size, quality and type using image processing methods. Using pattern recognition techniques, he classified oranges on a single color basis. He carried out his work using 160 orange images. He used only 4 image features to classify oranges into four classes based on maturity level and 3 classes based on size. It has achieved classification accuracies of up to 905 and 98% from Multi Seed Nearest Neighbor and Linear Regression methods, respectively (Jhawar, 2016). Arakeri et al. have proposed an automatic tomato grading system. The proposed method consists of two steps. In the first step, the software identifies the defects in the tomato. In the second stage, the maturity of the tomatoes was analyzed by image processing techniques. They achieved 96.47% classification accuracy with the method they suggested (Arakeri, 2016). Ramos et al. proposed a non-destructive method in order to estimate the number of fruit on the branches of the coffee tree with one-sided images of the branch. A total of 1018 coffee branch images were used. The images were collected in different numbers of trees, branches and times. In their experiments, they obtained an R2 result of over 0.93 (Ramos, Prieto, Montoya, & Oliveros, 2017). Sofu et al. have proposed an automatic apple grading and quality control system. They classified apples according to color, weight and size. With the method they recommend, they can also detect stains, crusts and rot. They used an industrial camera placed on a conveyor in a closed cabinet to analyze the image properties of apples. As a result, they were able to extract an average of 15 apples per second with the method they proposed. As a result of experimental studies, they achieved an average of 73%-96% separation accuracy (Sofu, Er, Kayacan, & Cetişli, 2016).

* Corresponding author email: *ytaspinar@selcuk.edu.tr*

Elhariri et al. proposed an image processing-based system to monitor the ripening processes of tomatoes. The proposed approach consists of three stages. They determined that it was tomato with pre-processing, feature extraction and classification stages. Since color is the most important feature in maturity, they used a colored histogram. They used Support Vector Machine and Principal Component Analysis (PCA) for classification. There are 230 images in total in the dataset they use. They divided the images into 5 classes according to their maturity stage. As a result of the experiments, they obtained 92.72% classification accuracy with the SVM method (Elhariri et al., 2014). Ohali proposed an image-processing-based system to rate the quality of dates by date. They identified a set of external quality characteristics that they identified. According to the features extracted from the date images, the dates are divided into 3 quality grades by experts. They achieved 80% classification accuracy in their classification with the back propagation neural network (Al Ohali, 2011). Zhang et al. proposed a hybrid classification model based on artificial bee colony and feedforward neural network to classify fruits. By removing the background of the fruit images they used, they extracted the color histogram, shape and texture properties of the images. They used PCA to reduce the number of features. They achieved the highest classification accuracy of 88.72% with a dataset of 1653 images in 18 classes (Zhang, Wang, Ji, & Phillips, 2014). Muhammad used a feature extraction based SVM classifier to classify palm images in his study. Local Binary Pattern (LBP) and Weber Local Descriptor (WLD) were used to extract features from images. It combined the obtained features. Fisher used the Discrimination Ratio (FDR) to reduce the dimensionality of the feature set. As a result of the classifications made with SVM, it has achieved more than 98% classification accuracy (Muhammad, 2015). Moallem et al. proposed a six-step computer vision-based apple classification method. As a result of all operations, they extracted statistical, geometric and textural properties of apples. Finally, SVM, Multilayer Perceptron (MLP) and k Nearest Neighbor (kNN) were used for classification. In their classifications, they achieved the highest classification accuracy of 92.5% with SVM (Moallem, Serajoddin, & Pourghassem, 2017).

Oo et al. proposed a simple and efficient image processing method for estimating strawberry shape and size. In their proposed method, diameter, apex angle and length properties are used for estimation. They achieved classification accuracy between 94% and 97% in their classification with artificial neural networks (Oo & Aung, 2018). Mim et al. classified mango fruits according to six maturity levels. They used more than 100 mango images in the experiments. 24 image features were classified by the decision tree method. As a result of the classifications, they achieved classification accuracy of up to 96% (Mim, Galib, Hasan, & Jerin, 2018). Wang et al. classified fungi according to their diameters by image processing methods. With the algorithms they proposed, they eliminated the effect of shadow and stem on the image. They achieved 97.42% classification accuracy in their experimental studies with OpenCV (Wang et al., 2018). Wan et al. used color values and ANN to determine the maturity level of fresh tomatoes. The diameter and color of the tomatoes were used to determine the maturity level. As a result, they achieved 99.31% classification accuracy (Wan, Toudeshki, Tan, & Ehsani, 2018).

When the studies in the literature are examined, it is seen that the types, ripe levels and quality of vegetables and fruits can be classified by image processing methods. Although there are studies on tomato, there are not many studies on the classification of the species. For this reason, in this study, the subject of classification of tomato species was studied. The steps in the article are as follows:

• A dataset containing a total of 6810 images of 9 different tomato species was used.

• For classification of tomato images, the dataset is divided into 5103 trains and 1707 test images.

• AlexNet, InceptionV3 and VGG16 pre-trained models were used for classification with transfer learning method.

• Confusion matrix tables were used to compare the classification performances of the models.

• Performance metrics were calculated using confusion matrix data for the detailed analysis of the performances of the models.

When other studies in the literature are examined, the contributions of this study to the literature are listed as follows:

• A 9-class tomato dataset, which is not included in other studies in the literature, was used.

• Classification of tomato images was made with 3 different CNN models and compared.

• Performance evaluation of AlexNet, InceptionV3 and VGG16 pre-trained models was made.

The study includes the material and method used in the 2nd section, the 3rd section experimental results and the 4th section results and recommendations.

2. Materials and Methods

In this section, the dataset used in the study, CNN, pre-trained CNN models and the methods used in performance evaluation are explained.

2.1. Tomato Dataset

The dataset used in the study includes images of 9 tomato species (Mureşan & Oltean, 2017; "Tomato Dataset,"). 5103 tomato images are reserved for train, 1707 tomato images are reserved for testing. The total number of images is 6810. Each image in the dataset is 100x100 pixels. The names and image numbers of the tomato classes in the dataset are given in Table 1. Example images according to the classes in the dataset are given in Figure 1.

