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Turkish Journal of Field Crops is the official publication of Society of Field Crops Science. It offers the latest developments in research to scientists active in field crops. The Journal includes only the original research papers on the following topics: Breeding and Genetics, Agronomy, Physiology, Forage Crops, Medicinal and Aromatical Plants, Biotechnology and Utilization

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Tables, graphs, and other material should be arranged separately in the text. Word editor should be used to create tables, for tables each item should be placed into a separate cell. Dimensions of tables must be maximum 16 × 20 cm and minimum 8 cm wide. Tables are to be numbered with Arabic numerals in the order in which they are referred to in the text and have a brief, but self-explanatory title. Abbreviations or symbols used in the tables must be explained either in the table title or as a footnote.

Figures should be submitted in TIFF or JPG format and dimensions of figures must be maximum 16 × 20 cm and minimum 8 cm wide. All photos, graphs, illustrations and diagrams should be referred to as a figure and numbered, continually according to the order in which they are included in the text, using Arabic numerals.

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The main text of scientific paper must be developed under the following headings: Introduction, Materials and Methods, Results and Discussion. Results and Discussion can be presented under one or separate heading.

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MATERIALS and METHODS

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Berdahl, J.D., A.C. Wilton and A.B. Frank. 1989. Survival and agronomic performance of 25 alfalfa cultivars and strains interseeded into rangeland. J. Range Manage. 42:312-316.

Books: Steel, R.G.D. and J.H. Torrie. 1980. Principles and Procedures of Statistics: A Biometrical Approach. 2. ed. New York: McGraw-Hill Publ. Company.

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PHYSIOLOGICAL AND BIOCHEMICAL RESPONSES OF CHICKPEA (*Cicer arietinum* L.) GENOTYPES TO DIFFERENT MOISTURE STRESSES

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ABSTRACT

Moisture stress influence seed germination, growth including physiological, biochemical attributes and yield of chickpea (*Cicer arietinum* L.). Genotypes may vary in their capacity to tolerate moisture stress. Therefore, the study was undertaken to evaluate physiological and biochemical responses of selected chickpea genotypes in the drought prone ecosystems. Relative water content and carotenoids content significantly decreased when stress imposed until pod formation stage. Moisture stress imposed during pre-flowering stage significantly decreased chlorophyll a and chlorophyll b content. Proline accumulation was higher in BD-6048 compared to other genotypes under all moisture stress conditions. Phosphorus, potassium and protein content were lower under moisture stress until pod formation stage. Under moisture stress conditions the genotypes BD-6048 had the highest yield compared to other genotypes. Moisture stress until pre-flowering and pod formation stage reduced seed yield more severe than that on flowering stage.

Keywords: Chlorophyll content, field capacity, relative water content and seed yield

INTRODUCTION

Agriculture and water resource sectors are commonly affected by moisture stress. It may cause significant economic losses in the agriculture part of developed countries through diminutions in crop yield or total failure of crops (Toker and Mutlu, 2011; Sweet et al., 2017). It can also cause human migration and food crisis in developing countries under certain circumstances (Gray and Mueller, 2012; Grolle, 2015). In irrigated agricultural systems moisture stress has great impact on crop production (Vidal-Macua et al., 2018) and also troubles for urban water supply, manufacturing needs, decreases of hydropower production, etc. (Balling and Gober, 2007; Jerez et al., 2013). About 35% of land of the world is in arid and semi-arid condition. Farmers have taken on low yield set of varieties for the chickpea (*Cicer arietinum* L.) crop in rain-fed areas. The uses of optimal inputs are restricted bythe adjustment of such type of genotypes cultivation (Jackson et al., 2007). The concept of moisture stress tolerance of chickpea genotypes becoming a novel assignment for researchers due to water shortage, climate change as well as alteration of irrigated land into household for over population (Fahadet al., 2017; Eckstein et al., 2019; Jamro et al., 2020).

Recently, chickpea is the third most important pulse crop of Bangladesh in area and production. Bangladesh grows chickpea on about 4812 ha producing 5347 tons of grains with an average yield of about 1111 kg ha^{-1} in 2019 (FAOSTAT, 2021), which constitute about 0.04% of the total chickpea production in the world. Chickpea is mainly grown in the dry zone of Bangladesh. It is grown under residual soil moisture in both lowland and upland conditions. In lowland areas, it is grown as a relay or sequential crop after rice, while in upland areas it is grown mostly on fertile soil with a good water holding capacity after sesame, maize, green gram or fallow. The main constraints in yield reduction of chickpea crop are long moisture stress and sudden rain existed in the dry region where the water shortage is the main difficulty. The breeding and selection of genotypes under drought are considered an effective method to minimize the ramification of moisture stress exposure (Toker and Mutlu, 2011; Eckstein et al., 2019; Jamro et al., 2020).

Plants react to moisture stress and become adapted through various physiological and biochemical changes including changes of water use efficiency, proline content, and photosynthetic activity (Farooq et al., 2009). Moisture stress tolerance is linked with high relative water content (RWC) and low excised-leaf water loss. There is modulation of the activities of antioxidant enzymes which leads to enhanced cellular protection during the crop experiences stress conditions (Kaur et al., 2012). Plant cells respond defensively to oxidative stress by maintaining antioxidant defense compounds and osmolytes. Proline is one of the familiar osmolytes which boost in plants under moisture stress and assist the plants to continue cell turgidity (Moayedi et al., 2011). The damage caused during stress finally stress yield. Studies on reaction of antioxidative and non-antioxidative defense systems have been reported earlier in chickpea, but changes in carotenoids, chlorophyll, phosphorus, potassium, protein and proline content due to drought stress is still lacking. Therefore, the present study aims to expose various physiological and biochemical adaptations of selected chickpea genotypes with yield attributes at different moisture stress.

MATERIALS AND METHODS

Site description

The experiments were conducted during rabi season of 2017 and 2018 in the pot yard of the Bangladesh Agricultural Research Institute (BARI), Joydebpur, Gazipur, Bangladesh. The experimental site was located at $23^0 59'$ latitude and $90^0 24'$longitude and at the elevation of 34.5 m above the sea level. The soil was clay loam/clay in texture. General fertility status of the soil was low having low status of organic matter, including low status of phosphorus (P), potassium (K), medium status of zinc (Zn) and boron (B). The pH of the soil is 5.6. The soil contained 1.12% organic matter, 0.054% total nitrogen, 7.6 meq phosphorus, 0.14 meq 100 g^{-1} potash, 11.4 µg g^{-1}sulphur, 0.74 µg g^{-1} zinc and 0.23 µg g^{-1} boron.

Experimental design and treatments

The experiment was laid out in factorial experimental design. All treatments including four genotypes and 11 moisture stress applications were performed as Complete Randomized Design (CRD) with three replications. Experiment was carried out at net house.

Genotypes and moisture treatments

Four genotypes (G_1- BD-6048, G_2 - BD-6045, G_3- BD-6090, G_4- BD-6092) of chickpea along with 11 moisture stresses including T_1- Control (without irrigation), T_2- 30% of Field Capacity (FC) until pre-flowering stage,T_3- 50% of FC until pre-flowering stage,T_4- 70% of FC until pre-flowering stage, T_5- 90% of FC until pre-flowering stage, T_6- 30% of FC until flowering stage,T_7- 50% of FC until flowering stage, T_8- 70% of FC until flowering stage, T_9- 90% of FC until flowering stage, T_{10}-30% of FC until pod formation stage,T_{11}- 50% of FC until pod formation stage were included in the study. Seeds were collected from Plant Genetic Resources Center (PGRC), BARI.

Pot preparation and seed sowing

At first 132 pots were set at the net house of BARI. The size of each pot was 20 cm depth and 9 cm radius. Then the prepared soil was filled in the plastic pots. Each plastic pot was contained 6.0 kg soil. After filling pots, seeds were sown in the plastic pot properly. Ten seeds were sown in each plastic pot by hand at 3 to 4 cm depth with plant spacing of about 8 cm. Before sowing the seeds, all the pots were pre-irrigated to make them in optimum soil moisture condition, necessary for germination. After the germination, three healthy seedlings per pot were kept for each pot.

Procedures of water management

An appropriate amount of water was applied to all the pots every day until the beginning of the treatments. The day before the starting of the treatments, 500 ml of water was applied to each pot so that the soil moisture content (percentage) of all the genotypes remained equal. The first water stress treatment was started on 15, December 2017 and 2018 until pre-flowering stage, 2nd water stress treatment was started on 15, February 2018 and 2019 until flowering stage and 3rd water stress treatment was started on 1st, March 2018 and 2019 until pod formation stage. The amount of water needed according to the treatments applied in each pot with the help of the measuring cylinder. Moisture was maintained on the basis of prevailing moisture of sun-dried soil in pots and pot weight every one-day interval taking consideration of weight of plants in pot.

Physiological parameters

Relative leaf water content was estimated according to the method of Weatherley (1950). 100 mg fresh leaves were kept in distilled water for 4 hours to obtained turgid weight. The turgid weight was recorded after blotting the excess water on the surface of the sample. Dry weight was obtained after drying the samples in oven at 70˚ C till constant weight occurred. The relative water content (RWC) was calculated by the formula:

$$\text{RWC (\%)} = \frac{\text{Fresh weight - Dry weight}}{\text{Turgid weight - Dry weight}} \times 100$$

Biochemical parameters

Samples for chlorophyll and carotenoids determination were taken from chickpea leaves using a 0.8 cm diameter cork borer, weighted quickly in pre-weighted clean glass vials and 5 cm^3 of 80% acetone was added to these samples. The leaf material was bleached and decanted off. The optical density (OD) was read at λ = 663 nm, 645 and 470 nm using 80% acetone as a blank by a spectrophotometer. Content of chlorophyll a, chlorophyll b and carotenoids were calculated according to Lichtenthaler and Wellburn (1983) using the following formula:

Chlorophyll a = 12.21 OD 663 nm – 2.81 OD 645 nm

Chlorophyll b = 20.13 OD 645 nm – 5.03 OD 663 nm

Carotenoids = (1000 OD 470 nm – 3.27 chlorophyll a - 104 Chlorophyll b) / 229

Proline content was measured according to the method of Bates et al. (1973). An aliquot amount of fresh green leaf of chickpea was homogenized in 10ml of 3% sulphosalicylic acid and the homogenate was centrifuged at 5000 rpm for 15 min. Two milliliters of the supernatant were reacted with 2 ml of acid ninhydrin (1.25 g ninhydrin dissolve in 30 ml of glacial acetic acid and 20 ml of 6 M phsphoric acid) and 2 ml of glacial acetic acid for 1 hr at 100 ^{0}C and the reaction was then terminated in an ice bath.

The colored reaction mixture was extracted with 4 ml of toluene and the absorbance was recorded at 520 nm. Proline content was calculated from a standard curve. The protein contents of seeds were estimated following the procedure of Microkjeldhal (AOAC, 1965). One g seed sample was kept in a digest ion flask, with a little quantity of catalyst mixture (K_2SO_4 + $CuSO_4$), 10 ml of 96% concentrate sulphuric acid was added and kept for complete digestion. Digested sample was distilled. The distilled amount of ammonia was titrated with 0.1 N H_2SO_4.Nitrogen and protein content were calculated as per following formula:

Nitrogen (%) =Normality of H_2SO_4× V of H_2SO_4× 1.4 x100

Weight of sample

Protein (%) = Percent of nitrogen × 6.25

The content of phosphorous in the leaf of chickpea was determined by the procedure of Jackson (1975). One milliliter of aliquot of plant diacid extract in 50 ml volumetric flask, mixed with 10 ml of vanadate molybdate reagent diluted to 50 ml with distilled water and mixed well. The color was read after 30 min at 470 nm. The phosphorus concentrations were calculated using the standard curve expressed as percent. The content of potassium in the leaves of chickpea was determined by the method of Chapman and Pratt (1961). The plant extract (diacid digested) was directly read on flame photometer or after appropriate dilution as that final concentration range between 0 to 50 mg potassium per liter. A blank without sample was also run simultaneously. Result was calculated using a standard reading from potassium solution and expressed as percent potassium.

Measurement of yield and yield components

At maturity, the whole plant was cut at the ground level with a sickle. The harvested crop from each pot was bundled separately and tagged appropriately. Finally, data on yield contributing parameters such as plant height, number of pods per plant and seed yield were recorded separately.

Data analyses

Data were assessed by analysis of variance and by Duncan’s multiple range test (Gomez and Gomez, 1984) with a probability $P \leq 0.05$. The values followed by the same letters are not significantly different and different letters within treatments indicate significant differences at the 0.05 probability level.

RESULTS AND DISCUSSION

Climatic condition during crop cycle

The climatic condition of the experimental plot is subtropical in nature characterized by heavy rainfall from June to September (78-92%) and scanty in winter (1-11%) and mean rainfall is around 2200 mm per year. Temperature starts rising from February and continues till September and then gradually falls from the month of October of the year. The monthly rainfall, maximum and minimum temperature sand humidity during the study period were detailed in Fig. 1 and Fig. 2.

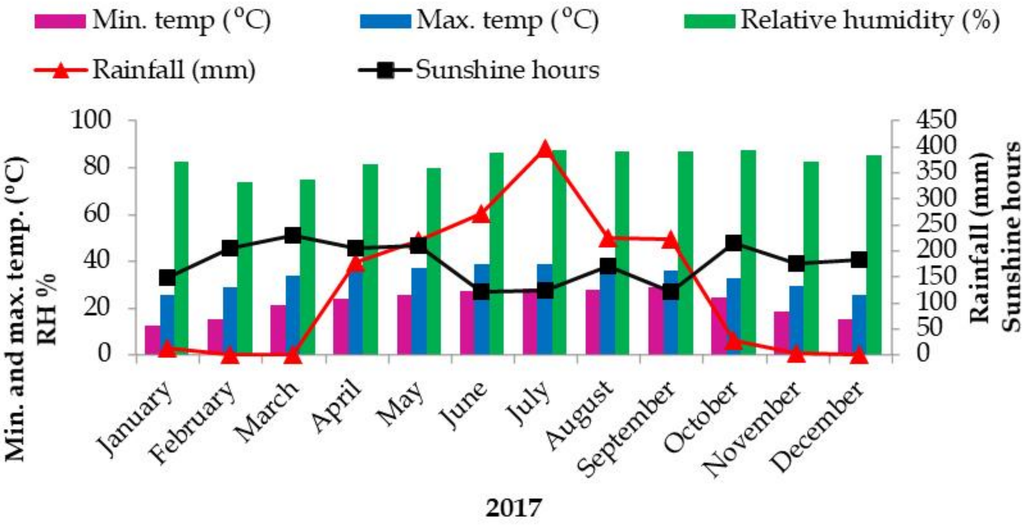

Fig. 1 Monthly average temperature, rainfall, relative humidity and sunshine hours of the experimental site in Gazipur during 2017

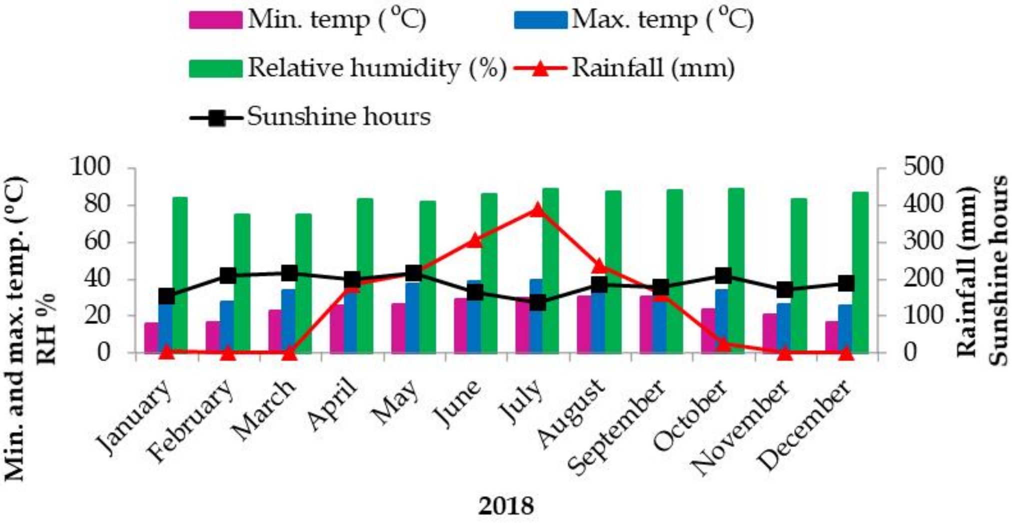

Fig. 2 Monthly average temperature, rainfall, relative humidity and sunshine hours of the experimental site in Gazipur during 2018

Interactions /Statistical significant analyses

Genotype by drought stresses and year interactions were found to be significant for RWC, carotenoids, chlorophyll a and b, protein and proline contents. ($P \leq 0.05$). Genotypic effect was significant for plant height, number of pod per plant and seed yield ($P \leq 0.05$).

Relative water content

Optimum relative water content is crucial for effective physiological functioning and growth processes of crop and is known as potential physiological marker in many crops. In the present study, RWC significantly decreased in all genotypes under moisture stress condition (Fig. 3). Among all the genotypes, BD-6048 showed maximum RWC (72.36 %) at 70% of FC until flowering stage and decreased when drought stress continued up to pod formation stage. This decline may be due to higher water loss through stomatal regulation during photosynthesis and ineffective water utilization assimilation under moisture stress (Lobato et al., 2008).

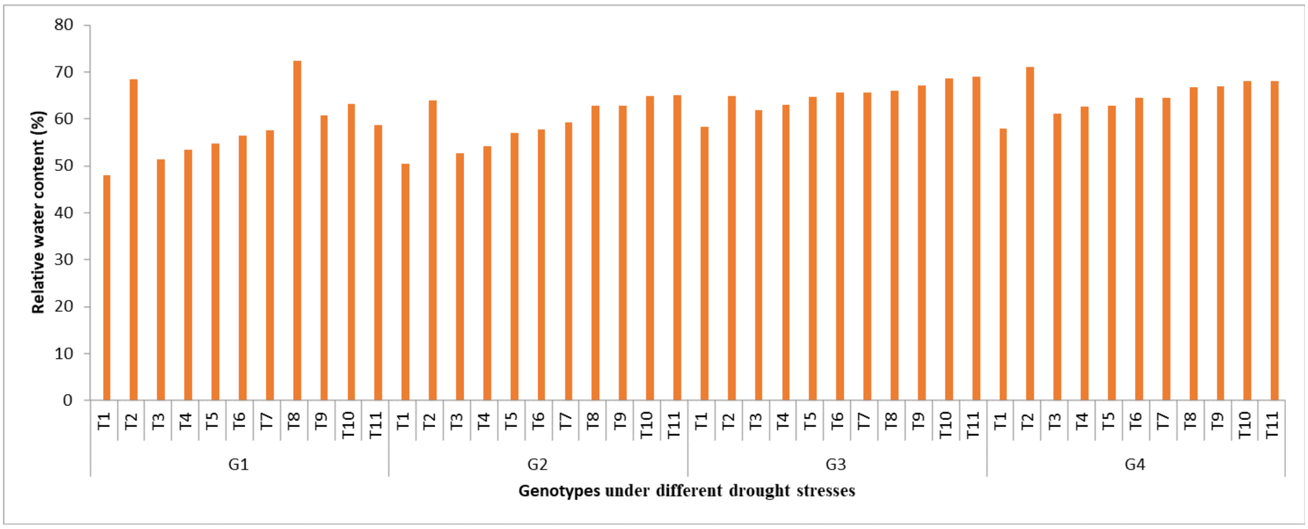

Fig 3 Relative water content (%) of genotypes under different drought stresses.

Carotenoids content

Leaf carotenoids content of chickpea under moisture stress differed in a genotype-dependent manner (Table 1). Carotenoids content of genotypes BD-6048 was highest compared to other genotypes. Moisture stress tolerant genotypes (‘Yazd’ and ‘Shiraz’) exhibited higher accumulation of carotenoids than moisture stress sensitive genotypes (Askari and Ehsanzadeh, 2015). While progressive increase in moisture stress level resulted in significant increases carotenoids content up to flowering stage and when chickpea was grown under moisture stress up to pod formation stage then it tended to decrease.

Table 1. Effect of interaction between genotypes and moisture stress on carotenoids content, chlorophyll a, chlorophyll b and potassium (%) of chickpea

Treatments	Carotenoids content (mgg^{-1})				Chlorophyll a (mgg^{-1})				Chlorophyll b (mgg^{-1})				Potassium (%)			
	G_1	G_2	G_3	G_4	G_1	G_2	G_3	G_4	G_1	G_2	G_3	G_4	G_1	G_2	G_3	G_4
T_1	1.28 xy	1.34vwx	1.94 ijk	1.10 z	0.62 za	0.62 za	0.76 rs	0.43 c	0.57 x	0.62z	0.75y-d	0.43C	3.16 zab	3.00 bc	2.62 d	6.02 j
T_2	2.28 e	2.03 gh	2.00 hij	1.86 lm	0.66 xy	0.66 xy	0.52 b	0.74 stu	0.67 x	0.69w	0.51z-a	0.79s	3.48 wx	3.14 zab	2.86 c	6.10 ij
T_3	1.29 xy	1.42 tuv	2.02 ghi	1.15 z	0.71 uv	0.61 za	0.59 a	0.83 p	0.73 v	0.73v	0.57y-b	0.83q	3.58 vw	3.27 yz	3.09 ab	6.19 hi
T_4	1.33 wx	1.46 stu	2.08 g	1.23 y	0.76 rs	0.62 z	0.63 yz	0.87 o	0.79 s	0.77t	0.63z	0.86p	3.69 uv	3.42 xy	3.04 b	6.29 h
T_5	1.40uvw	1.54 rs	2.18 f	1.27 xy	0.80 pq	0.70 vw	0.67 wx	0.91 mn	0.83 q	0.81r	0.66y	0.90o	3.89 st	3.59 vw	3.13 zab	6.53g
T_6	1.59 qr	1.59 qr	2.27 e	1.34vwx	0.91 lm	0.74 st	0.70 v	0.93klm	0.93 m	0.86p	0.69w	0.92mn	3.91 rst	3.69 uv	3.20 za	6.79f
T_7	1.64 pq	1.65 pq	2.32 de	1.44 tu	0.94 jkl	0.88 no	0.72 tuv	0.97 hij	1.09 g	0.92n	0.72v	0.96k	3.95 rst	3.71 uv	3.39 xy	7.00e
T_8	2.63 a	1.72 op	2.37 cd	1.49 st	1.35 a	0.96 ijk	0.78 qr	0.98fghi	1.29a	0.95l	0.76tu	0.98j	8.06a	3.80 tu	3.61 vw	7.26d
T_9	1.88klm	1.82 mn	2.45 bc	1.58 qr	0.99 fgh	1.03 e	0.82 p	1.01 efg	1.22c	0.99j	0.83q	1.02i	4.28 mn	4.05 pqr	3.97 qrs	7.60c
T_{10}	1.94 ijk	1.93 jkl	2.52 b	1.76 no	1.29 b	1.08 d	0.88 no	1.08 d	1.30b	1.10g	0.87p	1.07h	5.14 kl	4.25 no	4.16 nop	7.82b
T_{11}	1.72 op	2.01ghij	2.65 a	1.82 mn	0.98 ghi	1.15 c	1.01 ef	1.12 c	1.15d	1.13e	0.99j	1.11f	4.10 opq	5.04 l	4.42 m	5.23k
CV (%)	1.78	1.78	1.78	1.78	2.27	2.27	2.27	2.27	0.92	0.92	0.92	0.92	2.10	2.10	2.10	2.10
LSD ($_{0.05}$)	0.08	0.08	0.08	0.08	0.031	0.031	0.031	0.031	0.12	0.12	0.12	0.12	1.53	1.53	1.53	1.53

In a column, means followed by same letters are not significantly different at 5 % probability level by Duncan's Multiple Range Test (DMRT),G_1 = BD-6048, G_2 = BD-6045, G_3 = BD-6090, G_4 = BD-6092, T_1 = Control (without irrigation), T_2 = 30% of FC until pre flowering stage, T_3 = 50% of FC until pre flowering stage, T_4 = 70% of FC until pre flowering stage, T_5 = 90% of FC until pre flowering stage, T_6 = 30% of FC until flowering stage, T_7 = 50% of FC until flowering stage, T_8 = 70% of FC until flowering stage, T_9 = 90% of FC until flowering stage, T_{10} = 30% of FC until pod formation stage, T_{11} = 50% of FC until pod formation stage.

Chlorophyll a and chlorophyll b content

The interactions between genotypes by moisture stress treatments were significant for chlorophyll contents($P \leq 0.05$).Moisture stress imposed up to the pre-flowering stage, significantly decreased chlorophyll *a* and chlorophyll *b* content whereas moisture stress imposed up to flowering and pod formation stage also influenced these contents. The restricted water supply up to pre-flowering is liable for reducing chlorophyll content and during flowering stage and pod formation stage it had a mild effect on these contents. The moisture stress also indicated that chlorophyll *b* is not more sensitive to moisture stress than chlorophyll *a* (Table 1).The genotype BD-6048 showed a higher chlorophyll *a* and *b* content than the other genotypes up to flowering stage (Table 1). The results are accord with Nyachiro et al. (2001), depicted a significant decrease of chlorophyll *a* and *b* caused by water deficit in six *Triticum aestivum* L. cultivars. Decreased or unchanged chlorophyll level during moisture stress has been stated in other species, depending on the interval and severity of moisture stress (Kpyoarissis et al., 1995). A decrease of chlorophyll with moisture stress means a lowered capacity for light harvesting.

K and P content

The impact of moisture stress on P and K of the plant leaf was shown in Table 1 and 2.K content was higher when moisture stress imposed up to 70% of FC until flowering stage and tended to decrease at moisture stress up to pod formation stage. P contents showed considerable variation among the stress treatments and genotypes. Decreases in P content were larger for moisture stress up to pod formation stage compared to drought imposed at the pre-flowering and flowering stage. This might be expected with decreasing mobility of soil nutrients and plant water uptake as stress progressed. Novak and Voinovich (2000) stated that nutrient loss mechanism uptake of nutrients during the latter parts of the vegetation period was related to the differences in the estimated dry-weight biomass. In addition, Radersma et al. (2005) found that lower soil water contents caused less P uptake (through hampered diffusion) and decreased maize biomass growth.

Proline content

Differences in proline content or interactions between genotypes by moisture stress treatment were significant($P \leq 0.05$). The proline content increased where moisture stress imposed up to pre flowering stage than up to both flowering and pod formation stages in all genotypes of chickpea (Table 2). The proline content due to moisture stress was more in genotype BD-6048. The proline content depends on plant age, leaf age, leaf position or leaf part (Chiang and Dandekar, 1995). Under vegetative stage, moisture stress improved proline content, these increasing roles due to osmotic compatible and adjust osmotic potential which resulted in moisture stress avoidance in chickpea. Proline was found to be accumulated in a large level under moisture stress (Kalefetoglu, 2006; Tan et al.,2006; Ceyhan et al., 2012). Proline accumulation play adaptive roles in plant stress tolerance (Verbruggen and Hermans, 2008). For selection of stress tolerance, accumulation of proline has been considered as a parameter (Jaleel et al., 2007).

Protein content

Percent protein content was reduced with receding moisture stress (Table 2). At maintaining moisture stress up to flowering stage, the increase in leaf protein content was observed in BD-6048. With the further reduction in water levels, the protein content decreased in BD-6048 in comparison with control. Johal et al. (2020) elucidated that stress sensitive genotypes recorded maximal reduction (20.86 %) in comparison with tolerant accession at 75% receding moisture level.

Plant height and pod number per plant

Moisture stress had a significant effect on plant height and the number of pods per plant. Plants were generally tallest and had the highest number of pods when they were grown without moisture stress. Interactions between genotypes by moisture stress treatment were significant for plant height and pod number. The effect of the moisture stress on plant height up to pre flowering stage was severe and it had less effect when stressed up to flowering and pod formation stage (Table 3). Averaged across treatments BD-6048 showed the highest plant height. Pod number was also affected by moisture stress. The severity was more when stressed continued up to pod formation stage. BD-6048 had the highest pod numbers irrespective of treatments (Table 2). The yield of grain legumes grown under moisture stress conditions is largely depending on the number of pods plant per plant (Lopez et al., 1996; Pilbeam et al., 1992).

Seed yield per plant

The yield reaction to drought stress of chickpea is given in Table 3. The yield of all for genotypes of chickpea was affected by moisture stress. Interactions between cultivars by moisture stress treatment were significant. Plants stressed until pre-flowering stage and pod formation stage, gave a significantly lower yield than plants stressed during flowering stage. The highest yield was obtained from BD-6048 when stress imposed 70% of FC until flowering stage. Seed yield under moisture stress at 50% of FC until pod formation stage showed 19.94 % less than that under stress treatment at 50% of FC until flowering stage.

Table 2. Effect of interaction between genotypes and moisture stress on phosphorus (%), proline content and protein content (%) of chickpea

Treatments	**Phosphorus (%)**				**Proline content (mg100g^{-1})**				**Protein content (%)**			
	G_1	**G_2**	**G_3**	**G_4**	**G_1**	**G_2**	**G_3**	**G_4**	**G_1**	**G_2**	**G_3**	**G_4**
T_1	1.02 a	0.72 d	0.68 e	0.75 c	219.39a	214.52b	205.33d	193.38i	20.88 yz	22.42 n-r	21.09 yz	18.27 yzo
T_2	0.68 e	0.66 fgh	0.61 jkl	0.51 tuv	208.18c	202.37f	196.32h	186.39l	22.31o-s	22.98 i-m	21.62 vwx	20.81 z
T_3	0.74 cd	0.66 e-h	0.67 efg	0.67 ef	203.41e	196.33h	193.65i	183.44no	22.78 k-n	23.09 h-l	21.70 u-x	21.32 xy
T_4	0.85 b	0.61 jk	0.65 gh	0.68 e	199.30g	191.48j	191.46j	181.50q	22.84j-n	23.26 g-j	21.81 t-w	21.57 wx
T_5	0.74 cd	0.59 lmn	0.64 hi	0.65 h	195.35h	184.50m	188.71k	178.44s	23.82 def	23.40 f-i	22.05 r-v	21.89 s-w
T_6	0.68 e	0.58mno	0.62 ij	0.62 jk	191.26j	182.51opq	186.90l	176.68t	23.93 cde	23.54 e-h	22.14 q-u	22.20 q-t
T_7	0.54 rs	0.53 st	0.60k-n	0.61 jk	186.65l	177.46st	183.56mn	173.70u	24.13 cd	23.65 efg	22.26 p-s	22.26 p-s
T_8	0.31 a	0.51 uv	0.60j-m	0.58 nop	182.69nop	171.31v	181.69pq	171.46v	24.72 a	23.73 def	22.29 p-s	22.70 l-p
T_9	0.38 y	0.49 v	0.58 nop	0.56 opq	178.44s	168.67x	179.72r	168.57x	24.36 abc	23.77 def	22.59m-q	22.95 i-m
T_{10}	0.35 z	0.46 w	0.56 pqr	0.55 qrs	174.23u	163.28z	176.57t	166.38y	24.62 ab	23.67 efg	22.75 k-o	23.16 h-k
T_{11}	0.49 v	0.41 x	0.53 s	0.53 stu	170.24w	159.30a	174.28u	163.38z	24.18 bcd	23.10 h-l	22.82 j-n	23.26 g-j
CV (%)	2.35	2.35	2.35	2.35	0.34	0.34	0.34	0.34	1.22	0.34	1.22	0.34
LSD (0.05)	0.023	0.023	0.023	0.023	1.03	1.03	1.03	1.03	0.451	1.03	0.451	1.03

In a column, means followed by same letters are not significantly different at 5 % probability level by Duncan's Multiple Range Test (DMRT), G_1 = BD-6048, G_2 = BD-6045, G_3 = BD-6090, G_4 = BD-6092,T_1 = Control (without irrigation), T_2 = 30% of FC until pre flowering stage, T_3 = 50% of FC until pre flowering stage, T_4 = 70% of FC until pre flowering stage, T_5 = 90% of FC until pre flowering stage, T_6 = 30% of FC until flowering stage, T_7 = 50% of FC until flowering stage, T_8 = 70% of FC until flowering stage, T_9 = 90% of FC until flowering stage, T_{10} = 30% of FC until pod formation stage, T_{11} = 50% of FC until pod formation stage.

Table 3. Effect of interaction between genotypes and moisture stress on yield and yield contributing characters of chickpea

Treatments	**Plant height (cm)**				**Number of pod plant^{-1}**				**Seed yield plant^{-1} (g)**			
	G_1	**G_2**	**G_3**	**G_4**	**G_1**	**G_2**	**G_3**	**G_4**	**G_1**	**G_2**	**G_3**	**G_4**
T_1	43.07m-q	42.61pq	41.02r	39.21s	65.57rs	64.13stu	63.71tu	59.34w	15.06j-m	14.12mn	11.56qr	11.25r
T_2	44.10 h-n	43.15 m-q	42.95 n-q	40.52r	69.89j-n	66.46pqr	63.35uv	61.80v	16.50fgh	14.39lmn	12.43pq	11.80qr
T_3	44.13 h-n	43.71 k-p	43.05 m-q	42.48q	70.95 h-l	68.46no	65.13rst	63.14uv	16.01hij	15.72h-k	12.93op	12.17pqr
T_4	45.85 c-f	43.60 l-q	43.46 l-q	42.70opq	71.91ghi	69.64j-n	67.47opq	66.43pqr	16.45f-i	15.99hij	14.15mn	13.58no
T_5	45.95cde	44.08 h-n	43.75 k-p	43.47 l-q	74.71cde	70.31-m	65.15rst	65.88qrs	15.92h-k	14.87klm	13.67no	13.67no
T_6	46.10cd	45.01d-j	44.24 g-m	43.93 i-o	75.65bcd	73.43efg	70.36i-m	67.70op	18.62bc	17.42def	15.42i-l	15.49hijk
T_7	47.00bc	46.08cd	45.21d-h	45.27d-h	76.91ab	74.30de	71.30h-k	69.52lmn	19.46ab	17.76cde	15.50h-k	15.85hijk
T_8	48.70a	48.03ab	46.62c	45.92cde	78.37a	76.37bc	73.83ef	71.86ghi	20.37a	19.85a	18.46bcd	17.90cde
T_9	45.99cde	45.91cde	45.02d-j	43.94 i-n	76.91ab	74.42de	70.81h-m	70.84h-m	18.02cde	18.73bc	17.45def	17.28efg
T_{10}	45.35d-g	44.86 e-k	45.09d-i	44.66 f-l	71.36hij	72.46fgh	69.59k-n	69.70j-n	16.29ghi	17.99cde	15.71h-k	15.74h-k
T_{11}	44.40g-l	44.45 g-l	43.75 k-p	43.84 j-p	73.67ef	71.14 h-l	69.18mno	67.46opq	15.58h-k	16.49fgh	16.21ghi	14.24mn
CV (%)	3.93	3.93	3.93	3.93	1.55	1.55	1.55	1.55	4.17	4.17	4.17	4.17
LSD (0.05)	1.23	1.23	1.23	1.23	1.75	1.75	1.75	1.75	1.06	1.06	1.06	1.06

In a column, means followed by same letters are not significantly different at 5 % probability level by Duncan's Multiple Range Test (DMRT), G_1 = BD-6048, G_2 = BD-6045, G_3 = BD-6090, G_4 = BD-6092, T_1 = Control (without irrigation), T_2 = 30% of FC until pre flowering stage, T_3 = 50% of FC until pre flowering stage, T_4 = 70% of FC until pre flowering stage, T_5 = 90% of FC until pre flowering stage, T_6 = 30% of FC until flowering stage, T_7 = 50% of FC until flowering stage, T_8 = 70% of FC until flowering stage, T_9 = 90% of FC until flowering stage, T_{10} = 30% of FC until pod formation stage, T_{11} = 50% of FC until pod formation stage.

CONCLUSIONS

In conclusion, chickpea genotypes showed significant variation based on studied parameters for moisture stresses, which highlight the usability of these genotypes for future research programs. With the current outcome, it can be accomplished that moisture stress hinders the growth and metabolic action of chickpea genotypes. On the basis of analysis of chickpea genotypes, we concluded that there was considerable difference in physiological, biochemical attributes and seed yield. BD-6048 had higher RWC, chlorophyll, carotenoids and proline content in comparison to other genotypes up to moisture stress at pre-flowering stage. These parameters also demonstrated substantial variability under different moisture stress conditions. This research may assist to understand some adaptive mechanisms developed by chickpea genotypes and may useful to categorize valuable traits for chickpea breeding programs.

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THE INFLUENCE OF PRE-PLANT TREATMENTS ON SILAGE MAIZE (*Zea mays* L.) YIELD IN NO-TILLAGE SYSTEM

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ABSTRACT

No-tillage agricultural practices and pre-plant applications in agricultural systems have become quite common in recent years. In this study, the effect was examined of pre-plant applications on agronomic characteristics and yield in silage maize cultivation under no-tillage conditions. Plants of the forage legumes (common vetch, narbon vetch and fodder pea) and cereals (barley, triticale and annual ryegrass) were used as pre-plants materials and the values of plant height, green herbage yield, dry matter ratio, dry matter yield, leaf/stem ratio, peak tasselling time and core tasselling time were determined in silage maize. The data obtained demonstrated, that the green herbage yield and dry matter yield of maize was higher when the forage legume plants were used as pre-plants. Among the legumes, common vetch increased maize green herbage yield and dry matter yield more than other plants. On contrast, plants of the in cereals family caused a decrease in the maize yield. It was concluded that legume plants should be selected as pre-plant in no-tillage silage maize cultivation. Good results were obtained especially from common vetch, and the use of cereals as pre-plant had a negative effect on the maize for silage grown subsequently.

Key words: Cereals, legumes, maize, plant residue, stubble sowing.

INTRODUCTION

Soil cultivation is defined as mechanical application to the soil for the production of plants which has significant effects on soil properties such as soil water conservation, soil temperature, infiltration and evaporation (Busari et al., 2015). A good seed bed is prepared with conventional soil tillage systems and ensures the removal of many plant residues from the production area (Briones and Schmidt, 2017). However, conventional tillage systems have significant problems such as heavy machinery traffic, soil compaction and environmental pollution due to carbon emissions from fuel use (Bertolino et al., 2010; Krauss et al., 2010; Shahzad et al., 2016; Priya et al., 2019; Neugschwandtner et al., 2020).

Conservation soil tillage practices (such as reduced tillage and no tillage) provide minimum soil damage to conserve the soil water and improve water use efficiency (Wang and Shangguan, 2015). For example, no-tillage agriculture systems can increase water infiltration and reduce evaporation, thereby providing more soil water protection (Ranaivoson et al., 2019). Studies have shown that tillage technologies such as no tillage and reduced tillage can affect water use efficiency and reduce evapotranspiration in maize (Guo et al., 2019) and wheat (Liu et al., 2020). In other studies, the lint yield in cotton (DeLauna et al., 2020), nitrogen efficiency in soybean (Roy et al., 2019) and grain yield in maize (Ramos et al., 2019) have been shown to be affected by conservation tillage methods. However, reduced tillage becomes meaningful if it provides cost reduction without causing a decrease in yield (Faligowska and Szukala, 2015).

Another important agronomical practice in agricultural production is crop rotation and crop residues are of great importance in crop rotation systems. Mixing the organic crop residues from agricultural products into the soil both improves the physical properties of the soil and increases the organic matter and plant nutrients (Carranca, 2013; Kirkby et al., 2014; Langeroodi, 2015; Ebrahimian et al., 2016). Crop residues play an important role in facilitating the infiltration of rainwater into the soil and reducing evaporation from the soil, increasing the water holding capacity in production areas where irrigation is not provided and the need for water is met by rainwater alone (Verberg et al., 2012). In addition, plant residues (especially legume residues) positively affect soil bulk density, which is an important feature for soil functions (Singh et al., 2011; Nasta et al., 2020; Musa et al., 2020). Low soil mass density provides a good production environment for plants (Wang et al., 2016; Pan et al., 2017; Kool et al., 2019).