Table 1
Data counts by classes in the dataset

Class	Number of Images	
	Train	Test
Tomato 1	738	246
Tomato 2	672	225
Tomato 3	738	246
Tomato 4	479	160
Tomato Cherry Red	492	164
Tomato Heart	684	228
Tomato Maroon	367	127
Tomato Yellow	459	153
Tomato Not Ripened	474	158

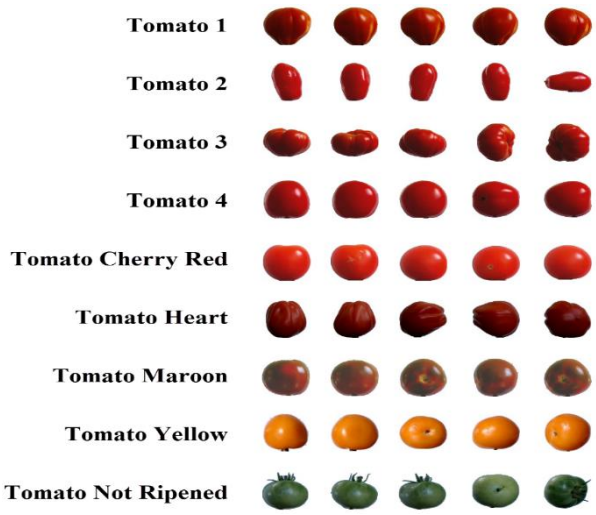


Figure 1
Example images of classes in the dataset

2.2. Convolutional Neural Network (CNN)

CNN is a deep learning model used in object detection, pattern recognition and image classification problems. There are different layers in the CNN structure. End-to-end classification processes can be performed with convolutional, pooling, activation and fully connected layers (Koklu, Kursun, Taspinar, & Cinar, 2021). Convolution is the layer where feature extraction operations are performed from the image. Feature maps are output from this layer (Singh et al., 2022). The pooling layer is used to get rid of data clutter in feature maps. This layer reduces the size of feature maps. Activation layers, on the other hand, ensure that the data is kept within a certain range. Classification operations of feature maps are performed with the fully connected layer. The number of layers in CNN may differ according to the problem (Unal, Taspinar, Cinar, Kursun, & Koklu, 2022). AlexNet, InceptionV3 and VGG16 architectures were used in this study.

2.3.Transfer Learning and AlexNet, InceptionV3, VGG16 Pre-trained Models

It is the use of machine learning methods to solve other problems. Pre-trained CNN models are trained with a large number of images. The information obtained as a result of the training can be stored in the network and used in the classification of new images (Taspinar et al., 2022). The advantage of transfer learning is that high accuracy can be achieved using less training data. At the same time, the model tends to be trained quickly with new images since it has been run on images before. A large amount of data may be needed to train models from scratch (Kishore et al., 2022). At the same time, high hardware features may be required. The models used in this study are proven models. The AlexNet pre-trained model has a depth of 8. The AlexNet pre-trained model includes 61M parameters. The Inception V3 model has a depth of 48 and contains 23.9M parameters. The VGG16 model has a depth of 16 and includes 138M parameters. For classification of all pre-trained models and images, the input layer and the penultimate layer, the fully connected layer, are set for this study. The number of fully connected layer outputs is set to 9, which is the number of classes in the dataset.

2.4. Confusion matrix and performance metrics

It is a table used to evaluate the performance of models used in solving confusion matrix classification problems (Taspinar, Cinar, & Koklu, 2021). There are True positive, True negative, False positive and False negative values on the table (Cinar & Koklu, 2022). The 9x9 confusion matrix used in the study and the calculation of these values are shown in Figure 2.

ACTUAL CLASS

PREDICTED CLASS	Tomato 1	Tomato 2	Tomato 3	Tomato 4	Tomato Cherry Red	Tomato Heart	Tomato Maroon	Tomato Yellow	Tomato Not Ripened
Tomato 1	TN	FN	TN	TN	TN	TN	TN	TN	TN
Tomato 2	FP	TP	...	...	...	...	...	...	FP
Tomato 3	TN	...	TN	TN	TN	TN	TN	TN	TN
Tomato 4	TN	...	TN	TN	TN	TN	TN	TN	TN
Tomato Cherry Red	TN	...	TN	TN	TN	TN	TN	TN	TN
Tomato Heart	TN	...	TN	TN	TN	TN	TN	TN	TN
Tomato Maroon	TN	...	TN	TN	TN	TN	TN	TN	TN
Tomato Yellow	TN	...	TN	TN	TN	TN	TN	TN	TN
Tomato Not Ripened	TN	FN	TN	TN	TN	TN	TN	TN	TN

Figure 2
9x9 confusion matrix

With these values, some metrics can be calculated to measure the performance of the models (Taspinar, Koklu, & Altin, 2021). The metrics used in the study are Accuracy, precision, recall, specificity and F1 Score. Accuracy is the rate of correct prediction. Precision shows how many of the predicted samples are actually correct. Recall is the metric that shows how many of the samples belonging to the positive class are correct. Specificity is the ratio of false positives to false positives and true negatives. F1 Score is the harmonic mean of precision and recall. This metric is an important metric that shows the strength of the model (Al-Doori, Taspinar, & Koklu, 2021). The formulas of these metrics used in the study are shown in Table 2.

Table 2
Performance metrics equations

Metrics	Equation
Accuracy	$\frac{TP+TN}{TP+TN+FP+FN} x100$
Precision	$\frac{TP}{TP+FP}$
Recall	$\frac{TP}{TP+FN}$
F1 Score	$2x\frac{Precision\ x\ Recall}{Precision+Recall}$
Specificity	$\frac{FP}{FP+TN}$

3. Experimental Results

In this section, classification results and analyzes made with tomato dataset are given. AlexNet, InceptionV3 and VGG16 pre-trained models were used to classify tomato images. As a result of the training and tests, a confusion matrix was obtained for each model. Performance metrics of the models were calculated with the obtained confusion matrix data. In the study, a computer with Intel® Core i7™ 12700K 3.61 GHz, NVIDIA GeForce RTX 3080Ti, and 64GB RAM was used. For training and testing the models, the dataset is divided into train 75% - test 25%. The classification processes of the images in the tomato dataset in the study are shown in Figure 3.

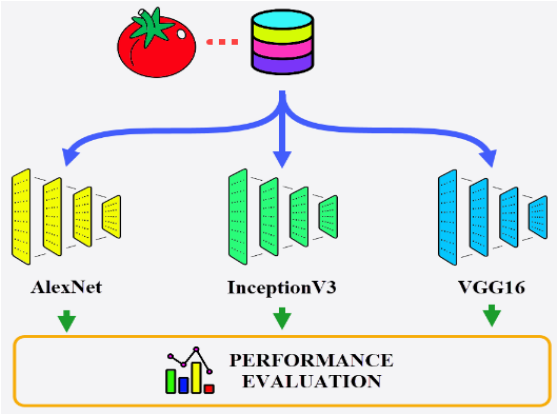


Figure 3
Tomato dataset classification process

Table 3
Confusion matrix of AlexNet model

PREDICTED CLASS \ ACTUAL CLASS	Tomato 1	Tomato 2	Tomato 3	Tomato 4	Tomato Cherry Red	Tomato Heart	Tomato Maroon	Tomato Yellow	Tomato Not Ripened
Tomato 1	246	0	0	0	0	0	0	0	0
Tomato 2	0	225	0	0	0	0	0	0	0
Tomato 3	0	0	246	0	0	0	0	0	0
Tomato 4	0	0	0	160	0	0	0	0	0
Tomato Cherry Red	0	0	0	0	164	0	0	0	0
Tomato Heart	0	0	0	0	0	228	0	0	0
Tomato Maroon	0	0	0	0	0	0	127	0	0
Tomato Yellow	0	0	0	0	0	0	0	153	0
Tomato Not Ripened	0	0	0	0	0	0	0	0	158

As a result of training and testing the images given as input to the AlexNet model, the confusion matrix in Table 3 was obtained. The confusion matrix of the InceptionV3 model is given in Table 4, and the confusion matrix of the VGG16 model is given in Table 5.