The aim of this study was to evaluate the effect of different pre-plant applications on the yield traits of maize plant in no-tillage conditions.

MATERIALS AND METHODS

The study was conducted in the research and application field of Akdeniz University Faculty of Agriculture, Department of Field Crops (36° 54′ N, 30° 38′ E) as a 2-year study in the 2016 and 2017 growing periods. The monthly average air temperature and total precipitation values of the research site are given in Table 1 (Anonymous, 2020). The research area soil has clay-loam texture, is slightly salty, high in lime and strongly alkaline. The soil has low organic matter and absorbable phosphorus and potassium content.

Table 1. Temperature and precipitation values of the study area in Antalya province

	Temperature (°C)			**Precipitation (mm)**		
	2015-2016	**2016-2017**	**Long term** (1930-2020)	**2015-2016**	**2016-2017**	**Long term** (1930-2020)
November	18.4	17.5	15.5	116.9	99.2	131.6
December	13.2	11.2	11.6	0.4	76.3	262.1
January	10.6	10.2	10.0	79.4	132.8	232.6
February	14.6	12.5	10.7	66.7	4.4	153.5
March	15.3	15.0	12.9	57.2	166.9	94.5
April	19.1	17.7	16.4	14.4	54.0	49.9
May	20.4	21.3	20.6	28.2	42.2	32.1
June	26.9	26.3	25.3	24.3	3.4	10.8
July	29.9	30.4	28.5	0.6	0.4	4.5
August	29.5	29.0	28.4	0.0	1.6	4.6

The experiment was performed with 3 replications according to the Randomized Complete Blocks Design (RCBD). Maize (*Zea mays* L. ‘Kilowatt’ (FAO 700)) was used as the main material. Plants from the legume family such as common vetch (*Vicia sativa* L. ‘Gulhan 2005’), narbon vetch **(***Vicia narbonensis* L. ‘Balkan’) and fodder pea (*Pisum arvense* L. ‘Tore’), and from the cereal family such as barley (*Hordeum vulgare* ‘Sladoran’), triticale (*xTriticosecale Wittmack* ‘Karma 2000’) and annual ryegrass (*Lolium multiflorum* ‘Trinova’) were used as pre-plant material.

Traditional soil tillage practices were applied while sowing the pre-plants. Fertilization (for legumes: 50 kg ha^{-1} N and 100 kg ha^{-1} P_2O_5, for cereals: 100 kg ha^{-1} N and 100 kg ha^{-1} P_2O_5) was applied before sowing these plants. All plants except annual ryegrass were planted on the same date, and annual ryegrass, which is a multiple cuts plant, was planted 2 weeks earlier in order to equalize the date of the second cutting with other plants. When the plants came to the shape period for green herbage, they were cut with a sickle at a height of 5 cm. In order to be able to sow the maize plant, which was the main subject of the study, on the same date in all applications the cutting time of some pre-plants was delayed.

Maize sowing was carried out after the pre-plants were cut. Seeds were sown on the 3rd of May in the first year and the 5th of May in the second year. The maize was sown directly into the stubble with no tillage applied. Rows were opened using a pickaxe, to imitate the direct seed drill, with 70 cm row spacing. Seeds were sown with 3-5 cm in-row spacing and thinned to 12 cm in-row spacing after emergence. At the time of sowing, for 100 kg ha^{-1} of nitrogen, phosphorus and potassium elements, 15*15*15 fertilizer was applied to the plots, and when the plants were 30-40 cm, 100 kg ha^{-1} nitrogen was applied with 33% ammonium nitrate fertilizer for upper fertilization.

During the trial, irrigation was applied as necessary, and weed removal was done manually without the use of herbicides. No other process was applied except these process. The peak tasselling time (day) and core tasselling time (day) were recorded during the growing season.

When the cobs reached the dough maturation period, the harvesting process was started. Plant height (cm) was measured in 10 plants from each parcel before the harvest. During the harvest, 1 row was removed from the edges of the parcels and 25 cm from the beginning and end of each row. The harvested plants were weighed and the parcel yield was calculated, and using the obtained value, green herbage yield (t ha^{-1}) was determined. In addition, 200 g samples were taken for each parcel and dried in a drying oven at 75 ° C for 48 hours (Shirvanian et al., 2004). The dry matter ratios (%) were determined, and dry matter yield (t ha^{-1}) was calculated using dry matter ratios and green herbage yield. In addition, the leaves and stems of 10 of the harvested plants were separated and the leaf / stem ratio per plant was determined (Yildiz and Erdogan, 2018).

Statistical analysis of the obtained data was made using the SAS statistics program and DUNCAN multiple comparison test was applied to the averages (Oten, 2017).

RESULTS

When the two-year results obtained from the study were compared in terms of legumes and cereals, it was seen that the applications had statistically significant effects on all characteristics except plant height and leaf / stem ratio (Table 2). When the green herbage yield values are evaluated, 45.14 t ha^{-1} of yield was obtained from legume applications and 40.18 t ha^{-1} from cereal applications. In terms of dry matter ratio and dry matter yield, the highest values were obtained with 33.79% and 15.34 t ha^{-1}, respectively, from legume applications, while the same values were determined as 32.00% and 12.97 t ha^{-1} in cereal applications. Higher values were recorded for legume applications then cereal applications in respect of the features of peak tasselling (64 vs 67 days) and core tasselling days (68 vs 70 days). Although the applications had an effect on plant height and leaf / stem ratios, these effects were not significant statistically.

Table 2. Agronomical characteristics of maize grown after legumes and cereals in the field trial run in 2016 and 2017 (means of two years)

	Green Herbage Yield (t ha^{-1})	**Plant Height (cm)**	**Dry Matter Ratio (%)**	**Dry Matter Yield (t ha^{-1})**	**Leaf/ Stem Ratio**	**Peak Tasseling Time (days)**	**Core Tasseling Time (days)**
Legumes	45.14 A	261 A	33.79 A	15.34 A	0.37 A	64 B	68 B
Cereals	40.18 B	257 A	32.00 B	12.97 B	0.36 A	67 A	70 A
F	7.72*	2.19	7.16*	9.94**	1.17	34.70**	13.21**
LSD ($p<0.05$)	3.85	5.28	1.80	1.80	0.02	0.97	0.85

F: * significant at the $p<0.05$ levels of probability, F: ** significant at the $p<0.01$ levels of probability
LSD: means with different letters indicates significant differences

When the legume plants were evaluated, it was determined that the pre-plant applications had statistically significant effects on all traits except plant height and core tasselling time (Table 3). While the maize green herbage yield values varied between 38.65 t ha^{-1} and 49.98 t ha^{-1}, the highest yield was obtained from parcels where common vetch was grown as pre-plant. The differences between plant heights according to the applications were not found to be statistically significant. Dry matter ratios varied between 30.23 and 36.13, with the lowest ratio determined for field pea, and the highest value was obtained from the application without pre-plant application, but this value was in the same group with the values obtained from common vetch and narbon vetch. Dry matter yield values were also similar to dry matter ratios. The lowest yield was obtained from the pre-plant application of field pea with 11.68 t ha^{-1}. The highest dry matter yield (17.38 t ha^{-1}) was determined in the common vetch application, but there was no statistical difference between this value and the values obtained from narbon vetch (16.05 t ha^{-1}) and without pre-plant applications (16.23 t ha^{-1}). The highest leaf / stem ratio values were determined as 0.38, 0.38 and 0.37 in common vetch, narbon vetch and fodder pea applications, respectively, but the differences between these values were not statistically significant. The lowest leaf / stem ratio 0.34 was obtained without pre-plant application. A similar situation was observed for peak tasselling time, with common vetch, narbon vetch and fodder pea applications in the same group at 63 days, and the highest value was determined as 67 days without pre-plant application. The core tasselling time, values varied between 68 and 70 days, with no statistical difference between the applications.

Table 3. Agronomical characteristics of maize grown after legumes in the field trial run in 2016 and 2017 (means of two years)

	Green Herbage Yield (t ha^{-1})	**Plant Height (cm)**	**Dry Matter Ratio (%)**	**Dry Matter Yield (t ha^{-1})**	**Leaf/ Stem Ratio**	**Peak Tasselling Time (days)**	**Core Tasselling Time (days)**
Common vetch	49.98 A	265A	34.76 A	17.38 A	0.38 A	63 B	68 A
Narbon vetch	47.13 AB	260A	34.05 A	16.05 A	0.38 A	63 B	68 A
Fodder pea	38.65 B	258A	30.23 B	11.68 B	0.37 A	63 B	68 A
Without pre-plant	44.85 AB	262A	36.13 A	16.23 A	0.34 B	67 A	70 A
F	4.21*	0.45	7.64**	5.89**	8.27**	6.76**	2.88
LSD ($p<0.05$)	5.86	39.49	3.73	3.15	0.03	3.42	2.61

F: * significant at the $p<0.05$ levels of probability, F: ** significant at the $p<0.01$ levels of probability
LSD: means with different letters indicates significant differences

According to the two-year averages, with the exception of peak tasselling time and core tasselling time in cereal pre-plant applications, the pre-plant applications were effective on all properties (Table 4). The green herbage yield of maize varied between 44.85 t ha^{-1} and 34.20 t ha^{-1} and while the highest yield was obtained without pre-planting treatment, the yield decreased in the pre-planted plots and the lowest yield was recorded in plots where annual ryegrass was used as a pre-plant. A similar situation was observed in plant height, with the highest plant height value of 262 cm recorded without pre-planting treatment and the lowest plant height value of 246 cm recorded in the annual ryegrass application. The highest dry matter ratio and dry matter yield values for maize were obtained with 36.23% and 16.23 t ha^{-1}, respectively, in plots without pre-plant, while the lowest values were obtained from annual ryegrass application with 28.74% and 9.85 t ha^{-1} for both properties. While annual ryegrass application gave the highest value of 0.39 leaf / stem ratio, other applications varied between 0.34 and 0.36, but the differences between these applications were not statistically significant. The features of peak tasselling time and core tasselling time were not affected by pre-plant applications

Table 4. Agronomical characteristics of maize grown after cereal crops in the field trial run in 2016 and 2017 (means of two years)

	Green Herbage Yield ($t\ ha^{-1}$)	Plant Height (cm)	Dry Matter Ratio (%)	Dry Matter Yield ($t\ ha^{-1}$)	Leaf/ Stem Ratio	Peak Tasselling Time (days)	Core Tasselling Time (days)
Annual ryegrass	34.20 B	246 B	28.74 B	9.85 B	0.39 A	67 A	70 A
Triticale	38.35 AB	262 A	31.91 AB	12.29 AB	0.36 B	66 A	71 A
Barley	43.33 AB	259 AB	31.23 AB	13.50 AB	0.36 B	67 A	70 A
Without pre-plant	44.85 A	262 A	36.13 A	16.23 A	0.34 B	67 A	70 A
F	4.17*	5.99**	4.49*	5.61**	10.73**	0.39	0.36
LSD ($p<0.05$)	6.79	5.71	3.15	2.61	0.02	2.47	3.16

F: * significant at the $p<0.05$ levels of probability, F: ** significant at the $p<0.01$ levels of probability
LSD: means with different letters indicates significant differences

DISCUSSION

The results obtained from this study, which was carried out to determine suitable pre-planting in maize cultivation in a no-tillage system, demonstrated that selecting legume family plants as the pre-plant provides good results in terms of green herbage yield, dry matter ratio, dry matter yield, peak tasselling time and core tasselling time

In conditions where nitrogen is limited, plants of the legume family have the ability to perform atmospheric nitrogen fixation through Rhizobium bacteria in their roots, and this is of great importance in legume-based production systems. Although there are variations according to species, legumes have a nitrogen fixation potential of between 100 and 300 kg ha^{-1} from the atmosphere. (Sun et al., 2008; Dwivedi et al., 2015; Dhanushkodi et al., 2018). Large quantities of mineral N can be released when legume residues are converted largely by nitrification and denitrification (Sant'Anna et al., 2018). Thus, for plants grown subsequently legume plants, may provide more mineral N than cereal residues due to their relatively high N content. In addition, the nitrogen element becomes more immobilized during the decomposition of cereal plant residues compared to the decomposition of legume plant residues, thus decreasing its usefulness (Hayat et al., 2008).

The roots of legume plants provide a high percentage of organic matter to the soil (Miheguli et al., 2018). Increasing organic matter improves many properties of the soil and facilitates the intake of nutrients. Another advantage of legume residues is that they have a lower C:N ratio than cereal residues (Palm et al., 2001; Lynch et al., 2016). In the C:N ratio, cellulose and lignin content are among the most important factors affecting mineralization of plant residues. A high C:N ratio usually leads to immobilization. In cases where the C: N ratio is high (> 25), nitrogen is immobilized by microorganisms during the decomposition of organic matter or is mineralized into the soil as ammoniacal nitrogen (Talgre et al., 2017). Decomposition is very slow in plant residues with high C:N, Lignin: N and polyphenol: N ratios (Lupwayi et al., 2011).

Plants grown after legumes can use 10% to 20% of the N amount in legume residues, and this ratio decreases to less than 10% in non-legume plant residues (Fillery, 2001). This contributes to the good nutrition of the plants grown after the pre-plant, especially during the seedling period, the formation of healthy seedlings and ultimately the increase in yield. In a study by Adeleke and Haruna (2012) the nitrogen content in the upper part of the soil was determined to increase by 250% in the lablab area, 200% in the peanut area, 170% in the feed pea field and 107% in the soybean area before maize planting. Arif et al. (2011), reported that chickpea cultivation, before maize, provided significant increases in maize yield compared to wheat cultivation. A similar result was found by Rajkumara et al. (2014) who stated that the application of 5 t ha^{-1} chickpea crop residue in no-tillage farming condition resulted in an increased maize yield. Amusan et al. (2011) reported that soybean residues found in the maize production area increased the maize yield by 7%. Singh et al. (2011) reported that legume residues in legume-wheat production systems caused an increase of 156% in the amount of soil microbial biomass carbon compared to the areas without residue. Gul et al. (2008) reported that legumes pre-planting is very good for maize, both increasing the dry matter yield of maize and reducing the nitrogen need of maize. Videnovic et al. (2013) determined that soybean, grown as a pre-plant, had a greater positive effect on maize grain yield compared to winter wheat.

In that study, when the legume plants were evaluated, while higher green herbage yield, dry matter ratio and dry matter yield values were obtained from common vetch, narbon vetch and without pre-plant application, lower values were recorded in fodder pea. Ozyazici and Manga (2000) used some legume plants, including common vetch, narbon vetch and fodder pea, as green manure plants, and when these three plants were compared in terms of maize green herbage yield, the highest yield was determined in the common vetch and narbon vetch plant treatments, and the lowest yield value was recorded in fodder pea treatment. In the same study, the dry matter yield from large to small was recorded in narbon vetch, common vetch and fodder pea, respectively. In another study, Kavut and Geren (2015) reported that the use of common vetch as a pre-plant in maize cultivation provided higher green herbage yield compared to fodder peas. Kalkan and Avci (2020) used narbon vetch, Hungarian vetch and fodder pea plants as pre-plants in maize cultivation and determined that the maize green herbage yield was higher in the narbon vetch applications compared to the fodder peas. Idikut and Kara (2011) used vetch and wheat plants as pre-plants in maize cultivation and reported that higher protein ratio and grain yields were obtained in maize grown after the vetch plant pre-planting. The findings of the current study are similar to these literature results.

Generally, the nitrogen contribution is expected to be higher in vetch species than in fodder peas. This is because in fodder peas, some of the nitrogen accumulates in the grains and leaves the system, but in the small grain vetches, nitrogen is added to the soil by remaining in the plant residues (Enrico et al., 2020). Sidaris et al. (1999) grew common vetch using different tillage methods and found that the nitrogen accumulation of the above-ground organs varied between 54.3 and 109.0 kg ha^{-1}, and that the same values varied between 73.3 and 173.3 kg ha^{-1} in the root. Ntatsi et al. (2019) reported biological nitrogen fixation of 45-125 kg ha^{-1} in peas, and also Cuttle et al. (2003) reported that although nitrogen fixation in peas ranged from 215 to 246 kg ha^{-1}, most of this was lost with the grain and the net gain of nitrogen was 106 kg ha^{-1}. These data show that vetch species provide more nitrogen to the soil for the next plant than peas.

When the cereals were evaluated, it was seen that the application without pre-planting positively affected the maize yield compared to the treatment where cereals were grown. Due to their high C:N ratio, cereals can inhibit the mobility of soil N, reduce the availability of N, and adversely affect the growth and yield of subsequent crops (Schomberg et al., 2004). Loomis et al. (2020) reported that the C:N ratio of barley varies between 54:1 and 80:1 depending on the growing conditions and location. Since the plants not included in the legume family have low N content and high C:N ratios, they have little or no positive effect on the crops grown after them, and may sometimes even have negative effects (Kramberger et al., 2009). This explains the yield decreases in the parcel where the cereal plants were used as pre-planting in the current study.

CONCLUSION

The results of this study demonstrated that pre-plant applications in maize cultivation under no-tillage conditions are effective on yield and some other agronomic characteristics. Especially when legume plants were used as pre-plants, higher yields were obtained compared to cereal pre-planting. In the evaluation of forage legumes, higher values were determined especially in terms of yield in the plots where common vetch was used as the pre-plant. When the results obtained from cereals were evaluated, pre-plant applications caused a decrease in yields, and higher yields were recorded in plots without pre-planting. It was concluded from the findings of this study that forage legumes should be preferred as the pre-plant in maize cultivation, common vetch in particular provides a significant increase in yield and the use of cereals as pre-plants causes a decrease especially in green herbage yield and dry matter yield. However, barley gave better results compared to triticale and annual ryegrass. Therefore, in regions where legumes cannot be cultivated it would be appropriate to use barley as a pre-plant.

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PERFORMANCE OF SOME DIFFERENT HYBRID DENT CORN (*Zea mays* L. *indentata* S.) VARIETIES UNDER CENTRAL ANATOLIAN CONDITIONS

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ABSTRACT

This study was conducted under Cumra ecological conditions between 2019 and 2020 in order to determine the performance of some KWS hybrid corn varieties (V1; Kerbanis, V2; Kefieros, V3; Kontigos, V4; Kefrancos, V5; 2572) belonging to different maturity groups. When the results obtained in both years of the study were evaluated together, it was seen that the highest leaf number, grain protein content, and the lowest stalk lodging values were obtained from the V3 variety. The highest first ear height and thousand grain weight values were obtained from the V2, V4, and V5 varieties. The highest ear weight and grain weight per ear values were obtained from the V2 and V5 varieties. The highest grain ear ratio, hectoliter, and the lowest grain moisture values were obtained from the V1 variety. The highest plant height, number of grains per ear, tasseling stage, ear length, ear diameter, stalk lodging, and grain moisture values were obtained from the V5 variety. However, obtaining the highest stalk lodging and grain moisture values from this variety in both research years eliminated the other positive characteristics of this variety. The highest grain yield values were obtained from the V2 variety. When the evaluation is done by taking into account the highest grain yield, lowest grain moisture, and stalk lodging values together, it is seen that the V3 variety is of a nature that can be recommended for Cumra-Konya ecological conditions.

Keywords: Corn, variety, yield components

INTRODUCTION

Corn is the most widely produced grain in the world with the highest yield obtained from a unit area. In 2020, there were 691 thousand 632 hectares of cultivation area, 9410 kg ha^{-1} yield, and 6.5 million tons of corn production in Turkey. Konya province ranks first in the country with a total grain corn production of 1 million 70 thousand tons (TURKSTAT, 2021).

In crop production, the yield is determined by the genetic potential of the variety, environmental factors (soil, climate), and cultivation techniques (Karasahin, 2021). The new hybrid grain corn varieties developed have a genetic yield potential of about 4 tons da^{-1}. The emergence of this potential varies depending on environmental stress factors, such as climate, planting time, plant density, soil structure and texture, amount of organic matter, pH, EC, water management, fertilization, weed control, and disease and pest presence (GRDC, 2017).

Theoretically, the ratio of C3 plants to convert solar energy into biomass under 30 °C and 380 ppm CO_2 conditions is 4.6%, while this ratio is 6% in C4 plants. In practice, the maximum rate reached is 2.4% for C3 plants and 3.7% for C4 plants. Within the total sunlight band, the photosynthetically active band (400-740nm) ratio is 48.7% (Zhu et al., 2008). The main function of the corn canopy is to absorb sunlight. If light falls on the soil instead of the corn canopy, it is absorbed by soil and lost, thereby a decrease in photosynthesis and yield is experienced. The use of this absorbed light as efficiently as possible is closely related to the leaf angle. Flat and horizontal leaves capture light best but use it quite inefficiently. Vertical leaves are quite unsuccessful in capturing light, but they use it most efficiently. For this reason, the ideal plant type is those that have horizontal lower leaves and vertical upper leaves. After the tasseling stage, the top 5 leaves of the corn canopy account for 26% of the total leaf area and realize 40% of dry matter production (Liu et al., 2020). More than 90% of the grain weight is obtained from photosynthesis, which takes place during the grain filling period, and during this period the photosynthetic products are transferred directly to the grains. Therefore, the production of dry matter after the emergence of the corn silk is very important for grain yield. It has been reported that shading practices in the silking period and before and after this period have different effects on varieties and cause a 12-50% reduction in grain yield (Liu, 2008; Kim et al., 2016).

Corn varieties mature at different times depending on their total temperature requirements. For this reason, by taking into account the dates of the last frost of spring and the first frost of autumn in the region where planting will be done, knowing the average total temperature values for many years during this vegetation period is one of the main elements that should be considered in the selection of varieties (Karasahin, 2021). Although late varieties are high in yield potential due to the length of maturation time, they even may not form ears if the total temperature requirements cannot be met. The ear kernels they form may not arrive at harvest maturity. Harvest becomes difficult due to high grain moisture, and because there will be problems with storing corn grains at high humidity, an obligation to sell the product at fairly low prices occurs due to high drying costs. Even, due to frost damage, low yield problems can be experienced. For these reasons, in the selection of varieties for growing the same varieties in different regions, the differences in maturation times must necessarily be taken into account.

Although the number of ears per unit area increases as the plant density increases, a decrease in ear weight occurs. For this reason, to be able to obtain high-yield varieties at high plant densities, the length of grain-filling time is important. The fact that grain-filling periods of varieties with a long maturation time are longer leads to high yield potential. As the maturation period increases, corn will receive more solar radiation and store more energy, and as a result of this, grain yield will increase (Sangoi, 2000). In addition, many studies conducted in our country and around the world have revealed that corn varieties with a long maturation period have higher yields (Tollenaar and Wu, 1999; Sangoi, 2000; Kocer, 2004). While the number of corn varieties registered in Turkey is approximately 307, this number increases to 938 along with registered corn lines. In addition, the number of production-permitted varieties that are at the stage of registration is 57 (TTSMM, 2021).

Today, on the one hand, new hybrid-corn varieties are being improved and released to the market, on the other hand, new varieties that have not been farmed before in our region are being brought and seeds are being sold to our farmers. Determining the suitability of registered varieties to their own ecology is of great importance. Problems of high grain moisture and low yield in harvest depending on the maturation period take place at the top of the most important problems in grain corn production in Konya region, where continental climate prevails. These problems will be solved by planting corn varieties with appropriate vegetation time at an appropriate density and applying cultural methods such as optimal irrigation and fertilization on site and on time (Karasahin and Sade, 2012).

In this study, it was aimed to determine the performance of some KWS hybrid corn (*Zea mays* L. *indentata* S.) varieties belonging to different maturity groups under the environmental conditions of Cumra.

MATERIALS AND METHODS

The research was carried out in the experimental fields of S.U. Cumra School of Applied Sciences between 2019 and 2020 under the ecological conditions of Cumra District of Konya province. Hybrid corn varieties (*Zea mays* L. *indentata* S.) belonging to V1 (Kerbanis; FAO 550), V2 (Kefieros; FAO 580), V3 (Kontigos; FAO 600), V4 (Kefrancos; FAO 650), V5 (2572; FAO 700) maturity groups were used as material.

Çumra-Konya district, where the research was conducted, has a typical Central Anatolian climate with cold and limited rainfall winters and hot and dry summers. Some meteorological data recorded in 2019 and in 2020 in Cumra-Konya district where the research was carried out and their average values for many years are given in Table 1. In order to determine the physical and chemical properties of the soil in which the study was conducted, samples were taken from depths of 0-30 cm and 30-60 cm and analyzed. Results of the analysis showed that they were in the clay structure class and poor in organic matter (Table 2).

Table 1. Some meteorological data of Cumra-Konya district

Months	**Precipitation (mm)**			**Mean Temperature (°C)**			**Mean Relative Humidity (%)**		
	2019	**2020**	**1972-2019**	**2019**	**2020**	**1972-2019**	**2019**	**2020**	**1972-2019**
May	2.0	13.8	37.3	18.4	17.0	15.7	44.7	49.0	57.5
June	36.2	6.8	20.3	21.5	20.7	19.8	54.5	47.4	53.5
July	6.4	10.0	6.8	22.3	24.6	22.9	47.7	43.3	48.2
August	4.6	0.0	4.5	22.7	23.1	22.4	48.9	39.2	49.1
September	6.4	12.2	10.2	19.1	22.0	18.3	47.8	48.0	52.1
October	9.2	6.2	31.1	15.5	17.1	12.6	56.4	49.2	62.8
November	43.0	10.0	35.8	8.8	6.0	6.2	71.6	69.8	71.2
Mean	-	-	-	18.3	18.6	16.8	53.1	49.4	56.3
Total	107.8	59.0	146	-	-	-	-	-	-

Table 2. Physical and chemical soil properties of the research field

Properties	Depth (0-30cm)	Depth (0-60 cm)	Properties	Depth (0-30 cm) (ppm)	Depth (0-60 cm) (ppm)
Organic matter (%)	1.24	0.62	Total N (%)	0.11	0.07
pH	7.58	8.14	P	49.6	6
EC (mS cm^{-1})	0.55	0.45	K	416	160
Lime ($CaCO_3$) (%)	19.8	19.0	Ca	6023	5960
Sand (%)	14.9	12.3	Mg	1193	1631
Silt (%)	26.0	26.7	Fe	4.22	3.38
Clay (%)	59.1	60.1	Zn	0.50	0.18
Texture class	Clay	Clay	B	0.64	0.31
Bulk density (g cm^{-3})	1.28	1.46	Mn	2.81	3.29
Field capacity (% v v^{-1})	34.43	35.98	Cu	1.69	1.63
Wilting point (%v v^{-1})	17.42	17.55	Na (meq l^{-1})	1.28	1.59
Infiltration rate (mm h $^{-1}$)	4.4	5.0	SO_4 (meq l^{-1})	0.11	0.51

The water used in the research as irrigation water was T2 (medium salt) A1 (low sodium) irrigation water (Table 3). The dynamic fertigation approach was used in fertilization. In this approach, plant nutrient uptake is associated with water intake. Plant nutrients are applied at a certain concentration of the applied irrigation water (Voogt et al., 2000). Based on this approach, TDR was used in the preparation of a soil-based irrigation program, and after determining the need for irrigation water, the amount of pure N amount required to reach the target product (20 tons ha^{-1}) was calculated by taking into account soil analysis. The determined amount (300 kg ha^{-1}) was proportionated to the total amount of water consumption of corn (7500 mm ha^{-1}) according to the average for many years, then the obtained result was multiplied by the amount of water to be applied per hour, and the amount of fertilizer to be given per hour was found. The amount of fertilizer to be required during the irrigation period was injected into the drip irrigation system through the venturi type dosing pump in the form of a solution to be prepared in a 1-ton water tank. A portion of nitrogen and all phosphorus and potassium were given with the base fertilizer. 500 kg ha^{-1} composite 13.24.12.10.1.1 (13% N, 24% P_2O_5, 12% K_2O, 10% SO_3, 1% Zn, and 1% Fe) fertilizer was used as the base fertilizer. The remaining amount of nitrogen (265 kg) was applied in each irrigation in the form of urea as described above (46% N). Above-ground drip irrigation method was used for irrigation of research plots. In this process, drip irrigation pipes of Akona Company, which have 22 mm diameter drippers with a 1.6 lt h^{-1} flow rate (dripping one at each 30 cm) were used.

Table 3. Irrigation water analysis report

Parameters	Results	Parameters	Results (ppm)
pH	7.3	Calcium	68
EC (mS cm^{-1})	0.58	Magnesium	17.99
Total hardness (Ca + Mg) (°F)	24.46	Bicarbonate	275.15
SAR	0.35	Chloride	25.17
Salinity and Alkalinity Class	T2-A1	Sulfate	28.80
Sodium (ppm)	12.65	Boron	0.14
Potassium (ppm)	2.73	Iron	0.18

The research was carried out as three replications based on the “randomized complete block design”. In the experiment, the plots were arranged as 5 x 2.8 m = 14.0 m^2 and in such a way that there are 4 rows in each plot. All plots were arranged in such a way that between rows and above rows were 70 x 15 cm (95238 plant ha^{-1}). The previous crop was dry bean. Planting was carried out with a pneumatic sowing machine on 25 May 2019 in the first year, and on 10 May 2020 in the second year. After the plants came out and the rows were clear, above-ground drip irrigation pipes were placed on the plots in such a way that they were in the middle of both rows.

In the research, full irrigation management was used. TDR (time domain reflectometry) device was used to measure soil moisture in the preparation of the irrigation program. For the first periods, by taking the average TDR readings for soil depth of 0 - 15 and 15 - 30 cm, irrigation was started when 40% of the soil useful moisture was used (TDR value 40), and the amount of water enough for bringing it to the field-capacity with a soil depth of 30 cm was applied by calculating. After the stem elongation period, irrigation was started by taking the averages of TDR readings for soil depth of 0 - 15, 15 – 30, 30 – 45 and 45 - 60 cm, and the amount of water enough for bringing it to the field-capacity with a soil depth of 60 cm was applied by calculating. The accuracy of the irrigation amount was checked by TDR measurements made 24 h after irrigation.

After physiological maturation, a row of edges was discarded from the edges of the plots as an edge effect.

The harvest was carried out on the remaining 7 m^2 area by manually collecting the cobs from the plants. Plant-based measurements were made on five plants randomly selected from each plot. Characteristics and methods studied in the research are as follows: stem diameter (mm), number of leaves, first ear height (cm), plant height (cm), number of grains per ear (piece ear^{-1}), tasseling stage (days), ear length (cm), ear diameter (mm), ear weight (g), grain weight per ear (g), grain ear ratio (%), stalk lodging (number), hectoliter weight (kg hl^{-1}), thousand grain weight (g), harvest grain moisture (%), and grain yield per unit area (kg ha^{-1}) (TTSMM, 2018). Analysis of raw protein content in grain (%) was done in an external laboratory according to AACC (2000). The obtained data was subjected to analysis of variance and the F test was performed (Steel and Torrie, 1980). The average values of the operations whose differences were detected were grouped according to the "HSD" importance test (JMP, 2007).

RESULTS AND DISCUSSION

Stem diameter, leaf number and first ear height

When the means of the two years were examined, it was observed that the highest stem diameter values (21.8 mm) were obtained from the V4 variety, while the lowest values were obtained from the V1 and V5 varieties (20.1 and 20.3 mm, respectively, $P<0.01$) and they took place in the same statistical group (b) (Table 4). Effects of the "year x variety interactions" were significant on stem diameter; the highest values were obtained from the first year x V4, second year x V4, and second year x V5 interactions (21.7, 21.9, and 20.0 mm respectively, $P<0.01$) and they took place in the same statistical group (a) (Table 4). It has been noted that varieties with a high stem diameter are more resistant to stalk lodging (Hondroyianni et al., 2000; Ma et al., 2014; Robertson et al., 2016).

Table 4. Means of stem diameter, leaf number and first ear height of varieties measured in the field trial run in 2019 and 2020

Varieties	**Stem Diameter (mm)****			**Leaf Number****			**First Ear Height (cm)****		
	2019	**2020**	**Mean**	**2019**	**2020**	**Mean**	**2019**	**2020**	**Mean**
V1	19.2 bc	21.0 ab	20.1b	17.1 a	16.4 bc	16.8 ab	96.5 d	119.0 a-d	107.7 b
V2	21.2 ab	19.5abc	20.4 ab	16.3 c	16.6 abc	16.4 bc	135.4 ab	124.4 abc	129.9 a
V3	20.8 abc	18.6 c	19.7 b	17.0 ab	16.9 abc	16.9 a	124.5 abc	117.3 bcd	120.9 ab
V4	21.7 a	21.9 a	21.8 a	16.4 bc	16.3 c	16.3 c	139.5 ab	124.6 abc	132.1 a
V5	20.7 abc	20.0 a	20.3 b	16.6 abc	16.5 abc	16.6 abc	140,8 a	110.2 cd	125.5 a
Mean	**20.7**	**20.2**		**16.7**	**16.5**		**127.3 a**	**119.1 b**	
CV (%)		9.8			3.5			15.9	
V		**			**			**	
Y		Ns			Ns			*	
V*Y Int.		**			**			**	

CV; Coefficient of variation, *; P <0.05, **; P <0.01, Ns; Not significant, V; Varieties, Y; Year

** Different letters indicate significant differences based on the LSD test at the p<0.05 level.

Regarding the mean of the two years, it was determined that the highest leaf number values (16.9) were obtained from the V3 variety ($P<0.01$). The year x variety interactions had significant effects on leaf number, and the highest values (17.1) were obtained from the first year x V1 interaction. On the other hand, the lowest values (16.3) were obtained from the second year x V4 interaction ($P<0.01$), (Table 4). There is a positive relationship between the number of leaves and the plant height. In general, the number of leaves decreases in short varieties, and this is largely affected by genetic factors (Hallauer and Miranda, 1987; Ozata et al., 2013).

In terms of the first ear height values, as the mean of the varieties, the difference between the years was statistically significant ($P<0.05$). The values of the first research year were obtained greater than the second year (127.3 and 119.1 cm, respectively). The highest first ear height values were obtained from the V2, V4, and V5 varieties in terms of the two-year mean data (129.9, 132.1, and 125.5 cm, respectively, $P<0.01$) and they took place in the same statistical group (a) (Table 4). Effects of year x variety interactions were significant on the first ear height values and the highest values (140.8 cm) were obtained from the first year x V5 interaction ($P<0.01$) (Table 4). Previous research has shown that the first ear height is a morphological character formed under the influence of ecological factors and the characteristic of the variety (Karasahin, 2008; Amanolahi-Baharvand et al., 2014). It has been reported that as the maturity group moves from early to late, the first ear height gradually increases in relation to this (Karasahin and Sade, 2012). Whereas the first ear height values of tall genotypes were found to be high, the first ear height values of short genotypes were found to be lower (Hallauer and Miranda, 1987).

Plant height, number of grains per ear, and tasseling stage

In terms of two-year data, the highest plant height values (321.5 cm) were obtained from the V5 variety ($P<0.01$). Year x variety interactions had significant effects on plant height values, and the highest values (334.6 cm) were obtained from the first year x V5 interaction ($P<0.01$). On the other hand, the lowest values (284.0 cm) were obtained from the second year x V3 interaction (Table 5). Although plant height in corn is largely under the influence of genetic factors, it is known to be widely affected by cultivation techniques and environmental conditions (Hallauer and Miranda, 1987; Koca, 2009). There is a positive relationship between plant height and first ear height, and the first ear heights of varieties with high plant height are also longer (Karasahin and Sade, 2012).

Table 5. Means of plant height, number of kernels ear^{-1} and tasselling stage of varieties measured in the field trial run in 2019 and 2020

Varieties	Plant Height (cm)**			Number of Grains Ear^{-1}**			Tasselling Stage (Days)**		
	2019	2020	Mean	2019	2020	Mean	2019	2020	Mean
V1	294.1 bc	295.8 bc	294.9 c	480.2 ef	552.7 cd	516.4 d	65	75	70 e
V2	292.3 bc	302.6 bc	297.4 bc	520.6 de	641.6 ab	581.1 b	68	78	73 d
V3	295.3 bc	284.0 c	289.6 c	503.4 def	618.3 ab	560.8 bc	70	80	75 c
V4	307.9 b	308.6 b	308.3 b	462.3 f	599.2 bc	530.7 cd	75	85	80 b
V5	334.6 a	308.5 b	321.5 a	623.5 ab	662.1 a	642.8 a	80	90	85 a
Mean	**304.8**	**299.9**		**518.0 b**	**614.8 a**		**71.6 b**	**81.6 a**	
CV (%)		5.5			8.0			0.001	
V		**			**			**	
Y		Ns			**			**	
V*Y Int.		**			**			Ns	

CV; Coefficient of variation, *; P <0.05, **; P <0.01, Ns; Not significant, V; Varieties, Y; Year

** Different letters indicate significant differences based on the LSD test at the p<0.05 level.

Considering the means of varieties, it is seen that the number of grains per ear values were obtained more in the second research year compared to the first year (614.8 and 518.0 respectively, P<0.01). The highest number of grains per ear values (642.8) were obtained from the V5 variety in terms of the mean of the two-year data (P<0.01). Effects of year x variety interactions were significant on number of grains per ear values, and the highest values (662.1) were obtained from the second year x V5 interaction (P<0.01). On the other hand, the lowest values (462.3) were obtained from the first year x V4 interaction (Table 5). There is a positive relationship between the number of grains per ear and grain yield, which are among the corn yield elements (Tollenear et al., 1992; Kara, 2001).

Considering the means of the varieties, it is seen that higher tasseling stage values were obtained in the second research year compared to the first year (81.6 and 71.6 respectively, P<0.01). In terms of the means of the two years, the highest tasseling stage values (85) were obtained from the V5 variety (P<0.01), (Table 5). The tasseling period in corn depends on genotype and environmental conditions, especially air temperature (Daughtry et al., 1984). Tasseling and maturation are delayed if factors such as moisture, nitrogen, and lighting are negative during the tasseling period (Shaw, 1988; Ozata et al., 2013).

Ear length, ear diameter and ear weight

In terms of the mean data of the varieties, higher ear length values were obtained in the second research year compared to the first year (18.0 and 16.7 respectively, P<0.01). Considering the two-year mean values, the highest ear length values (18.9 cm) were obtained from the V5 variety (P<0.01). Year x variety interactions had significant effects on ear length values, and whereas the highest values were obtained from almost all interactions (P<0.01), the lowest values (13.3 cm) were obtained only from the first year x V1 interaction (Table 6).

Table 6. Means of ear length, diameter and weight of varieties measured in the field trial run in 2019 and 2020

Varieties	Ear Length (cm)**			Ear Diameter (mm)**			Ear Weight (g)**		
	2019	2020	Mean	2019	2020	Mean	2019	2020	Mean
V1	13.3 b	18.1 a	15.7 c	51.1 bcd	50.5 cd	50.8 c	209.5 d	315.6 abc	262.6 b
V2	17.6 a	18.9 a	18.3 ab	52.3 abc	51.9 abc	52.1 ab	312.5 bc	374.0 a	343.3 a
V3	17.1 a	16.9 a	17.0 bc	52.4 abc	49.1 d	50.8 c	279.3 c	282.0 c	280.6 b
V4	16.7 a	17.1 a	16.9 bc	52.6 ab	49.7 d	51.1 bc	285.5 c	307.5 bc	296.5 b
V5	18.8 a	19.0 a	18.9 a	53.9 a	51.0 bcd	52.4 a	330.9 abc	356.1 ab	343.5 a
Mean	**16.7 b**	**18.0 a**		**52.5 a**	**50.4 b**		**283.5 b**	**327.0 a**	
CV (%)		11.9			3.4			16.9	
V		**			**			**	
Y		**			**			**	
V*Y Int.		**			**			**	

CV; Coefficient of variation, *; P <0.05, **; P <0.01, Ns; Not significant, V; Varieties, Y; Year

** Different letters indicate significant differences based on the LSD test at the p<0.05 level.

Regarding the means of the varieties, higher ear diameter values were obtained in the first research year compared to the second year (52.5 and 50.4 mm respectively, P<0.01). The highest ear diameter values (52.4 mm) were obtained from the V5 variety in terms of the two-year means (P<0.01) (Table 6). Effects of year x variety interactions were significant on ear diameter values, and the highest values were obtained from the first year x V5 interaction (P<0.01). On the other hand, the lowest values were obtained from the second year x V3 and second year x V4 interactions (49.1 and 49.7 mm, respectively), and they took place in the same statistical group (d) (Table 6). Ear length, which is an important yield element in corn, is under the influence of environmental and genetic factors. Variation among genotypes for ear diameter could be related to the genetic background of cultivars (Kusaksiz and Kutlu Kusaksiz, 2018). In general, it has been observed that the ear lengths and diameters of high-yielding varieties are also high. This indicates that there is a strong relationship between the length and diameter of the ear (Tekkanat and Soylu, 2005).

Considering the means of the varieties, it is seen that higher ear weight values were obtained in the second research year compared to the first year (327.0 and 283.5 g respectively, P<0.01). Regarding the means of the both years, the highest ear weight values were obtained from the V2 and V5 varieties (343.3 and 343.5 g respectively, P<0.01). Year x variety interactions had significant effects on ear weight values, and the highest values (374.0 g) were obtained from the second year x V2 interaction (P<0.01) (Table 6).

Grain weight per ear, grain ear ratio, and stalk lodging

In terms of the mean data of the varieties, higher grain weight per ear values were obtained in the second research year compared to the first year (282.1 and 236.8 g respectively, P<0.01). Considering the means of the both years, it is seen that the highest grain weight per ear values were obtained from the V2 and V5 varieties (288.9 and 292.2 g respectively, P<0.01). Year x variety interactions had significant effects on grain weight per ear values, and the highest values (317.9 g) were obtained from the second year x V2 interaction (P<0.01) (Table 7). There is high positive correlation between grain yield with ear length, ear diameter, number of kernel per ear, ear weight and thousand grain weight (Kokten and Akcura, 2017).

Table 7. Means of grain weight ear^{-1}, grain ear ratio and stalk lodging of varieties measured in the field trial run in 2019 and 2020

Varieties	Grain Weight Ear^{-1} (g)**			Grain Ear Ratio (%)**			Stalk Lodging (number)**		
	2019	2020	Mean	2019	2020	Mean	2019	2020	Mean
V1	182.5 d	275.1 abc	228.8 b	87.1 a	87.2 a	87.1 a	1.3 b	5.0 b	3.2 b
V2	259.9 bc	317.9 a	288.9 a	83.2 c	85.0 b	84.1 c	1.7 b	5.5 ab	3.6 b
V3	233.4 cd	244.1 c	238.8 b	83.5 c	86.6 a	85.1 b	0.0 c	2.0 c	1.0 c
V4	231.0 cd	265.8 bc	248.4 b	80.9 d	86.4 a	83.7 c	2.0 b	5.5 ab	3.8 b
V5	277.0 abc	307.3 ab	292.2 a	83.8 c	86.3 a	85.0 b	4.3 a	6.5 a	5.4 a
Mean	**236.8 b**	**282.1 a**		**83.7 b**	**86.3 a**		**1.9 b**	**4.9 a**	
CV (%)		16.9			1.2			12.4	
V		**			**			**	
Y		**			**			**	
V*Y Int.		**			**			**	

CV; Coefficient of variation, *; P <0.05, **; P <0.01, Ns; Not significant, V; Varieties, Y; Year
** Different letters indicate significant differences based on the LSD test at the p<0.05 level.

Regarding the means of the varieties, it is observed that higher grain ear ratio values were obtained in the second research year compared to the first year (86.3 and 83.7 % respectively, P<0.01). The highest grain ear ratio values (87.1 mm) were obtained from the V1 variety in terms of the two-year means (P<0.01) (Table 7). Effects of year x variety interactions were significant on grain ear ratio values, and the highest values were obtained from the first year x V1, second year x V1, second year x V3, second year x V4, and second year x V5 interactions, and they took place in the same statistical group (a) (87.1, 87.2, 86.6, 86.4, and 86.3 % respectively, P<0.01) (Table 7).

According to the means of the varieties, higher stalk lodging values were obtained in the second research year compared to the first year (4.9 and 1.9 respectively, P<0.01). When the means of the two years are considered, it is seen that whereas the highest stalk lodging values (5.4) were obtained from the V5 variety (P<0.01), the lowest stalk lodging values (1.0) were obtained from the V3 variety. Year x variety interactions had significant effects on stalk lodging values, and the highest values were obtained from the first year x V5 and second year x V5 interactions, and they took place in the same statistical group (a) (4.3 and 6.5 respectively, P<0.01). In both research years, the lowest stalk lodging values were obtained from the first year x V3 and second year x V3 interactions, and they took place in the same statistical group (c) (0.0 and 2.0 respectively) (Table 7). Genetic characters of varieties are the most important factor on stalk lodging rates (Butzen, 2013). Since plant stems are weaker at high plant densities, the rate of stalk lodging increases (Echezona, 2007). It has been observed that tall varieties are more prone to have stalk lodging under the influence of gravity (Ransom, 2005). In late varieties, the mechanical strength of the stalk is weaker due to the fact that they have high humidity. Therefore, more stalk lodging is observed in this type of varieties. Elements such as drought, low light intensity, and disease and harmful presence negatively affect photosynthesis, and since the carbohydrate needs of corn grains that develop in such cases cannot be met by photosynthetic activities, these needs are provided from carbohydrates accumulated in the root and stalk. For this reason, the mechanical strength of the stalk reduced and lodging occurs. Loses between 5 and 50% are experienced in grain yield due to the stalk lodging (Li et al., 2015; Xue et al., 2020; Wang et al., 2020).

Hectoliter, grain protein content and thousand grain weight

In terms of the means of the varieties, higher hectoliter values were obtained in the first research year compared to the second year (77.3 and 76.2 kg hl^{-1} respectively, P<0.01). The highest hectoliter values (78.5 kg hl^{-1}) were obtained from the V1 variety in terms of the two-year means (P<0.05) (Table 8). Year x variety interactions had significant effects on hectoliter values. The highest values were obtained from the second year x V1 and the first year x V2 interactions and they took place in the same statistical group (a) (79.9 and 79.4 kg hl^{-1} respectively, P<0.01) (Table 8). Ozmen (2008) reported that effects of genotype and environmental interaction were significant on hectoliter weight.

Table 8. Means of hectoliter, grain protein and thousand grain weight of varieties measured in the field trial run in 2019 and 2020

Varieties	**Hectoliter (kg hl^{-1})****			**Grain Protein Content (%)****			**Thousand Grain Weight (g)****		
	2019	**2020**	**Mean**	**2019**	**2020**	**Mean**	**2019**	**2020**	**Mean**
V1	77.1 ab	79.9 a	78.5 a	7.3 c	8.0 a	7.7 b	369.7 c	410.5 cd	390.1 b
V2	79.4 a	72.6 d	76.0 b	6.9 de	6.7 e	6.8 d	475.0 a	434.0 bc	454.5 a
V3	76.7 abc	78.7 ab	77.7 ab	7.5 bc	8.3 a	7.9 a	435.0 abc	378.0 d	406.5 b
V4	75.0 bcd	76.9 ab	75.9 b	6.8 de	7.7 b	7.2 c	448.0 abc	460.5 ab	454.3 a
V5	78.3 ab	73.0 cd	75.6 b	7.4 c	7.0 d	7.c c	434.0 bc	452.5 ab	443.3 a
Mean	**77.3 a**	**76.2 b**		**7.2 b**	**7.5 a**		**432.3**	**427.1**	
CV (%)		1.7			1.0			3.2	
V		**			**			**	
Y		*			**			Ns	
V*Y Int.		**			**			**	

CV; Coefficient of variation, *; P <0.05, **; P <0.01, Ns; Not significant, V; Varieties, Y; Year

** Different letters indicate significant differences based on the LSD test at the p<0.05 level.

Considering the means of the varieties, it is seen that higher grain protein content values were obtained from the second research year compared to the first year (7.5 and 7.2 % respectively, P<0.01). In terms of two-year means, the highest grain protein content values (7.9 %) were obtained from the V3 variety (P<0.01). Year x variety interactions had significant effects on grain protein content values, and the highest values were obtained from the second year x V1 and second year x V3 interactions (8.0 and 8.3 % respectively, P<0.01) (Table 8).

In terms of the mean data of the two years, the highest thousand grain weight values were obtained from the V2, V4, and V5 varieties (454.5, 454.3, and 443.3 g respectively, P<0.01). Effects of year x variety interactions were significant on thousand grain weight values, and the highest values (475.0 g) were obtained from the first year x V2 interaction (P<0.01) (Table 8). Since multiplication of the number of grains obtained from the unit area and the thousand grain weight directly determine the grain yield, the number of grains and weight constitute the main yield elements (Battaglia et al., 2017).

Grain moisture and grain yield

In terms of the mean data of the varieties, higher grain moisture values were obtained in the first research year compared to the second year (22.8 and 22.3 % respectively, P<0.01). When the means of the two years were examined, it was seen that whereas the highest grain moisture values (27.6 %) were obtained from the V5 variety (P<0.01), the lowest grain moisture values (17.3) were obtained from the V1 variety. Effects of year x variety interactions were significant on grain moisture values, and the highest values (28.0 %) were obtained from the first year x V5 interaction (P<0.01). In both research years, the lowest grain moisture values were obtained from the first year x V1 and second year x V1 interactions, and they took place in the same statistical group (e) (17.6 and 17.0 respectively) (Table 9). When previous studies on grain moisture at harvest were examined, it was observed that in general, the harvest grain moisture in early varieties was found to be lower than in late varieties (Kapar and Oz, 2006; Vartanli and Emekliler, 2007). The decrease in moisture in the grain after physiological maturation is closely related to physical factors. Climate factors such as temperature, humidity, and wind speed affect the moisture decrease in the grain. Variety characteristics also play an important role in moisture loss after maturation. For example, characteristics such as the way corn ear husk wraps the ear, the size and number of ear husks, the permeability of the kernel shell, and the oblique or upright posture of the ear are important variety characteristics that affect moisture drop (Nielsen, 2006; Demirci, 2009). Due to the limited vegetation time of the Central Anatolia region, grain moisture in harvest is very important in terms of production costs. If grain moisture is high at harvest, the cost of drying will bring a serious burden and negatively affect producers today, when energy prices are increasing day by day (Karasahin and Sade, 2012; Karasahin, 2021).

Table 9. Means of grain moisture and grain yield of varieties measured in the field trial run in 2019 and 2020

Varieties	Grain Moisture (%)**			Grain Yield (kg ha^{-1})**		
	2019	2020	Mean	2019	2020	Mean
V1	17.6 e	17.0 e	17.3 d	15715 e	16650 d	16183 d
V2	21.9 c	26.5 b	24.2 b	19370 b	20410 a	19890 a
V3	20.0 d	19.5 d	19.8 c	17655 c	17925 c	17790 c
V4	26.6 b	21.2 c	23.9 b	17355 cd	17670 c	17513 c
V5	28.0 a	27.2 b	27.6 a	18070 c	19500 b	18785 b
Mean	**22.8 a**	**22.3 b**		**17633 b**	**18431 a**	
CV (%)		1.1			1.5	
V		**			**	
Y		**			**	
V*Y Int.		**			**	

CV; Coefficient of variation, *; P <0.05, **; P <0.01, Ns; Not significant, V; Varieties, Y; Year

** Different letters indicate significant differences based on the LSD test at the p<0.05 level.

When the means of the varieties were examined, it was observed that higher grain yield values were obtained in the second research year compared to the first year (18431 and 17633 kg ha^{-1} respectively, P<0.01). The highest grain yield values (19890 kg ha^{-1}) were obtained from the V2 variety in terms of the two-year means (P<0.01), (Table 9). Effects of year x variety interactions were significant on grain yield values, and the highest values (20410 kg ha^{-1}) were obtained from the second year x V2 interaction (P<0.01) (Table 9). Grain yield in the corn plant is genetic and a complex character that occurs from planting to harvest as a result of ecology and the common effects of cultivation techniques (Hallauer and Miranda, 1987). It has been stated that in variety preference, all parameters, such as grain yield, grain moisture, and the rate of moisture loss after physiological development should be evaluated together (Sade, 1999).

There are similarities and differences between the values obtained from the results of this research and the values obtained from other research studies related to the issue. It is considered that these similarities and differences stem from the type and dose of mineral fertilizers, the genetic characteristics of cultivated varieties, climate and soil properties, and applied cultivation techniques (soil preparation, the depth and density of planting, pest and weed control, irrigation, and fertilization).

CONCLUSIONS

When the results obtained in both years of the study are evaluated together, it is seen that the highest stem diameter values were obtained from the V4 variety. The highest leaf number, grain protein content and the lowest stalk lodging values were obtained from the V3 variety. The highest first ear height and thousand grain weight values were obtained from the V2, V4 and V5 varieties. The highest ear weight and grain weight per ear values were obtained from the V2 and V5 varieties. The highest grain ear ratio, hectoliter and the lowest grain moisture values were obtained from the V1 variety. The highest plant height, number of grains per ear, tasseling stage, ear length, ear diameter, stalk lodging and grain moisture values were obtained from the V5 variety. However, obtaining the highest stalk lodging and grain moisture values from this variety in both research years eliminated the other positive characteristics of this variety. The highest grain yield values were obtained from V2 variety. When the evaluation is done by taking into account the highest grain yield, lowest grain moisture, and stalk lodging values together, it is seen that the V3 variety is of a nature that can be recommended for Cumra-Konya ecological conditions.

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EVALUATION OF FORAGE TURNIP + CEREAL MIXTURES FOR FORAGE YIELD AND QUALITY TRAITS

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ABSTRACT

The aim of current study was to investigate the effects of intercropping forage turnip “FT” with cereals (barley, “B”, wheat “W” and oat “O”) for improving forage yield and nutritive value in Bilecik conditions in 2019 and 2020 growing periods. The percentage of forage turnip and cereals in mixtures was 100+0%, 75+25%, 50+50%, 25+75%, and 0+100%, respectively. Experiments were arranged in a randomized complete block design with three replications. The hay yield, crude protein yield, relative feed value (RFV), condensed tannin, total phenolic, total flavonoid, and mineral contents (K, P, Ca, and Mg) were determined. The hay yield of treatments was ranged between 5.45-10.27 t ha^{-1}. The highest crude protein yield was obtained mixture of 50FT+50O% (1.80 t ha^{-1}). The pure forage turnip, 75FT+25B%, 75FT+25O%, and 50FT+50B% mixtures were statistically in the same group as 50FT+50O%.

The condensed tannin was ranged between 2.00-2.84%. The highest RFV was calculated for 75FT+25O% (136.29), and the 50FT+50O% mixture (125.42) was statistically in the same group as mixture of 75FT+25O%. The present study showed that intercropping of forage turnip with barley and oat improved the hay yield and quality. The best results regarding for forage yield and quality were obtained from the FT+B and FT+O mixtures with seed rates of 75:25% and 50:50%.

Keywords: Bilecik, cereals, forage turnip, hay yield, nutritive value.

INTORDUCTION

The roughages are one of the indispensable feed sources of livestock. In Turkey, there is a serious shortage of roughages for livestock. Acar et al. (2020) reported that the deficit of quality roughage was 55 million tons in Turkey. To meet this requirement, it is necessary to give importance to different forage plants and especially to the production of intermediate forage crops. Turkey has very different soil, climate, and production designs through which it is possible to successfully grow many forage plants, however, very few forage plant species and varieties are cultivated.

Leaf-type forage turnip (Lenox) is a very high protein rate and contains rich vitamins, therefore, it increases the efficiency of animals. Lenox is an easily digested plant. It is especially consumed by sheep, goats, cattle, and dairy cows with great appetite (Geren, 2002). In recent years, forage turnip, which is used as a source of roughage in ruminant feeding to close the roughage deficit gap, and has become attractive due to its rapid growth ability and not needing irrigation in the winter season. Besides, the forage turnip is resistant to frost in autumn and early winter and maintains its high nutrients.

Intercropping, which is the cultivation of plants belonging to more than one species in the same area, is considered one of the sustainable farming techniques (Bauman et al., 2002). While intercropping provides an increase in total product and income, it enables more efficient use of soil, water, and labor resources and inputs. In addition, it has important advantages in terms of compatibility with ecological agriculture and less damage to the environment (Hook and Gascho, 1988; Akman and Kara, 2001; Bauman et al., 2002).

The present study was aimed to explore the potential of forage turnip-cereals intercropping systems with different mixture ratio and the effects on yield and chemical composition of fodder in Bilecik-Turkey.

MATERIALS AND METHODS

The experiments were conducted during 2019-2020 and 2020-2021 winter growing season at the Agricultural Practice and Research Area, Bilecik Seyh Edebali University, Turkey. Soil proporties of the experiment field taken from 30 cm depth were clay-loam type with pH of 7.71 and 7.82% $CaCO_3$, 257.2 kg ha^{-1} phosphorus, 1605.0 kg ha^{-1} potassium, and 1.25% organic matter. The Table 1 shows the meteorological data of the experiment area during growth season (December – May), including monthly average temperature, monthly total precipitation and avarage moisture. During to growing season, total precipitation was 322.0 mm at the long-term, it was 342.3 mm for 2019-2020 and 338.3 mm for 2020-2021 (Table 1).

Table 1. Meteorological data of experiment area in the longterm and studied years*

Months	**Temperature (˚C)**			**Precipitation (mm)**			**Moisture (%)**		
	LT**	2019-20	2020-21	LT**	2019-20	2020-21	LT**	2019-20	2020-21
November	9.0	12.7	8.3	37.2	27.6	3.6	71.1	63.0	72.0
December	4.5	5.6	7.9	55.9	78.4	9.7	76.0	78.0	71.5
January	2.4	2.4	5.6	50.1	45.4	78.3	76.5	74.0	58.6
February	3.7	5.2	5.7	42.0	65.6	37.7	73.2	72.1	68.0
March	6.4	8.6	5.1	47.3	34.1	101.0	69.3	68.8	72.1
April	11.5	10.8	11.4	41.8	36.0	73.0	64.2	61.0	67.0
May	16.1	16.7	17.5	47.7	55.2	35.0	64.5	62.0	60.1
Average	**7.7**	**8.9**	**8.8**				**70.7**	**68.4**	**67.0**
Total				**322.0**	**342.3**	**338.3**			

* Tukish State Meterogical Service; **: Long-term

Forage turnip *(Brassica rapa* L. cv. Lenox), barley (*Hordeum vulgare* L. cv. Ramata), wheat (*Triticum aestivum* L. cv. Reis) and oat (Avena sativa L. cv. Cekota) were sown in pure and in mixtures (forage turnip:cereals respectively as; 75:25, 50:50 and 25:75) on 21 November, 2019 and 20 November, 2020. Experiment was arranged in a randomized complete block design with three replications.The plots were formed 6 rows with 20 cm space and 5 m length. In pure sowings, 10 kg ha^{-1} for seed was used for forage turnip, 220 kg ha^{-1} for barley, 200 kg ha^{-1} for wheat, 200 kg ha^{-1} for oat. The P fertilizer (P_2O_5) 80 kg ha^{-1} was uniformly applied to the soil with sowing. Pure forage turnip and mixtures were harvested at the flowering stage based on forage turnip, while the cereals were harvested at milk-dough stages (Harvest was determined using Zadoks scale 73) (Zadoks et al., 1974; Mut et al., 2015; Mut et al., 2018). All treatments were manually harvested and then the species were separated as forage turnp and cereal.

Plant samples were dried 65 °C until constant weight to determine hay yield. Crude protein ratio (CP), acid detergent fiber (ADF), neutral detergent fiber (NDF), potassium (K), phosphorus (P), calcium (C) and magnesium (Mg) content of hay was determined by using Near Reflectance Spectroscopy (NIRS, ‘Foss 6500’) with software package program ‘IC-0904FE’.

Relative feed value (RFV) was estimated according to the following equations adapted from Rohweder et al. (1978);

RFV = (DDM% * DMI%)/1.29; DDM%= 88.9 - (0.779 x ADF%);

DMI% = 120/NDF%,

DDM = Dry matter digestibility,

DMI = Dry matter intake.

The total phenolic contents (TP) of samples were determined with slight modification according to the Folin-Ciocalteu reagent (FCR) method of Singleton et al. (1999). Samples (200 µL) were mixed with diluted FCR (200 µL) and shaken vigorously for 3 min. Then, 200 µL sodium carbonate (Na_2CO_3) solutions (20%) were added. Then samples absorbance of each sample was measured at a spectrophotometer at the absorbance value of 760 nm after incubating in dark at room temperature for 2 h. The total phenolic contents were expressed as mg equivalents of gallic acid (GAE) g^{-1} dry weight (DW) according to the equation obtained from the standard gallic acid graph and calculated from the calibration curve (R^2= 0.9994).

The total flavonoid content (TF) was determined by using Arvouet-Grand et al. (1994) with some modifications. Each sample (200 µL) was mixed with 100 µL of aluminum nitrate (10%) and 100 µL of potassium acetate (1 M). The total volume of the solution was adjusted to 5mL with ethanol. Similarly, a blank was prepared by adding methanol in place of the sample. Absorbance measurements were read at a spectrophotometer at the absorbance value of 417 nm after 40 min incubation at room temperature in dark conditions. Total flavonoid content was expressed as mg equivalents of quercetin (QE) g^{-1} DW according to the equation obtained from the standard quercetin graph and calculated from the calibration curve (R^2= 0.9994).

A 6 ml of tannin solution was added to 0.01 g of ground sample then placed in a tube and mixed on a vortex. The tubes were tightly capped and kept at 100 °C for 1 hour, and the samples were allowed to cool. Then, they were read at a spectrophotometer at the absorbance value of 550 nm (Bate-Smith, 1975). Condensed tannins (CT) were calculated by the following formula: Absorbance (550 nm x 156.5 x dilution factor) / Dry weight (%).

The data was analyzed in separate and combined years. ANOVA was performed by using SPSS 22.0 package program and, means were grouped with Duncan's multiple-range test.

RESULTS AND DISCUSSION

In combined years, botanical composition of forage turnip + cereal mixtures was given Figure 1. It is difficult to maintain the desired botanical composition in intercropping. Accordingly, it was observed that cereals were more dominant in the present study. This is due to the cereals being earlier germination than the forage turnip. Therefore, they are become dominant by tillering.

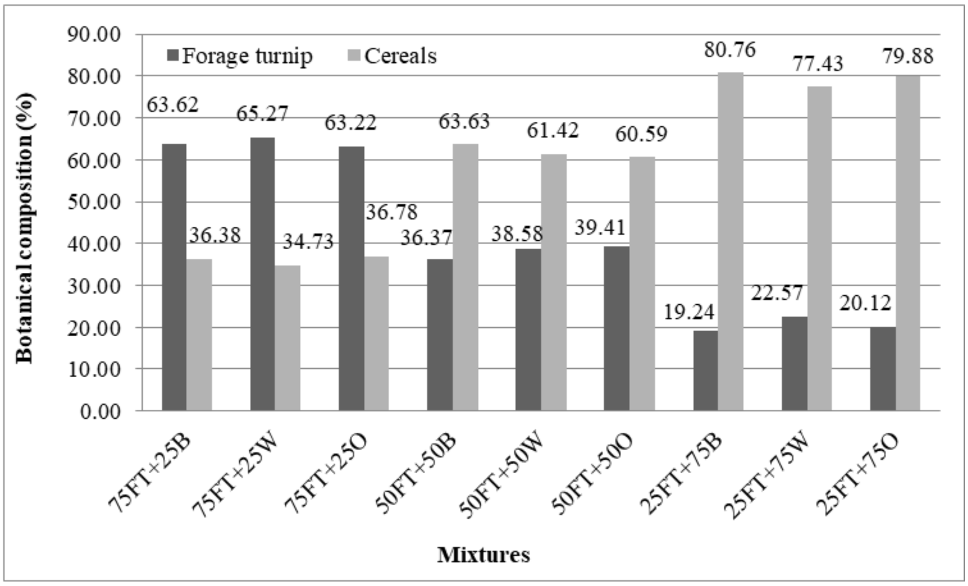

Figure 1. Botanical composition of forage turnip + cereal mixtures averaged over two years **(**FT: Forage turnip; B: Barley; W: Wheat; O: Oat)

Hay and crude protein yield of forage turnip + cereal mixtures were given in Table 2. The effect of intercropping on hay yield was significant (P<0.01) in the first (2019-2020) and combined years, while the second year (2020-2021) was not significant. It was determined that significant (P<0.01) differences between treatments in both separate and combined years in terms of crude protein yield. Besides, it was no significant differences among years in terms of hay and crude protein yield.

Table 2. Hay and crude protein yield of forage turnip + cereal mixtures

Treatments	**Hay yield (t ha^{-1})**			**Crude protein yield (t ha^{-1})**		
	2019-20**	**2020-21^{ns}**	**Mean****	**2019-20****	**2020-21****	**Mean****
100^{FT}	5.77 cd	7.24	6.50 cde	1.23 bc	1.64 ab	1.44 a-d
100^{B}	8.84 ab	8.40	8.62 a-d	1.08 c	0.93 c	1.01 de
100^{W}	7.50 abc	7.23	7.37 b-e	1.02 c	0.91 c	0.97 e
100^{O}	9.09 ab	8.16	8.62 a-d	1.20 bc	1.07 bc	1.14 cde
75^{FT}+25^{B}	8.38 ab	9.43	8.91 ab	1.62 ab	1.85 a	1.74 ab
75^{FT}+25^{W}	4.96 d	5.94	5.45 e	0.97 c	1.19 abc	1.08 cde
75^{FT}+25^{O}	8.56 ab	8.79	8.67 abc	1.66 ab	1.82 a	1.74 ab
50^{FT}+50^{B}	9.21 ab	8.64	8.93 ab	1.56 ab	1.44 abc	1.50 abc
50^{FT}+50^{W}	5.80 cd	6.92	6.36 de	1.04 c	1.22 abc	1.13 cde
50^{FT}+50^{O}	9.96 a	10.58	10.27 a	1.73 a	1.86 a	1.80 a
25^{FT}+75^{B}	9.72 ab	9.00	9.35 ab	1.41 abc	1.24 abc	1.33 b-e
25^{FT}+75^{W}	7.34 bcd	8.01	7.67 b-e	0.98 c	1.20 abc	1.09 cde
25^{FT}+75^{O}	9.39 ab	9.62	9.50 ab	1.23 bc	1.46 abc	1.35 b-e
Mean	***8.04^{ns}***	***8.30^{ns}***		***1.29^{ns}***	***1.37^{ns}***	

FT: Forage turnip; B: Barley; W: Wheat; O: Oat; ^{ns} is not significant, ** is significant at $P \leq 0.01$.

In combined years, the highest hay yield was determined in 50FT+50O% (10.27 t ha^{-1}). The pure barley (8.62 t ha^{-1}), pure oat (8.62 t ha^{-1}), 75FT+25B% (8.91 t ha^{-1}), 75FT+25O% (8.67 t ha^{-1}), 50FT+50B% (8.93 t ha^{-1}), 25FT+75B% (9.35 t ha^{-1}), and 25FT+75O% (9.50 t ha^{-1}) mixtures were statistically in the same group as 50FT+50O%. The lowest hay yield was determined in mixture of 75FT+25W% (5.45 t ha^{-1}) (Table 2). The cereal seed ratio in the mixture had a positive effect on hay yield. Accordingly, the hay yield of mixtures was increased with increasing cereal seed ratio excepted the 25FT+75O% mixture. Besides, the hay yield of forage turnip + barley and forage turnip + oat mixtures were higher than mixture of forage turnip + wheat. This is due to the hay yield of barley and oats have higher than wheat. When pure sowings were compared, barley and oats were in the same statistical group and had a higher hay yield than wheat and forage turnip. Zeybek (2017) reported that hay yield of forage turnip and companion crops (Hungarian vetch, forage pea, common vetch, and oat) mixtures ranged between 4.27 and 9.25 t ha^{-1}. The hay yields obtained in the present study are similar to the findings by Zeybek (2017).

In combined years, the highest crude protein yield (1.80 t ha^{-1}) was obtained from the mixture containing 50% forage turnip and 50% oat, and the lowest protein yield was obtained from the pure wheat (0.97 t ha^{-1}) sowing (Table 2). Copur Dogrusoz et al. (2019) reported that the crude protein yield of turnip-legume mixtures ranged between 0.41-1.09 t ha^{-1}. In the current study, the crude protein yield of forage turnip and cereal mixtures was higher than the findings by Copur Dogrusoz et al. (2019). Environmental conditions, cultural applications, and the cultivars used in the trials could cause such differences. Besides, the high hay yield could be another reason for the difference.

The relative feed values (RFV) and condensed tannin contents of forage turnip + cereal mixtures were given in Table 3. The effect of intercropping on RFV and condensed tannin content was significant (P<0.01) in separate and combined years. Besides, the effect of years was a significant at 5% level on RFV and condensed tannin.

Table 3. Relative feed values and condansed tannin content of forage turnip + cereal mixtures

Treatments	**Relative Feed Value**			**Condensed tannin content (%)**		
	2019-20**	**2020-21****	**Mean****	**2019-20****	**2020-21****	**Mean****
100^{FT}	107.1 def	110.44 c	108.66 cd	2.29 b	2.04 bcd	2.17 bcd
100^{B}	91.4 gh	84.97 e	88.06 e	2.85 a	2.83 a	2.84 a
100^{W}	82.7 h	87.27 e	84.96 e	2.87 a	2.74 a	2.80 a
100^{O}	100.8 fg	93.29 de	96.78 de	2.91 a	2.59 a	2.75 a
$75^{FT}+25^{B}$	117.7 bcd	124.57 bc	121.00 bc	2.26 b	2.10 bcd	2.18 bcd
$75^{FT}+25^{W}$	111.7 c-f	125.49 bc	118.34 bc	2.26 b	1.99 bcd	2.13 bcd
$75^{FT}+25^{O}$	130.0 a	143.28 a	136.29 a	2.10 bc	2.14 bc	2.12 bcd
$50^{FT}+50^{B}$	114.3 cde	113.89 bc	114.03 bc	2.25 b	2.12 bc	2.19 bcd
$50^{FT}+50^{W}$	119.5 abc	116.51 bc	117.94 bc	2.27 b	2.18 bc	2.23 bc
$50^{FT}+50^{O}$	128.3 ab	123.03 bc	125.42 ab	2.30 b	2.20 bc	2.24 b
$25^{FT}+75^{B}$	104.8 ef	113.09 bc	108.76 cd	1.79 c	2.21 b	2.00 d
$25^{FT}+75^{W}$	117.4 bcd	108.29 cd	112.60 bc	2.14 b	1.91 cd	2.02 c
$25^{FT}+75^{O}$	109.5 c-f	129.52 ab	119.04 bc	2.20 b	1.82 d	2.01 d
Mean	***110.40 B****	***113.36 A****		***2.34 A****	***2.22 B****	

FT: Forage turnip; B: Barley; W: Wheat; O: Oat; * is significant at P< 0.05; ** is significant at P< 0.01.

In combined years, The highest RFV was calculated for 75FT+25O% (136.29), however, the 50FT+50O% (125.42) mixture was statistically in the same group as 75FT+25O% (Table 3). When pure sowings were compared, the highest RFV was calculated in pure forage turnip compared to the pure cereals. This is due to the forage turnip having a lower ratio of ADF and NDF. Generally, the RFV of the forage turnip + oat mixture was higher than other mixtures. Yılmaz et al. (2015) reported that RFV values decrease with increasing ratio of cereals in the mixtures. Similar results were obtained in the current study. The relative feed value (RFV) is the widely used index of feed quality worldwide and based on estimates of feed intake from NDF content and digestibility from ADF content. Acordingly, the RFV value for beginning quality standard is > 151, for the first quality standard is 151–125, for the second quality standard is 124–103, for the third quality standard is 102–87, for the fourth quality standard is 86–75 and for the fifth quality standard is < 75 represented the forage quality (Rohweder et al., 1978). The RFV values determined in the study ranged between fourth and first quality class of fodder.

It is estimated that ¼ of the methane gas released into the atmosphere is produced in the digestive system of ruminants (Lascano and Cardenas, 2010). Condensed tannins are inhibited some hydrogen-producing protozoans and methane-producing organisms that use hydrogen directly in the rumen, and reduce greenhouse gas emissions. (Martin et al., 2016). Besides, the condensed tannins show an anthelmintic effect, reduce animal internal parasites and increase productivity in the animals (Luscher et al., 2016). Barry (1987) indicated that plants with low tannin content have a beneficial effect as they reduce protein degradation in the rumen, while Kumar and Singh (1984) stated that high amounts of condensed tannin negatively affect protein digestion and microbial and enzyme activities. Onal Asci and Acar (2018) indicated that the feeds with low condensed tannin led to increase in protein content of milk Accordingly, the condensed tannin content of plants is required to be 2-3% or less. In the present study, the condensed tannin content ranged between 2.00-2.84% in the combined years and below the critical level (Table 3). In previous studies, the condensed tannin content of different forage crops ranged from 0.21% to 0.45% (Bal et al., 2006; Kokten et al., 2017; Yıldız et al., 2021).

There were significant (P<0.01) differences between mixture rates (P<0.01) and years (P<0.05) in terms of total phenolic and flavonoid content (Table 4). In combined years, the highest total phenolic and flavonoid contents were determined in the mixture of 25 FT+75O% (8.36 mg GAE g-1 and 5.32 mg QE g-1, respectively) (Table 4). The studies of ruminant nutrition have shown that flavonoids and phenolic compounds are very important for rumen health and animal productivity (Rochfort et al., 2008; Patra et al., 2016; Lee et al., 2017). These compounds have antioxidant and antimicrobial effects, and they have significant potential to improve animal yield and quality (O'Connell and Fox, 2001; Robbins, 2003; Santos Neto et al., 2009; Frozza et al., 2013). Besides, the positive effect of flavonoids and phenolic compounds on the productivity and health of animals as well as rumen fermentation and control of nutritional stress such as bloat and acidosis have been demonstrated in several studies (Seradj et al., 2014; Paula et al., 2016). Kuppusamy et al. (2018) reported that the total the phenolic and flavonoids content of *Lolium multiflorum* was determined as 3.90 mg GAE g^{-1} and 6.83 mg QE g^{-1}, respectively.

Table 4. Total phenolic and flavonoid contents of forage turnip + cereal mixtures

Treatments	Total phenolic content (mg GA g^{-1})			Total flavonoid content (mg QE g^{-1})		
	2019-20**	2020-21**	Mean**	2019-20**	2020-21**	Mean**
100^{FT}	6.20 bcd	6.93 b	6.56 cde	3.31 de	3.65 c	3.48 fg
100^{B}	5.25 e	3.23 d	4.24 h	2.47 f	1.14 e	1.80 i
100^{W}	5.73 cde	6.24 b	5.99 f	3.08 ef	2.44 d	2.76 h
100^{O}	5.11 e	4.74 c	4.93 g	2.47 f	2.21 d	2.34 ı
$75^{FT}+25^{B}$	6.44 bc	7.18 b	6.81 cd	4.73 b	5.00 b	4.86 b
$75^{FT}+25^{W}$	6.22 bcd	6.29 b	6.26 def	3.05 ef	4.46 b	3.76 ef
$75^{FT}+25^{O}$	6.33 bc	9.01 a	7.67 b	4.63 b	5.95 a	5.29 a
$50^{FT}+50^{B}$	6.23 bcd	6.02 b	6.12 ef	3.30 de	4.89 b	4.10 de
$50^{FT}+50^{W}$	5.65 cde	6.24 b	5.94 f	4.04 bcd	4.86 b	4.45 cd
$50^{FT}+50^{O}$	5.59 cde	7.10 b	6.35 c-f	2.78 ef	4.80 b	3.79 ef
$25^{FT}+75^{B}$	5.37 de	4.30 c	4.83 g	3.55 cde	3.10 c	3.33 g
$25^{FT}+75^{W}$	6.72 b	7.06 b	6.88 c	4.21 bc	4.88 b	4.55 bc
$25^{FT}+75^{O}$	7.83 a	8.90 a	8.36 a	5.48 a	5.16 b	5.32 a
Mean	6.05 B*	6.40 A*		3.62 B*	4.04 A*	

FT: Forage turnip; B: Barley; W: Wheat; O: Oat, * is significant at P <0.05; ** is significant at P <0.01.

Mineral matter content of hay including potassium (K) and phosphorus (P) were significantly (P<0.01) different among treatments and between years (Table 5). In combined years, K and P contents were ranged between 1.747-3.056% and 0.355-0.514%, respectively. K and P contents of the mixtures were higher than pure treatments. Mut et al. (2017) reported that K and P content of maize + legume mixtures were ranged between 1.79-2.33% and 0.25-0.32%, respectively. Kidambi et al. (1989) and Anonymous (1971) reported that the requirements for diary cattle are 0.8% for K, 0.20% for P. Within this respect, in this study, ratios of K and P were very high (Table 5).

Table 5. Potassium and phosphorus raito of forage turnip + cereal mixtures

Treatments	Potassium (%)			Phosphorus (%)		
	2019-20**	2020-21**	Mean**	2019-20**	2020-21**	Mean**
100^{FT}	2.230 c	2.503 d	2.366 cd	0.413 c	0.473 b	0.443 c
100^{B}	2.129 c	2.497 d	2.313 d	0.389 d	0.354 d	0.371 e
100^{W}	1.840 d	1.653 e	1.747 e	0.359 e	0.352 d	0.355 f
100^{O}	2.633 b	2.429 d	2.531 c	0.423 e	0.399 c	0.411 d
$75^{FT}+25^{B}$	2.556 b	3.218 abc	2.887 ab	0.471 b	0.525 a	0.498 b
$75^{FT}+25^{W}$	2.685 ab	2.979 bc	2.832 b	0.480 ab	0.534 a	0.507 ab
$75^{FT}+25^{O}$	2.551 b	3.124 abc	2.837 b	0.487 ab	0.521 a	0.504 ab
$50^{FT}+50^{B}$	2.633 b	3.362 a	2.997 ab	0.493 ab	0.533 a	0.513 ab
$50^{FT}+50^{W}$	2.870 a	3.024 abc	2.947 ab	0.476 ab	0.528 a	0.502 ab
$50^{FT}+50^{O}$	2.723 ab	3.228 abc	2.976 ab	0.487 ab	0.536 a	0.512 ab
$25^{FT}+75^{B}$	2.866 a	3.245 ab	3.056 a	0.475 ab	0.533 a	0.504 ab
$25^{FT}+75^{W}$	2.583 b	3.029 abc	2.806 b	0.484 ab	0.545 a	0.514 a
$25^{FT}+75^{O}$	2.847 a	2.890 c	2.868 ab	0.482 ab	0.536 a	0.509 ab
Mean	2.550 B**	2.860 A**		0.455 B**	0.490 A**	

FT: Forage turnip; B: Barley; W: Wheat; O: Oat; ** is significant at P <0.01.

The calcium (Ca) and magnesium (Mg) content of forage turnip + cereal mixtures were significant differences among treatments (p<0.01) and between years (p<0.05). The highest Ca (1.131%, 0.984%, and 1.057%) and Mg (0.326%, 0.308%, and 0.317%) contents were determined in pure forage turnip in both separate and combined years. Tajeda et al. (1985) indicated that forage should contain 0.30% Ca and between 0.12-0.20% Mg. In the current study, Ca and Mg values of all treatments were higher than suggested by Tajeda et al. (1985) (Table 6).