Table 4
Confusion matrix of InceptionV3 model

PREDICTED CLASS \ ACTUAL CLASS	Tomato 1	Tomato 2	Tomato 3	Tomato 4	Tomato Cherry Red	Tomato Heart	Tomato Maroon	Tomato Yellow	Tomato Not Ripened
Tomato 1	242	0	0	0	0	1	0	0	0
Tomato 2	0	225	0	0	0	0	0	0	0
Tomato 3	0	0	246	0	0	21	0	0	0
Tomato 4	0	0	0	160	0	0	0	0	0
Tomato Cherry Red	0	0	0	0	164	0	0	0	0
Tomato Heart	0	0	0	0	0	206	0	0	0
Tomato Maroon	0	0	0	0	0	0	127	0	0
Tomato Yellow	0	0	0	0	0	0	0	153	0
Tomato Not Ripened	0	0	0	0	0	1	0	0	157

Table 5
Confusion matrix of VGG16 model

PREDICTED CLASS \ ACTUAL CLASS	Tomato 1	Tomato 2	Tomato 3	Tomato 4	Tomato Cherry Red	Tomato Heart	Tomato Maroon	Tomato Yellow	Tomato Not Ripened
Tomato 1	246	0	2	0	0	0	0	0	0
Tomato 2	0	225	0	0	0	12	0	0	0
Tomato 3	0	0	244	0	0	35	0	0	0
Tomato 4	0	0	0	160	0	7	0	0	0
Tomato Cherry Red	0	0	0	0	164	1	0	0	0
Tomato Heart	0	0	0	0	0	170	0	0	0
Tomato Maroon	0	0	0	0	0	0	127	0	0
Tomato Yellow	0	0	0	0	0	3	0	153	6
Tomato Not Ripened	0	0	0	0	0	0	0	0	152

According to the confusion matrix data obtained from the AlexNet model given in Table 3, FP, FN and TP values in all classes are zero. All classes have been classified with 100% accuracy. In the confusion matrix of the InceptionV3 model given in Table 4, the FP value of the Tomato 1 class is 1, the FP value of the Tomato 3 class is 21 and the FP value of the Tomato Not Ripened class is 1. The FN value of the Tomato Heart class is 24. According to these data, Tomato 3 and Tomato Heart are classes that are confused with each other by the model. In the confusion matrix of the VGG16 model given in Table 5, the FP value of Tomato 1 class is 2, Tomato 2 class FP value is 12, Tomato 3 class FP value is 35, Tomato 4 class FP value is 7, Tomato Cherry Red class FP value is 1 and Tomato Yellow FP value is 9. The high FN value of Rn belongs to the Tomato Heart class and is 58. The most confused class in InceptionV3 and VGG16 models are Tomato Heart and Tomato 3 classes. Performance metrics calculated using confusion matrix data of all models are shown in Table 6.

Table 5
Performance metrics of AlexNet, InceptionV3 and VGG16 models

	Accuracy (%)	F1 Score	Precision	Recall	Specificity
AlexNet	100	1	1	1	1
InceptionV3	98.4	0.984	0.985	0.984	0.997
VGG16	96.1	0.96	0.965	0.961	0.994

When the data in Table 6 is examined, it is seen that the model with the highest classification accuracy is AlexNet. It is seen that the model with the lowest classification accuracy is the VGG16 model. Accuracy metric values show parallelism with other metric values. Although the model with the lowest depth was Alexnet, the highest classification accuracy was obtained from this model. In Figure 4, the column chart of the classification accuracy of all models is shown in Figure 4.

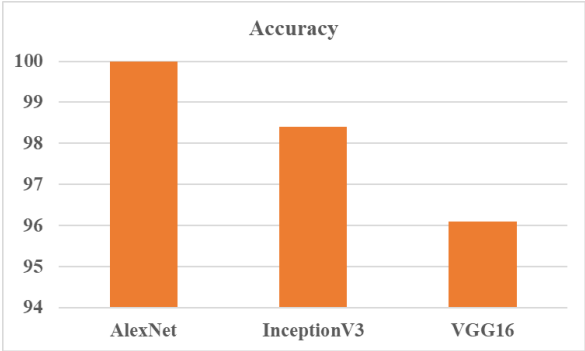


Figure 4
Comparison of classification accuracy of models

According to the graph in Figure 4, the AlexNet model with the smallest model size showed the highest classification accuracy. VGG16 with the highest model size showed the lowest classification accuracy. Image

4. Conclusions

Three different values were obtained as a result of the classification of images of nine different tomato species with AlexNet, InceptionV3 and VGG16 models. AlexNet model achieved 100% classification accuracy, InceptionV3 model 98.4% and VGG16 model 96.1% classification accuracy. In measuring the performance of the models, the dataset was divided into train-test at a rate of 75%-25%. It has been determined that the number of images in the dataset is sufficient for training and testing the models. Although the AlexNet model has the smallest size, it has achieved the highest classification accuracy. Although the VGG16 model has the largest size, it has the lowest classification accuracy. From these results, it has been seen that models with high depth and complexity cannot achieve high accuracy in every dataset. This situation may vary according to the datasets. For this reason, detailed analyzes are required in image classification problems regardless of the size and depth of the models. This also applies to the training and testing times of the models.

The usability of the proposed models in the classification of tomatoes is high. Tomato types can be separated from each other by image processing in automatic sorting machines. In addition, more detailed classification analyzes can be made by increasing the number of tomato species and creating new datasets. With all these developments, tomatoes will be able to be sorted non-destructively and quickly.

5. Acknowledgements

This project was supported by the Scientific Research Coordinator of Selcuk University with the project number 22111002. No funding was received for this study.

6. References

Al-Doori SKS, Taspinar YS, Koklu M (2021). Distracted Driving Detection with Machine Learning Methods by CNN Based Feature Extraction. *International Journal of Applied Mathematics Electronics and Computers*, *9*(4), 116-121.

Al Ohali Y (2011). Computer vision based date fruit grading system: Design and implementation. *Journal of King Saud University-Computer and Information Sciences*, *23*(1), 29-36.

Arakeri MP (2016). Computer vision based fruit grading system for quality evaluation of tomato in agriculture industry. *Procedia Computer Science*, *79*, 426-433.