Table 6. Calcium and magnesium raito of forage turnip + cereal mixtures

Treatments	**Calcium (%)**			**Magnesium (%)**		
	2019-20**	**2020-21****	**Mean****	**2019-20****	**2020-21****	**Mean****
100^{FT}	1.131 a	0.984 a	1.057 a	0.326 a	0.308 a	0.317 a
100^{B}	0.213 g	0.182 ı	0.198 g	0.154 g	0.130 f	0.142 h
100^{W}	0.282 g	0.262 hı	0.272 fg	0.157 g	0.176 e	0.167 h
100^{O}	0.344 fg	0.292 ghı	0.318 f	0.221 def	0.179 e	0.200 g
$75^{FT}+25^{B}$	0.667 bcd	0.601 cd	0.634 bc	0.251 bcd	0.242 cd	0.247 cde
$75^{FT}+25^{W}$	0.772 b	0.661 c	0.717 b	0.266 b	0.260 bcd	0.263 bc
$75^{FT}+25^{O}$	0.697 bc	0.808 b	0.753 b	0.267 b	0.290 ab	0.279 b
$50^{FT}+50^{B}$	0.480 ef	0.482 def	0.481 de	0.222 def	0.231 d	0.226 efg
$50^{FT}+50^{W}$	0.721 bc	0.636 cd	0.678 bc	0.248 b-e	0.254 bcd	0.251 b-e
$50^{FT}+50^{O}$	0.575 cde	0.595 cd	0.585 cd	0.254 bc	0.262 bcd	0.258 bcd
$25^{FT}+75^{B}$	0.489 ef	0.406 fgh	0.447 e	0.208 f	0.223 d	0.215 fg
$25^{FT}+75^{W}$	0.542 de	0.436 efg	0.489 de	0.217 ef	0.245 cd	0.231 def
$25^{FT}+75^{O}$	0.467 ef	0.561 cde	0.514 de	0.226 c-f	0.282 abc	0.254 b-e
Mean	***0.568 A****	***0.531 B****		***0.232 B****	***0.237 A****	

FT: Forage turnip; B: Barley; W: Wheat; O: Oat; * is significant at P <0.05; ** is significant at P< 0.01.

CONCLUSION

The current study showed that intercropping forage turnip with barley and oat improved the forage yield and quality compared to their monocrops. Besides, it was determined that the seed rate selection was important in intercropping in this study. Accordingly, mixtures of forage turnip with barley and oat at seed rate of 25+75% and 50+50% exhibited superior performance compared to other treatments in Bilecik ecological condition.

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EFFECT OF SALINITY STRESS ON ANTIOXIDANT ACTIVITY AND GRAIN YIELD OF DIFFERENT WHEAT GENOTYPES

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ABSTRACT

In order to evaluate the antioxidant activity of wheat in salinity stress conditions, an experiment with 27 wheat genotypes grown on two types of soil was conducted: solonetz (increased salinity) and chernozem (control), during two vegetation seasons (2015/2016 and 2016/2017). Analysis of DPPH radical scavenging activity and phenolic content (PC) were performed in different phenophases of wheat (tillering, stem elongation and heading). Genotypes showed significantly higher DPPH radical scavenging activity (9.82 mg trolox equivalents (TE) per mg of dry matter (d.m.)) and PC (8.15 mg gallic acid equivalents (GAE) per mg d.m.) under salinity stress conditions compared to values obtained on control (8.52 mg TE mg^{-1} d.m. and 7.13 mg GAE mg^{-1} d.m., respectively). All analyzed factors (genotype, soil type and year) had the highly significant influence on phenotypic variation of grain yield. Salinity stress reduced grain yield by 30%, whereas drought stress in 2016/2017 vegetation season reduced grain yield by 20%. Highly significant and positive correlations are present between grain yield and parameters of antioxidant activity in all growth stages of wheat and both soil conditions. Therefore, it could be possible to select salinity tolerant genotypes in early growth stages. DPPH scavenging activity and total phenolic content are in highly significant and positive correlation in all growth stages, which indicates that antioxidant activity is highly derived by phenolics.

Key words: Chernozem, correlation, DPPH, phenolic content, solonetz

INTRODUCTION

As one of the staple cereals in human nutrition, wheat has always occupied a central place in agricultural production. Due to the growth rate of the world population, the demand for wheat is expected to increase by 40% by 2030 (Dixon et al., 2009). Soil salinity is one of the main stress factors, especially in arid or semi-arid areas, which can significantly limit agricultural production (Borzouei et al., 2012), reducing the yield of the most important crops up to 50% (Dimitrijević et al., 2012). Approximately one billion hectares (or 7% of the total terrestrial area in the world) is affected by some form of salinity (Hossain, 2019). Soil type of solonetz occupies 150 million hectares in the world, and most of it is located in steppe where climate is characterized by annual precipitation that does not exceed 400 to 500 mm (Chesworth, 2008). Halomorphic soils in the European Union cover a total of 21,585 km^2, where the solonchak takes 11,728 km^2, and the solonetz 9,857 km^2 (Tóth et al., 2008).

Salinity stress could cause the production of Reactive oxygen species (ROS) in the plant (Hasanuzzaman et al., 2021). Production of ROS leads to oxidative stress in the plant, where the formed free radicals can cause cell extinction due to present oxidative processes, such as cell membrane lipid peroxidation, protein oxidation, enzyme inhibition and damage to DNA and RNA (Ashraf, 2009; Azizpour et al., 2010; Hasanuzzaman et al., 2021). All that was noticed affects the inhibition of plant growth and grain yield (Hendawy et al., 2017; Khokhar et al., 2017; Mansour et al., 2020). To avoid harmful effects of ROS production, plants have developed the antioxidant defense systems which contain enzymatic and non-enzymatic antioxidants that reduce the level of oxidative stress in plant cells (Ashraf, 2009; Sharma et al., 2012). Non-enzymatic components of the antioxidant system include major cellular redox buffers, ascorbate, glutathione and other thiol proteins, as well as tocopherols, carotenoids, and phenolic compounds (Sharma et al., 2012). Phenolic compounds are considered to be a major group of compounds that contribute to the antioxidant activity

(Sytar et al., 2018). Analysing the tolerance of wheat and barley genotypes to salinity based on grain yield is expensive and time-consuming, and therefore it's suggested that evaluation of tolerance to salinity is carried out at the early stages of plant development for these species (El-Hendawy et al., 2011; Turkyilmaz et al., 2011). Results obtained by El-Hendawy et al. (2011), Khayatnezhad and Gholamin (2010) and Turki et al. (2012) showed that wheat in the early stages of vegetative growth was sensitive to salinity stress. In addition, parameters in terms of salinity tolerance measured at early growth stages were correlated with salt tolerance parameters at maturity stage (El-Hendawy et al., 2011; Turki et al. 2012). El-Hendawy et al. (2017) state that the establishment of field trials is of great importance in wheat breeding programs aimed to increase the tolerance to soil salinity.

The aim of this study was to determine the influence of salt stress on grain yield and parameters of antioxidant activity analyzed in different growth stages. Furthermore, the goal is to establish the connection between the antioxidant activity and grain yield in different growth stages of wheat and to investigate the possibility of selecting salt tolerance genotypes at early growth stages.

MATERIALS AND METHODS

Experiment design and plant material

Field experiment was obtained in Randomized Complete Block Design with three replications during 2015/2016 and 2016/2017 vegetation seasons at two localities: Kumane (Banat, Vojvodina, 45.522ºN, 20,195ºE) and Rimski Šančevi (Novi Sad, Bačka, Vojvodina, 45,322ºN, 19,836E). The locality of Kumane was chosen due to solonetz soil type, while locality of Rimski Šančevi, where chernozem soil type was present, was chosen as a control locality. The experimental material consisted of 27 wheat genotypes (*Triticum aestivum* ssp. *vulgare*). The genotypes included in this study (local landrace Banatka, old Hungarian variety Bankut 1205, varieties created at Institute for Field and Vegetable Crops in Novi Sad: Jugoslavija, NS-rana 5, Renesansa and Pesma) were chosen on the basis of previous studies that tested the adaptability of these genotypes to abiotic stress, expressed at the Kumane locality. In order to increase genotypic variability, this research included old and modern varieties of the Centre for Small Grains in Kragujevac, as well as the local landrace Grbljanka grown in Montenegro (Table 1).

Table 1. Wheat genotypes included in investigation

Genotype code	Genotype	Year of approval	Genotype code	Genotype	Year of approval
G1	Banatka	Local landrace	G15	Jugoslavija	1980.
G2	Grbljanka	Local landrace	G16	Oplenka	1982.
G3	Bankut 1205	1953.	G17	Ljubičevka	1985.
G4	Kragujevačka 75	1966.	G18	Srbijanka	1986.
G5	Šumadija	1968.	G19	Šumadinka	1988.
G6	Kosmajka	1971.	G20	NS-rana 5	1991.
G7	Gružanka	1972.	G21	Renesansa	1994.
G8	Morava	1972.	G22	Pesma	1995.
G9	Zastava	1973.	G23	Aleksandra	2007.
G10	Kragujevačka 56	1975.	G24	Perfekta	2009.
G11	Orašanka	1976.	G25	Harmonija	2012.
G12	Kragujevačka 58	1977.	G26	Rujna	2013.
G13	Kragujevačka 78	1978.	G27	Premija	2013.
G14	Lepenica	1980.			

At both soil types, the analyzed genotypes were sown with continuous sowing, where the arrangement of genotypes was organized according to a randomized complete block design. The size of the basic plot was 2 m^2, the space between rows was 10 cm, and the distance between the plots was 25 cm. The usual agronomic practices for wheat production were applied at both localities. Harvesting was performed at the stage of physiological maturity, when the grain moisture was below 14%. The grain yield was determined by measuring the grain weight per square meter, on each plot, and converting into t ha^{-1}.

Soil conditions

Solonetz is an alkaline soil type with high clay content in the subsurface horizon and a high proportion of adsorption of sodium and / or magnesium ions (Tóth et al., 2008). The highest content of adsorbed Na+ was found in Bt,na horizon (13.45 cmol kg^{-1}), at a depth of 58 to 85 cm, while the lowest content of adsorbed Na was recorded in the surface layer (0.88 cmol kg^{-1}), Belic *et al*. 2014. Due to the high sum of exchangeable cations, and content of clay and exchangeable Na (containing more than 15% of adsorbed Na), as well as the alkalinity (pH>9) in Bt,na horizon, the solonetz is characterized as a soil of unfavorable physical and chemical properties (Belić et al., 2006). On the other hand, chernozem is considered an ideal soil type for agricultural production with a favorable, loamy, mechanical composition (Miljković, 1996). It is characterized by a good crumbly structure, stable aggregates and good water permeability. Also, it has a sufficient organic matter (3 to 4%) and plant nutrients and is characterized by a favorable water-air and heat regime (Hadžić et al., 2002).

Meteorological conditions

The mean monthly temperature at both localities (Kumane and Rimski Šančevi) during analyzed vegetation seasons (2015/2016 and 2016/2017) was similar. In both vegetation seasons, the average sum of monthly precipitation was higher at the locality of Rimski Šančevi (603 mm in 2015/2016 and 428 mm in 2017/2018), compared to amount of precipitation recorded at locality of Kumane (527.3 mm in 2015/2016 and 289.9 mm in 2016/2017). Warmer weather and small amount of precipitation affected the earlier ripening of plants in the 2016/2017 vegetation season, especially at Kumane locality (RHMZ, 2020), Figure 1.

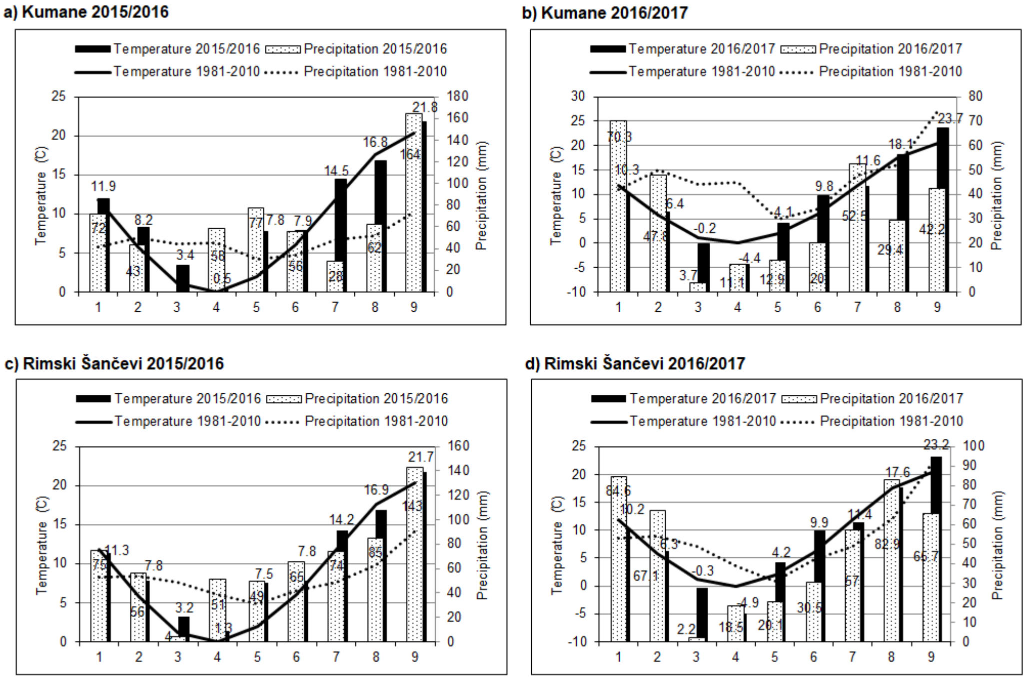


Figure 1. Monthly mean temperatures and sum of precipitation in Kumane (a, b) and Rimski Šančevi (c, b) in two vegetation seasons

Methods to evaluate the antioxidant activity

The antioxidant activity was analyzed in dry plant material. Sampling of the leaves from whole plant was carried out in three different wheat phenophases: tillering, stem elongation and heading. The average sample was made from each plot.

The plant material (0.2 g) was extracted with 70% acetone (10 mL) during 24 h in a dark. After extraction, the macerate was filtered through a qualitative filter paper. The extracts are stored in a refrigerator at a temperature of a 4°C, during 24h (until use for further analysis).

Determination of DPPH˙ scavenging activity. Free radical scavenging ability was measured using the stable DPPH (2.2-diphenyl-1-picrylhydrazine) radical (Lee et al., 1998). Extract (40 µL) was mixed with DPPH reagent (2 mL). The absorbance was measured at 517 nm, using UV/VIS spectrophotometer (Thermo Scientific Evolution 220, USA). Calibration curve was made using trolox as standard. The activity of removing the DPPH radical is expressed in mg equivalents of trolox per g of dry matter (mg TE g^{-1} d.m.). All measurements were performed in triplicate.

Determination of phenolic content. Phenolic content (TPC) of the wheat extract was determined according to the Folin-Ciocalteu method (Hagerman et al., 2000). Plant extract (20 µL) was mixed with 1.8 mL of deionized water, 0.2 mL of 20% sodium carbonate and 0.1 mL of Folin-Ciocalteu reagent, which was previously diluted (1:2) with distilled water. After incubation at room temperature for 30 min, the absorbance of the reaction mixture was measured at 720 nm using an UV/VIS spectrophotometer (Thermo Scientific Evolution 220, USA). The phenolic content was determined using a standard curve with gallic acid (0.005 to 0.05 mg ml^{-1}). The data were expressed in mg of gallic acid equivalents (GAE) per g of dry matter (mg GAE g^{-1} d.m.). All measurements were performed in triplicate.

Statistical analysis

The data obtained in this study were expressed as the mean of triplicate determinations. The analysis of variance was performed, with genotype and environment (phenophase, year and soil type) as fixed effects. All data and means were compared by the LSD test, at p<0.01 and p<0.05 levels of significance (Steel and Torrie, 1980).

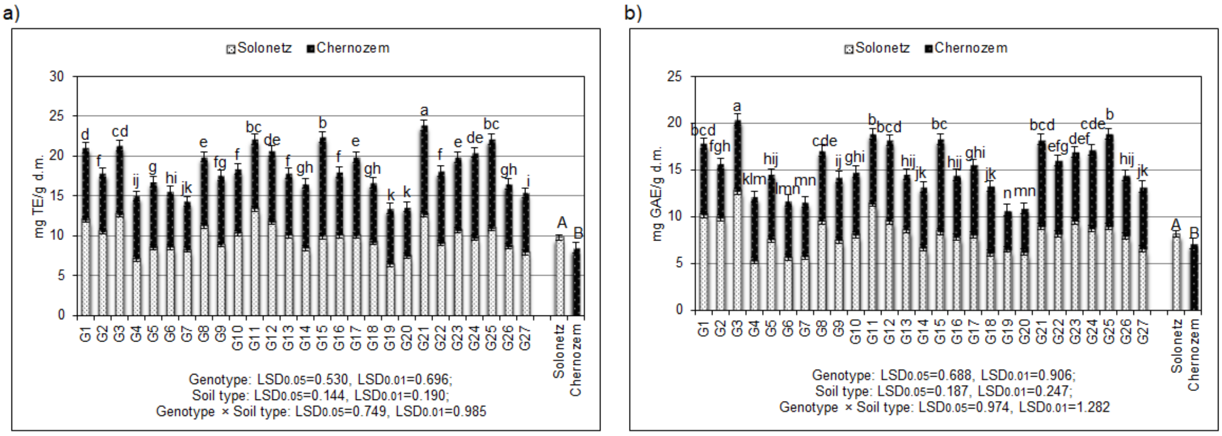
Correlations between analyzed characteristics were calculated in each environment and each phenophase and both localities (agro-ecological environments). Statistical analysis was performed using IBM SPSS Statistics 22.0, trial version (https://www.ibm.com/analytics/spss-trials).

RESULTS

DPPH$^{\bullet}$ scavenging activity and phenolic content

DPPH$^{\bullet}$ scavenging activity and phenolic content significantly varied under the influence of soil type (Figure 2). In average for both vegetation seasons, the significantly higher ($p<0.01$) DPPH$^{\bullet}$ scavenging activity and phenolic content was obtained on solonetz (9.82 mg TE g^{-1} d.m. and 8.15 mg GAE g^{-1} d.m.) in relation to the chernozem (8.53 mg TE g^{-1} d.m. and 7.13 mg GAE g^{-1} d.m).

Factor of genotype had a significant effect on analyzed parameters. The highest DPPH$^{\bullet}$ scavenging activity and phenolic content were examined by the genotypes Orašanka (G11) (13.41 mg TE g^{-1} d.m, 10.24 mg GAE g^{-1} d.m.) and Bankut 1205 (G3) (12.71 mg TE g^{-1} d.m., 12.74 mg GAE g^{-1} d.m.) when grown on solonetz. On chernozem soil type, the highest DPPH$^{\bullet}$ scavenging activity and phenolic content were measured in genotypes Jugoslavija (G15) (12.42 mg TE g^{-1} d.m., 9.89 mg GAE g^{-1} d.m.), Renesansa (G21) (11.22 mg TE g^{-1} d.m., 9.30 mg GAE g^{-1} d.m.) and Harmonija (G25) (11.22 mg TE g^{-1} d.m., 9.91 mg GAE g^{-1} d.m.), Figure 2.



Bars marked with different letters differ significantly at the level of 5%

Figure 2. Influence of genotype, soil type and genotype × soil type interaction on variation of DPPH$^{\bullet}$ scavenging activity (a) and phenolic content (b)

Factor of year had a dominant effect ($p<0.01$) on the variation of DPPH$^{\bullet}$ scavenging activity, while the phenolic content was not affected by this factor ($p>0.05$). In 2016/2017 vegetation season, DPPH$^{\bullet}$ scavenging activity was 12.05 mg TE g^{-1} d.m. and was twice as high as the value measured in 2015/2016 vegetation season (6.30 mg TE g^{-1} d.m.), while almost equal values of phenolic content were recorded in both vegetation seasons (Figure 3).

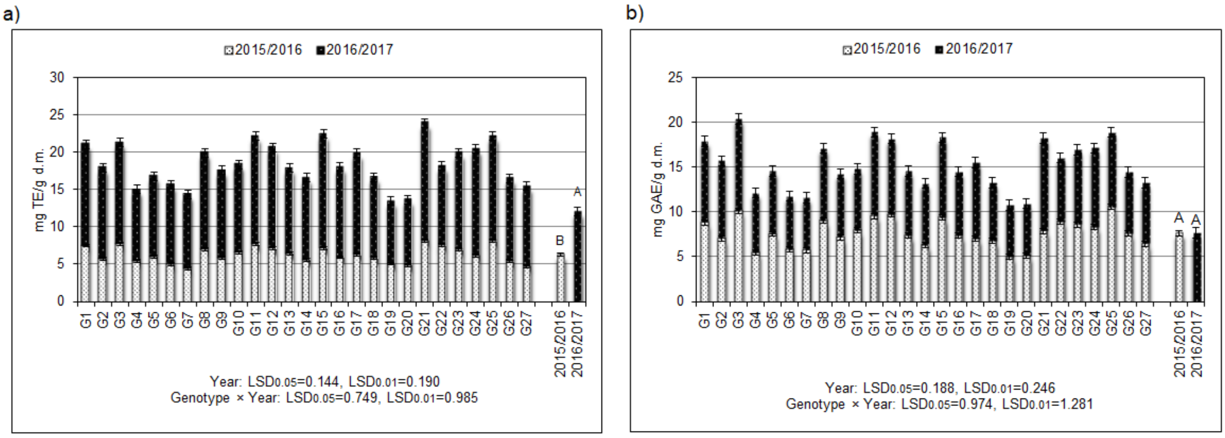


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Figure 3. Influence of year and genotype × year interaction on variation of DPPH$^{\bullet}$ scavenging activity (a) and phenolic content (b)

Factor of phenophase had siginificant effect on DPPH$^{\bullet}$ scavenging activity and phenolic content variation. The highest DPPH$^{\bullet}$ scavenging activity was measured in phenophase of stem elongation (10.87 mg TE g^{-1} d.m.), while the highest value of phenolic content was recorded in phenophase of heading (7.86 mg GAE g^{-1} d.m.). Also, interactions of phenophase × soil type and phenophase × year were statistically significant ($p<0.01$) for both analyzed parameters (Figure 4). The highest differences between soil types in measured values of analyzed parameters were observed in phenophase of stem elongation, while the lowest differences were recorded in phenophase of tillering. The highest difference in DPPH$^{\bullet}$ scavenging activity between vegetation seasons was established in phenophase of tillering, while the highest difference in phenolic content was observed in phenophase of heading. The DPPH$^{\bullet}$ scavenging activity significantly ($p<0.01$) varied under the influence of soil type × year ineraction, where the difference between soil conditions in DPPH$^{\bullet}$ scavenging activity was more pronounced in the 2015/2016 vegetation season. Interaction of soil type × year did not significantly ($p>0.05$) affect the variation of phenolic content (Figure 4).

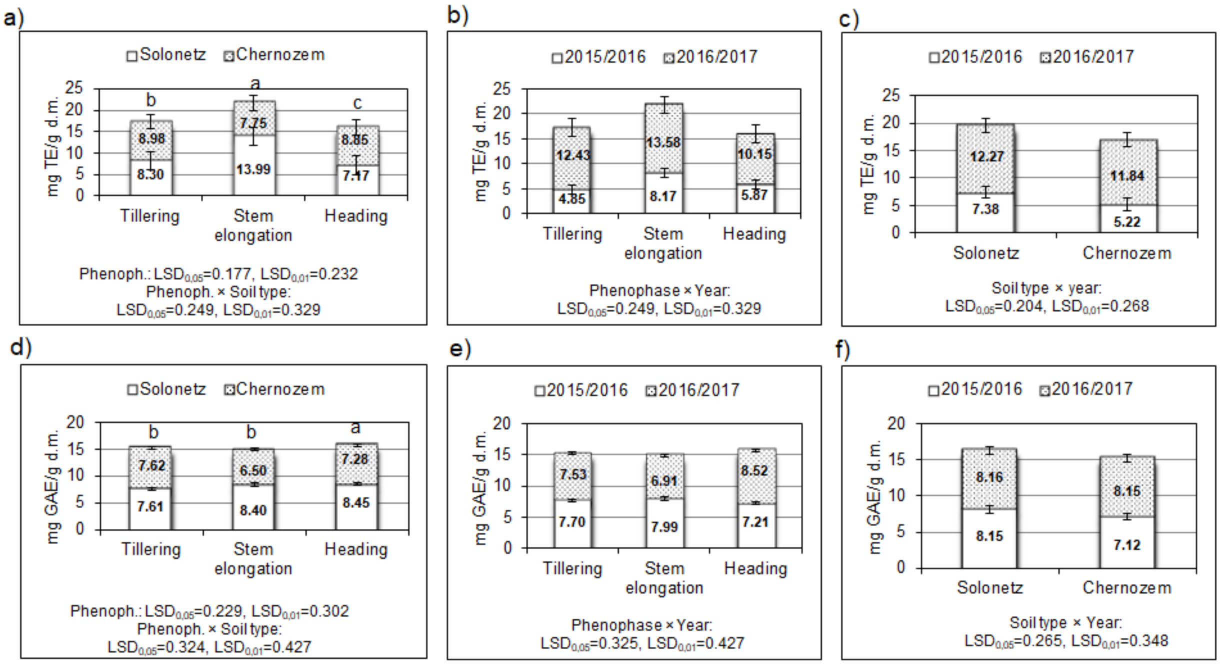

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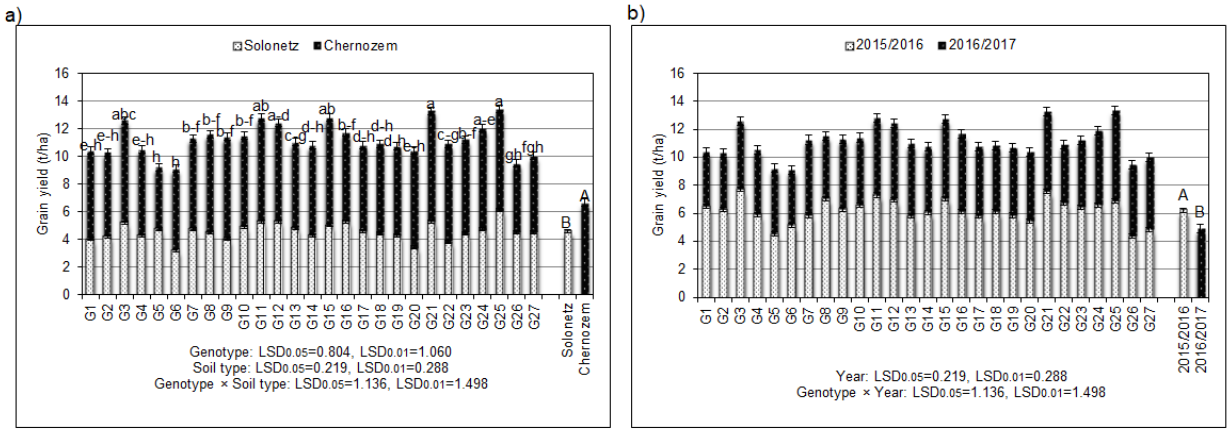
Figure 4. Influence of phenophase, phenophase × soil type, phenophase × year (b) and soil type × year on variation of DPPH$^{\bullet}$ scavenging activity (a, b, c) and phenolic content (d, e, f)

Grain yield

The grain yield significantly varied under the influence of genotype, soil type, year and their interactions (Figure 5). The factor of soil type had the highest effect on grain yield variation. The result indicates that salinity led to reduction in grain yield by 30% (4.5 t ha^{-1} on solonetz, 6.6 t ha^{-1} on chernozem). Similar effect on grain yield variation had factor of year, where grain yield in 2015/2016 vegetation season (6.2 t ha^{-1}) was significantly higher ($p<0.01$) in relation to the grain yield achieved in 2016/2017 vegetation season (4.9 t ha^{-1}). Interaction of genotype × soil type and genotype × year significantly affected the grain yield variation as well. On solonetz, genotype Harmonija achieved the highest grain yield (6.1 t ha^{-1}). This genotype had the highest value in 2016/2017 vegetation season, while in 2015/2016 the highest value of grain yield was observed at genotype Jugoslavija (7.1 t ha^{-1}). On chernozem soil type, the highest average value of grain yield was found in genotype Renesansa (G21) (8.0 t ha^{-1}), Figure 5.



Bars marked with different letters differ significantly at the level of 5%

Figure 5. Influence of genotype, soil type, genotype × soil type (a) and year, genotype × year (b) on variation of grain yield

Correlation between antioxidant activity parameters and grain yield

In order to analyse the relationship of grain yield and parameters of antioxidant activity, single correlations were conducted for both soil conditions separately during two vegetation seasons (Table 2). The values of grain yield and DPPH scavenging activity were in a significant and positive correlation, in all phenophases and in both soil conditions. On solonetz, the highest value of correlation coefficient between grain yield and DPPH scavenging activity is present in the phenophase of stem elongation (0.528^{**}), followed by the value in phenophase heading (0.481^{**}) and tillering (0.444^{**}). On chernozem, grain yield had the strongest and the most positive correlation with DPPH scavenging activity in phenophase of heading (0.633^{**}), then in phenophase of stem elongation (0.485^{**}) and in phenophase of tillering (0.329^{*}), Table 6. Significant and positive correlations between grain yield and phenolic content were found in all phenophases and on both soil types during two vegetation seasons (Table 2). The highest value of correlation coefficient was observed in stem elongation (0.456^{**} on solonetz and 0.485^{**} on chernozem), while the lowest correlation was found in growth stage of tillering (0.383^{*} on solonetz and 0.322^{*} on chernozem), Table 2.

Table 2. Correlation coefficients between grain yield and parameters of antioxidant activity in three phenophases of wheat genotypes grown on two soil types and two vegetation seasons

Solonetz		**Chernozem**		**Solonetz**		**Chernozem**	
DPPH scavenging activity	Grain yield	DPPH scavenging activity	Grain yield	Phenolic content	Grain yield	Phenolic content	Grain yield
Tillering	0.444^{**}	Tillering	0.329^{*}	Tillering	0.383^{*}	Tillering	0.322^{*}
Stem elongation	0.528^{**}	Stem elongation	0.485^{**}	Stem elongation	0.456^{**}	Stem elongation	0.485^{**}
Heading	0.481^{**}	Heading	0.633^{**}	Heading	0.423^{**}	Heading	0.411^{**}

*Significant at the p<0.05; **Significant at the p<0.01

DISCUSSION

In order to avoid harmful effects of salinity, due to the production of reactive oxygen species (ROS), plants have developed an antioxidant defense system which includes enzymatic and non-enzymatic components. Therefore, changes in the content of antioxidant levels may act as a signal for ROS scavenging processes (Hasanuzzaman et al., 2021). Phenolic compounds are among the main non-enzymatic components of the plant antioxidant system (Ashraf, 2009; Sytar et al., 2018). Accumulation of phenolic compounds could be one of the cellular adaptive mechanisms for scavenging oxygen free radicals during stress (Mohamed and Aly, 2008). The analyzed genotypes showed significantly higher DPPH scavenging activity and phenolic content when grown on high saline soil, such as solonetz, compared to chernozem. Furthermore, the results obtained by Kumar et al. (2017) and Kiani et al. (2021), confirmed that the salinity significantly increases phenolic content in the wheat plant. Also, salinity stress caused significant increases the DPPH scavenging activity in wheat leaf (Kiani et al., 2021).

The highest share in total variation of DPPH radical scavenging activity had vegetation season.

In the dry 2016/2017 vegetation season, antioxidant activity was twice as high as the antioxidant activity in the more favorable 2015/2016 vegetation season. Sarker and Oba (2012) noted that DPPH radical scavenging activity significantly increased in *Amaranthus tricolor* with the increased of drought stress. The variation in phenol content was mostly influenced by the factor of genotype, while the factor of vegetation season did not have a significant influence. Predominant effect of genotype on the accumulation of total phenolic acid in wheat grains, under elevated temperatures, was found by Shamloo et al. (2017).

In condition of salinity stress, the highest values of DPPH radical scavenging activity and phenolic content were shown in the leaves of old varieties Bankut 1205 and Orašanka, such as in local landrace Banatka. On the other hand, newer varieties Harmonija, Renesansa and Jugoslavija had the highest DPPH radical scavenging activity and phenolic content on chernozem. Banjac (2015) got similar results at the same locality, on the solonetz soil type, where he stated that the genotype Bankut 1205 had the highest phenolic content in the phenophase of heading and the highest DPPH scavenging activity in the phase of milk maturity. The same author states that the local landrace Banatka was characterized by higher neutralization ability of free DPPH radicals in relation to newer varieties.

Soil salinity significantly reduced the grain yield of the examined genotypes by 30%. This is in accordance to the results obtained by Dimitrijević et al. (2012), where they found that increased soil salinity on solonetz, reduced grain yield by 35 to 50% in relation to yield achieved on nonsaline soil (chernozem). Also, Khokhar et al. (2017), Nadeem et al. (2020) and Mansour et al. (2020) found a significant reduction in wheat grain yield under the influence of salinity stress. Phenolic components are the main group of compounds that contribute to the antioxidant activity of cereals (Zendehbad et al., 2014). Therefore, the difference in antioxidant activity between wheat genotypes may be due to the different composition of the phenolic compounds present.

Parameters of antioxidant activity were in highly significant and positive correlations with grain yield in all analyzed phenophases. Therefore, these antioxidant parameters could be used as selection criteria, even in the early growth stages of plant growth, especially in conditions of high salinity. Similar results have been reported by El-Hendawy et al. (2011), where they found that rank of wheat genotypes for salt tolerance in terms of parameters measured at seedling stages matched with their ranking in terms of grain yield at final harvest. The same authors emphasized that analyzed genotypes were very sensitive to salinity stress at early growth stage, and that it is possible to select salt tolerance of wheat genotypes in seedling stage. Turki et al. (2012) reported similar results, where they found a highly positive correlation between measured parameters in maturity and tillering stage of wheat at high salinity conditions.

CONCLUSION

The analyzed genotypes had a significantly higher values of DPPH scavenging activity and the phenolic content on solonetz soil type (in conditions of increased salinity), compared to the values achieved on chernozem soil type. The old genotypes Bankut 1205, Orašanka and Banatka had the highest antioxidant activity on solonetz, while on chernozem, the highest antioxidant activity was shown by newer genotypes - Harmonija, Renesansa and Jugoslavija. Thus, this shows that even older genotypes can serve as a desirable genetic resource for breeding for salinity tolerance. Also, this is of practical importance for wheat producers in areas of increased soil salinity. Highly significant and positive correlations are present between grain yield and parameters of antioxidant activity measured in all phenophases of wheat. The correlation between grain yield and antioxidant activity in early growth stages indicates that the parameters of antioxidant activity could be used as a selection criteria, especially in stressful conditions due to salinity. The association of phenolic content and the DPPH radical scavenging activity in tested genotypes indicates that phenolic compounds have a large share in the antioxidant activity of wheat.

ACKNOWLEDGEMENTS

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ASSOCIATION MAPPING OF GERMINATION AND SOME EARLY SEEDLING STAGE TRAITS OF A TURKISH ORIGIN OAT COLLECTION

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ABSTRACT

In recent years, oat plant especially for hay yield is on high demand in Turkey. In this study, 167 Turkish oat landraces were evaluated for germination and some early seedling stage traits and genotyped by 6K SNP Chip assay to detect candidate markers using association mapping analysis. The variation in oat genotypes for germination and other investigated traits was significant 5%, except for germination rate (GR). In the research, principal component (PC) 1 and PC2 value was found 41.5% and 21.5%, respectively, explaining the 63% of the total variation. According to the results of the association mapping analysis a total number of 33 candidate markers were observed, eight candidate markers for germination rate, five candidate markers for germination ratio (GP), five markers for radicle length (RL), five markers for coleoptile length (CL), four markers for plumula length (PL), and six markers for seed vigor index (VI). These candidate markers identified in the study for germination and early seedling stage traits could be used in future studies.

Keywords: Association mapping, germination, principal components, oats

INTRODUCTION

Turkey is one of the important gene centers of oats that containing many wild and cultivated varieties (Dumlupinar et al., 2011a). Local varieties are important natural genetic resources that offer opportunities and innovations to plant breeders in the development of high yielding and high quality new varieties.

The success of seed germination in a plant and the frequency of normal seedling management are tremendously decisive factors in the production of crop plants, which are of both economic and ecological importance. Germination is considered one of the most critical sages in the life cycle of the plants, since it is under the influence of several biotic and abiotic factors. The process of germination begins with the water uptake of the mature dry seed and ends with the emergence of the radicle from the seed coat (Rajjou et al., 2012). SNP markers identify the locus resulting from single nucleotide differences when two alleles are observed in a population. SNPs represent the most common type of sequence polymorphism in plant and animal genomes. SNP markers have become widely used in many plant species due to their stability, ease of use, extremely low mutation rate, and high genotyping capacity. The use of SNPs allows for the easy selection of many traits in a large breeding population, turning selection into profit with less risk. Today, many molecular plant-breeding programs are based on SNP markers, and germplasm, gene sources represent the power of genetic interactions essentially to differences in the genome in mapping genes that control certain traits in breeding lines (Morgil et al., 2020).

The complex phenotypic variations or background architecture of many agricultural traits are effected by different environments. Association mapping, also known as linkage disequilibrium (LD), is a powerful and effective tool for revealing the inheritance mechanism of complex traits in identifying candidate genes responsible for the association between phenotypic and genotypic data by revealing historical and ancestral recombination events at the population level. As a new alternative approach to traditional relationship mapping, association mapping has three advantages: 1) increased map density, 2) natural linkage with a wide and unique diversity without the need for the more costly and effort-intensive mapping populations (F_2, BC and RIL) used in traditional linkage analyses, 3) greater allele diversity (Yu and Buckler, 2006). Based on the size and point of interest of a particular study, association mapping falls into two subcategories. These approaches are candidate gene-based association mapping, which associates causal polymorphisms in candidate genes that are stated to be involved in controlling phenotypic variation for specific traits, and genome-wide association studies known as GWAS, which search for genetic variation in the whole genome to find association peaks and points for various traits.

Association mapping is an application that focuses on LD to examine the relationship between phenotypic variation and genetic polymorphism (Mackay, 2001). Association mapping uses large mixed samples from the natural population sets, which are populations developed from multi-parental hybrids of related species, or collections of breeding material, including cultivars. Samples should be capable of as much genetic variation as the current population collection is useful in practice. In this study, it was aimed i) to reveal germination and early seedling stage traits of Turkish origin oat genotypes obtained from various gene banks, ii) to find out relationships among investigated traits with principal component analysis, iii) to determine candidate markers related with germination traits via association mapping analysis using 6K SNP Chip assay.

MATERIALS AND METHODS

Plant Materials

In the study, 167 local oat genotypes across Turkey obtained from USDA-ARS-National Gene Bank (United States Department of Agriculture- Agricultural Research Service, National Small Grains Collection), and four commercial cultivars, Arslanbey, Kahraman, Kirklar and Yeniceri were used as plant materials. The pedigree of the landraces were presented in Comertpay et al. (2018).

SNP Genotyping

Single nucleotide polymorphism (SNP) markers developed by Oliver et al. (2011) were used for association mapping analysis in current study. The 6K SNP Chip was applied using Illumina Infinium Technology by General Mills, Inc., Minneapolis, MN, and USDA–ARS. Details about SNP development might be launched in former studies (Oliver et al. 2011; 2013)

Phenotyping

The research was arranged in an augmented experimental design with six replications of commercial cultivars in order to examine germination and some early seedling stage traits under laboratory conditions and to identify candidate markers related to these characteristics in 2020. For this purpose, seeds were surface sterilized with 1% NaOCl (sodium hypochlorite) solution. Twenty-five seeds of each genotype were placed into double-layer sterile filter papers in autoclaved petri dishes and put in the culture room at 25 °C for 8 days for germination. Seeds were considered germinated when the radicle of the seeds reached at least 2 mm.

Germination rate was measured in the 4th day of the experiment; on the 8th day; germination and some early seedling stage characteristics such as germination ratio, coleoptile length, plumula length, radicle length, seed vigor index were phenotyped and the data were recorded.

Data Analysis

The phenotypic data were subjected to analysis of variance (F test) according to the augmented experiment design. The Duncan test was used to compare mean data. Principal component values were examined using the biplot analysis approach over the average data of the features. All phenotypic data analysis was performed using the JMP 15.1 statistical package program (SAS Institute Inc, 2020). Significance was determined as 5% likelihood level unless else indicated.

GAPIT Version 2 (Tang et al., 2016) software was used to determine the relationship between some germination characteristics in oats and SNP markers in the 6K chipset. In the study, those with a minor allele frequency value of less than 5% (MAF < 0.05) were extracted and high quality robust SNPs were obtained. Heterozygosity frequency was calculated both between individuals and between markers. Afterwards, in the same program, the marker density, which is an important criterion for calculating the LD (linkage disequilibrium) between the markers, was determined, and the comparison between the LD decay at physical distance and the marker density was presented in the plot graphic.

In the LD analysis, the r^2 value of the LD decrease, which indicates the mutual mutation and recombination of the markers with each other, was calculated in the spread graph. In the association study, PCA + K data was used in the Compressed Mixed Linear Model (CMLM) method, which gathers the genotypes into groups and links genetic values of clusters as random effects in the design, thus promotes statistical effectiveness and saves time on large examples, and candidate gene mapping analysis was performed (Zhang et al., 2010). Because of association analysis, the genomic position of the markers on different chromosomes on the x-axis, the logarithmic ($-\log_{10}(p)$) representation of the p-values on the y-axis are presented in the Manhattan plot. In addition, following the method of VanRaden (2008), a heat map showing the genetic and kinship relationship based on the kinship matrix of the genotype individuals and observed and expected and observed a quantile-quantile (Q-Q) plot was created.

RESULTS AND DISCUSSION

Phenotypic Traits

The phenotypic data obtained as a result of the study carried out to determine the performance of local oat genotypes on germination and some early seedling stage traits examined were statistically evaluated and the data for these traits are shown as histogram (Figure 1). The variation in local oat genotypes was found significant at the 5% significance level, except for germination rate.

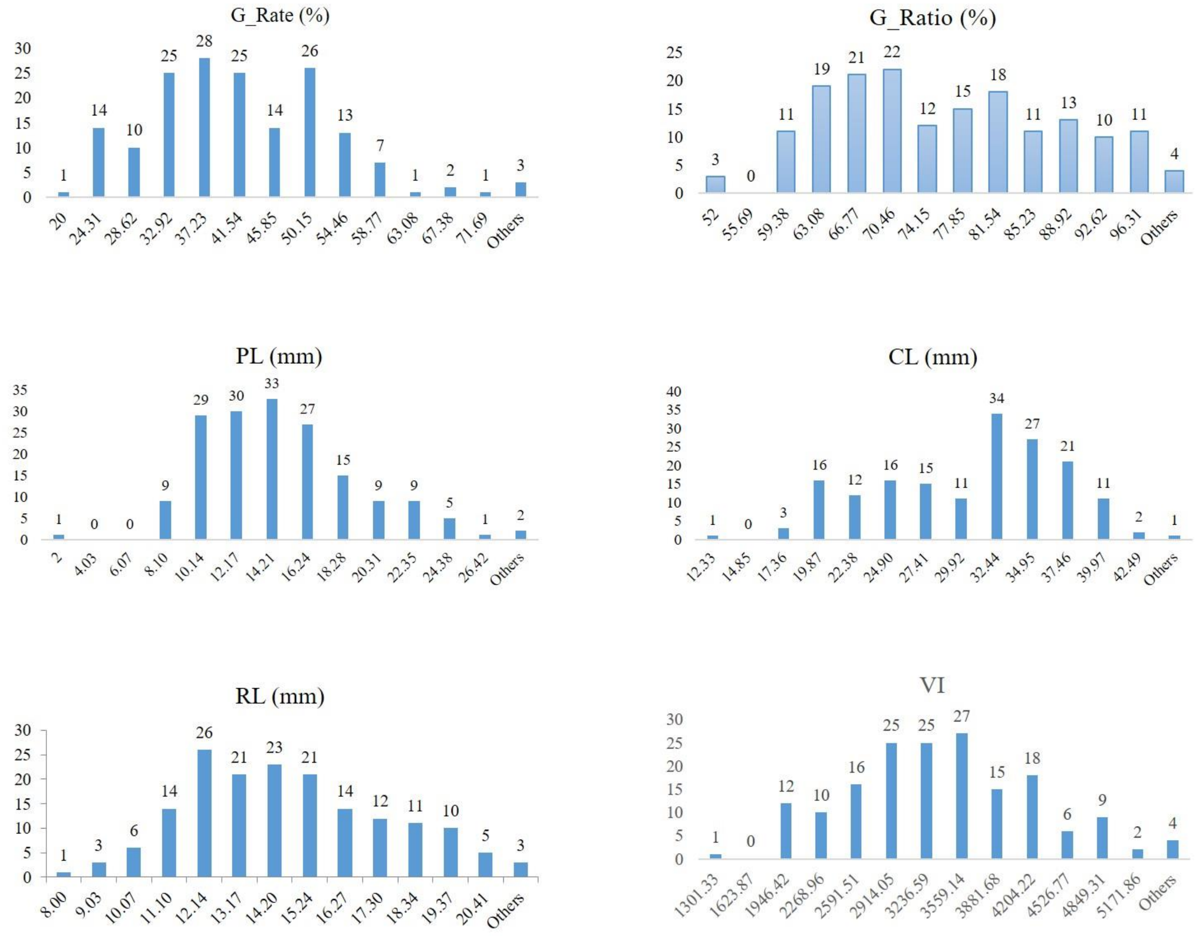

Figure 1. A histogram was created for investigated traits of oat genotypes

Germination rates of local genotypes varied between 20-76%, and between 39-43% among the control varieties (Figure 1). While TL97 reached the highest germination rate value (76%), TL55 got the lowest value (20%). Akyol (2014) reported that there was a significant variation among oat cultivars in terms of germination rate and the germination rate was significantly low, as they require a long period to germinate. Germination ratio among local oat genotypes were found between 52-100% and 80-83% among the control varieties (Figure 1). Genotypes TL10, TL11 and TL149 had the highest germination ratio values (100%), while TL113, TL119 and TL252 had the lowest values (52%). Dumlupinar et al. (2011b) reported significant variations in oat germination ratio and they reported that the hull rates and other seed characteristics of the genotypes played an important role on germination and emergence rates after sowing. Coleoptile length values varied between 12.33-45.00 mm and between 23.65-25.95 mm among the control cultivars (Figure 1). While TL53 genotype reached the highest coleoptile value with 45 mm, TL7 genotype had the lowest value with 12.33 mm. Radicle length among tested local oat genotypes varied between 8.00 and 21.44 mm, and between 12.72 and 13.49 mm in control cultivars. Among the oat genotypes, TL343 had the longest radicle length with 21.44 mm, while TL7 genotype had the shortest radicle length with 8.00 mm. Oner et al. (2018) reported that radicle length showed a significant change depending on to the severity of stress conditions. Plumula length values were observed between 2.00-28.45 mm, and between 14.79-17.13 mm in control cultivars. While TL326 genotype had the highest plumula length with 28.45 mm, TL128 genotype had the lowest value with 2 mm. Oner et al. (2018) found significant variations in oat genotypes in terms of plumula length, in agreement with our results. Seed vigor index ranged from 1301.33-5360.00 and 2986.26-3171.88 among the control varieties (Figure 1). In terms of seed vigor indices, TL149 genotype had the highest indices with 5360, while TL 7 genotype had the lowest with 1301.33. Gungor et al. (2017) stated that oat genotypes showed significant differences in terms of seed vigor index, and they reported that the cultivar Yeniceri had the highest seed vigor index.

Principal Component Analysis

Principal component (PC) biplot analysis plays an important role in selecting genotypes with superior adaptability in plant breeding programs, but they also actively benefit not only in the adequate evaluation of genotypes, but also in determining the positive or negative relationships of traits with each other. (Yan and Tinker, 2006). In our study, the PC1 value was found 41.5% and the PC2 value was 21.5%, explaining 63% of the total variation. It was determined that there were strong positive relationships between germination rate and germination ratio, radicle length and coleoptile length, seed vigor index and coleoptile length, and between seed vigor index and radicle length in the local oat genotypes investigated in the study. In addition, significant and negative correlations were determined for plumula and coleoptile lengths, and plumula and radicle lengths (Figure 2). Gungor et al. (2021) stated a 50.3% of the total variation, with 33.9% PC1 and 16.4%PC2 values, which is consistent with our findings.

Figure 2. Principal component biplot analysis of oat genotypes and investigated traits

Association Mapping Analysis

In the study, the 4599 SNP markers obtained from 6K SNP assay were filtered down to 2306 SNP markers and the heterozygosity frequencies of both were determined. (Figure 3a). Next, the marker density, which is an important criterion for calculating the LD between marker pairs, was tested and the histogram plot showed whether the markers were adequately suited to a good LD density by comparison between the marker density and LD degradation (Figure 3b). When the distribution states of the SNPs in the spread graph are examined, their frequencies continue in an increasing trend after 1000 and in 2000; the accumulation frequencies were determined to start from 0.6 and end at 1.0. In order to determine the population structure, the eigenvalue, where the PC value is determined as optimal K, by entering the R scripts with the current parameter settings in GAPIT, the heat map plot based on the kinship matrix was created with the EMMA algorithm based on the model of 3D PC and Van Raden (2008). While there was a significant change in the break point of the 4th PC from the variance components, this situation showed that the first 4 main components reflected the accumulation frequency well and explained 50% of the variance components (Figure 3c). In the 3D PC pilot, it was determined that a significant part of the genotypes were tightly clustered close to PC2, while the other parts were located on the left side close to PC3 and diverged from each other (Figure 3d). Considering the mating status of genotypes in the bidirectional kinship matrix; It was determined that the population structure was divided into 4 different groups considering their ancestral origins, and a similar result was obtained from the heat map of the kinship matrix analysis (Figure 4). Wang et al. (2017) observed a significant change in the first 8 PC values of the variance components in the population structure and kinship analyzes performed as a result of genotyping with 90K Illumina SNP markers. It was determined that their findings were different from ours by revealing that their wheat genotypes were separated into different clusters in the three-dimensional plot and that the panel was classified according to three groups in the dendrogram of the heat map of the kinship matrix.

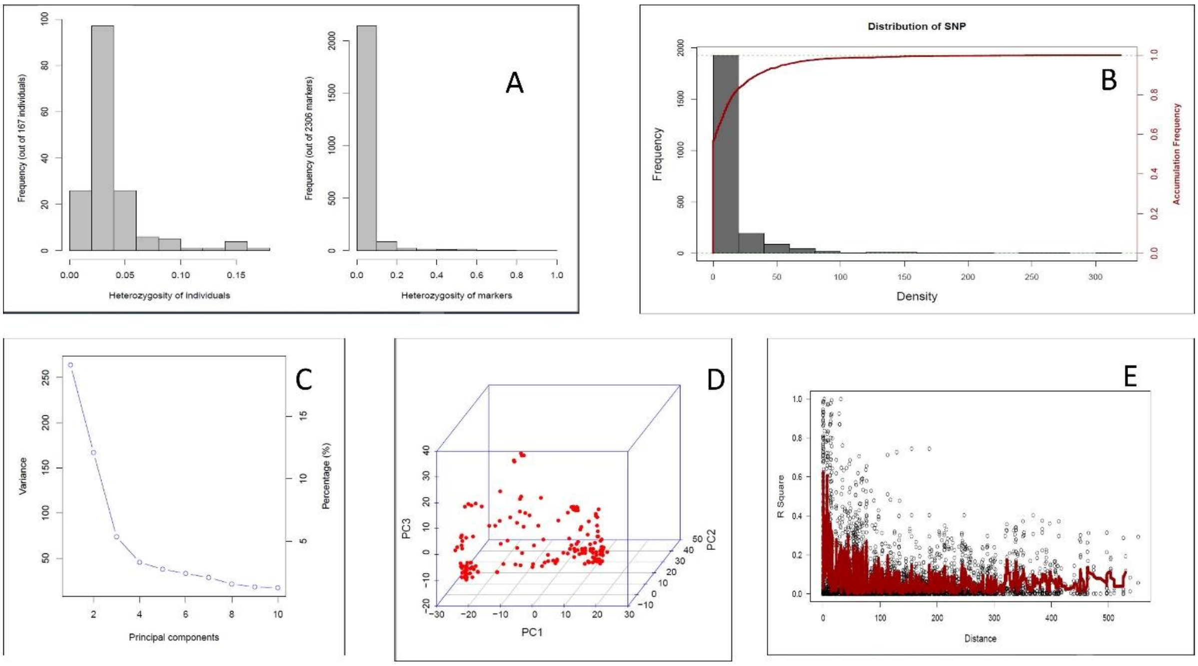

Figure 3. Marker properties of SNP markers (a) Heterozygosity frequencies of 6K SNP chip markers and 167oat individuals, (b) Histogram of marker densities and degradations, (c) principal components of the marker variables, (d) 3D principal components of the SNP markers, (e) graphic of the LD degradations

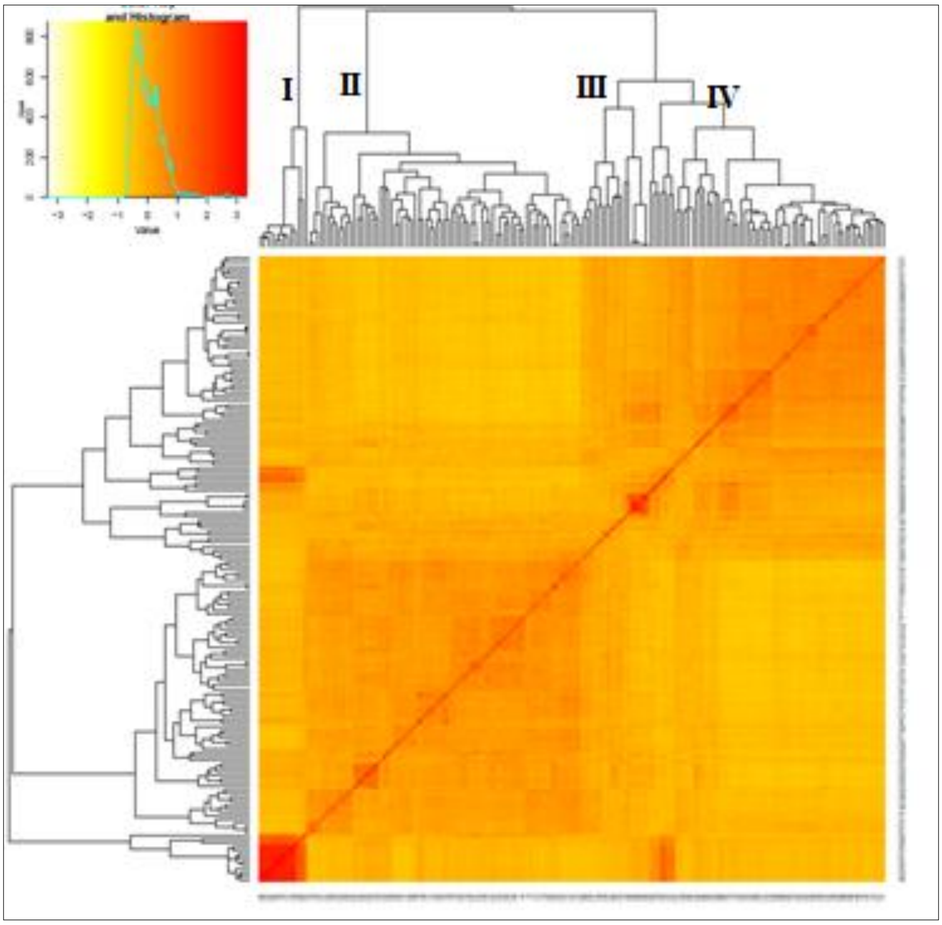

Figure 4. Heat map of genetic and kinship relationship based on the kinship matrix

Linkage Disequilibrium

The concept of LD, first described by Jennings in 1917 and first expressed numerically by Lewtonin in 1964, is a phenomenon that describes the non-random relationship of alleles at different loci in individuals in a population (Abdurakhmonov and Abdukarimov, 2008). Genetic drift, selection, mutation rate, autogamy, epistatic interaction, genetic isolation, population size, genomic rearrangements in chromosomes are important factors responsible for LD increase, as well as being followed in an attitude that varies from population to population, from individual to individual, from species to species. It has different traces as a shareholder in the success of association mapping (Flint-Garcia et al., 2003; Nadeem et al., 2018).

In the spread graph of LD degradation shown here, the coverage of the SNP markers in the area covered and their relationship with each other might be seen. The LD decrease was found to average when it fell to half of the maximum r^2 value. In the research findings, the LD decreased from 0.6 to 0.3 at a short distance (Figure 3e).

Candidate Gene Association Mapping Analysis

While establishing the relationship between phenotype and genotype in candidate gene association mapping analysis, the single locus-based compressed mixed linear model statistical method, which is one of the most popular methods reduces the type-I error (false positives) caused by kinship and population structure. As it is a common problem, and ensures the reliability of the test with the inevitability of a large amount of computational power, it has opened the door to the search for different algorithms (Sakiroglu, 2020). By taking the average of the data of the six traits examined in the study, the correlation analysis was carried out in the CMLM model. The candidate markers associated with each trait, the locations of the QTLs in the chromosomal regions were observed in the Manhattan plot graphs. The observed and expected P-values were presented in Q-Q plot graph (Figure 5). The significance threshold range of each of these markers was determined on the basis of $-\log_{10}(p) \geq 2.50$, and a false discovery rate (FDR) described by Benjamini and Hochberg (1995) was used to determine statistical significance threshold (α=0.05) using SAS Multtest procedure (SAS Institute Inc, 2020).

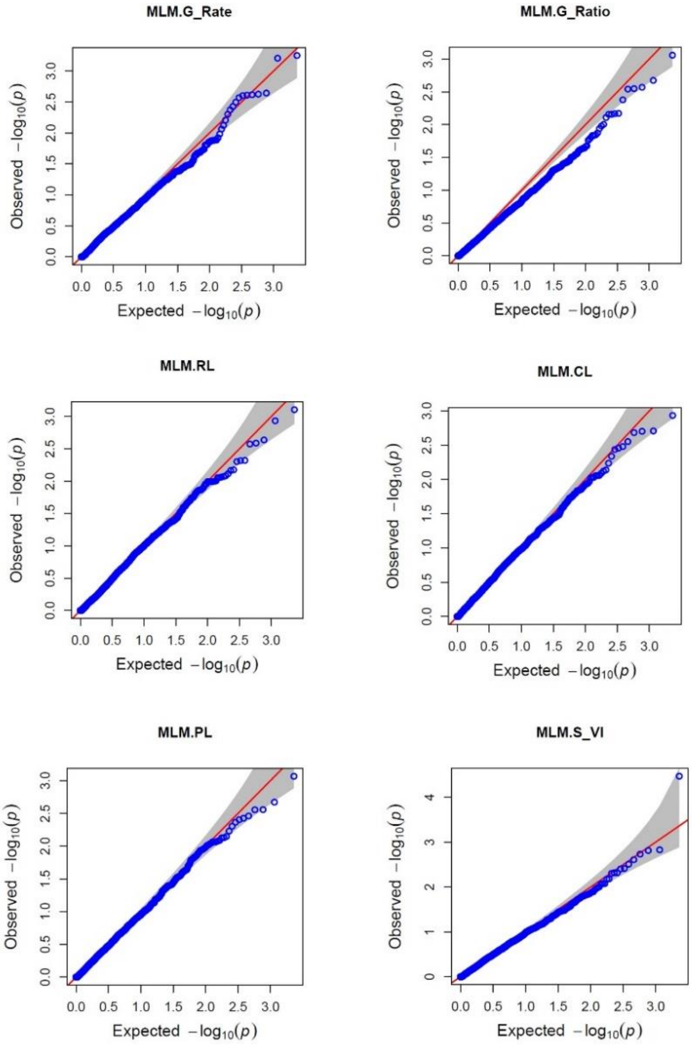

Figure 5. Q-Q Plot of the observed and expected $-\log_{10}(p)$ values

As a result of the candidate gene association analysis conducted out with the CMLM model using the mean data for the germination rate of the germination traits, eight candidate markers (values $-\log_{10} \geq 2.50$ and $p<0.01$) were determined (Table 1 and Figure 6a). The phenotypic variance (r^2) of the germination rate, which explains the percentage of the marker, varied between 0.10 and 0.12, and the minor allele frequency (MAF) value between 0.01 and 0.40 for all markers (Table 1). The candidate markers were located on chromosomes 19, 15, 6, 11, 6, 8, 11 and 6 located in positional distances of 221, 164, 1023, 88, 1023, 1628, 37 and 1002 cM longs, respectively. GMI_DS_LB_1076 marker was taken place in Mrg19 linkage group which is consisted with Chaffin et al. (2016). The GMI_GBS_50325 marker was localized in the Mrg15 linkage group, which was positioned in the Mrg15 linkage group in framework consensus map and in the Mrg11 linkage group in oat reference consensus map (Bekele et al., 2018). The locations of candidate markers previously reported that associated with germination rate in Mrg6, Mrg8, Mrg11, Mrg15 and Mrg19 linkage groups were also in harmony with our findings (Lin et al., 2014; Chaffin et al., 2016; Brooke, 2017; Bekele et al., 2018).

Table 1. Associated loci with germination and root traits identified in 167 local oat genotypes

	SNP	Chromosome	Position	P Value	r^2	MAF	FDR
Germination Rate	GMI_DS_LB_1076	19	221	0.00562	0.12	0.03	0.0031
	GMI_GBS_50325	15	164	0.00622	0.11	0.24	0.0031
	GMI_ES14_c2965_193	6	1023	0.002275	0.10	0.06	0.0033
	GMI_ES02_c24507_561	11	88	0.002364	0.10	0.22	0.0033
	GMI_ES_LB_9817	6	1023	0.002415	0.10	0.05	0.0033
	GMI_GBS_13916	8	1628	0.002454	0.10	0.12	0.0033
	GMI_ES14_c5428_351	11	37	0.002503	0.10	0.40	0.0033
	GMI_GBS_95238	6	1002	0.002694	0.10	0.01	0.0033
Germination Ratio	GMI_ESI_c12749_234	21	1720	0.000875	0.07	0.43	0.0028
	GMI_ES05_c1006_442	23	232	0.00212	0.06	0.17	0.0028
	GMI_ES05_c2760_657	1	866	0.002698	0.06	0.41	0.0028
	GMI_GBS_95238	6	1002	0.00283	0.06	0.01	0.0028
	GMI_ES14_ lrc18344_662	20	2491	0.002873	0.06	0.46	0.0028
Radicle Length	GMI_ES_LB_11026	20	2035	0.000782	0.11	0.46	0.0026
	GMI_ES_LB_11028	20	2035	0.001162	0.11	0.45	0.0026
	GMI_ES01_c10216_255	13	714	0.002302	0.10	0.46	0.0026
	GMI_ES05_c2715_265	20	2051	0.002573	0.10	0.48	0.0026
	GMI_GBS_50940	9	983	0.002665	0.10	0.41	0.0026
Coleoptile Length	GMI_ ES15_ c5315_ 156	6	1045	0.001165	0.08	0.14	0.0036
	GMI_ES_15_lrc19156_98	18	231	0.001961	0.07	0.41	0.0036
	GMI_ES22_c9827_183	33	380	0.001986	0.07	0.26	0.0036
	GMI_DS_LB_9600	23	1106	0.002074	0.07	0.07	0.0036
	GMI_GBS_78545	8	814	0.002805	0.07	0.13	0.0036
Plumula Length	GMI_DS_LB_4204	21	777	0.000858	0.11	0.06	0.0039
	GMI_ES01_c3435_183	5	1251	0.002128	0.10	0.26	0.0039
	GMI_ES15_c1855_452	8	1296	0.002768	0.10	0.18	0.0039
	GMI_DS_LB_10835	21	777	0.002786	0.10	0.11	0.0039
Seed Vigor Index	GMI_ESI_c12749_234	21	1720	3.39E-05	0.11	0.43	0.00027
	GMI_ES22_c7747_621	21	1747	0.001483	0.06	0.39	0.0037
	GMI_ES14_lrc18344_662	20	2491	0.001536	0.06	0.46	0.0037
	GMI_ES22_c9230_196	33	820	0.00185	0.06	0.21	0.0037
	GMI_GBS_95238	6	1002	0.002459	0.05	0.01	0.0039
	GMI_ES_CC10682_318	21	1783	0.003098	0.05	0.23	0.0039

r^2: Variation explained by the marker, P Value: Significant threshold of the markers, MAF: Minor Allele Frequency, FDR: False Discovery Rate

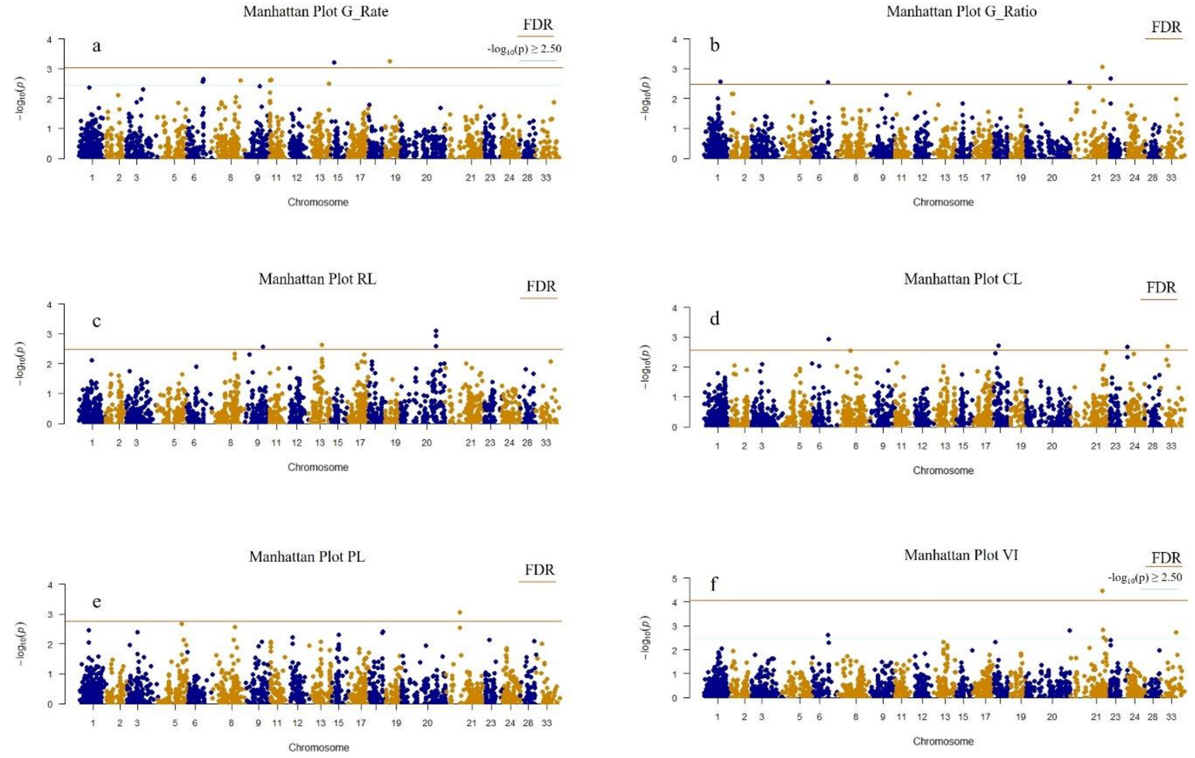

Figure 6. Manhattan plots for germination and root traits using the compressed mixed linear model (CMLM). The orange horizontal lines represent the false discovery rate (FDR) of 5%. (a) Germination rate (FDR=0.0031), (b) germination ratio (FDR=0.0028), (c) radicle length (FDR=0.0026), (d) coleoptile length (FDR=0.0036), (e) plumula length (FDR=0.0039), (f) seed vigor index (FDR=0.00027).

Five candidate markers at $-\log_{10} \geq 2.50$ and $p<0.01-0.001$ were determined for germination ratio (Table 1 and Figure 6b). The phenotypic variance (r^2) of the germination ratio, changed between 0.06 and 0.07, and the MAF value was between 0.01 and 0.46 for all markers. The candidate markers were located on chromosomes 21, 23, 1, 6 and 20 fell into positional distances of 1720, 232, 866, 1002 and 2491 cM longs, respectively (Table 1). Of the candidate markers, GMI_ES01_c12749_234 was located in the Mrg21, GMI_ES05_c2760_657 was located in Mrg01, and GMI_GBS_95238 was located in Mrg06 linkage group. Those findings are in consistent with the previous works, while GMI_ES05_c1006_442 took place in the Mrg23 in our study in contrast to previously reported Mrg05 linkage group maps (Lin et al., 2014; Chaffin et al., 2016; Bekele et al., 2018). On the other hand, there was no report for the GMI_ES14_lrc18344_662 candidate marker.

In terms of radicle length, five candidate markers at the significance of $-\log_{10} \geq 2.50$ and $p<0.01-0.001$ were determined via association analysis (Table 1 and Figure 6c). The phenotypic variance (r^2) ranked between 0.10 and 0.11, and the MAF value ranged from 0.41 and 0.48 for all markers (Table 1). These candidate markers were located on chromosomes 9, 13 and 20 and with the positional distances of 2035, 2035, 714, 2051 and 983 cM longs. GMI_ES_LB_11026, GMI_ES_LB_11028 and GMI_ES05_c2715_265 markers were located in Mrg20 linkage group in consistent with Oat Consensus map 2018 (Bekele et al., 2018). GMI_ES01_c10216_255 marker is located in Mrg13 as reported by Chaffin et al. (2016). GMI_GBS_50940 marker was also reported in Oat Consensus maps 2016 and 2018 located in Mrg9 linkage group, which is similar with our results (Chaffin et al., 2016; Bekele et al., 2018).

Five candidate markers ($-\log_{10} \geq 2.50$ and $p<0.01$) were determined for coleoptile length (Table 1 and Figure 6d). The phenotypic variance (r^2) for the coleoptile length was determined between 0.06 and 0.08, and the MAF value between 0.03 and 0.41 for all markers. Those candidate markers were located on chromosomes 6, 18, 33, 23 and 8 positional distances were 1045, 231, 380, 1106 and 814 cM longs, respectively.

The GMI_ES15_c5315_156 candidate marker is located in Mrg06, GMI_ES22_c9827_183 in Mrg18, GMI_DS_LB_9600 in Mrg23 and, GMI_GBS_78545 in Mrg8 linkage groups, which were also previously reported (Oat-2014-CrownRust, Oat-2016-AxM, Oat-2016-Consensus, Oat-2016-PxB, Oat-2018-Consensus, 1045 cM long) (Lin et al., 2014; Chaffin et al., 2016; Bekele et al., 2018). On the other hand, GMI_ES_15_lrc19156_98 marker located in Mrg18 was not reported previously in the literature.

Four candidate markers ($-\log_{10} \geq 2.50$ $p<0.01-0.001$) were determined as a result of the candidate gene association mapping analysis for plumula length (Table 1 and Figure 6e). The phenotypic variance (r^2) varied between 0.10 and 0.11, and the MAF value between 0.06 and 0.26 for all markers (Table 1). The candidate markers are located on chromosomes 21, 5, 8, and 21 and fell into positional distances of 777, 1251, 1296 and 777 cM longs respectively. GMI_DS_LB_4204 candidate marker was located in the Mrg21 linkage group (Lin et al., 2014; Chaffin et al., 2016; Bekele et al., 2018). Kilinc (2020) reported a pleiotropic effect for GMI_DS_LB_4204 on vegetative period and days to maturity in oat. GMI_ES01_c3435_183 marker was located in Mrg05 linkage group, which is indicated by Chaffin et al. (2016) related with P_{C71} gene. Mrg05, Mrg08 and Mrg21 linkage groups related to plumula length were detected in our study, which is also reported by Mohler, (2021). GMI_ES15_c1855_452 (Mrg8) and GMI_DS_LB_10835 (Mrg21) markers were also indicated in Oat Consensus Map 2018 (Bekele et al., 2018).

Six candidate markers ($-\log_{10} \geq 2.50$ and $p<0.01$-0.0001) were determined for the seed vigor index (Table 1 and Figure 6e). The phenotypic variance (r^2), varied between 0.05 and 0.11, and the MAF value between 0.01 and 0.46 for all markers (Table 1). The candidate markers are located on chromosomes 21, 21, 20, 33, 6 and 21 with the positional distances of 1720, 1747, 2491, 820, 1002 and 1783 cM longs, respectively. The candidate marker GMI_ES01_c12749_234, GMI_ES22_c7747_621 and GMI_ES14_lrc18344_662 were located in the Mrg21, Mrg21 and Mrg20 linkage groups, respectively and, they were presented in previously published oat reference consensus maps, which is in agreement with our results (Oat-2014-CrownRust, Oat-2016-AxM, Oat-2016-Consensus, Oat-2018-Consensus, 1720 cM long) (Lin et al., 2014; Bekele et al., 2018). GMI_ES22_c9230_196 and GMI_GBS_95238 markers were located in the same linkage groups (Mrg33 and Mrg6, respectively) which was reported by Chaffin et al. (2016), while GMI_ES_CC10682_318 marker was located in Mrg21 linkage group, which was also reported by Kilinc (2020).

In addition, Huang et al. (2020) reported robust approach associated (FarmCPU) with seed vigor traits in two different locations by establishing a new phenotyping pipeline in 650 elite oat breeding lines from CORE (Collaborative Oat Research Enterprise) material. They determined the candidate marker and QTL for the first time by GWAS mapping based on strong statistical method, 2 of 41 SNP genomic regions determined for root traits affecting seed vigor were not mapped, other markers were distributed in 16 linkage groups, 16 associated with stem traits, which are other seed vigor factor, were not mapped. They stated that although one of the SNPs could not be mapped, the other markers fell into 10 linkage groups.

CONCLUSION

As a result of the principal component and biplot analysis based on the average data of the examined traits and genotypes, the PC1 component value was determined as 41.5% and the PC2 component value was determined as 21.5%, and it was determined that it explained 63% of the total variation, reflecting a value above the average. In terms of germination parameters examined in oat genotypes, strong positive relationships were determined between germination rate and germination ratio, radicle length and coleoptile length, seed vigor index and coleoptile length and radicle length.

According to candidate gene association mapping analysis performed with the CMLM model, 33 genomic regions were identified consisted of eight QTLs associated with germination rate, five QTLs associated with germination ratio, five QTLs associated with radicle length, five QTLs associated with coleoptile length, four QTLs associated with plumula length, and six for seed vigor index. Some of these QTLs are similar to those in the studies in the literature, and newly identified genomic regions have also been found for the first time. As a result, the candidate markers of these QTLs determined in the study were validated in future studies both in multi-field trials and with the currently developed multi-locus model algorithms and integrated into approaches such as haplotype analysis and genomic selection by determining whether they are true QTLs.

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Conflict of Interest

The authors declare no conflicts of interest.

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THE EFFECT OF DIFFERENT PHOSPHORUS DOSES ON SEED YIELD AND QUALITY PARAMETERS OF BLACK CUMIN (*Nigella* sp.)

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ABSTRACT

Phosphorus is an important element that affects the generative development, seed and fruit quality of plants. This study was carried out under the ecological conditions of Kahramanmaras province of Turkey to determine the effects of different phosphorus doses on the vegetation, yield, and quality characteristics of black cumin. The experiment was carried out in the field of the Field Crops Research and Application Department of Kahramanmaras Sutcu Imam University Faculty of Agriculture in the winter growing seasons of 2017-18 and 2019-20 periods, according to the Randomized Complete Block Design (RCBD) in 3 replications arranged in split plots. Two *Nigella* genotypes, one of which is a registered variety (Cameli variety), and five different P doses (0, 3, 6, 9, 12 kg da^{-1}) were used in the study. According to the research findings, while phosphorus doses did not have a significant effect on plant characteristics such as number of branches, number of capsules, plant height, it was determined that it affected the number of seeds in the capsule and seed yield. The highest number of seeds per capsule (128.23 capsule^{-1}) was obtained from the highest P (12 kg da^{-1}) application, and the highest seed yield (136.04 kg da^{-1}) was obtained from the 6 kg da^{-1} P application. Significant differences were observed in doses and genotype × dose interaction in terms of quality characteristics.

Keywords: Cameli variety, *Nigella sp*, Phosphorus dose, quality characteristics, yield

INTRODUCTION

Medicinal plants have been used for centuries for food, seasoning, medicine, and healing purposes (Bayram et al., 2010). *Nigella sativa* is an annual herbaceous plant of the Ranunculaceae family. There are 20-24 species of *Nigella* in the world and it is stated that 12 to 15 of them are found in the flora of Turkey (Baser, 2010; Ayhan, 2012). *Nigella* is one of the high value-added medicinal and aromatic plants that can be easily grown in the climatic conditions of Turkey, and is used in folk medicine, in kitchens as spices, odorants, and flavor enhancers, and as raw material in the food industry (Akgul, 1993; Ceylan, 1997). It has a hairy and upright structure, which can usually grow up to 35-70 cm in height. *Nigella* seeds are black in color and 2.5-4 mm in size (Baydar, 2013). Comprehensive studies have been carried out on *N. sativa* by many researchers and its antidiabetic, immunomodulatory, analgesic, antimicrobial, anti-inflammatory (Ramadan, 2015; Saleh et al., 2018), anticancer, spasmolytic, bronchodilator, renal protective, gastro-protective, antioxidant properties have been determined. It has been widely used for therapeutic purposes for its antihypertensive, digestive, liver tonics, diuretic, analgesic, antibacterial, antidiarrheal, appetizing, properties and in skin disorders (Ahmad et al., 2013). The purpose of commercial medicinal plant cultivation is to obtain high-seed, essential, and fixed oil yield per hectare. *N. sativa* seeds contain crude fiber (8.4%), protein (26%), ash (4.8%), and carbohydrates (25%). The World Health Organization (WHO) attaches importance to the research of medicinal plant species for the benefit of human health and systems. It has been reported that the effective use of *N. sativa* for therapeutic and commercial purposes will largely depend on yield (bioactive compounds-essential oil, seeds) and quality (Yimam et al., 2015). The basic condition for success in plant production is to obtain high yield and quality products. Adequate and balanced fertilization is one of the important cultural practices that affect yield and quality increase (Anonymous, 2016). Therefore, the amount of fertilizer needed by the plant, the application time of the fertilizer, and the application method should be determined by the studies for each plant species and the region where production will be made. Phosphorus is one of the most deficient elements after nitrogen for the soils of our country in increasing the yielding of plants (Turan, 2014). The P contents of the plants generally vary between 0.05% and 1.0% according to the dry matter principle. Plants contain much less phosphorus compared to nitrogen (N), magnesium (Mg), calcium (Ca), and potassium (K). Inorganic phosphorus compounds (Pi) entering the root are stored in the root or transported to the top organs of the plant. After various chemical reactions, inorganic P turns into various organic compounds, including enzymes, nucleic acids, and proteins. In addition to those called adenosine monophosphate (AMP), adenosine diphosphate (ADP) and adenosine triphosphate (ATP), other organic-phosphate groups play an important role in transferring the energy needed in metabolic events in plants. The importance of phosphorus in photosynthesis, carbon fixation, and assimilation is undeniable. Phosphorus plays an important role in the formation of genes and chromosomes in plants. Phosphorus plays a role in the transmission of genetic codes from one generation to the next. Phosphorus requirement is very high in meristematic tissues for cell division and growth. It has been determined that phosphorus significantly affects seed and fruit formation, and in the absence of sufficient phosphorus, seed and fruit quality decreases significantly (Kacar, 2015). Phosphorus (P) is among the most limiting nutrients needed for good plant growth and higher yield, and is stored in seeds (White and Veneklaas, 2012; Seyyedi et al., 2017). There is no standard definition of P that is suitable for plant in soil. For this reason, a wide variety of extraction methods are used in the estimation of P for the plant in the soil (Buczko et al., 2018). In plant production, more or less fertilization has negative effects on growth and yield. Black seed quality and production can be severely affected by P deficiencies in the soils (Seyyedi et al., 2017). Mohammed et al. (2000) reported that P and N fertilizers significantly improved growth parameters, yield and yield components as well as nutrient contents in *Nigella sativa.* It was reported that high phosphorus and nitrogen levels cause significant increases in fixed oil, essential oil, protein and phosphorus, and this increase may be because of the effect of phosphorus on root development, flower formation, seed and fruit development (Hammo, 2008). This study aims to determine the effects of different phosphorus doses on seed yield and quality characteristics of black cumin in Kahramanmaras conditions.