Cinar I, Koklu M (2022). Identification of Rice Varieties Using Machine Learning Algorithms. *Journal of Agricultural Sciences*, 9-9.

Elhariri E, El-Bendary N, Fouad MMM, Platoš J, Hassanien AE, Hussein AM (2014). Multi-class SVM based classification approach for tomato ripeness. In *Innovations in Bio-inspired Computing and Applications* (pp. 175-186): Springer.

Jhawar J (2016). Orange sorting by applying pattern recognition on colour image. *Procedia Computer Science*, *78*, 691-697.

Kishore B, Yasar A, Taspinar YS, Kursun R, Cinar I, Shankar VG, Koklu M, Ofori I (2022). Computer-Aided Multiclass Classification of Corn from Corn Images Integrating Deep Feature Extraction. *Computational Intelligence and Neuroscience*, *2022*.

Koklu M, Kursun R, Taspinar YS, Cinar I (2021). Classification of date fruits into genetic varieties using image analysis. *Mathematical Problems in Engineering*, *2021*.

Mim FS, Galib SM, Hasan MF, Jerin SA (2018). Automatic detection of mango ripening stages–An application of information technology to botany. *Scientia horticulturae*, *237*, 156-163.

Moallem P, Serajoddin A, Pourghassem H (2017). Computer vision-based apple grading for golden delicious apples based on surface features. *Information processing in agriculture*, *4*(1), 33-40.

Muhammad G (2015). Date fruits classification using texture descriptors and shape-size features. *Engineering Applications of Artificial Intelligence*, *37*, 361-367.

Mureşan H, Oltean M (2017). Fruit recognition from images using deep learning. *arXiv preprint arXiv:1712.00580*.

Oo LM, Aung NZ (2018). A simple and efficient method for automatic strawberry shape and size estimation

and classification. *Biosystems engineering*, *170*, 96-107.

Ramos P, Prieto FA, Montoya E, Oliveros CE (2017). Automatic fruit count on coffee branches using computer vision. *Computers and Electronics in Agriculture*, *137*, 9-22.

Singh D, Taspinar YS, Kursun R, Cinar I, Koklu M, Ozkan IA, Lee HN (2022). Classification and Analysis of Pistachio Species with Pre-Trained Deep Learning Models. *Electronics*, *11*(7), 981.

Sofu MM, Er O, Kayacan M, Cetişli B (2016). Design of an automatic apple sorting system using machine vision. *Computers and Electronics in Agriculture*, *127*, 395-405.

Taspinar YS, Cinar I, Koklu M (2021). Prediction of Computer Type Using Benchmark Scores of Hardware Units. *Selcuk University Journal of Engineering Sciences*, *20*(1), 11-17.

Taspinar YS, Dogan M, Cinar I, Kursun R, Ozkan IA, Koklu M (2022). Computer vision classification of dry beans (Phaseolus vulgaris L.) based on deep transfer learning techniques. *European Food Research and Technology*, *248*(11), 2707-2725.

Taspinar YS, Koklu M, Altin M (2021). Classification of flame extinction based on acoustic oscillations using artificial intelligence methods. *Case Studies in Thermal Engineering*, *28*, 101561.

Tomato Dataset. Retrieved from https://github.com/Horea94/Fruit-Images-Dataset

Unal Y, Taspinar YS, Cinar I, Kursun R, Koklu M (2022). Application of Pre-Trained Deep Convolutional Neural Networks for Coffee Beans Species Detection. *Food Analytical Methods*, *15*(12), 3232-3243.

Wan P, Toudeshki A, Tan H, Ehsani R (2018). A methodology for fresh tomato maturity detection using computer vision. *Computers and electronics in agriculture*, *146*, 43-50.

Wang F, Zheng J, Tian X, Wang J, Niu L, Feng W (2018). An automatic sorting system for fresh white button mushrooms based on image processing. *Computers and electronics in agriculture*, *151*, 416-425.

Zhang Y, Wang S, Ji G, Phillips P (2014). Fruit classification using computer vision and feedforward neural network. *Journal of Food Engineering*, *143*, 167-177.

Selcuk Journal of Agriculture and Food Sciences SJAFS

http://sjafs.selcuk.edu.tr/sjafs/index

(2022) 36 (3), 521-526

e-ISSN: 2458-8377

DOI:10.15316/SJAFS.2022.068

Research Article

A Research on Food Safety in Turkey after the Covid-19 Pandemic

Günel MİKAİLSOY

Sakarya University, Institute of Business Administration, Department of Production Management and Marketing, Sakarya, Türkiye

ARTICLE INFO

Article history:
Received date: 17.11.2022
Accepted date: 20.12.2022

Keywords:
Consumer preferences
Food safety
Knowledge
Non-parametric tests

ABSTRACT

The aim of the study is to investigate the food safety perception of consumers during and after the covid 19 pandemic. Surveys were administered to a total of 1000 people from all regions of Turkey via the internet. The survey data were defined as frequency and percentage, and analyzed by nonparametric test methods for dependent and independent groups. As a result of the analysis, it has been determined that the knowledge and awareness levels of consumers about food safety are generally high, and that there are significant differences according to education, age and region. It was observed that the participants did not differ much between their food preferences before and during COVID-19, but they were more careful about health and packaging safety. In other words, the anxiety level of the participants increased during COVID-19. In addition, it was found that with the increasing of education level and age, the awareness and knowledge of the food preferences of the participants also increased. As another factor affecting consumer behavior, the result of the anxiety level measurement that can be evaluated within the scope of perceived risk, it was determined that 76.4% of the participants would prefer more packaged food products during and during the pandemic period. The results of the study support that consumers' preferences in situations of uncertainty are shaped according to their risk perceptions and there is an increase in their anxiety levels.

1. Introduction

According to the research conducted by Euromonitor, a worldwide strategic market research organization, in the post-coronavirus pandemic period, there has been a faster than expected change in consumer trends. Among the trends that have been transformed to suit multiple purposes, the virus is in the focus of consumers, and this is the source of their fears (BBC, 2020a). So, what are the effects of this process when consumers are shopping for food of animal origin? At this point, what kind of role does the consumer awareness level play in the choice of food products of animal origin? For example, although there are various viruses transmitted from food products of animal origin, no such findings about coronavirus have yet been encountered. However, in one state in the USA, most of the meat factories and slaughterhouses have been closed due to an pandemic. Because some of the workers working here have tested positive for COVID-19 (BBC, 2020b).