MATERIALS AND METHODS

In this study, black cumin seeds were sown in the winters of 2018 and 2020. Due to the unsuitable land and climatic conditions in the 2019 planting season, the second year of the experiment was carried out in the 2020 winter growing season.

Experimental Site and Conditions

The research was carried out in the Field Crops Research and Application land of Kahramanmaras Sutcu Imam University, Faculty of Agriculture. According to Table 1, the long-term average amount of precipitation was 650.80 mm. While the total precipitation in the vegetation period of 2017-18, in which the research was conducted, was 522.80 mm, which was below the long-term average of total precipitation, the total precipitation in the vegetation period of 2019-20, was above the long-term average of the total precipitation with a value of 652.30 mm. While the long-term average temperature was 12.60°C, the average temperature of the vegetation period of 2017-18, in which the study was conducted, was 14.74°C, and the average temperature of the vegetation period of 2019-20 was 13.25°C, which was above the long-term average. According to Table 1, the average temperature values in both years were higher than the long-term average temperature, and the average temperature was higher in the first year than in the second year. The average relative humidity in Kahramanmaras is 63.04% compared to the long-term average. Although the average relative humidity in the study period of 2017-18 was 59.98%, and the average relative humidity in the study period of 2019-20 was higher than the first year with 61.91%, it was seen that the average relative humidity values of both years in which the study was carried out were lower than the long-term average relative humidity value. Soil samples taken from 0-30 cm depth in the experimental field were analyzed at the University-Industry-Public Cooperation Development Application and Research Center (USKIM). Some physical and chemical properties of the soil are given in Table 2. Properties of the soil sample in 2018 and 2020 are as follows, clayed loamy (72.00 and 69.96), slightly alkaline (7.66 and 7.71), low salinity (0.86%) and salt-free (0.05%), moderately calcareous (3.91 and 6.09%), low in organic matter (1.66 and 1.58%), high in potassium (53.00 and 55.51 kg da^{-1}), moderately phosphorus (6.29 kg da^{-1}), and very low (2.84 kg da^{-1}), respectively.

Table 1. Precipitation temperature, and relative humidity values of experimental years and long-term growing seasons in Kahramanmaras Province (Anonymous 2020a)

		Months								
Climatic Factory	Year	November	December	January	February	March	April	May	June	Total or Avarage
Precipitation (mm)	2017-2018	89.60	33.70	149.90	63.10	47.40	71.60	28.10	39.40	522.80
	2019-2020	39.10	198.50	88.00	72.70	173.40	61.80	18.50	0.30	652.30
	Long years	87.50	116.60	125.40	108.30	93.40	69.80	41.20	8.40	650.80
Average Temperature (°C)	2017-2018	12.20	8.90	7.40	9.70	14.20	18.40	21.70	25.40	14.74
	2019-2020	13.50	8.40	6.30	6.10	12.50	15.90	15.90	24.50	13.25
	Long years	11.50	6.80	4.90	6.40	10.60	15.50	20.30	25.30	12.60
Relative humidity (%)	2017-2018	64.20	69.00	69.50	69.40	60.80	45.30	52.60	49.10	59.98
	2019-2020	56.20	81.90	69.30	68.30	67.30	58.20	47.20	46.90	61.91
	Long years	66.68	79.85	69.99	65.62	60.00	57.59	54.95	49.67	63.04

Table 2. Some chemical and physical properties of the study area soils (Anonymous 2020b)

Year	Texture class	Organic matter (%)	$CaCO_3$ (%)	EC (dS m-1)	pH	P_2O_5 (kg da-1)	K_2O (kg da-1)
2018	Clay (72)	1.66	3.91	0.86	7.66	6.29	53
2020	Clay-loam (69.96)	1.58	6.09	0.05	7.71	2.84	55.51

Experimental Material

In the study, *N. sativa* species (Genotype 1-G1) and registered Cameli variety (Genotype 2-G2) seeds obtained from the GAP Agricultural Research Institute Directorate and Eskisehir Transitional Zone Agricultural Research Institute, were used in the study as materials. In the study, Triple Super Phosphate (43-45%) fertilizer for phosphorus doses and Ammonium Nitrate fertilizer (33%) for N dose were used. In the experiment, 5 different doses of phosphorus, 0, 3, 6, 9, and 12 kg da^{-1} were applied. Nitrogen was calculated to be 6 kg pure N per decare, and a half was applied during planting and the remaining half was applied in spring during the bolting period.

Design and Cultural Practices

In the land where the soil preparation was made, the rows were opened with the help of a marker with a planting depth of 2-3 cm, and a spacing of 30 cm in 6 rows, and the genotypes were divided into the main plots, the phosphorus doses were placed in the sub-plots, and the plots were carried out in three replications according to the experimental design. The length of the plot was 3 m and the width was 1.80 m, the distance between the plots was 0.5 m and the distance between the blocks was 2.5 m. Plants were thinned so that the row spacing was 10 cm. Irrigation was done by the furrow method twice in both years and weed control was done by hand. The trial was established in November in both years. The thinning process of the plants was carried out in March, the weed control was carried out once in April with the thinning process. Irrigation was performed twice, in May and June (Table 3). No diseases or pests were detected during the vegetation period of the plants. After removing the edge effect of the plants which grew ripe (one row from the top and bottom, and 50 cm from the beginning and end of the plot), the plants were harvested by hand on 18 June 2018 and 25 June 2020. The following characteristics were measured in 10 randomly selected plants from the plots after harvest; the number of branches, plant height (cm) in a plant (piece plant^{-1}), the number of capsules in a plant (capsule plant^{-1}), the number of seeds in a capsule (piece capsule^{-1}), thousand-grain weight (g), seed yield (kg da^{-1}), protein ratio (%), essential oil content (%), essential oil yield (L da^{-1}), fixed oil content (%), and fixed oil yield (kg da^{-1}). After 25 g of seeds samples were ground, the hydro-distillation process was performed in Neo-Clevenger device for 3 hours, and then the essential oil content (%) was volumetrically determined. In addition, fixed oil content (%) was determined by extraction with petroleum ether for 6 hours in the Soxhlet apparatus in 5 g of ground seeds.

Table 3. Data on cultural activities and practice times

Years	Sowing date	Plant thinning	Weed control	Irrigation 1	Irrigation 2
2018	16 November 2018	20 March 2018	22 April 2018	15 May 2018	2 June 2018
2020	14 November 2020	15 March 2020	20 April 2020	28 May 2020	5 June 2020

Statistical Analyses

Statistical analyzes of the examined yield and quality-related characteristics were analyzed using the SAS 9.1 package program according to the Randomized Complete Block Design (RCBD) arranged in split-plots. The significant differences were compared by the LSD multiple comparison test at the P<0.05 and P<0.01 levels of significance (Steel and Torrie, 1980).

RESULTS AND DISCUSSION

In this study, the effects of phosphorus applications on yield, and quality characteristics in two different black cumin genotypes were investigated.

Plant height (cm)

While the difference between genotype, dose, and genotype × dose interaction was not found to be significant in terms of the effect of phosphorus application on plant height, the application years were found to be significant. The average plant height value was 75.35 cm in 2018 and 60.06 cm in 2020 (Table 4). Since the amount of precipitation in 2018 was higher than 2020 in April-May, which was the period of plant bolting, there was a difference in plant height between the years (Table 1). Kizil et al. (2008) reported that higher precipitation amount in the first year of the study contributed positively to number of branches, plant height, and capsules number per plant. Yimam et al. (2015) reported plant height as 45.7-72.5 cm, Tuncturk et al. (2011) as 30.7- 35.3 cm, Muhammad et al. (2017) as 50.02-53.51 cm, Shirmohammadi et al. (2014) as 23.0-32.1 cm, Kosar and Ozel (2018) reported plant height as 47.8-68.6 cm, Kilic and Arabaci (2016) 78.9 cm, and Abadi et al. (2015) as 19.04-23.43 cm. Kizil et al. (2008) reported plant height in the range of 50.5-59.0 cm in winter planting and that there was no difference between doses. Similarly, no differences were detected in plant height between doses in this study. The increased plant height may actually be because of the greater availability of nutrients in the soil in the cultivation area (Yimam et al., 2015). The ecology, soil, and agricultural practices in which the plant grows had important effects on plant height. The differences between the results of other studies in terms of plant height were because of soil, genotype, and ecological conditions.

Table 4. Two-year averages and LSD groupings of applied phosphorus doses on yield and vegetative properties in black cumin

		PH	NB	NC	NSC	TSW	SY
Genotype	G_1	68.66	8.43 a	16.95 a	120.49	2.58 b	125.31
	G_2	66.76	7.56 b	14.79 b	123.29	2.70 a	129.56
Doses (kg da^{-1})	0	67.77	7.87	15.59	119.86 bc	2.62	121.77 b
	3	67.90	7.80	16.42	116.04 c	2.61	128.46 ab
	6	66.23	7.91	16.13	122.78 b	2.64	136.04 a
	9	68.46	8.06	15.59	122.53 b	2.63	124.57 b
	12	68.18	8.30	15.64	128.23 a	2.69	126.34 b
Year	2018	75.35 a	9.76 a	18.76 a	111.63 b	2.67 a	154.34 a
	2020	60.06 b	6.23 b	12.98 b	132.15 a	2.61 b	100.54 b
Genotype x Doses (G × D)	$G_1 \times P_0$	68.26	8.13	16.98	120.62	2.53	116.91
	$G_1 \times P_3$	68.68	8.33	17.61	114.73	2.53	118.40
	$G_1 \times P_6$	66.67	8.20	16.86	121.52	2.56	138.23
	$G_1 \times P_9$	70.83	8.82	17.12	117.00	2.60	127.27
	$G_1 \times P_{12}$	68.87	8.67	16.21	128.60	2.69	125.76
	$G_2 \times P_0$	67.28	7.62	14.21	119.12	2.73	126.65
	$G_{2\times}$x P_3	67.12	7.28	15.24	117.35	2.70	138.54
	$G_2 \times P_6$	65.80	7.63	15.40	124.05	2.73	133.86
	$G_2 \times P_9$	66.10	7.32	14.07	128.07	2.67	121.88
	$G_2 \times P_{12}$	67.50	7.95	15.07	127.87	2.70	126.93
Mean		67.71	7.99	15.87	121.9	2.64	127.44
CV		5.89	6.8	7.9	4.77	3.00	7.24
LSD for years		2.09**	0.28**	0.66**	3.06**	0.04*	4.85**
LSD for genotype		ns	0.28**	0.66*	ns	0.04**	ns
LSD for doses		ns	ns	ns	4.83**	ns	7.67**
LSD for G × D		ns	ns	ns	ns	ns	ns

PH: Plant height (cm), NB: Number of branches (branch plant^{-1}), NC: Number of capsules (capsule plant^{-1}), NSC; Number of seeds per capsule (seed capsule^{-1}), TSW: Thousand seeds weight (g), SY: Seed yield (kg da^{-1}), ns: No significant, *$P<0.05$; **$P<0.01$

Number of branches (number plant^{-1})

Genotypes were significant at a level of 1% in terms of the number of branches in the plant, while the interaction between dose and genotype × dose was found to be insignificant. While the number of branches was 8.43 per plant^{-1} in genotype 1 (G1), it was 7.56 per plant^{-1} in genotype 2 (G2) (Table 4). The difference between years was significant and the average number of branches was 9.76 in the first year, while it was 6.23 in the second year (Table 4). Tuncturk et al. (2011) reported the number of branches per plant as 3.76 to 4.10, and Kizil et al. (2008) as 5.1 to 6.2 in winter sowing period. Faizy (2019) determined the number of branches as 7.18-7.88 branch plant^{-1}. Kizil et al. (2008) reported that more rainfall in the first year of the trial contributed positively to number of branches, plant height, and number of capsules per plant. Similarly, in the second year of the study, plant growth was weaker, and therefore, the number of branching decreased in the second year because of the fact that the precipitation in March and April, which is the plant development period, was less than in the first year (Table 1). The response of the plant to phosphate fertilizer can be highly variable from one year to the next because of many factors affecting P availability and crop growth (Kizil et al., 2008). This may cause differences in the data of the plant observations in different years.

Number of capsules per plant (capsule plant^{-1})

While genotypes were significant at the 5% level in terms of the number of capsules in the plant, no difference was observed in the interaction of genotype x doses. Genotype 1 (16.95 capsule plant^{-1}) has a lower value than genotype 2 (17.79 capsule plant^{-1}) in terms of the number of capsules. While the number of capsules per plant was 18.76 capsule plant^{-1} in the first year, it was determined as 12.98 capsule plant^{-1} in the second year (Table 4). The fact that Geren et al. (1997) did not find a difference between doses in the February sowing in the second year supports our study. Kizil et al. (2008) determined the number of capsules per plant in winter sowing as 12.4-15.5 capsules per plant^{-1} and did not find any difference between doses and years. Yimam et al. (2015) determined the number of capsules per plant as 21.5-45.9 (60/40 N/P), Datta (2004) obtained the highest number of capsules at 20 and 40 kg ha^{-1} phosphorus (5.68-5.61), the lowest number of capsules (4.68) in the control group. Tuncturk et al. (2011) reported 4.68-5.68 capsules per plant^{-1}, Faizy (2019) 34.40-38.46 capsules per plant^{-1}, Kosar and Ozel (2018) 4.03-7.63

capsules per plant^{-1}, Kilic and Arabaci (2016) 16.17 capsules per plant^{-1}.

Number of seeds (piece capsule^{-1})

While it was found that the effect of phosphorus doses on the number of seeds in the capsule was at a significance level of 1%, the interaction between genotype and genotype × dose was not significant. While the average number of seeds per capsule in G1 was 120.49 per capsule, it was determined as 123.29 in G2. In terms of doses, the highest value was obtained from the 12 kg da^{-1} application with 128.23 seeds per capsule at, while the lowest value was determined as 119.86 and 116.04 seeds per capsule from 0 kg da^{-1} and 3 kg da^{-1} applications, respectively. While the number of seeds in a capsule was 111.63 seeds per capsule in the first year, it was determined as 132.15 seeds per capsule in the second year (Table 4). Yimam et al. (2015) determined the number of seeds per capsule in the range of 55.1-91.6, Tuncturk et al. (2011) determined the number of seeds in a capsule in the range of 52.2-56.2 seeds per capsule^{-1} and reported that there was no statistical difference between the doses. Rana et al. (2012) reported that there were significant differences between the varieties in terms of the number of capsules per plant (29.53-30.30) and the number of seeds per capsule (58.47-60.33), and increasing fertilizer doses increased these values at significant level. This increase may be because of the increased nutrient uptake by the root system after the fertilizer, increased chlorophyll content, and increased protein and photosynthesis (Rana et al., 2012). It was reported that there are significant differences between the concentrations on the number of seeds in the capsule in black cumin (Ali et al., 2015b), and this may be because of the higher enzymatic activity and increased metabolite levels (Shah and Samiullah, 2007).

Thousand-seed weight (g)

It was observed that *Nigella* genotypes were significant at the level of 1% in terms of thousand-seed weight, but the effect of dose and genotype × dose interaction was not significant. The thousand-seed weight of G2 (2.70 g) was higher than that of G1 (2.58 g). The thousand-seed weight was determined as 2.67 g in the first year, and as 2.61 in the second year (Table 4). Yimam et al. (2015) reported that although there was a numerical difference (2.10-2.30 g) in the thousand-seed weight, it was insignificant, Ali et al. (2015a) reported that thousand-seed weight was an important indicator of yield and that fertilizer levels in *Nigella* genotypes were not very important in thousand-seed weight. Tuncturk et al. (2011) reported the thousand-seed weight as between 2.28-2.48 g and found it significant. Kosar and Ozel (2018) determined the thousand-seed weight as 1.81-3.16 g. Tuncturk et al. (2011) and Yimam et al. (2015) reported that a wide variety of factors such as variety, climatic factors, soil properties, and growing conditions affect thousand-seed weight. Yimam et al. (2015) reported that the P and N interaction was very important ($p<0.01$) in the yield parameters and growth parameters other than the thousand-seed weight of black cumin.

Seed yield (kg da^{-1})

Considering the effect of increasing phosphorus doses on seed yield, it was seen that the effects of genotype and genotype × dose interaction were not significant, while the doses were significant at the level of 1%. Since genotypes were mainly formed from the same species (*N. sativa*), although there was a numerical difference between the values in terms of seed yield, the difference was found to be statistically insignificant. According to Table 4, the seed yield of G1 was 125.31 kg da^{-1}, while the seed yield of G2 was determined as 129.56 kg da^{-1}. while the highest seed yield was obtained from the 6 kg da^{-1} application, the lowest seed yield was obtained from the 0 kg da^{-1} (121.77 kg da^{-1}), 9 kg da^{-1} (124.57 kg da^{-1}), and 12 kg da^{-1} (126.34 kg da^{-1}). The difference between years was significant. The seed yield was determined as 154.34 kg da^{-1} in the first year and 100.54 kg da^{-1} in the second year. According to Geren et al. (1997), although the seed yield of the P dose of 0 kg da^{-1} (12.6 kg da^{-1}) was statistically significant in the same group as 8 kg da^{-1} dose (11.8 kg da^{-1}) in February sowing in the second year, the control dose was found to be higher than the 8 kg da^{-1} dose in numerical value. Kizil et al. (2008) reported that the highest seed yield in winter sowing was obtained from the P application of 120 kg ha^{-1} with a value of 1534 kg ha^{-1}. Yimam et al. (2015) reported that the seed yield was in the range of 639.6-1336.7 kg ha^{-1} and the highest value was obtained from the 60/40 kg ha^{-1} (N/P) application. Tuncturk et al. (2011) obtained a seed yield value of 507-568 kg ha^{-1} in the first year, and a value of 549-626 kg ha^{-1} in the second year, and the highest yield from the P application of 40 kg ha^{-1}. It was reported that the difference between the years may be due to different precipitation regimes and differences in temperature. On the other hand, Ozguven and Sekeroglu (2007) applied 0, 3, 6 and 9 kg da^{-1} N and 0, 3 and 6 kg da^{-1} P doses to *Nigella* in Cukurova conditions, and reported that the highest seed yield (100.6 kg da^{-1}) was obtained from 6 kg N da^{-1} and 6 kg P da^{-1} fertilizer applications and that fertilizer doses did not significantly affect the oil content in the seed. While Turan (2014) obtained the highest seed yield as 96.64 kg da^{-1} in Cameli variety, and the highest seed yield at 2 kg P_2O_5 da^{-1} dose (111 kg da^{-1}). In this study, a higher seed yield was obtained from the Cameli variety with a value of 129.56 kg da^{-1} and the highest seed yield was obtained from 6 kg da^{-1} P application (136.04 kg da^{-1}). According to literature data, seed yields obtained as a result of P doses applied to *Nigella* vary. Accordingly; Tuncturk et al. (2011) from the 4 kg da^{-1} P application with a value of 62.6 kg da^{-1}, Shirmohammadi et al. (2014) obtained the lowest seed yield from the control group (451 kg ha^{-1}), and the highest seed yield (735 kg ha^{-1}) from biological phosphate + 40 kg ha^{-1} phosphorus application. Geren et al. (1997) from the 8 kg da^{-1} P application with a value of 60 kg da^{-1} and Kizil et al. (2008) from the 12 kg da^{-1} P application with a value of 1534 kg ha^{-1}. Turan (2014) reported that the highest seed yield at different phosphorus doses was closely related to climate and genotypic effects. Phosphorus for the plant is a nutrient element of great importance because it increases resistance to diseases and pests, ensures root development, plant maturation, early seed formation, and fertilization

(Bilen and Sezen, 1993). Yield components such as capsules and the number of branches affect the seed yield in field crops directly (Ozguven and Sekeroglu, 2007). Yimam et al. (2015) the application of P and N doses together in black cumin causes an increase in plant development, number of leaves and photosynthesis rate. Accordingly, an increase in the number of capsules may lead to an increase in seed yield. Seyyedi et al. (2015) reported that soils that contained high amounts of calcium carbonate affect the phosphorus uptake of the plant negatively, which will also negatively affect the phosphorus uptake in black cumin. Differences in seed yield in similar studies can be explained by the difference in soil structure and phosphorus content in the trial soils of the mentioned studies. According to the study that was conducted by Kacar and Katkat (2009), it was reported that phosphorus has positive effects on the reproductive period and seed maturity in cultivated plants.

Fixed oil content (%)

While there was no difference between genotypes in terms of fixed oil content, it was seen that the doses were significant at the 5% level and the genotype ×dose interaction at the 1% level. While the fixed oil content of genotype 1 was 37.33%, genotype 2 was 35.71%. The highest fixed oil content was obtained from the phosphorus doses of 6 kg da^{-1} (37.63%) and 9 kg da^{-1} (37.54%), which were statistically significant in the same group, while the lowest fixed oil content was obtained from the 12 kg da^{-1} (35.27%) dose. Kizil et al. (2008) obtained the maximum fixed oil content (37.4%) from the 16 kg da^{-1} P winter application, which was in the same statistical group as the 12 kg da^{-1} P application. Kara et al. (2015) reported the fixed oil content as 29.5%- 28.4% in the first and second year. Mamun and Absar (2018) reported fixed oil content as 25% and oleic acid content as 11.7%, Muhammad et al. (2017) as 21.92-22.95%, Faizy (2019) as 35.74-38.46%, Kosar and Ozel (2018) as 36.42-40.17%, Kilic and Arabaci (2016) as 38.17% in black cumin. According to Figure 1A, the highest fixed oil content was observed at the 3 kg da^{-1} (G1×P3) dose of genotype 1, and the lowest fixed oil content at the 3 kg da^{-1} (G2×P3) dose of G2. When compared by years, while the fixed oil content was 34.16% in the first year, it was 38.88% in the second year (Table 5). Aytac et al. (2017) determined that the fixed fat ratio was higher in the first year (43.2%) than in the second year (32.6%). Acetyl-CoA is a starting material for the formation of fatty acids. The glucose that is produced by photosynthesis is firstly converted into pyruvic acid and then into Acetyl-CoA in pyruvic acid. The oils are accumulated in the storage cells that are called oleasomes in the endosperm after the formation of the ovule after the fertilization in the plant. Oils are mostly accumulated in seeds in annual plants (Baydar and Erbas, 2014). Some responses may be associated with the role of phosphorus as a component of phosphatidylcholine, a biosynthetic intermediate and a carrier of acyl chains in plant seeds (Bates et al., 2013). Previous studies reported that P deprivation can lead to low phosphatidylcholine levels (Okazaki et al., 2013). Fredeen et al. (1990) reported that proper P nutrition could improve the photosynthetic process that would allow increased carbon fixation in the production of carbohydrates and lipids. It was reported that phosphorus affects the fixed oil rate and its components because it is found in the structure of many important enzymes such as nucleic acids, coenzymes, nucleotides, phytates, phospholipids, and sugar phosphates in plants playing active roles in photosynthesis.

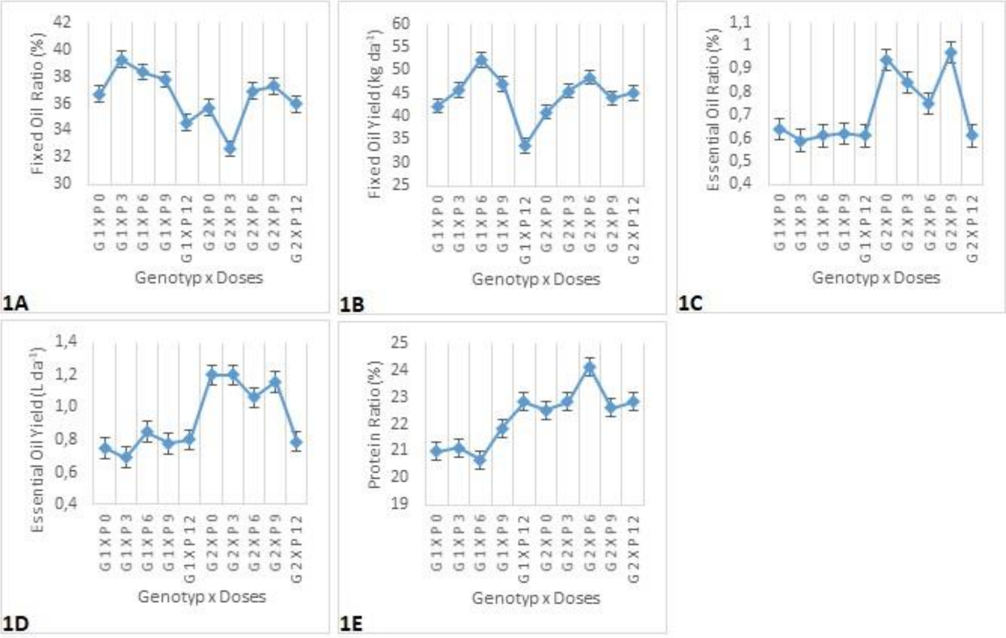

Figure 1. Two-year average values of genotype × dose interaction, which is statistically significant in the investigated characteristics

Table 5. Two-year averages and LSD groupings of applied phosphorus doses for properties examined in black cumin

		FOC (%)		FOY (kg da^{-1})		EOC (%)		EOY (L da^{-1})		PC (%)	
Genotype	G1	37.33		44.18		0.61	b	0.77	b	21.48	b
	G2	35.71		44.78		0.82	a	1.08	a	22.97	a
Doses (kg da^{-1})	0	36.21	ba	41.58	c	0.78	a	0.97	a	21.73	d
	3	35.96	ba	45.63	b	0.71	b	0.94	a	21.96	cd
	6	37.63	a	50.36	a	0.68	b	0.95	a	22.41	b
	9	37.54	a	45.49	b	0.79	a	0.96	a	22.22	cb
	12	35.27	b	39.37	c	0.61	c	0.79	b	22.82	a
Year	2018	34.16	b	50.02	a	0.77	a	1.19	a	22.37	a
	2020	38.88	a	38.95	b	0.66	b	0.66	b	22.08	b
Genotyp × Doses (G×D)	$G_1 \times P_0$	36.71	a-c	42.23	ab	0.64	de	0.75	bc	20.97	de
	$G_1 \times P_3$	39.27	a	45.77	a	0.59	e	0.69	c	21.12	de
	$G_1 \times P_6$	38.34	ab	52.29	a	0.61	e	0.85	b	20.68	e
	$G_1 \times P_9$	37.79	ab	47.06	a	0.62	e	0.78	bc	21.84	cd
	$G_1 \times P_{12}$	34.58	bc	33.60	b	0.61	e	0.80	bc	22.82	b
	$G_2 \times P_0$	35.72	a-c	40.95	ab	0.94	ab	1.20	a	22.50	bc
	$G_2 \times P_3$	32.66	c	45.49	a	0.84	bc	1.20	a	22.81	b
	$G_2 \times P_6$	36.93	a-c	48.43	a	0.75	cd	1.06	a	24.14	a
	$G_2 \times P_9$	37.29	a-c	43.93	a	0.97	a	1.16	a	22.61	b
	$G_2 \times P_{12}$	35.97	a-c	45.14	a	0.61	e	0.79	bc	22.83	b
Mean		36.52		44.48		0.71		0.92		22.23	
CV		5.50		8.86		5.88		6.33		1.54	
LSD for years		1.05**		2.07**		0.02**		0.03**		0.18**	
LSD for genotype		ns		ns		0.02**		0.03**		0.18**	
LSD for doses		1.67*		3.28**		0.03**		0.04**		0.28**	
LSD for G x D		7.89**		15.49**		0.16**		0.23**		1.34**	

FOC: Fatty oil content, FOY: Fatty oil yield, EOC: Essential oil content, EOY: Essential oil yield, PC: Protein content, ns: No significant, *$P<0.05$; **$P<0.01$

Fixed oil yield (kg da^{-1})

While there was no difference between genotypes for fixed oil yield, it was observed that dose and genotype × dose interaction were significant at the 1% level (Table 5). The fixed oil yield was determined as 44.18 kg da^{-1} in G1 and 44.78 kg da^{-1} in G2. The highest fixed oil yield was observed in the 6 kg da^{-1} P application (50.36 kg da^{-1}), while the lowest was found in the 0 kg da^{-1} P (41.58 kg da^{-1}) and 12 kg da^{-1} P (39.37 kg da^{-1}) doses, which were statistically significant in the same group. According to Figure 1B, the lowest fixed oil yield was observed at 12 kg da^{-1} P dose of genotype 1 (33.60 kg da^{-1}), while the highest was observed at 0 kg da^{-1} and 12 kg da^{-1} P doses of G1, and at the doses, which were statistically significant in the same group as those of G2 except for the 0 kg da^{-1} P dose. In terms of years, the fixed oil yield was determined as 50.02 kg da^{-1} in the first year and 38.95 kg da^{-1} in the second year (Table 5). Akgoren (2011) reported the fixed oil yield between 18.85-41.08 kg da^{-1}. Telci (1995) determined the average fixed oil yield as 44.70-49.81 kg da^{-1}, which is consistent with the fixed oil yield values in the study. According to Turan (2014), the highest oil yield in the Bilecik population was obtained from 4 kg P_2O_5 da^{-1} application (42.97 kg da^{-1}), while the highest oil yield (46.34 kg da^{-1}) in Cameli variety was obtained from 2 kg P_2O_5 da^{-1} application. In this study, the fixed oil yield of the Cameli variety (Genotype 2) was statistically significant in the same group at all doses except for the control dose. Phosphorus is an important element playing roles in cell division, increasing the resistance of plants by promoting the uptake of potassium by plants, root development and maturation (Brohi and Aydeniz, 1994). With the increasing root development after phosphorus applications, the contact surface of the root in the soil expands, and the rate of utilization of other nutrients by plants increases (Marschner, 1995). Phosphorus fertilizers are very important in increasing the seed quality as well as the seed yield. Climatic conditions such as suitable temperature and sufficient precipitation are very important in black seed cultivation. Also, better root development, branching, and flowering (Bilen and Sezen, 1993), as well as increased number of capsules and the amount of seeds are observed (Seyyedi et al., 2017) with the administration of appropriate phosphorus doses. The effect of phosphorus on seed yield and fixed oil ratio directly affects fixed oil yield because fixed oil yield is calculated by using fixed oil ratio (%) and seed yield (kg da^{-1}) values.

Essential oil content (%)

Genotype, dose, and genotype × dose interaction were significant at the 1% level for essential oil content (Table 5). While the essential oil content of G1 was 0.61% and G2 was 0.82%. While the maximum essential oil content was obtained from 0 kg da^{-1} P and 9 kg da^{-1} P doses, which were statistically significant in the same group, the lowest was obtained from the 12 kg da^{-1} P dose. Considering the genotype × dose interaction according to figure 1C, it is seen that the maximum essential oil content was obtained from the 9 kg da^{-1} P dose of genotype 2, while the lowest content was obtained from the doses of genotype 1 except for the 0 kg da^{-1} P dose, which was in the same group as the 12 kg da^{-1} P dose of genotype 2 (Table 5). According to years, it was found to be 0.77% in the first year and 0.66% in the second year. Kizil et al. (2008) obtained the maximum essential oil content (0.60%) from the control dose (0 kg ha^{-1} P), which is similar to our results. Kara et al. (2015) determined the average essential oil content as 0.34% in the first year and 0.29% in the second year. Faizy (2019) determined the essential oil ratio in the range of 0.79-0.82%. Essential oil amount could be varied due to extraction methods applied and ecological factors such as fertilization, irrigation, seed type and climatic conditions etc (Aksu et al., 2021). As reported by Salem et al. (2020), fertilization has an important role in the growth, flowering, fruit, seed and oil yields of plants as well as in their biochemical components.

Essential oil yield (L da^{-1})

Genotype, dose, and genotype × dose interaction were found to be significant at the 1% level in terms of essential oil yield (Table 5). While the essential oil yield was 0.77 L da^{-1} in genotype 1, it was determined as 1.08 L da^{-1} in genotype 2. When the doses were examined in terms of essential oil yield, it was seen that the lowest essential oil ratio was obtained from the 12 kg da^{-1} P application and that all other doses were statistically significant in the same group (Table 5). According to figure 1D, while the highest essential oil yield was observed at the doses of genotype 2 other than 12 kg da^{-1} P dose, the lowest value was observed at 3 kg da^{-1} P dose of genotype 1. Looking at the years, it was determined as 1.19 L da^{-1} in the first year and 0.66 L da^{-1} in the second year.

Protein content (%)

According to Table 5, genotype, dose, and genotype × dose interaction are significant at the 1% level in terms of protein content. The protein content was found to be higher in genotype 2 (22.97%) than in genotype 1 (21.48%). Considering the doses, the highest protein content was obtained from the 12 kg da^{-1} P application (22.82%), while the lowest protein content was obtained from the control application (21.73%). According to figure 1E, the maximum protein content was obtained from the 9 kg da^{-1} P application of genotype 2, while the lowest protein content was obtained from the 9 kg da^{-1} P dose of genotype 1. While the protein content was 22.37% in the first year, it was 22.08% in the second year. According to Rana et al. (2012), the protein content in the applied fertilizer doses was between 15.42% and 23.18%, and the highest value (23.18%) was obtained from the 60/120 kg ha^{-1} N/P application, which was the highest dose. Takruri and Dameh (1998) found a protein content between 19.9-24.1% and obtained the highest value from the Indian source. Kabir et al. (2019) determined the protein content as 20.3%. The values in this study are seen to be compatible with the literature data.

CONCLUSION

This study was carried out under Kahramanmaras ecological conditions during 2018 – 2020 periods to determine the effect of increasing phosphorus doses in black cumin on yield and quality characteristics. Increasing phosphorus doses were found to be significant for the number of seeds in the capsule, seed yield, fixed oil content, fixed oil yield, essential oil content, essential oil yield, and protein content. The highest seed yield was obtained from the 6 kg da^{-1} P application (136.04 kg da^{-1}). While the seed yield increased up to the 6 kg da^{-1} P application, the 9 kg da^{-1} P and 12 kg da^{-1} P applications were in the same statistical group as the control. The fixed oil content, fixed oil yield, and essential oil yield, in a similar way, reached the highest value with the 6 kg da^{-1} P application, while the highest essential oil content was obtained from the 9 kg da^{-1} P application, and the highest protein content was obtained from the 12 kg da^{-1} P application. As a result, the difference between phosphorus doses on number of capsules per plant, plant height, number of branches per plant, and thousand-seed weight was found to be insignificant, and the phosphorus dose coming forward for seed yield and quality properties was 6 kg da^{-1}.

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EFFECTS OF SUPPRESSION APPLICATIONS ON SUMMER ASPHODEL (*Asphodelus aestivus* Brot.) DENSITY, BOTANICAL COMPOSITION, FORAGE YIELD AND QUALITY OF AEGEAN RANGELANDS

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ABSTRACT

Summer asphodel (*Asphodelus aestivus* Brot.) has an underground storage organ that enables the plant to survive extreme conditions and contains alkaloids that are toxic to the livestock in rangelands. For this reason, the experiment was carried out in the rangeland of Aydin province (Turkey) in order to determine the most effective methods of weed management where there is an increase in the summer asphodel (*Asphodelus aestivus* Brot.) population. In the study, the effects of 8 different control methods (control, mowing, fertilization, paraquat, glyphosate, 2,4-D, 2,4-D + Picloram, grubbing) on yield, quality and botanical composition were investigated. Result of the findings in this study shows that the grasses increased more in vegetation. The highest values of hay yield were observed in paraquat and fertilization applications. High values of crude protein yield were found in fertilization and grubbing applications. The lowest population of summer asphodel (*Asphodelus aestivus* Brot.) was obtained grubbing and paraquat applications. Together with these results, grubbing, fertilization, paraquat and glyphosate applications come to the fore. However, due to the high workforce, it is significant to choose an application considering the size of the rangeland and the population of the indicator weed species.

Keywords: *Asphodelus aestivus*, forage quality, herbicide, range improvement, weed control

INTRODUCTION

In addition to many ecosystem services, natural rangelands provide forage for livestock and wildlife (Barnes et al., 1995; USDA, 2018). However, mismanagement mainly overgrazing is the main problem of the rangelands in Turkey. It was claimed that because of mismanagement practices, the rangelands of Turkey have lost approximately 90% of their original vegetation (Genckan et al., 1990; Cetiner et al., 2012; Turan et al., 2015; Gokkus, 2020; Koc et al., 2021). As a result of these conditions, an increase in invasive weed populations has been observed in rangelands. Impacts of this increase to the livestock industry not included in the estimate are the negative effects of invasive plants on yield and quality of forage, livestock poisoning, interference with grazing, supplemental costs associated with managing and producing livestock, and land values. Also, invasive weeds can decrease wildlife and plant biodiversity (Mack et al., 2000; DiTomaso et al., 2010).

The genus *Asphodelus* is native to temperate Europe, the Mediterranean, Africa, the Middle East, and the Indian Subcontinent, and now naturalized in other places (New Zealand, Australia, Mexico, southwestern United States, etc.). It reaches its maximum diversity in the Mediterranean rangelands (Malmir et al., 2018). The family consists of three subfamilies: *Asphodeloideae* Burnett (including 13 genera), *Hemerocallidoideae* Lindley (including 19 genera) and *Xanthorrhoeoideae* M.W. Chase (with only one genus) (Malmir et al., 2018). As with other geophytes, *Asphodelus aestivus* Brot. is a dominant species in some degraded Mediterranean ecosystems. These regions are sometimes referred to as "asphodel deserts" (Sawidis et al., 2005).

A severe neurological syndrome accompanied by intense neuronal pigmentation in sheep in Turkey was observed after the ingestion of summer asphodel (Calis et al., 2006). The high silicon (Si) content of mature leaves contributes to its unpalatability, whereas the tubers are protected from herbivores through the accumulation of defense substances, such as alkaloids that are harmful to sheep and goats (Rhizopoulou et al., 1997).

Currently, mechanical, cultural and chemical methods are used in weed management. These methods include such as grubbing, mowing and herbicides (DiTomaso et al., 2010). Fertilization can increase the density of grasses and other families in the rangeland areas and restrict some species such as *Asphodelus aestivus* from dominating (Aydin and Uzun, 2000; Masters and Sheley, 2001; Yavuz et al., 2008). Grubbing is a mechanical treatment, but the high cost of this treatment limits its use to control rangeland weeds. Mowing is often used to control annuals but can occasionally reduce seed production and provide suppression of biennials and perennials (Rinella et al., 2001; DiTomaso et al., 2010). On rangelands, herbicides are the most frequently used tool for the control of invasive and dominant species. The most used herbicides on rangelands are 2,4-D (auxin-like growth regulator that selectively controls broadleaf species), glyphosate (a non-selective foliar-applied systemic herbicide), paraquat (photosystem 1 energized cell membrane disrupter contact herbicide) (Gokkus and Koc, 1996; Masters and Sheley, 2001; DiTomaso et al., 2010).

The density of asphodel is gradually increasing in the rangeland areas where it is mismanaged for a long time in the region. Different control methods have been tried in order to suppress this species from being more dominant than other species in the rangeland. Due to the lack of research on summer asphodel control in the region, the study was designed and performed for 3 years.

MATERIALS AND METHODS

The experiment was carried out in Cakmar rangelands (37^o 45' N, 27^o 45' E) in Kocarli district of Aydin, which has an altitude of 60 m with Mediterranean climate, for a period of 3 years between 2016-2018. When the climate data of the experimental area were viewed, fluctuations were observed between years according to the average of the three years, while the average temperature was similar. Decreases in rangeland production were observed due to lower amounts and irregular distribution patterns of precipitation. The most obvious difference was recorded rainfall in January (2017) compared to the long-term average (Figure 1.).

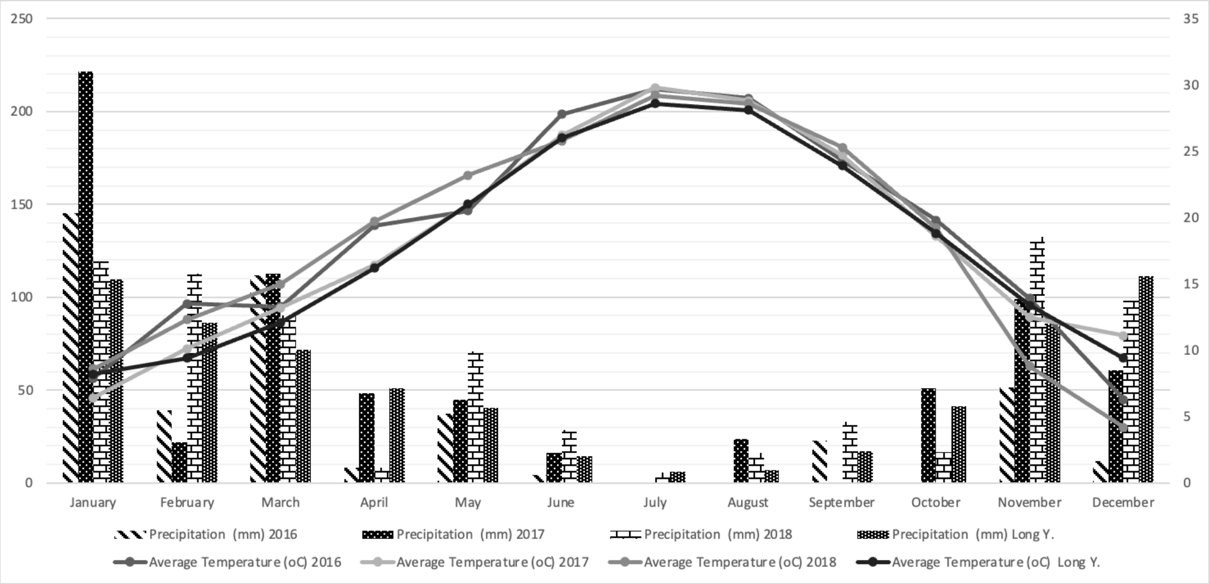

Figure 1. The average monthly meteorological datas for the years of 2016-18 and long years of Aydin province (Turkish State Meteorological Service, 2019).

The soil samples of the experimental area taken from 0-30 cm depth were analyzed at the laboratories of the Department of Soil Science and Plant Nutrition at the Faculty of Agriculture, Aydin Adnan Menderes University. According to the results, it was observed that the experimental area soils have sandy loam texture, a slightly alkaline with pH of 7.54 and rich in soil organic matter with 3.26%. According to other macro and micro-element analyzes, it was determined that except for Na (30 ppm) and Mg (80 ppm), the other nutrient elements were sufficient or high.

The experiment was established in the randomized complete block design. Each plot was designed in size of 4 x5 m. In the experiment, control, mowing, fertilization, paraquat (N, N′-dimethyl-4,4′-bipyridinium dichloride), glyphosate (N-(phosphonomethyl)glycine), 2,4-D (2,4-Dichlorophenoxyacetic acid), 2,4-D + Picloram and grubbing were performed. The mowing process was carried out at the time when the budding or beginning of the flowering in which the stage of summer asphodels has the lowest energy in storage organs (Altin, 1992). Fertilizer application consisted of 50 kg ha^{-1} P_2O_5 and 100 kg ha^{-1} N. Herbicides are applied directly to summer asphodels in the 1st and 2nd year in 3-5 leaves growing stage (Darrell, 2005). The grubbing was practiced to remove all of summer asphodel (*Asphodelus aestivus* Brot.) tubers in the parcel. Asphodel density was determined by counting plants in 2 quadrats (0.50 x 0.50 m) in each parcel. In the experiment, harvesting operations were carried out in each of the parcels during the beginning of the flowering period of common grasses, leaving 5 cm stubble in 4 (0.50 x 0.50 m) quadrats. Samples taken from each plot were separated considering functional plant groups. In the laboratory, the samples were dried in the oven (Mikrotest, MST) for 48 hours at 70 °C and weighted to determine botanical composition (Cook and Stubbendieck, 1986). The samples, whose dry weight was measured, were then ground in the grinding mill and prepared for chemical analysis. Neutral Detergent Fiber (NDF), and Acid Detergent Fiber (ADF) were measured using ANKOM (ANKOM^{200}, Ankom Technology, Fairport, NY) for fiber analysis (Van Soest et al., 1991). Nitrogen determination was made by using Kjeldahl method and the nitrogen content was multiplied with 6.25 coefficient to calculate the crude protein ratio (AOAC, 1990). Following the measurements, crude protein yield (kg ha^{-1}) was calculated by proportioning with hay yield. Relative feed value (RFV) was calculated using the formulas (Horrocks and Vallentine, 1999).

An arc-sine transformation was applied to botanical composition and summer asphodel values. ANOVA was performed for all data considering repeated measurement and means were compared using Duncan multiple range tests using SAS statistical software (SAS Institute, 1998).

RESULTS AND DISCUSSION

Management practices cause significantly changes in the botanical composition of natural rangelands. In the study, we observed the effects of different applications on legumes, grasses, and other families percentages (Table 1.). The data showed that all applications (years and treatments) and their interaction effects also significantly affect functional plant group percentage. While grasses percentage was about 17% in first two years it increased significantly in 3rd year. Whereas legume percentage decreased significantly in the second year and increased again in the third year. The other families percentage was 65.25% and it increased significantly in the second year and then decreased sharply.

Table 1. Percentage of botanical composition by weight of families in different suppression methods (%)

	Grass (%)	Legume (%)	Other Families (%)
Year			
2016	17.18 b	17.54 a	65.27 b
2017	17.52 b	2.85 c	79.61 a
2018	50.71 a	16.32 b	32.96 c
Applications			
Control	6.13 f	22.34 a	71.51 b
Mowing	40.94 a	3.78 e	55.27 d
Fertilization	24.90 d	17.92 b	57.16 d
Paraquat	26.29 d	10.41 c	63.29 c
Glyphosate	19.77 e	5.20 de	75.01 a
2,4-D	42.04 a	6.40 d	51.55 e
2,4-D+Picloram	31.06 c	17.92 c	57.70 d
Grubbing	36.64 b	20.60 a	42.74 f
Mean	28.47	12.23	59.28
Year	**	**	**
Applications	**	**	**
Y*A	**	**	**

*: P≤0,05 **: P≤0,01 ns: non-significant

All treatments caused significantly increases in grasses percentage compared to control, however, 2,4-D and moving caused the highest increases. The increase in grasses, which has increased since the beginning of the experiment, was evident especially in the 3rd year. However, fertilization and 2,4 D + Picloram applications caused higher increases in grasses percentage. (Figure 2.). Except for grubbing, all treatments caused significantly decreases in legume percentage, but the decreases were more pronounced in moving, glyphosate and 2,4-D treatments (Table 1.). In terms of legumes, the rate changed greater in the first year in the areas where the application took place, but a decrease was observed in all applications in the second year. By the 3rd year of the experiment, the increase in legumes seen in 2,4 D + Picloram applications was more than in other applications (Figure 3.).

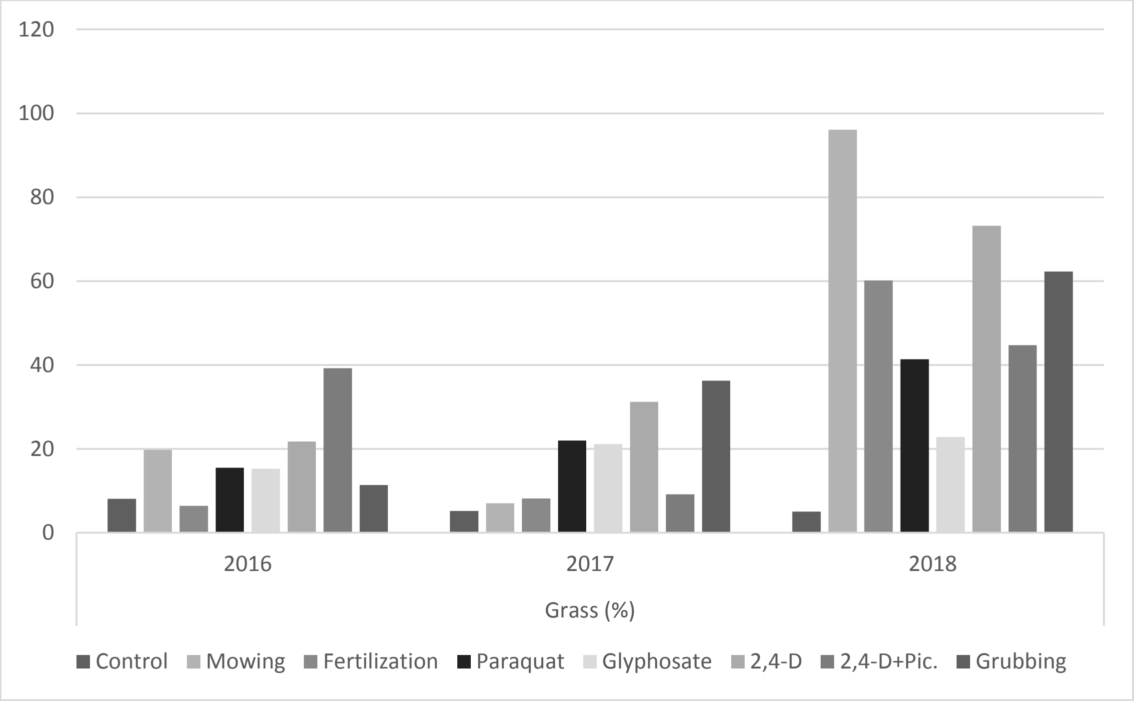

Figure 2. Change in grass ratio depending on years and practices

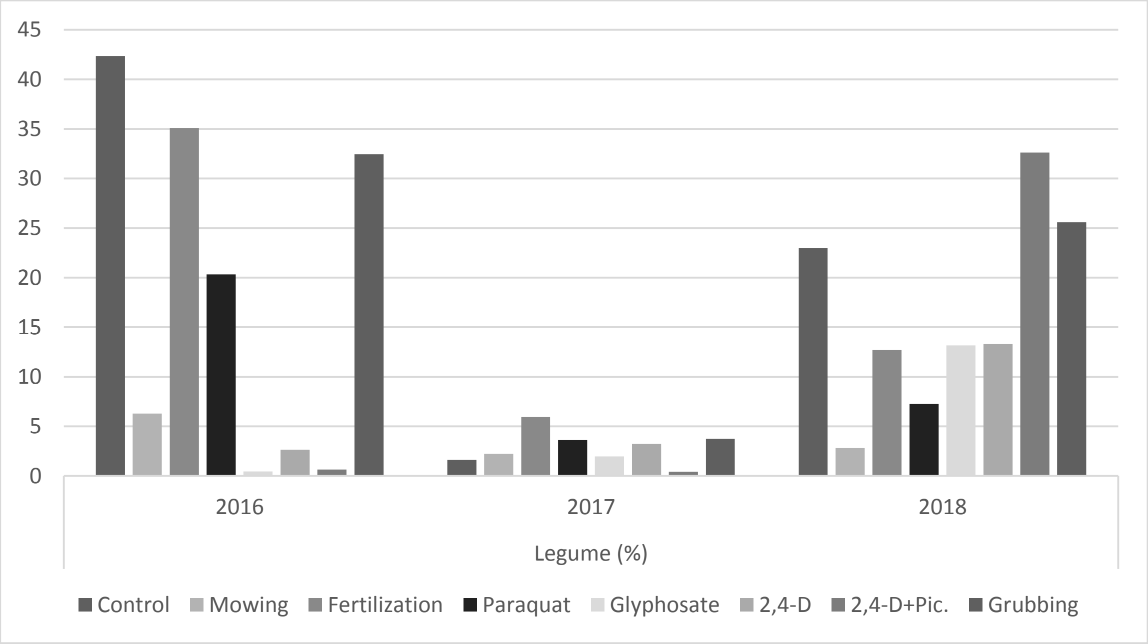

Figure 3. Change in legume ratio depending on years and practices

Except for glyphosate treatment, the other families percentage decreased significantly depending on treatments compared to control. However, the highest decreases were observed in grubbing treatment (Table 1.). In terms of other families, while the 2nd year values increased in general, a decrease was observed in most of the applications in the 3rd year. Among them, the fertilization application effect was clear in the 3rd year (Figure 4.).

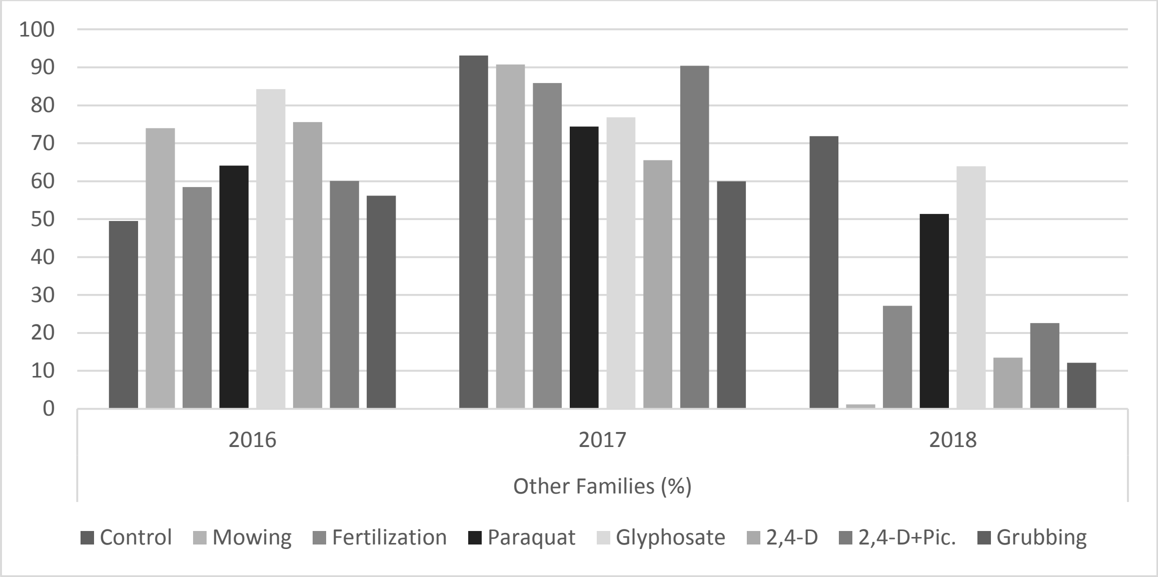

Figure 4. Change in other families ratio depending on years and practices

The summer asphodel population decreased in the 2nd and 3rd years of the experiment in all suppression methods except control. While it was determined that the highest decrease in the summer asphodel density was in the 2nd year of paraquat application, the least change was seen in the fertilization applications. Paraquat and glyphosate applications have been the most effective methods for reducing summer asphodel (Figure 5.).

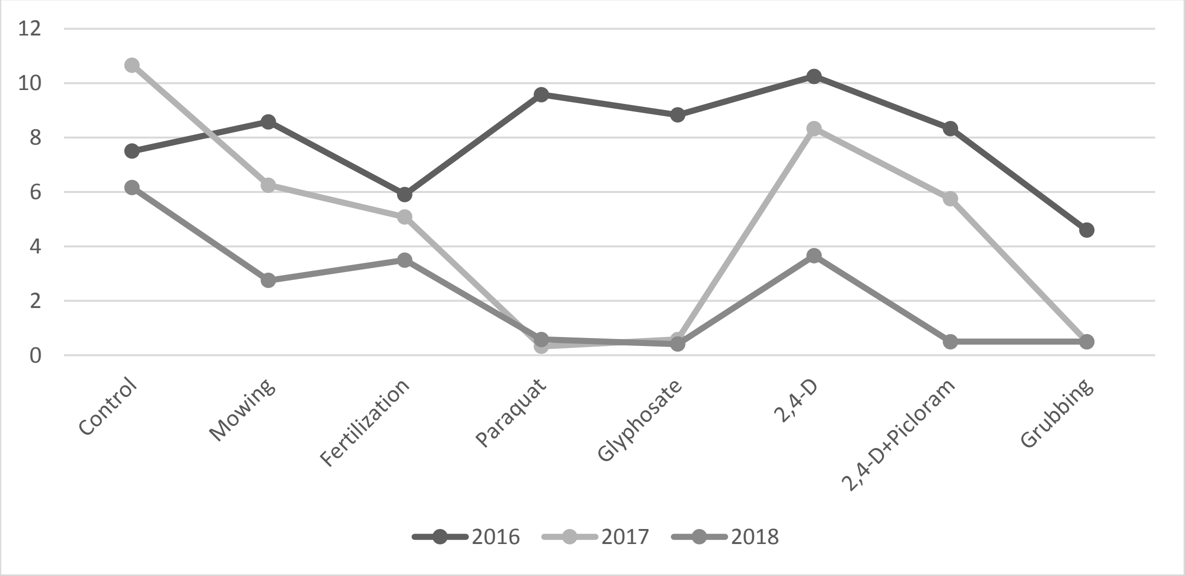

Figure 5. Changes in *Asphodelus aestivus* Brot. population density depending on years and applications

In all applications, decreases in hay yield were observed as of the 2nd year. However, paraquat has been the application in which the yield increased rapidly after the 2nd year (Figure 6). While yield decreases were observed in other applications, the change in botanical composition caused the yield to be affected. The highest hay yield was determined in paraquat application with 4443.3 kg ha^{-1}. While other applications did not reach this efficiency, the lowest efficiency was seen in the glyphosate application (Table 2.). It is seen that this systemic herbicide with low yields depending on the years in glyphosate application, causes a decrease in yield due to a decrease in the population. The crude protein ratio has increased over the years due to the decrease in yield and the increase of legume species in the botanical composition.

Table 2. Hay yield (kg ha^{-1}), Crude Protein Ratio (%) and Crude Protein Yield (kg ha^{-1}) averages of different suppression methods

	Hay yield (kg ha^{-1})	**CPR (%)**	**CPY (kg ha^{-1})**
Year			
2016	4798.3 a	15.98 ab	808.5 a
2017	2709.6 b	14.59 b	388.6 b
2018	2484.6 b	16.70 a	327.8 b
Applications			
Control	3403.3 bc	17.78 a	592.2 ab
Mowing	2881.1 cd	14.93 bc	428.4 bc
Fertilization	3590.0 b	17.60 a	637.7 a
Paraquat	4443.3 a	17.12 ab	561.8 ab
Glyphosate	2411.1 d	12.24 d	332.9 c
2,4-D	3734.4 b	13.88 cd	518.2 ac
2,4-D+Picloram	2687.8 d	13.70 cd	366.4 c
Grubbing	3495.6 b	18.82 a	628.7 a
Mean	3330.8	15.76	508.3
Year	**	*	**
Applications	**	**	**
Y*A	**	**	**
	*: P≤0,05 **: P≤0,01 ns: non-significant		

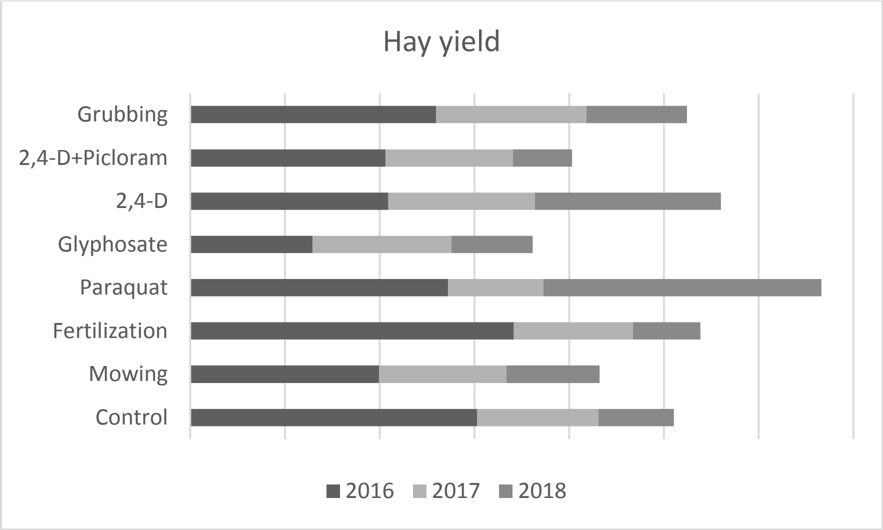

Figure 6. Change in hay yield (kg ha^{-1}) depending on years and practices

Grubbing, control, fertilization and paraquat applications are the applications with the highest crude protein ratios, 18.82, 17.78, 17.60 and 17.12%, respectively. Especially in the 3rd year of the experiment, the increase in the rate of legumes caused this situation. (Table 2.) While the sudden decrease in grubber application was remarkable, there was a continuous increase in mowing application (Figure 7.). Crude protein yield is also among those that show a decrease similar to hay yield. Crude protein yield is also among those that show a decrease similar to hay yield. Crude protein yield, which had an average of 808.5 kg ha^{-1} in the first year of the experiment, decreased to 327.8 kg ha^{-1}in the 3rd year of the experiment. Among the applications, the most decreases depending on the years are in fertilization and control applications (Figure 8.). Despite the increase in crude protein ratio, the decrease in the ratio of other families, which have an important role in yield, was also effective in crude protein yield. 2,4-D + Picloram, glyphosate and applications other than mowing had high values, while the highest values were obtained from fertilization. While the increase in crude protein ratio caused a decrease in fiber properties such as NDF and ADF, the difference between applications was not significant in terms of NDF (Table 3). Only a downward trend was observed over the years. The increase in yield and the decrease in crude protein ratio in Paraquat application caused an increase in ADF ratio. After all these, an increase is observed over the years according to the relative feed value obtained. While the value of 99.98 was obtained in the first year of the experiment, it increased to 112.78 in the third year. While there was a general increase among the applications, the highest increase was detected in the glyphosate application (Figure 9.).

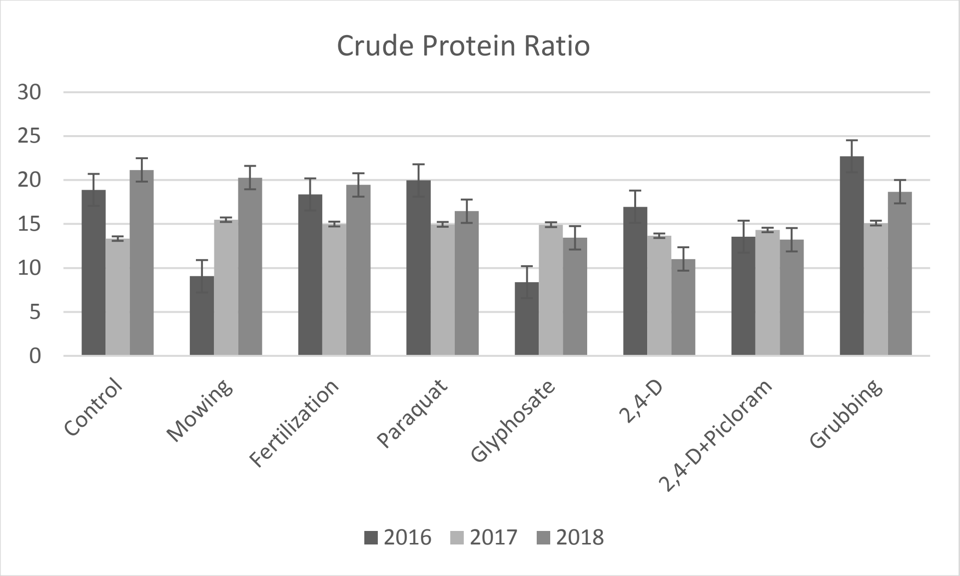

Figure 7. Change in crude protein ratio (%) depending on years and practices

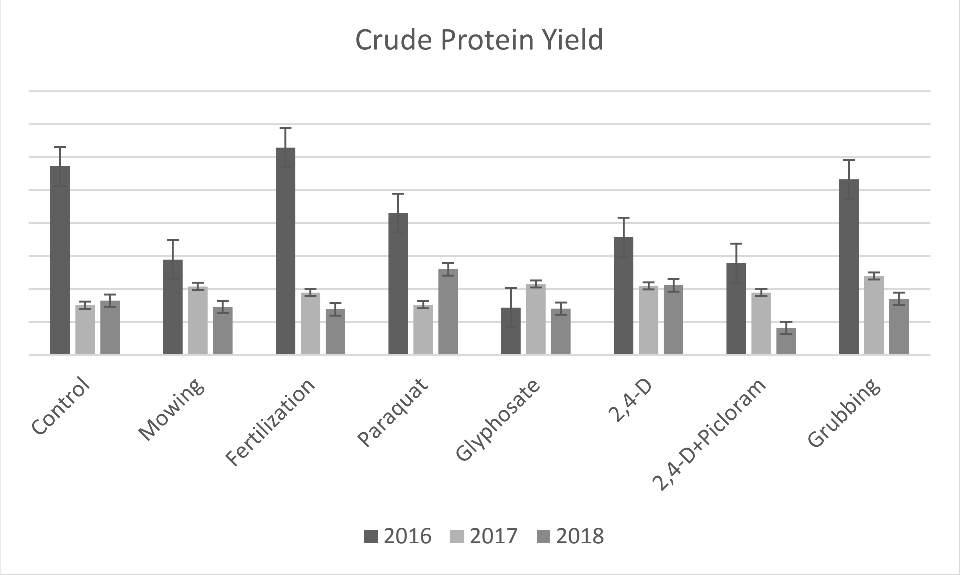

Figure 8. Change in crude protein yield (kg ha^{-1}) depending on years and practices

Table 3. NDF(%), ADF(%) and relative field value averages of different suppression methods

	NDF(%)	ADF(%)	RFV(%)
Year			
2016	52.72 a	41.78 a	99.98 b
2017	51.54 a	41.22 a	102.67 b
2018	50.03 b	36.29 c	112.78 a
Applications			
Control	49.27	38.88 c	110.70 a
Mowing	51.91	41.17 ab	102.36 cd
Fertilization	52.49	38.71 c	104.77 bd
Paraquat	51.82	42.04 a	100.85 d
Glyphosate	51.79	39.80 bc	104.61 bd
2,4-D	51.55	40.96 ab	103.41 bd
2,4-D+Picloram	51.59	38.34 c	106.50 ac
Grubbing	51.03	38.22 c	107.96 ab
Mean	51.43	39.77	105.14
Year	**	**	**
Applications	ns	**	**
Y*A	**	**	**

*: P≤0,05 **: P≤0,01 ns: non-significant

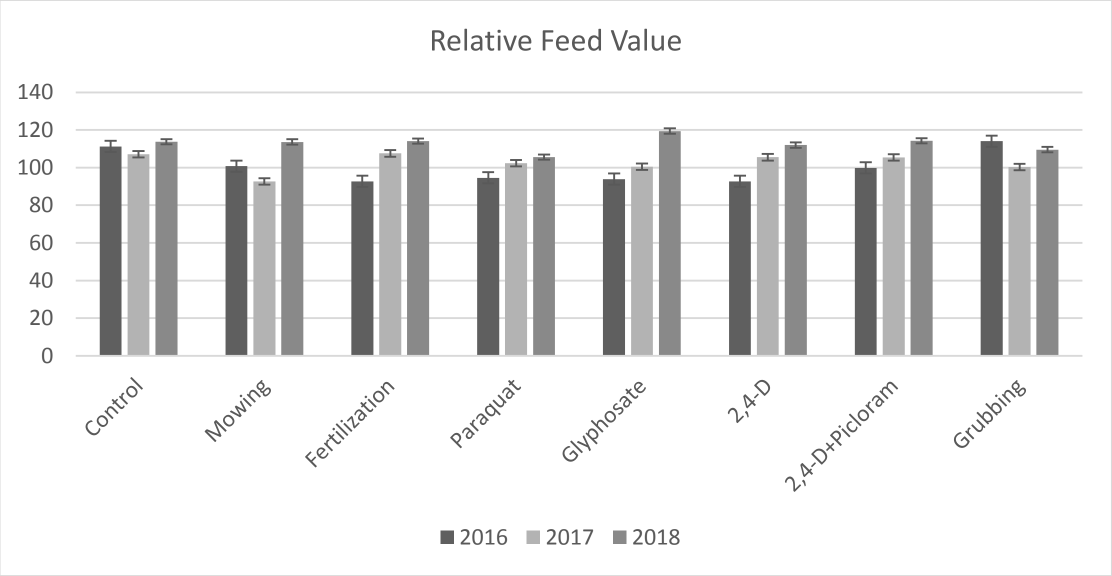

Figure 9. Change in relative feed value depending on years and practices

According to the effects of the applications, some researchers mentioned that fertilization has led to an increase in grass density and decreased legume density in the botanical composition (Kir, 1997; Tranel, 2000; Turk et al., 2005; Mut et al., 2010). whereas, some researchers claim that herbicide application reduces the weed population and encourages the grass population (Vallentine, 1989; Sheley et al., 2001).

Alaturk et al. (2018) reported that herbicide use could decrease the summer asphodel population. In addition to herbicide application to remove unwanted plants, fertilization can be done to increase plant growth and desired development can be achieved (Altin and Tuna, 1991).

Fertilization and the herbicide to be applied have the effect of increasing the rangeland yield. However, the cost calculation should be done while performing the applications (Tranel, 2000; Balabanli et al, 2010; Kowaljow et al., 2010; Mut et al., 2010; Sahinoglu and Uzun, 2016).

Suppression applications, which were carried out in the rangeland areas, have led to changes in yield, quality and botanical composition. Considering the characteristics that determine yield and quality, such as crude protein yield and relative feed value, it has been observed that glyphosate and paraquat applications have positive effects. In addition to these, it is clear that these herbicide applications are the most effective ways to remove the target species from the environment. Considering the positive contributions of other applications, these applications can be preferred to control this species (summer asphodel), which has a widespread problem in the Mediterranean rangeland. However, due to the large size of rangeland areas, it will be evaluated from an economic point of view and the most reasonable application will be preferred, which will facilitate the control of this species.

In the experiment, it was found that there were significant changes in crude protein content with applications. Changes in botanical composition due to applications and the disappearance of grazing pressure caused a decrease in crude protein content after the first year, while an increase in many applications in the following year. This is an expected situation when examined considering the botanical composition (Dovel, 1996; Severoglu and Gullap, 2020). Depending on the applications, the decrease and increase of the legume ratio in botanical composition brought about changes in the crude protein ratio.

ADF and NDF contents are indicators of the digestibility of forage crops (Ball et al., 2001; Rayburn, 2004) and it depends on the plant species (Ball et al., 2001). Changes in botanical composition resulted in significant decreases in ADF and NDF (Severoglu and Gullap, 2020). Forage quality declines with the advancing maturity because of the proportion of leaves in forage. As the CP concentration increases the ADF and NDF contents decrease together with the change of botanical composition (Erkovan et al., 2009).

Relative feed value is an important quality characteristic for determining the quality of forage crops. It is closely related to the fiber content, especially depending on the maturity (Jerenyama and Garcia, 2004). Depending on the decrease in ADF and NDF content, an increase in the RFV rate was observed in the trial. This negative relationship caused significantly changes in the years of the experiment (Table 3).

CONCLUSION

According to the results obtained from the experiment, environmental factors such as climate and management practices have significant effects on botanical composition change. However, there have been obvious decreases in weeds in applications without control plots and these have been replaced by desired species. Among the results obtained, depending on the years of change and not under the pressure of grazing in the rangeland, the grass population increased. When the crude protein yield, relative feed value and weed density are examined, the best application is grubbing. However, fact that grubbing requires intensive human power, thus, this application is not economical for broad-scale applications. Glyphosate and paraquat herbicide applications, which will be applied locally as an alternative to this application, have been determined as applications to increase the yield and quality of rangeland. The proper management after the applications are made will make these areas sustainable and will enable them to be used for many years.

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GENETIC AND ENVIRONMENTAL VARIABILITY, HERITABILITY AND GENETIC ADVANCE IN POD YIELD, YIELD COMPONENTS, OIL AND PROTEIN CONTENT OF PEANUT VARIETIES

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ABSTRACT

In this study, genetic and environmental variability, broad-sense heritability, genetic advance and correlation coefficients of pod yield, yield components, oil and protein content of ten registered peanut varieties were examined. Year (Y), location (L), year x location interaction (Y x L), genotype (G), genotype x year interaction (G x Y), genotype x location interaction (G x L) and genotype x year x location interaction (G x Y x L) were significant, but G x Y for shelling percentage was not significant. Genotypic and phenotypic variances were highest for pod yield followed by hundred pod weight, whereas all investigated traits of peanut varieties were significantly different. Broad sense heritability estimates ranged from moderate level to high. Heritability values were estimated to be maximum for shelling percentage (95.4%), hundred kernel weight (91.6%), hundred pod weight (88.3%), while moderate for pod number (63.8%), pod weight (60.4%), first quality pod ratio (63.3%), pod yield (63.2%), oil content (52.0%) and protein content (52.5%). High heritability for shelling percentage, hundred kernel weight and hundred pod weight indicated that these characteristics were affected less than the others by the environmental conditions. The magnitudes of genetic advance were observed to be very high (>50%) for hundred pod weight, 100 kernel weight, pod weight and pod yield; moderate (20-50%) for pod number, first quality pod ratio, shelling percentage and low (<20%) for oil and protein content. Significant and positive relationships were found the pod yield and pod number, pod weight, hundred pod weight and hundred kernel weight.

Keywords: Groundnut, genotypic and phenotypic variance, genetic advance

INTRODUCTION

Groundnut (*Arachis hypogaea* L.), which is generally planted in the tropics and semi-arid tropics, has found wide use in human nutrition (Kassa et al., 2009), animal husbandry (Savage and Keenan, 1994) and many branches of industry. Peanut is a legume crop and its seeds are highly nutritious due to its high oil and protein content. It is used in many areas such as peanut butter, roasted-salted peanuts and confectionery. Peanut seed contain 45-55% oil and 20-35% protein. Refined peanut oil is used in frying oil, salad oil, margarine, and mayonnaise, and for sauce production. Peanut oil is widely consumed in India, China and the USA. The antioxidant tocopherol substance found naturally in peanut oil increments the shelf life and stability of the oil (Akhtar et al., 2014; Zahran and Tawfeuk, 2019).

Peanut is one of the important oilseeds used in the oil and snack industry in the world. In Turkey, it is used completely as a snack. Peanut cultivation is carried out on an area of 55 thousand hectares in Turkey and 216 thousand tons of peanuts are produced annually (Anonymous, 2020). Peanut plant is very sensitive to changing environmental factors such as temperature, humidity and precipitation. Climate, soil, insect, disease and cultural practices differ from region to region. Therefore, genetic and environmental variability for yield, yield components, oil, and protein content should be estimated under different environmental conditions for successful breeding programs in peanuts. Yield and quality in peanuts are features that can be highly affected by environmental conditions (Prasad et al., 2000; Gulluoglu et al., 2018). Determining the response of cultivars and cultivar candidates to environmental conditions in terms of yield and quality characteristics is one of the important steps of plant breeding (Marfo and Padi, 1999; Ozturk and Yildirim, 2014). Seed yield and yield components of groundnut plant are influenced by several factors including environmental factors (temperature, rainfall, humidity, soil type, diseases and pests, etc.), genetic factors (cultivar or genotype), and interaction between genotype and environmental factors (Ku et al., 1998; Upadhyaya and Nigam, 1999; Dwivedi et al., 2000; Andersen and Gorbet, 2002; Isleib et al., 2008).

Genetic variations are significant for breeding strategies. The existence and magnitude of genetic variation allow the selection of new genotypes with different characteristics. Heritability indicates the degree to which characters are passed on to offspring. Genetic progression, on the other hand, describes the degree of progression reached in a particular variety by a given selection pressure. A high genetic progression value increases the probability of selecting plants with the most suitable traits. The objective of this study was to estimate the genotypic and phenotypic variance components, heritability, genetic advance and correlation coefficients for pod yield, yield components, oil and protein content of peanut genotypes.

MATERIALS AND METHODS

Study field description

The experiment was set up at three locations (Adana, Osmaniye and Kahramanmaras) and for two years (2018 and 2019) during the main crop peanut growing season. Adana, which is located in the Mediterranean region between 36°51'28.95" north latitude and 35°20'50.36" east longitude, has a slope of 1-2%. Its height above sea level is 12 m. Osmaniye, which is located in the Mediterranean region between 37°04'31" north latitude and 36°14'56" east longitude, has a slope of 1-2%. Its height above sea level is 130 m. Kahramanmaras, which is located in the Mediterranean region between 37°35'40.77" north latitude and 36°48'51.43" east longitude, has a slope of 3-5%. Its height above sea level is 491 m. The distance between Adana and Osmaniye, Osmaniye and Kahramanmaras is 102 km and 105 km, respectively.

The soils of the experimental field of all three locations have a clayey-loamy texture and the pH is slightly alkaline. Soils of the Adana location (18.54%) is more calcareous than those of the Osmaniye (9.47%) and Kahramanmaras (2.19%), and the organic matter contents of the soils is low at all locations (Table 1) (Anonymous, 2019a). The climate data for the experiment years and long years are given Table 2. All locations have a Mediterranean climate, with hot and dry summers and cold and rainy winters. The monthly average air temperature during the research period (April-October in 2018 and 2019) was 17.00 to 24.20°C in Adana, 16.30 to 23.10°C in Osmaniye and 14.20 to 21.30°C in Kahramanmaras. The total rainfall in 2018 and 2019 was 131.00 mm and 112.80 mm, 310.00 mm and 243.80 mm, 255.00 mm and 126.90 mm during the growing seasons in Adana, Osmaniye and Kahramanmaras, respectively (Anonymous, 2019b).

Table 1. Results of soil analysis of experimental areas at three locations.

Characteristics	L_1*	Explanation	L_2*	Explanation	L_3*	Explanation
Texture (% Sat.)	58.30	Clay - loam	57.20	Clay - loam	59.40	Clay - loam
Salinity (%)	0.10	Unsalted	0.07	Unsalted	0.13	Unsalted
Organic matter %	1.58	Very Low	1.29	Very Low	2.65	Low
Lime $CaCO_3$ (kg da^{-1})	18.54	High limy	9.47	Medium limy	2.19	Limy
Total nitrogen (%)	0.09	Insufficient	0.06	Insufficient	0.08	Insufficient
Phosphorus (mg kg^{-1})	13.80	Sufficient	8.64	Low	5.78	Low
Potassium (mg kg^{-1})	576.50	High	68.50	Low	112.10	Low
pH	7.66	slightly alkaline	7.63	slightly alkaline	7.53	slightly alkaline

(*) L_1: Adana location; L_2: Osmaniye location; L_3: Kahramanmaras location

Experimental materials description

Ten peanut varieties (Arioglu-2003, Batem-5025, Batem-Cihangir, Brantley, Flower-22, Halisbey, NC-7, Osmaniye-2005, Sultan and Wilson) used as the plant material in the experiment were obtained from Department of Field Crops of Agricultural Faculty of Cukurova University. Some characteristics of peanut varieties used in this research are given in Table 3.

Field management and experimental design

The field experiments at all locations were regulated in randomized complete block design with 3 replications. Before sowing, experimental plots had been fertilized with 300 kg ha^{-1} diammonium phosphate (DAP) (18% N, 46% P_2O_5) at three locations. Then, 150 kg ha^{-1} of urea was used as top fertilizer. Before sowing, seed spraying was done against crown rot disease and underground pests. In addition, spraying was applied against thrips damage. At all locations, seeds of the peanut cultivars were sown in the last week of April and first week of May in 2018 and 2019 respectively, in four rows of 5 m in length, with a sowing density of 70x15 cm (Kurt et al., 2016). In Adana, 3 times tractor anchors, twice hand anchors and 7 times sprinkler irrigation were performed. Two tractor anchors, two hand hoes, and 9 times irrigation were made in Kahramanmaras, and 2 times tractor and 2 times hand hoe, and irrigation was done 7 times in Osmaniye.