In recent years, due to the increased sensitivity of consumers to food production and consumption, food safety, which is one of the determining factors in the food products demand is always on the agenda and has become more important in the period during the COVID–19 pandemic. Consumers' food shopping preferences have always been affected by crises, necessarily or arbitrarily. Consumer behavior affected by the external environment has a great place in perceptions from meeting basic needs to shopping preferences. The effect of environmental factors may vary depending on the purpose of purchasing, the internal state of the consumer and other situational factors (Odabaşı and Barış, 2002; Güzel et al, 2018). Due to all these factors, depending on the level of education and culture, and with their rise the consumer wants to know what is in the food products they eat, and it is seen that the consumer cares about food safety (Özmetin, 2006). The concept of conscious consumer is increasing in importance day by day within the scope of consumer behavior. With the emergence of the conscious consumer concept over time, there have been changes in the purchasing behavior of the consumer and as a result, there have been differences in product preferences (Gülse et al, 2019). The level of awareness of consumers is directly proportional to their level of protection against the dangers they may encoun-

* Corresponding author email: *gunel.mikailsoy@ogr.sakarya.edu.tr*

ter. The conscious consumer takes precautions beforehand in his shopping preferences and is deemed to have protected himself from a number of adverse situations that pose a risk. Consumer behavior is affected by the reactions of the consumer to environmental factors in the context of the events that the consumer experiences while purchasing the product or service (İslamoğlu and Altunışık, 2017). Today, the behavior of the consumer of food products is on a line of balance and rationality. This situation is based on inherited or acquired traditions and food habits, which have effects on meeting the nutritional needs of the individual's body, but without neglecting modernity and innovation specific to the nutritional field (Petroman et al, 2015). As the perspectives and perceptions of different social classes vary in consumer behavior, it is normal for their preferences to also differ. Consumer behavior resulting from the relative effect of sociological factors on psychological factors (Güzel et al, 2018; Karabulut, 1981) health concerns of individuals and increased health awareness shape their lifestyles and food preferences (Dutta-Bergman, 2004; Bakke et al, 2016; Pakmak and Koçoğlu, 2019).

In this study, it was aimed to investigate the effect of the COVID–19 pandemic on the food consumption of consumers, which can be evaluated in the context of a situation factor affecting the whole world.

2. Materials and Methods

Material

Participants are grouped based on their native provinces data by the seven geographical regions of Turkey. The survey questions are designed based on the literature and consist of two parts. These are dimensioned in accordance with demographic data and the purpose of the research as awareness and knowledge and anxiety levels.

Method

The simple random sampling formula Eq.(1) was used in the study to estimate the sample size (Yamane, 1967). In general, according to this formula, when the population size exceeds 100 000, with $\alpha = 0.05$ significance level, $p = 0.5$ probability and $d = \pm 5\%$ margin of error, the sample number does not exceed 400. The fact that the number of surveys made is 2.5 times this number increases the power of the sampling.

$$n = \frac{Nz^2 pq}{(N-1)d^2 + z^2 pq} \quad (1)$$

In addition to the descriptive statistics of the data, the nonparametric Wilcoxon sign test for the analysis of the difference between the two dependent variables, the Mann Whitney U test to determine the difference between two independent groups, and the Kruskal Wallis H-test to compare more than two independent groups were used in the study. Spearman Rank - Order correlation was used to determine the relationship between some features (Cebeci, 2019).

3. Results and Discussion

The demographic information of the participants in the study is included in Table 1.

Table 1
Frequency table of some demographic information of the participants

Demographic Characteristics		n	%
Gender	Female	553	55.3
	Male	447	44.7
Education	Pimary school	26	2.6
	Middle school	36	3.6
	High school	179	17.9
	University	759	75.9
Age	Under 20	70	7.0
	21-30	364	36.4
	31-40	381	38.1
	41-50	141	14.1
	51-60	32	3.2
	Over 61	12	1.2
Hometown (as Regions)	Mediterranean	123	12.3
	Eastern Anatolia	80	8.0
	Aegean	58	5.8
	Southeastern Anatolia	452	45.2
	Central Anatolia	141	14.1
	Black Sea	78	7.8
	Marmara	123	12.3
Marital status	Married	584	58.4
	Single	416	41.6
Average monthly income	Less than 2500 TL	266	26.6
	Between 2500–5000 TL	395	39.5
	More than 5000 TL	339	33.9
Average monthly food expenditure	Less than 600 TL	152	15.2
	Between 600–1000 TL	269	26.9
	Between 1000–1500 TL	241	24.1
	More than 1500TL	338	33.8

As seen in Table 1, 55.3% of the participants are women and 44.7% are men. 58.4% of them are married and 41.6% are single. 2.6% of the participants' education level is primary school, 3.6% secondary school, 17.9% high school and 75.9% university graduates. Considering the age of the participants, 7% are under 20 years old, 36.4% are 21-30, 38.1% are 31-40, 14.1% are 41-50, 3.2% are 51-60 years old and 1.2%. is over 61 years old. The distribution of the respondents according to regions is as follows: Mediterranean Region 12.3%, Eastern Anatolia Region 8%, Aegean Region 5.8%, Southeastern Anatolia Region 45.2%, Central Anatolia Region 14.1%, Black Sea Region 7.8% and Marmara Region 12.3%. It has been determined that 26.6% of the participants' monthly income is less than 2500 TL, 39.5% is between 2500-5000 TL and 33.9% is more than 5000 TL. It is seen that 15.2% of participants' average monthly food expenditure is less than 600 TL, 26.9% is between 600-1000 TL, 24.1% is between 1000-1500 TL and 33.8% is more than 1500 TL.

Knowledge level

Consumer awareness is based on information and when evaluated from a marketing point of view, it can be said that consumers who have a high level of knowledge and awareness of their legal rights regarding products and producers will positively affect the dynamics of the market as conscious consumers (Pakmak and

Koçoğlu, 2019). In this context, the answers to the questions asked about the knowledge levels of the participants are included in Table 2.

Table 2
Frequency and percentages about the knowledge and awareness level of the participants

N	Questions	Yes		No	
		n	%	n	%
Knowledge level					
1.	When you encounter a spoiled or faulty food product, do you report your complaint to the place where you bought it?	833	83.3	167	16.7
2.	Do you pay attention that the food products you buy are approved by the Ministry of Agriculture?	653	65.3	347	34.7
Level of Consciousness					
3.	Have you heard of the concept of food safety before?	766	76.6	234	23.4
4.	I have information about Alo 174 Food Line	547	54.7	453	45.3
5.	I have information about Alo 175 Consumer Line	644	64.4	356	35.6
6.	When buying a food product, I always look at the expiration date.	922	92.2	78	7.8

Food safety for the purpose of ensuring full consumer safety should pass through the stage of protecting the products from biological, chemical and physical hazards in the process until they reach the end user, the consumer. According to this, it is seen that 76.6% of the participants have heard about the concept of food safety before, 54.7% have information about the Alo 174 Food Line and 64.4% have information about the Alo 175 Consumer Line. It has been determined that 65.3% of the participants pay attention to the fact that the purchased food products are approved by the Ministry of Agriculture, and 83.3% of the participants complain to the place where the product was purchased when a defective or faulty food product was encountered (Table 2). When the table above is examined, it is seen that the rate of reading the expiry date is the highest (92.2%), and the level of knowledge and awareness about food safety and consumer rights is followed by high rates. In another study, it was determined that 68.6% of consumers in Tokat province heard the concept of food safety and defined it correctly (Onurlubaş and Gürler, 2016). Taşdan et al. (2014) found the rate of knowledge of food safety to be 80% in their study. In the study conducted by Yalçın and Kızılaslan (2013) in Samsun city center, this rate was found to be 84%. According to the report published by the Ministry of Commerce in 2018 (Güzel et al, 2018), it was determined that the rates of consumers with high level of knowledge (36%) and medium level of knowledge (35.5%) are close to each other. The rate of consumers with low level of knowledge is 28.5%. In the study conducted by Yuzbaşıoğlu et al. (2018), it was stated that the food safety inspection of 69.7% of the participants in Sivas province should be done by the Ministry of Agriculture and Forestry. Similarly, in the study conducted by Mutlu (2007) in Adana, consumers stated the Ministry of Health (35.5%) and the Ministry of Agriculture and Forestry (22.7%) as the institution or organization conducting food safety inspection in the context of food safety knowledge level. In a study conducted in Denizli, it was determined that the participants did not have sufficient information about consumer rights (Pakmak and Koçoğlu, 2019). In the study conducted by Tüyben (2018), it was determined that 79.0% of individuals with the habit of reading food labels pay attention to the information of ingredients ($p<0.05$), and the rate of those who do not have information within the scope of the consumer line was found to be 23.5%.

It is observed that the rate of the participants who sometimes read the information on the packaging in the food they buy with labels is 38.5%, and the rate of those who do not read at all is 1.2% (Figure 1).

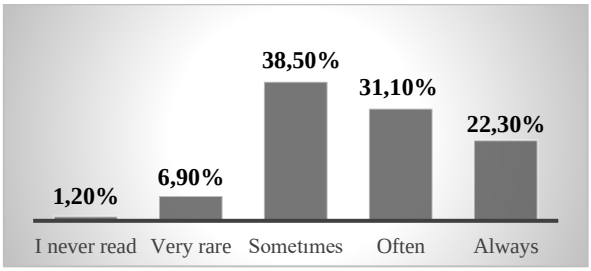


Figure 1
The percent of the participants who read the information on the packaging for the food they buy with labels

When evaluating in general, it can be said that the rate of reading the information on the packaging is high. When looking at the field information, it is seen that consumers generally read the information on the food products they buy packaged. Similarly, in Ogur's (2020) study with consumers in Bitlis, this rate is 64%. Güzel et al. (2018) stated in a research report that most of the participants generally read the label information when purchasing food products, but 8.7% of the participants never read it. Another study was conducted by Topuzoğlu et al. (2007) to examine the knowledge and attitudes of people who applied to a health center about purchasing food products and the rate of reading the label information was observed as 52.1%. In general, the rate of reading tags in our study coincides with the rates of other studies. In the study of Babaoğul et al. (2016), the rate of tag reading was always 20.5% and never 30.5%. In the research conducted by Çınar and Sağlık (2006), it was determined that 75% of the consumers partially read the label information and 25% never read it. These examples differ with our study in terms of not reading labels.

According to Duesenberry's theory of relative income, families influence each other in terms of consumption. The consumption of the individual within the family or damily as whole depends on the relative income of the families with which this family has a relationship. In addition, consumption habits that vary according to income and consumption activities depending on the cultural and social characteristics of the families also affect the country's economy (Özsungur and Güven, 2017). In this respect, the consciousness level scale was used together with the questions (1, 2, 3) measuring the knowledge level of the participants in our study. According to this scale:

Consciousness level score: Up to 8 "Weak",

Consciousness level score: Those between9-10 are "Medium",

Consciousness level score: Those who are 11 and above are classified as "High" information.

The conscious consumer aims to get the maximum benefit from it while purchasing a good or service. While purchasing, it is consciously organized, taking into account its real needs, shopping with plan and documentation, while taking into account the quality, standard, healthy, safe, environmentalist features of the product/service, it is a socio-economic element that attaches importance to saving by choosing the most suitable product for its budget and has the feature of directing the economy to efficiency over time (Topuzoğlu et al, 2007). The inability of consumers to know the organizations that control food production is an obstacle for them to play the conscious consumer role in this sector and to control it. Eryılmaz and Kılıç (2020) in their study had found that consumers with a high level of education and income level are more conscious about the food safety matter. In European Union countries, where the standard of living and social awareness is higher, there is no linear relationship between the education and income levels of consumers and their food safety awareness. In our study, statistically significant results were found as a result of the Spearman Sequential correlation analysis performed between the educational status, average monthly income and level of consciousness of the participants (Table 3).

According to the analysis results, a positive correlation was found between the level of consciousness and age, education level and average monthly income ($p<0.01$). In other words, as the age, education level and average monthly income increase, the awareness level of the consumers also increases. In the study of Erdoğan and Çiçek (2015), it was determined that household income contributes positively to the consumption level of some foods of animal origin. In the statistical analysis, positive correlations were found between the increase in income and consumption of drinking milk, yoghurt, beef-veal, fish meat, eggs and honey. Education level and income were observed to be effective in food preferences of animal origin. Demanding white meat more than red meat, keeping criteria such as production and expiration date, brand, packaging, and fat ratio in purchased products can be shown as evidence for this effect.

Tablo 3
Correlation coefficients between the education level, average monthly income and level of awareness of the participants

	Age	Education level	Average monthly income
Education level	0.041	-	-
Avarage monthly income	0.397**	0.302**	-
Consciousness level	0.141**	0.156**	0.148**

**:p<0.01

Significant results were found as a result of the statistical testing of the difference between knowledge and awareness levels according to socio-demographic characteristics (Table 4-5).

Table 4
Mann-Whitney U test results to determine the difference between the knowledge and awareness levels of the participants according to sex and marital status

N	Questions	Sex	Marital status
		Z value	
Knowledge level			
1.	Do you pay attention that the food products you buy are approved by the Ministry of Agriculture?	-4.793**	-2.377*
2.	When you encounter a spoiled or faulty food product, do you report your complaint to the place where you bought it?	-2.105*	-1.982*
Consciousness level			
3.	Have you heard of the concept of food safety before?	-0.39	-1.765
4.	I have information about Alo 174 Food Line	-3.639**	-2.261*
5.	I have information about Alo 175 Consumer Line	-1.576	-0.281
6.	Do you read the information on the packaging of the food you buy with labels?	-3.278**	-2.591*
7.	When buying a food product, I always look at the expiration date.	-4.299**	-1.328

*:p<0.05; **:p<0.01

Table 5
Kruskal Wallis test results for determining the difference between the consciousness levels of the participants according to the regions, age and education level

NQuestions	Regions	Age	Education level
	χ^2 value		
Knowledge level			
1. Do you pay attention that the food products you buy are approved by the Ministry of Agriculture?	4.454	12.226*	7.947
2. When you encounter a spoiled or faulty food product, do you report your complaint to the place where you bought it?	2.967	9.814	23.222**
Level of Consciousness			
3. Have you heard of the concept of food safety before?	13.481*	29.919**	56.436**
4. I have information about Alo 174 Food Line	28.882**	29.281**	4.908
5. I have information about Alo 175 Consumer Line	3.354	11.383*	18.955**
6. Do you read the information on the packaging of the food you buy with labels?	7.601	16.440**	29.920**
7. When buying a food product, I always look at the expiration date.	6.087	51.965**	24.367**

*:p<0.05; **:p<0.01

That is to say, the level of consciousness about the food line differs significantly by gender ($p<0.05$) and

marital status ($p<0.05$). We see that there is a statistically significant ($p<0.01$) difference between men and women in paying attention to the expiration date (Table 4). In different studies (Losasso et al., 2012; Eryılmaz & Kılıç, 2020), it has been determined that women are more concerned and more conscious about food safety than men. We see that there are important differences related to the knowledge-awareness level, according to the regions, age and education level (Table 5). That is to say, while there is a statistically significant difference in terms of only the concept of food safety and having knowledge about the food line, there are statistical differences in many concepts according to age and education level.

Anxiety level

Perceived risk, which is one of the factors affecting the purchasing behavior of consumers, causes an increase in the level of anxiety in an environment of uncertainty. Perceived risk is defined as the degree of uncertainty that consumers feel before and after purchasing a product or service. In other words, consumers' evaluations about products, selection decisions and purchasing behaviors will be affected by the uncertainty (Campbell and Goodstein, 2001; Deniz and Erciş, 2008).

Numerical data determining the anxiety level of the participants about food consumption in the COVID–19 process are given in Table 6.

Table 6
Descriptive statistics on anxiety level

Questions	Yes		No	
	n	%	n	%
When consuming food products, I now prefer packaged products more	764	76.4	236	23.6
I started to consume more organic agricultural products	698	69.8	302	30.2

76.4% of the participants stated that they preferred packaged products more in this period, 69.8% started to consume more organic products.

Richardson et al. (1994) conducted a survey by sending random letters to people in England to determine the red meat consumption habits, in their study. As a result of the findings obtained from the analyzes, 28.3% of the consumers reported that they reduced their meat consumption, the main reason for this was health concerns.

Significant differences were found between the anxiety levels of the participants by region, age, gender, education level and marital status (Table 7). We see that age and education level are very effective in these differences.

Table 7
Kruskal Wallis and Mann-Whitney U test results to determine the difference between the anxiety levels of the participants by demographic characteristics

N	Question	Regions	Age	Educational level	Gender	Marital status
		χ^2 value			Z value	
1.	Has there been any change in your life in terms of food consumption in general during the COVID-19 pandemic?	6.688	12.955*	4.904	-1.465	-1.804
2.	When consuming food products, I prefer packaged products now	9.747	15.301**	4.014	-0.375	-1.688
3.	I started to consume more organic agricultural products	12.476	7.993	10.938*	-3.878**	-1.727
4.	I'm more worried now while consuming	8.354	22.799**	13.179*	-1.583	-2.948**

*:$p<0.05$; **:$p<0.01$

3. Results and Discussion

In this study, before and during COVID-19 pandemic food consuming habits of people living is 7 regions of Turkey were examined and it was investigated how this findings change depending on such demographic characteristics as people's gender, age, education, marital status and average monthly income, as well as knowledge and anxiety level. In our study, the knowledge level of the participants showed statistically significant differences according to gender and marital status. The level of knowledge also varied by region, age and education level. Since information is the basis of consumer awareness, those who are conscious consumers affect the dynamics of the market positively, and in the study, it was found that the majority are those who look at the expiration date while purchasing a food product. When faced with a spoiled and faulty food product, it was determined that the majority of consumers complained to where they bought them. There was a difference between the anxiety levels of the participants and regions, age, education level, gender and marital status. It has been observed that the percentage of the responses of the participants that they will prefer more packaged and organic products during the COVID-19 period. Finally, it is recommended people to be conscious consumers and that necessary studies should be carried out to reduce their anxiety in any pandemic.

4. References

Babaoğul M, Şener A, Buğday EB (2016). Tüketici profili ve bilinç düzeyi araştırması. T.C. Gümrük ve Ticaret Bakanlığı Tüketici Akademisi Projesi.

Bakke A J, Shehan CV, Hayes JE (2016). Type of milk typically consumed, and stated preference, but not

health consciousness affect revealed preferences for fat in milk. *Food Quality and Preference*, 49: 92-99.

BBC (2020a). How is the coronavirus epidemic changing consumer habits? https://www.bbc.com/turkce/haberler-dunya-52448919 Accessed: 2020.

BBC (2020b). Coronavirus: There is a shortage of meat in the USA due to the epdemic.https://www.bbc.com/turkce/haberler-dunya-52557247 Accessed: 2020.

Campbell M, Goodstein R (2001). The moderating effect of perceived risk on consumers' evaluations of product incongruity: preference for the norm. *Journal of Consumer Research*, 28 (12): 439–449.

Cebeci Z (2019). R ile parametrik olmayan istatistik analiz. Abaküs Kitap Yayıh Evi. İstanbul(in Turkish)

Çınar R, Sağlık E (2006). Perakende gıda ürünlerinde etiketin önemi, tüketiciler üzerindeki etkileri ve Erzurum ölçeğinde bir alan araştırması. *Pazarlama Dünyası Dergisi*, 1-10.(in Turkish)

Deniz A, Erciş A (2008). Kişilik özellikleri ile algılanan risk arasındaki ilişkilerin incelenmesi üzerine bir araştırma. *İktisadi ve İdari Bilimler Dergisi*, 22(2): 301-330.(in Turkish)

Dutta-Bergman, MJ (2004). Primary sources of health information: comparisons in the domain of health attitudes, health cognitions, and health behaviors. *Health Communication*, 16(3): 273–288.

Erdoğan N, Çiçek H (2015). Hayvansal gıdaların tüketim yapısı ve tüketici tercihlerinin belirlenmesi üzerine bir araştırma: Afyon Kocatepe Üniversite personeli örneği. *Kocatepe Veteriner Dergisi*, 8(1): 25-31.

Eryılmaz AG, Kılıç O (2020). Türkiye ve Avrupa Birliği'nde tüketicilerin gıda güvenliğiyle ilgili tutum ve davranışları. *Food and Health*, 6(1): 57-66.

Gülse Bal HS, Kaplan E, Kaplan K (2019). Yaş meyve ve sebze tüketiminde gıda güvenliği algısı, tüketici endişesi ve bilinci: Tokat ili Merkez ilçe örneği. *III. International Eurasian Agriculture and Natural Sciences Congress*, 96-104.

Güzel Y, İpek G, Yıldırım Yılmaz T, Babaoğul M, Şener A, Buğday EB, Yücel S (2018). Tüketici profili ve bilinç düzeyi araştirma raporu. Türkiye Tüketicinin Korunması ve Piyasa Gözetimi Genel Müdürlüğü. Türkiye Cumhuriyeti Ticaret Bakanlığı. Ankara.

İslamoğlu AH, Altunışık R (2017). Tüketici Davranışları. Beta Basım, İstanbul.

Karabulut M (1981). Tüketici davranışları yeniliklerin kabülü ve yayılışı, Minnetoğlu Yayınları, İstanbul.

Losasso C, Cibin V, Cappa V, Roccato A, Vanzo A, Andrighetto I, Ricci A (2012). Food safety and nutrition: Improving consumer behaviour. *Food Control*, 26: 252–258.

Mutlu S (2007). Gıda güvenirliği açısından tüketici davranışları (Adana kentsel kesimde kırmızı et tüketimi örneği). Doktora tezi. Çukurova Üniversitesi Fen Bilimleri Enstitüsü, Adana.

Odabaşı Y, Barış G (2002). Tüketici davranışı. 2. Basım İstanbul: Mediacat.

Oğur S (2020). Bitlis'teki tüketicilerin gıda güvenliği ve gıda hijyeni konusundaki bilgi ve tutumları. *Bitlis Eren Üniversitesi Fen Bilimleri Dergisi*, 9(2): 780-796.

Onurlubaş E, Gürler AZ (2016). Gıda güvenliği konusunda tüketicilerin bilinç düzeyini etkileyen faktörler. *Journal of Agricultural Faculty of Gazi-osmanpasa University*, 33 (1): 132-141.

Özmetin S (2006). Gıda tüketim alışkanlıklarındaki değişim üzerine bir araştırma. Yüksek Lisans Tezi. Sakarya Üniversitesi, Sosyal Bilimler Enstitüsü, İşletme Anabilim Dalı, Sakarya.

Özsungur F, Güven S (2017). Tüketici davranışlarını etkileyen sosyal faktörler ve aile. *International Journal of Eurasian Education and Culture*, 3: 127-142.

Pakmak AS, Koçoğlu D (2019). Tüketici hakları farkındalığı ve tüketici bilinç düzeyi: Denizli örneği. *Pamukkale Journal of Eurasian Socioeconomic Studies*, 6(1): 37-58.

Petroman C, Bidireac IC, Petroman I, Sucan M, Marin D, Turc B, Merce I, Constantin EC (2015). The impact of education on the behaviour of the consumer of animal origin food products. Procedia – *Social and Behavioral Sciences*, 190: 429-43.

Richardson N, Macfie H, Sepherd R (1994). Consumer attitudes to meat eating. *Meat Science*, 36(1-2): 57-65.

Taşdan K, Albayrak M, Gürer B, Özer OO, Albayrak K, Güldal HT (2014). Geleneksel gıdalarda tüketicilerin gıda güvenliği algısı: Ankara ili örneği. *Uluslararası Davraz Sempozyumu*, Süleyman Demirel Üniversitesi, Isparta.

Topuzoğlu A, Hıdıroğlu S, Ay P, Önsüz F, İkiışık H (2007). Tüketicilerin gıda ürünleri ile ilgili bilgi düzeyleri ve sağlık risklerine karşı tutumları. TSK *Koruyucu Hekimlik Bülteni*, 6(4): 253-258.

Tüyben EE (2018). Tüketicilerin gıda güvenilirliği yönünden etiket okuma alışkanlığı ve algısının değerlendirilmesi. Y. Lisans tezi, Hacettepe Üniversitesi, Ankara.

Yalçın A, Kızılaslan H (2013). Tüketicilerin gıda güvenliği ile ilgili tutum ve davranışları (Samsun ili örneği). Tarımsal Ekonomi ve Politika Geliştirme Enstitüsü, 213.

Yamane T (1967). Statistics, an introductory analysis, 2nd Ed., New York: Harper and Row.

Yüzbaşıoğlu R, Kızılaslan H, Kızılaslan N, Kurt E (2018). Gıda güvenilirliği açısından tüketici davranışlarının incelenmesi: Sivas kentsel ve kırsal kesimde kırmızı et tüketimi. *Gaziosmanpaşa Bilimsel Araştırma Dergisi,* 7(3): 49-60.

Selcuk Journal of Agriculture and Food Sciences SJAFS

http://sjafs.selcuk.edu.tr/sjafs/index

(2022) 36 (3), 527

e-ISSN: 2458-8377

Erratum

Erratum to: The Effects of Iron Application Foliarly at Different Times and Amounts on Agricultural Characteristics in Some Peanut Varieties

Nursel ÇÖL KESKİN[1,*], Gülsüm KAYIŞOĞLU[2]

[1]Selcuk University, Faculty of Agriculture, Department of Field Crops, Konya, Türkiye

[2]Selcuk University, The Graduate School of Natural and Applied Science, Department of Plant Breeding and Genetic, Konya, Türkiye

ERRATUM INFO

Erratum to: SJAFS
(2022) 36 (2), 197-202
e-ISSN: 2458-8377
DOI:10.15316/SJAFS.2022.025

Erratum history:
Received date: 18.11.2022
Accepted date: 26.12.2022

ERRATUM

Published version:

It was conducted at doses: control, 400 kg da^{-1}, 500 kg da^{-1}, 600 kg da^{-1}.

Table 2 and Table 3 was typed D_0: kontrol, D_1: 400 kg da^{-1}, D_2: 500 kg da^{-1}, D_3: 600 kg da^{-1}.

The doses given in the summary were as follows: control, 400 kg da^{-1}, 500 kg da^{-1}, 600 kg da^{-1}.

The recommended dose in the summary and the conclusion section was as follows: 400 kg da^{-1}.

Reported corrected version:

It should be typed at doses: control, 400 g da^{-1}, 500 g da^{-1}, 600 g da^{-1}.

Table 2 and Table 3 should be typed D_0: control, D_1: 400 g da^{-1}, D_2: 500 g da^{-1}, D_3: 600 g da^{-1}.

The doses given in the summary should be as follows: control, 400 g da^{-1}, 500 g da^{-1}, 600 g da^{-1}.

The recommended dose in the summary and the conclusion section should be as follows: 400 g da^{-1}.

Acknowledgement We thank Ozden Ozturk for spotting the typographical error in the original paper

* Corresponding author email: *nurselcol@selcuk.edu.tr*; The online version of the original article can be found under doi: 10.15316/SJAFS.2022.025