Table 2. Climatological data for long years and experiment years of three locations.

		Total rainfall (mm)			Average temperature (°C)			Average humidity (%)		
	Months	L_1*	L_2*	L_3*	L_1	L_2	L_3	L_1	L_2	L_3
Long years	April	50.27	81.93	59.02	18.50	17.78	16.45	63.46	63.46	52.08
	May	52.38	101.55	51.89	21.75	20.75	20.30	67.94	67.94	53.71
	June	25.09	27.90	8.25	26.28	25.33	26.03	67.60	67.60	46.43
	July	5.71	10.21	1.12	29.23	28.28	29.63	68.85	68.85	44.20
	August	6.26	8.23	0.95	29.50	28.83	29.50	69.58	69.58	48.76
	September	25.73	37.47	14.35	26.73	25.65	25.78	64.58	64.58	45.41
	October	29.52	65.17	37.90	22.43	21.10	19.63	58.17	58.17	48.18
	Total/Mean	194.96	332.46	59.02	24.92	23.96	23.90	65.74	65.74	48.40
	Experiment years									
2018	April	33.00	41.00	46.80	20.10	18.90	18.40	61.20	62.60	45.30
	May	29.20	64.80	52.90	24.40	23.10	21.70	62.80	65.70	52.60
	June	23.40	111.20	39.40	26.40	25.10	25.40	70.20	74.70	49.10
	July	0.00	1.80	0.30	29.10	27.80	28.60	69.80	73.30	46.20
	August	0.00	0.00	0.00	29.60	28.60	29.10	68.80	70.90	43.80
	September	22.80	0.00	0.60	27.90	26.90	27.20	63.60	64.80	38.40
	October	22.60	91.20	115.00	22.90	21.90	19.80	58.60	58.00	51.50
	Total/Mean	131.00	310.00	255.00	25.77	24.61	24.31	65.00	67.14	46.70
2019	April	59.40	46.60	78.40	17.00	16.30	14.20	67.00	69.80	61.80
	May	2.60	2.50	4.00	24.10	23.30	23.10	57.60	56.60	44.00
	June	13.80	72.30	6.20	27.10	26.00	27.20	68.70	71.00	48.00
	July	28.00	52.90	0.10	28.40	27.10	28.40	68.80	72.50	47.20
	August	0.00	8.20	0.10	29.60	28.50	29.50	68.00	69.80	47.70
	September	0.00	14.50	1.50	27.30	26.10	26.30	62.10	61.30	41.20
	October	22.80	46.80	36.60	24.20	23.10	21.30	61.60	61.60	55.10
	Total/Mean	112.80	243.80	126.90	25.38	24.34	24.28	64.83	66.08	49.28

(*) L_1: Adana location; L_2: Osmaniye location; L_3: Kahramanmaras location

Table 3. Groundnut cultivars and their some properties.

Varieties	Market type	Origin	Kernel size	Growing period
Arioglu-2003	Virginia	Turkey	Large	Semi-spreading
Batem-5025	Virginia	Turkey	Large	Semi-spreading
Batem-Cihangir	Virginia	Turkey	Large	Semi-spreading
Brantley	Virginia	USA	Large	Spreading
Flower-22	Virginia	Chine	Large	Semi-spreading
Halisbey	Virginia	Turkey	Large	Semi-spreading
NC-7	Virginia	USA	Large	Spreading
Osmaniye-2005	Virginia	Turkey	Large	Semi-spreading
Sultan	Virginia	Turkey	Large	Semi-spreading
Wilson	Virginia	USA	Large	Semi-spreading

Data collection

Peanut plants at all locations were harvested by machine after the physiological maturity period in October. Harvested plants were turned upside down and left to dry. After three days of drying, peanut pods were collected by hand. Pod number and weight was found by counting and weighing all pods of 20 harvested plants and dividing them by the number of plants (Gulluoglu et al., 2017). First quality pod ratio was determined by counting the large, plump and two-seeded pod of 20 plants harvested from each plot and proportioning to the total number of pods. One hundred randomly-selected pods with four replications were weighed and the average of a hundred pods' weight was determined. Pod yield was determined from an area 1.4 m wide and 4 m long of the center two rows of each plot (Gulluoglu et al., 2018). Shelling percentage for each cultivar was calculated from a 200 g randomly selected pod sample as the proportion of shelled seed weight to the total weight of the unshelled pods (Arioglu et al., 2018; Daudi et al., 2021). Additionally, a hundred seed weight for each cultivar was determined as an average weight of four samples of 100 randomly selected kernels per plot (Arioglu et al., 2018; Daudi et al., 2021). The oil and protein contents of the seeds were studied according to the Association of Official Analytical Chemists (AOAC, 2010).

Statistical analysis

All obtained data were subjected to analysis of variance (ANOVA) by using MSTATC statistical software. According to the method proposed by Burton and Devane (1953) and Johnson et al. (1955), genotypic and phenotypic variances and their corresponding coefficients of variation were estimated using the respective mean square expectations. In addition, broad-sense Heritability (H), genetic advance (GA) were calculated using the procedure of Hanson et al. (1956) and Allard (1999). Correlation coefficients were conducted following the procedure developed by Wright (1921).

RESULTS AND DISCUSSION

The variance analysis for pod yield, pod number, pod weight, first quality pod ratio, 100-pod weight, shelling percentage, 100-kernel weight, oil and protein content of peanut varieties over 2 years (2018 and 2019) and 3 locations (Adana, Osmaniye and Kahramanmaras) indicated that there were significant variations among the cultivars (G), years (Y), locations (L), Y x L interactions, G x Y interactions, G x L interactions and G x Y x L interactions for all traits except shelling percentage (Table 4). G x Y interaction for shelling percentage was not significant. The existence of G x Y, G x L and G x Y x L interactions for investigated characteristics in this study indicates that the effect of years and locations was different for peanut genotypes. Thus, additional attention to the response of cultivars is needed in peanut production systems where un-predictable environmental factors (temperature, rainfall, relative humidity) changed from one place of the region to another or from year to year.

Table 4. Combined analysis of variance results (mean square) for investigated traits of peanut varieties over 2 years and 3 locations.

Source	DF	PN	PW	FPR	HPW	PY	SP	HKW	OC	PC
Year (Y)	1	1224.9**	318.9**	1463.9**	3761.7**	4048425.2**	41.7**	147.9**	264.7**	270.9**
Location (L)	2	1448.2**	584.9**	1549.5**	40268.5**	12947.9**	144.7**	3043.2**	456.1**	709.7**
Y x L	2	5123.9**	2131.8**	386.2**	35695.4**	138137.5**	403.8**	1543.7**	341.2**	250.4**
Genotype (G)	9	158.5**	561.9**	457.9**	15111.5**	40717.7**	193.3**	1631.2**	35.9**	17.8**
G x Y	9	49.8**	169.5**	58.7*	366.2**	13771.4**	5.8^{ns}	45.2*	8.0**	7.6**
G x L	18	50.0**	58.8**	184.7**	1737.2**	4678.6**	9.1*	136.2**	19.1**	4.6**
G x Y x L	18	42.5**	88.3**	104.8**	547.2**	3485.5**	8.7*	49.2**	11.8**	3.7**
Error	108	7.4	19.8	22.8	107.8	265.3	4.3	20.8	0.9	0.3

DF= Degree of freedom, PN= Pod number (no. plant^{-1}) , PW= Pod weight (g plant^{-1}), FPR= First quality pod ratio (%), HPW= Hundred pod weight (g), PY= Pod yield (kg ha^{-1}), SP= Shelling percentage (%), HKW= Hundred kernel weight (g), OC= Oil content (%), PC= Protein content (%)
ns, *, ** = not significant, significant at 5% and 1% level, respectively.

The estimated variance components ($\sigma^2 g$, $\sigma^2 gy$, $\sigma^2 gl$, $\sigma^2 gyl$, $\sigma^2 e$ and $\sigma^2 p$), phenotypic (PCV) and genotypic (GCV) coefficients of variation, Heritability (H) and Genetic Advance (GA) in Table 5 were calculated using the data in Table 4. Error variances for all traits were smaller than those of the other components. Genotype-year ($\sigma^2 gy$) interaction did not occur for first quality pod ratio, hundred pod weight, shelling percentage, hundred kernel weight and oil content, but it was very high for pod yield (1142.8) followed by pod weight (16.6). The fact that the $\sigma^2 gy$ value is zero for first quality pod ratio, hundred pod weight, shelling percentage, hundred kernel weight and oil content shows that the year effect is not important and more emphasis should be placed on locations in genotype-environment interaction studies. Genotype-location ($\sigma^2 gl$) and genotype-year-location ($\sigma^2 gyl$) interaction variances were high in terms of pod yield and hundred pod weight. Genotypic and phenotypic variances were high for pod yield, hundred pod weight and hundred kernel weight. The genetic variance was higher than error variance, for pod weight, hundred pod weight, pod yield, shelling percentage, hundred kernel weight and oil content. This is an indication that these features were less affected by environmental conditions (Hamidou et al., 2012; Oteng-Frimpong et al., 2017). For all tested parameters, the Phenotypic Variance Coefficient (PCV) is higher than the Genotypic Variance Coefficient (GCV). This fact shows the great importance of environmental conditions on the expression of the trait. Similar results of higher PCV were observed by Patil and Bhapkar (1987), Mahalaxmi et al. (2005), John et al. (2006), Kadam et al. (2007) and Jakkeral et al. (2014). The magnitude of GCV and PCV for the investigated traits in the current study was between 0.9 and 1.1 for pod yield, and 10.2 and 10.8 for 100-pod weight, respectively. It indicates that there is no wide genetically based variation. Studies conducted by Patil and Bhapkar (1987), Patil et al. (2014) indicated that genotypes by environmental interactions were significant for pod yield and yield components. Kebede and Tana (2014) reported a highly significant effect of genotypes, environments, and their interactions on pod yield. Kushwah et al. (2017) stated that the coefficient of phenotypic and genotypic variation was high for pod yield and kernel yield per plant, number of pods and kernels per plant, and hundred kernel weight.

Table 5. Genetic parameters for investigated traits of peanut varieties.

Traits	Mean	Variance components [a]						Coefficient of variation (%)[b]		H [c] (%)	GA[d] Mean (10%)
		$\sigma^2 g$	$\sigma^2 gy$	$\sigma^2 gl$	$\sigma^2 gyl$	$\sigma^2 e$	$\sigma^2 p$	PCV	GCV		
PN	32.2	5.6	0.8	1.2	11.7	7.4	8.8	9.2	7.3	63.8	30.6
PW	57.5	23.4	16.6	6.5	22.8	19.8	38.7	10.8	8.4	60.4	71.7
FPR	68.7	17.7	0	13.3	27.3	22.8	28.0	7.6	6.1	63.3	45.4
HPW	264.4	728.6	0	198.3	146.4	107.8	825.2	10.8	10.2	88.3	485.0
PY	4189.7	1430.7	1142.8	198.8	1073.4	265.3	2262.1	1.1	0.9	63.2	60.1
SP	67.5	10.4	0	0.1	1.4	4.3	10.8	4.8	4.7	95.4	27.1
HKW	110.6	83.3	0	14.5	9.4	20.8	90.8	8.6	8.2	91.6	132.4
OC	52.9	1.1	0	1.2	3.6	0.9	2.2	2.8	2.0	52.0	3.8
PC	27.1	0.5	0.4	0.1	0.3	1.2	0.9	3.6	2.6	52.5	3.4

a = genotypic variance ($\sigma^2 g$), genotype – year variance ($\sigma^2 gy$), genotype – location variance ($\sigma^2 gl$), genotype – year – location variance ($\sigma^2 gyl$), error variance ($\sigma^2 e$), phenotypic variance ($\sigma^2 p$)

b = phenotypic coefficient of variability (PCV), genotypic coefficient of variability (GCV)

c = broad-sense heritability (H)

d = genetic advance (GA) at 10% selection intensity

PN= Pod number (no. plant^{-1}) , PW= Pod weight (g plant^{-1}), FPR= First quality pod ratio (%), HPW= 100-pod weight (g), PY= Pod yield (kg ha^{-1}), SP= Shelling percentage (%), HKW= 100-kernel weight (g), OC= Oil content (%), PC= Protein content (%)

Broad sense heritability estimates ranged from 52.0% (oil content) to 95.4% (shelling percentage) (Table 5). The heritability estimates for pod number (63.8%), pod weight (60.4%), first quality pod ratio (63.3%), and pod yield (63.2%), oil (52.0%) and protein content (52.5%) were at moderate levels (50-65%). On the other hand, heritability degrees were estimated to be at high levels (>65%) for 100 pod weight (88.3%), shelling percentage (95.4%) and 100-kernel weight (91.6%). High value of broad sense heritability for 100-pod weight, shelling percentage and 100-kernel weight is due to the high share of the genotypic variance component (Yildirim et al., 1979). These characters are highly heritable in peanut plant. The magnitude of the genotypic variance indicates that are significant variations within peanut varieties in terms of pod and seed weight and kernel ratio. High values of broad-sense heritability were reported for seed weight (87-93%), 100-pod weight (88-91%), pod yield (71-74%) and 100-kernel weight (87-96%) by Tossim et al. (2020). Rao et al. (2014) reported high heritability for shelling percentage (92.2%), pod number (76.9%), kernel weight (97.4%) and pod yield (95.5%). High heritability for hundred kernel weight, pod number, and pod yield was reported by Savaliya et al. (2009), John et al. (2007) and Khote et al. (2009).

The magnitudes of genetic advance were observed to be high (> 20%) for pod number, pod weight, first quality pod ratio, 100-pod weight, pod yield, shelling percentage and 100-kernel weight and low (< 10%) for oil and protein content (Table 5). The high magnitude of genetic advance was reported earlier by Mahalaxmi et al. (2005) and Jakkeral et al. (2014) for pod yield and its components. A high level of genetic advance helps in determining the appropriate character for selection. Heritability estimation along with genetic progression provides insight into the genetic makeup of the population. High or moderate heritability accompanied by high genetic advance for pod number, pod weight, first quality pod ratio, 100-pod weight, pod yield, shelling percentage and 100-kernel weight suggested that selection can be effective for these traits based on phenotypic expression. These results explain the additive gene action and indicate phenotypic selection to be effective (Kushwah et al., 2017). Similar results of moderate to high heritability coupled with moderate to high genetic advance were observed earlier for these traits by Azad and Hamid (2000), Dashora and Nagda (2002), Mahalaxmi et al. (2005), Cholin et al. (2010), Shinde et al. (2010) and Jakkeral et al. (2014). The low magnitude of heritability and low magnitude of genetic advance was observed for oil and protein content. This indicated that there was narrow genetic variability within peanut varieties for oil and protein content. Due to the narrow genetic diversity, the success of these varieties in peanut breeding (selection or hybridization) for oil and protein ratio will be low.

Simple correlation coefficients calculated among examined characteristics are shown in Table 6. Significant and positive correlations of pod yield with pod number (0.35), pod weight (0.95), 100-pod weight (0.56) and 100-kernel weight (0.56) were found. Pod number exhibited significant positive correlation with pod weight (0.40), 100-pod weight (0.46), shelling percentage (0.25), 100-kernel weight (0.36), oil content (0.36) and protein content (0.21). There were positive and significant correlations between pod weight, 100-pod weight, shelling percentage and 100-kernel weight. The first quality pod ratio was positively and significantly correlated with 100-kernel weight and protein content. The correlations between oil content and 100-pod weight, shelling percentage, 100-kernel weight were found to be positive and significant.

Table 6. Simple correlation coefficients for investigated traits of peanut varieties.

Traits $^+$	PN	PW	FPR	HPW	PY	SP	HKW	OC
PW	0.40**	-						
FPR	0.08	-0.30**	-					
HPW	0.46**	0.57**	0.12	-				
PY	0.35**	0.95**	-0.28**	0.56**	-			
SP	0.25**	0.20**	0.02	0.06	0.13	-		
HKW	0.36**	0.54**	0.22**	0.95**	0.56**	0.12	-	
OC	0.36**	0.13	-0.19**	0.37**	0.14	0.25**	0.28**	-
PC	0.21**	-0.23**	0.32**	-0.15	-0.22**	0.04	-0.08	0.05

*, ** Significant at 5% and 1% level respectively.
+ = Pod number (PN), pod weight (PW), first quality pod ratio (FPR), 100-pod weight (HPW), pod yield (PY), shelling percentage (SP), 100-kernel weight (HKW), oil content (OC), protein content (PC)

Pod yield is a complex character that can be directly or indirectly affected by many characteristics. For this reason, taking into account the important characters that affect the yield in the selection will significantly increase the success. There was positive and significant correlations between pod yield, kernel yield, pod number per plant and hundred kernel weight (Rao et al., 2014). Chishti et al. (2000) reported positive and significant correlations between pod yield and pod number, pod weight, 100-kernel weight and oil content. Deshmukh et al. (1986) reported a positive and significant correlation between pod yield and the number of pods in peanut plants. Pod number per plant, pod weight, 100-pod weight and 100-kernel weight contributed significantly to pod yield. It is concluded that successful selection can be made on these characteristics for pod yield.

CONCLUSIONS

In this study, genetic variability, broad-sense heritability, genetic advance, and correlation coefficients among 10 peanut cultivars over 2 years and 3 locations were estimated for nine agro-morphological and quality traits. Genotypic and phenotypic variances were high for pod yield, hundred pod and kernel weight. Broad-sense heritability were estimated to be high for hundred pod weight, shelling percentage and hundred kernel weight. The magnitudes of genetic advance were observed to be high for pod number, pod weight, first quality pod ratio, hundred pod weight, pod yield, shelling percentage and hundred kernel weight. Significant and positive associations of pod yield with pod number, pod weight, hundred pod and kernel weight were found. Pod number, pod weight, hundred pods, and kernel weight were found to be the most significant characteristics that should be considered during selection.

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THE EFFECTS OF DIFFERENT SOWING TIMES AND HARVEST STAGES ON FORAGE YIELD AND QUALITY IN BUCKWHEAT (*Fagopyrum esculentum* Moench)

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ABSTRACT

This study was carried out to determine the effects of different sowing times and harvest stages on the yield and forage quality of buckwheat in Marmara's conditions. The experiment was conducted in 2018 and 2019 in Bursa Uludag University Agricultural Application and Research Area using the Gunes variety as plant material. The experiment arranged randomized complete block design with split plot arrangement having three replications. Four different sowing times (15 April, 1 May, 15 May and 1 June) and three different harvest stages (flowering, milky and dough) were considered in the main plot and sub-plot, respectively. Results showed that higher plant height and dry matter yield values were obtained from plants sown on 15 April and harvested during the milky or dough stages. The highest crude protein ratio, crude protein yield and relative feed value, and the lowest acid detergent fiber and neutral detergent fiber values were determined in plants that were sown early and harvested during flowering stage. In general, delaying the harvest stage increased the yield of buckwheat but negatively affected the forage quality.

Keywords: Buckwheat, dry matter yield, harvest stage, quality, sowing time

INTRODUCTION

Buckwheat production worldwide is estimated to be at 3.8 million tons, with the majority of this production occurring on the Asian continent (Bicer, 2019). China, Russia, Ukraine and Kazakhstan are among the countries where the buckwheat plant is grown the most (Er, 2018). Although buckwheat is a very new plant for our country, it has many uses. Particularly due to the plant's rapid vegetative growth and high dry matter yield, it has been considered as an alternative roughage source in recent years (Amelchanka et al., 2010; Kalber et al., 2012). This plant has recently been used into ruminant rations in a variety of forms, including fresh, silage, and grain (Amelchanka et al., 2010; Keles et al., 2017). Kara and Yuksel (2014) reported that the feed quality of buckwheat is poorer than alfalfa and sainfoin, but is comparable to that of corn, sorghum, sunflower and triticale. While buckwheat harvested during the early flowering period (5-6 weeks) contains a 15-20% crude protein content, this value may decrease to as little as 9% depending on maturation (Björkman and Chase, 2017).

Turkey's agricultural-ecological structure is suitable to successfully growing a variety of forage plants that can meet the demand for high-quality roughage. Buckwheat is one of the most important species among the alternative plants that could be produced in Turkey to increase the amount of high-quality roughage. It can reach milky stage in a short time due to its rapid growth. Therefore, it is possible to grow buckwheat as roughage in early spring and autumn in locations with a Mediterranean climate where the fields were previously uncultivated (Yavuz, 2014; Er, 2018).

Determining the appropriate sowing date to achieve high yield and quality is a critical factor for not only buckwheat, but for all plants. The climatic factors of the region should be considered while determining the optimal sowing date for buckwheat because buckwheat is a sensitive plant that is susceptible to both frost and high temperatures during flowering (Kaya, 2018). Harvesting is an important stage that affects yield and quality of buckwheat to be used as roughage. It has been determined that buckwheat grass harvested at 100% flowering period yields more but has a lower quality compared to the that harvested at 50% flowering period (Surmen and Kara, 2017). Keles et al. (2012) reported a 5501-5900 kg ha^{-1} dry matter yield for buckwheat grown in Konya's conditions and harvested at the milky stage. Kara (2014) determined that 8530 kg ha^{-1} dry matter yield was obtained from the harvest collected during the stage when the grains had turned 50% brown, but the harvest was more suitable in terms of forage quality and economy during full flowering. Gullap et al. (2021) reported that the highest dry matter yield (4784 kg ha^{-1}) was recorded when the buckwheat was harvested at the full flowering stage.

This study was conducted to determine the optimal sowing time and harvesting stage for buckwheat grown under the ecological conditions of Bursa for high forage yield and quality.

MATERIALS AND METHODS

Experimental materials

The research was conducted in 2018 and 2019 in the experimental area of Bursa Uludag University’s Faculty of Agriculture Agricultural Application and Research Center (40° 11ˈ N, 29° 04ˈ E). Gunes cultivar, which was developed by Bahri Dagdas International Agricultural Research Institute, was employed as plant material in the research. The Bahri Dagdas International Agricultural Research Institute registered the Gunes variety as the first buckwheat variety in Turkey in 2014. Although is highly adaptable to all regions of Turkey, the variety demands for a humid and cool climate with low temperature for optimal yield. This variety has a short vegetation period (8-14 weeks), white flowers, and an average height range of 85-100 cm (Anonymous, 2014).

Bursa province generally has a temperate climate, although both the mild and warm climate of the Sea of Marmara in the north and the harsh climate of Uludag in the south are occasionally encountered. The hottest months are July-August, while the coldest are December-January in the province (Anonymous, 2020). The monthly total precipitation (mm), mean temperature (°C), relative humidity (%) values of 2018, 2019, and the long period (1975-2014) are given in Table 1. When the six months (April-September) of Bursa Province’s long-term average data are examined, it is seen that the total precipitation amount is 218.2 mm, the average temperature is 20.4 °C, and the relative humidity is 60.5%. While the total precipitation in May and June 2018 (the time of the experiment) was higher than in 2019, the total precipitation in August 2019 was much higher than the average of both 2018 and many other years. When the temperature values for the 6 months are examined, it was seen that the average temperature for the years during which the experiment was conducted was similar to the average of many years. Some soil properties of the experimental region from 0 to 20 cm depth are given in Table 2. The findings of the soil analysis were compared to the reference values (Muftuoglu et al., 2014).

Table 1. Precipitation, mean temperature and relative humidity in 2018, 2019 and long-term in Bursa

	Mean temperature (°C)			Precipitation (mm)			Relative Humidity (%)		
Months	LT	2018	2019	LT	2018	2019	LT	2018	2019
April	13.0	15.8	12.8	66.0	14.2	43.6	66.1	70.8	69.7
May	17.4	19.9	19.8	43.4	89.8	48.6	62.0	76.5	65.9
June	22.5	23.5	24.5	36.5	59.2	31.0	57.8	70.1	65.4
July	24.8	26.1	24.8	17.7	15.4	21.2	56.2	63.2	59.7
August	24.5	26.4	25.2	13.8	2.0	31.4	57.3	59.7	62.3
September	20.2	21.8	21.5	40.8	46.6	12.4	63.8	67.6	63.2
Total/Avg.	20.4	22.3	21.4	218.2	227.2	188.2	60.5	68.0	64.4

LT: Long-term (1975-2014).

Table 2. Properties of the experimental area soils in 2018 and 2019.

	2018		2019	
Properties	Value	Class	Value	Class
Sand, %	36.8	Soil texture Clay (C)	28.0	Soil texture Clay (C)
Loam, %	17.3		19.2	
Clay, %	45.9		52.8	
pH	7.675	Slightly alkaline	7.522	Slightly alkaline
EC, µS cm^{-1}	721.2	Low	813.7	Low
$CaCO_3$, %	4.10	Medium	3.75	Medium
Organic matter, %	2.08	Low	2.29	Low
Total nitrogen (N), %	0.098	Medium	0.195	High
Available phosphorus (P), mg kg^{-1}	21.15	Sufficient	28.78	High
Available potassium (K), g kg^{-1}	0.632	Very high	0.589	Very high
DTPA- iron (Fe) mg kg^{-1}	13.63	High	12.77	High
DTPA- copper (Cu), mg kg^{-1}	4.95	Sufficient	5.46	Sufficient
DTPA- zinc (Zn), mg kg^{-1}	1.71	Sufficient	1.56	Sufficient
DTPA- manganese (Mn), mg kg^{-1}	66.25	High	53.91	High

Experimental design

The experimental design was the Randomized Complete Blocks with three replications arranged in Split Plots. Sowing times (15 April, 1 May, 15 May and 1 June) were placed into the main plots and the harvest stages (flowering, milky and dough) into the subplots. The harvest times made depending on the sowing times and harvest periods in 2018 and 2019 are given in Table 3. Row spacing was set at 25 cm, the sowing rate was 80 kg ha^{-1} and sowing depth was 3-4 cm. In the experiment, the size of the plots were 7.5 m^2 (5 m x 1.5 m) and 6 rows were sown manually in each plot. Fertilizers were added as such: 60 kg N ha^{-1} and 60 kg P_20_5 ha^{-1} at sowing date. Ammonium sulfate (21%N) was used as a nitrogen fertilizer source and Triple Super Phosphate (TSP-44% P) was employed as a phosphorus source. Following sowing, a roller was rolled through the experiment area and drip irrigation was utilized to ensure germination and emergence of the seeds. After emergence, drip irrigation was applied at the beginning of flowering, during the intensive flowering stage, and during the milk and dough stages. Weeds were manually removed through hand hoeing.

Table 3. Harvest times of buckwheat in 2018 and 2019.

Sowing time	2018			2019		
	Harvest stage			Harvest stage		
	Flowering	Milk	Dough	Flowering	Milk	Dough
15 April	29 May	12 June	19 June	23 May	10 June	17 June
1 May	4 June	19 June	25 June	10 June	18 June	25 June
15 May	19 June	4 July	22 July	18 June	2 July	19 July
1 June	9 July	22 July	10 August	5 July	23 July	6 August

Measurements, harvest, and analyses

The average plant height was determined by measuring 10 randomly selected plants from the soil level to the tip of the plant. The leaves of these plants were then plucked and dried separately, and the percentage of leaves per plant was calculated by proportioning the leaves to the total plant weight. The plants were harvested from ground level using a sickle in the middle two rows of the plots after removing 0.5 m from both edges of the rows (2 rows x 0.25 m x 4 m=2 m^2). The samples were dried at 70 °C for 48 hours and weighed to calculate dry matter yield and the dry samples were grounded in preparation for analysis. The Kjeldahl method was used to determine the nitrogen (N) content, and the crude protein content was calculated using the formula N x 6.25 (AOAC, 1997). The amount of neutral detergent fiber (NDF) and acid detergent fiber (ADF), which form the cell walls in the ground samples, was determined according to the method described by Van Soest et al. (1991). The relative feed value (RFV) was determined using the equations developed by Van Dyke and Anderson (2000). To begin calculating the relative feed value, dry matter digestion (DMD %) is calculated from the ADF value (DMD % = 88.9 - (0.779 x % ADF)). Depending on the live weight of the animal, dry matter consumption (% DMI) is calculated from the NDF value (DMI % = 120 / NDF). Finally, DMD % and DMI % values are incorporated into the formula to calculate the RFV.

$$RFV = DMD\ \% \times DMI \times 0.775$$

All data were subjected to analysis of variance using the JUMP-7 package program in accordance with the 'Randomized Complete Blocks Design'. The significance tests employed probability levels of 1% and 5%, 5% probability levels were used in the determination of different groups, and the LSD test was used to determine distinct groups.

RESULTS AND DISCUSSION

The data obtained in this study on the effects of different sowing times and harvest stages on the forage yield and quality of buckwheat were subjected to analysis of variance. First order (two-way) and second order (three-way) interactions were significant for the analyzed traits (Table 4). Therefore, the following tables were arranged considering these significant interaction components.

Table 4. The results of analysis of variance combined over two years.

Source of Variation	**df**	**PH**	**SD**	**LR**	**DMY**	**CP**	**CPY**	**ADF**	**NDF**	**RFV**
Year	**1**	**	**	**	ns	**	**	ns	ns	ns
Sowing time (ST)	**3**	**	**	**	**	**	**	ns	*	*
Y X ST	**3**	**	**	**	**	ns	ns	ns	*	*
Harvest stage (HS)	**2**	**	ns	**	**	**	ns	**	**	**
Y x HS	**2**	**	ns	**	ns	ns	ns	*	ns	*
ST X HS	**6**	**	ns	**	*	**	**	ns	ns	
Y X ST X HS	**6**	ns	*	**	**	ns	ns	**	*	**

PH: Plant height, SD: Stem diameter, LR: Leaf rate, DMY: Dry matter yield, CP: Crude Protein, CPY: Crude protein yield, ADF: Acid detergent insoluble fiber, NDF: Neutral detergent insoluble fiber, RFV: Relative feed value

* Significant at $p<0.05$; ** Significant at $p<0.01$, ns: Non-significant

Plant height

The effects of the interactions year x sowing time, year x harvest stage, and sowing time x harvest stage on plant height were significant (P≤ 0.01). The greatest plant height (78.42 cm) was observed in a plant sown on 1 May 2019. This value was followed by one sown on 15 April (77.53 cm) which was statistically in the same group as the plant with the highest height. The lowest value of plant height (41.49 cm) was found for the plant sown on 1 June 2018 (Table 5). Kaya (2018) reported that plant height gradually decreased with the delay of sowing time in buckwheat, and that the height of plants sown on May 21 decreased by approximately 26% compared to those sown on April 20 in their study. Similarly, Gunes et al. (2012) and Acar (2019) also reported that plant height in buckwheat was influenced by sowing time and shortened as the sowing time was delayed. This may be the result of an increase in temperature and light intensity with later sowings as Jung et al. (2015) reported that the growth and yield of buckwheat were closely related to temperature, precipitation, and sunshine duration. In the study, while the plant height decreased by 42.80% from the first to the last sowing time in 2018, it decreased by 13.88% during the same period in 2019, making the interaction of year x sowing time a significant factor (Table 4 and Table 5). In terms of harvest stages, the greatest plant height (86.24 cm) was measured during the dough stage in 2019, and the lowest plant height (64.68 cm) during the dough stage in 2018 (Table 5). A rapid increase in height generally continues up until the flowering stage in plants. In plants with simultaneous flowering, height growth stops with flowering. However, in species that continue to grow after flowering, such as buckwheat, growth continues (Gullap et al., 2021). Polat (2019) reported that the greatest plant height was 89.79 cm with a 75% seed setting period under Konya’s ecological conditions. When the interaction of sowing time x harvest stage is examined, the greatest plant height of 87.32 cm was obtained from plants sown on 1 May and harvested during the dough stage. This was followed by plants sown on 15 April and harvested during the dough stage, which were statistically in a similar group (Table 6).

Table 5. The means of plant height measured at different sowing times and harvest stages in the field trial run with significant year x sowing times and year x harvest stage interaction.

Sowing times	Year		Harvest stage	Year	
	2018	2019		2018	2019
15 April	72.54 b	77.53 a	Flowering	49.90 e	57.58 d
1 May	71.94 b	78.42 a	Milk	64.82 c	79.16 b
15 May	53.22 d	74.58 ab	Dough	64.68 c	86.24 a
1 June	41.49 e	66.77 c			

**: significant at P ≤ 0.01. Means followed by the same letters are not different for P ≤ 0.05 according to LSD test.

Table 6. The means of plant height measured at different sowing times and harvest stages in the field trial run with significant sowing time x harvest stage interaction.

	Harvest stage		
Sowing time	Flowering	Milk	Dough
15 April	53.99 e	84.39 a	86.74 a
1 May	60.38 d	77.84 b	87.32 a
15 May	57.72 de	66.27 c	67.70 c
1 June	42.87 f	59.44 d	60.08 d

**: significant at P ≤ 0.01. Means followed by the same letters are not different for P ≤ 0.05 according to LSD test.

Stem diameter

The stem diameter was significantly affected by the combination and interaction of the year x sowing time x harvest stage (P≤ 0.05). The highest stem diameter of 5.21 mm obtained from plants sown on 15 April 2018 and harvested during the milk stage. On the other hand, the lowest stem diameters (3.79 mm and 3.84 mm) were obtained from plants sown on 1 June 2018 and harvested during the dough stage, and on 1 June 2018 and harvested during the flowering stage (Table 7). In the same harvest stage of 2018, there was a bigger difference between stem diameter development than in 2019, depending on the sowing times. In 2018, stem diameter decreased by 27 % from the first to the last sowing time in the dough stage, depending on the sowing times, but this decrease was only by 6 % in 2019. In this case, the interaction of year x sowing time x harvest stage was significant. Contrary to our study’s results, Karatas et al. (2020) reported that sowing times had no effect on the stem diameter.

Leaf rate

The effects of the interactions year x sowing times x harvest stage interaction on the leaf rate were found to be significant (P≤ 0.01). The highest leaf rate (48.30%) was determined in plants sown on 1 June in 2018 and harvested during the flowering stage, while the lowest leaf rate (18.13%) was determined in plants sown on 15 April in 2019 and harvested during the dough stage. In all other sowing occasions in the study except for the 15 May sowing in 2018, the leaf rate decreased gradually due to the progress of the harvest stage. Contrary to our study’s results, Gullap et al. (2021) reported that the leaf ratio of buckwheat increased depending on the delaying of harvest stage. On the other hand, Alkay and Kokten (2020) reported that the leaf rate of buckwheat gradually decreased as a result of later sowing, and therefore the highest leaf rate (15.91%) was obtained by sowing on April 25. In our study, the rate of leaves detected at the milky stage was 30.62% was higher than that reported by Keles et al. (2012). This may be related to the difference of genotypes, as well as ecological factors and different cultural practices. In the first year of the experiment, while the leaf rate was 36.36% at the first sowing time during the flowering stage, it was 48.30% at the last sowing time. In 2019, the leaf rate decreased by about 4% in the same stages, and in this case, the interaction of year x sowing time x harvesting stage was significant (Table 7).

Table 7. The means of stem diameter and leaf rate measured at different sowing times and harvest stages in the field trial run with significant year x sowing times x harvest stage interaction.

Years	Sowing times	Stem diameter (mm)			Leaf rate (%)		
		Harvest stage			Harvest stage		
		Flowering	Milk	Dough	Flowering	Milk	Dough
2018	15 April	4.67 e-h	5.21 a	5.19 ab	36.36 e	21.80 kl	19.80 lm
	1 May	5.19 ab	4.88 b-f	5.13 a-d	39.61 cd	25.61 ıj	23.84 jk
	15 May	4.34 ıj	4.47 g-ı	4.28 ıj	31.49 f	30.51 fg	36.58 e
	1 June	3.84 k	4.02 jk	3.79 k	48.30 a	44.54 b	28.21 gh
2019	15 April	4.96 a-e	4.93 a-e	5.14 a-c	42.07 bc	20.00 lm	18.13 m
	1 May	4.75 e-g	4.79 ef	4.83 d-f	30.49 fg	26.08 h-j	21.92 kl
	15 May	4.59 f-ı	4.93 a-e	4.98 a-e	36.64 e	26.73 hı	24.31 ı-k
	1 June	4.78 e-g	4.43 hı	4.82 d-f	38.19 cd	26.41 hı	25.98 h-j

Means followed by the same letters are not different for P ≤ 0.05 according to LSD test.

Dry matter yield

Dry matter yield was significantly affected by the interaction of year x sowing time x harvest stage (P≤ 0.01). The highest dry matter yield (4560.9 kg ha^{-1}) was obtained from plants sown on 15 April 2019 and harvested during the milky stage. The lowest dry matter yield (1459.6 kg ha^{-1}) was obtained from plants sown on 1 June 2018 and harvested during the flowering stage (Table 8). When Table 8 is examined, it is seen that while the dry matter yield obtained from plants sown on 1 May and harvested during the milky stage decreased by 22.56% by 15 May in 2018, it increased by 4.79% during the same period in 2019. In addition, while the dry matter yield from milky stage to dough stage increased by 10% in the first sowing time in 2018, it decreased by 14% during the same period in 2019. This may be due to the significance effect of the interaction of year x sowing time x harvest stage. In general, the yield may have differed as a result of the cooler and wetter July and August experienced in 2019 compared to 2018. This statement is supported by Temel and Yolcu (2020), having reported that sowing and harvesting times can have different effects on plant development periods depending on the annual changes in climatic conditions. In studies on this subject, it has been established that dry matter yield varies significantly depending on sowing times. For example; Omidbaigi and Mastro (2004) reported that in Iranian conditions, the highest dry forage yields were obtained by sowing on 5 July (25.2 g plot^{-1}) and 5 August (24.6 g plot^{-1}), Koksal (2017) reported that in Yozgat's conditions, the highest hay yield was obtained by sowing on 5 July (4420 kg ha^{-1}) in the first year and on 5 August (2410 kg ha^{-1}) in the second year of the experiment. In our study, dry matter yield had a moderate increase depending on the harvesting stage, but this increase differed significantly according to the sowing times. Some studies investigating the effect of harvesting stages discovered that dry matter yield increased gradually depending on the harvest periods (Kara, 2014; Polat, 2019; Gullap et al., 2021). For instance, Kara (2014) reported that dry matter yield in buckwheat varies year to year and increases approximately 6.3 times from the beginning of flowering to the period at which 50% of the grains have turned brown. Surmen and Kara (2017), on the other hand, reported that in Aydın's ecological conditions, the dry matter yield of pure buckwheat was 5497.5 kg ha^{-1} at 50% flowering period and 5085.2 kg ha^{-1} at 100% flowering period. The dry matter yields obtained in our research were lower than the yields obtained in previous studies on this subject. This may be owing to ecological, cultivar, and agronomic differences.

Crude protein

The effect of the sowing time x harvest stage interaction on the crude protein content of buckwheat showed statistically significant differences in the results (P≤ 0.01). As for the sowing time x harvest stage interaction, the highest crude protein ratio (22.66 %) was obtained from plants sown on 15 May and harvested during the flowering stage, while the lowest crude protein ratio (14.75 %) was obtained from plants sown on 15 May and harvested during the dough stage (Table 9). Different results were obtained from studies investigating the effect of sowing time on the crude protein ratio of buckwheat. The results obtained in our research overlapped with some of these studies and differed greatly from others. For example; Koksal (2017) reported that in Yozgat's conditions, the highest crude protein ratio (15.81%) was obtained from sowing on 19 May in the first year of the experiment, and that the effects of sowing times on crude protein ratio were insignificant in the second year. On the other hand, Alkay and Kokten (2020) reported that sowing time significantly affected the crude protein ratio in buckwheat, the highest crude protein ratio (9.88 %) was obtained from the plot sown on 5 May, and that the lowest value (8.76 %) was obtained from that sown on 25 May. Björkman and Chase (2017) determined that the crude protein ratio in buckwheat varies between 15 and 20 % prior to flowering and that these values later decrease to 9 % depending on maturity. Surmen and Kara (2017) reported that in Aydın's ecological conditions, the crude protein rate in pure buckwheat was 15.89 % in the 50 % flowering period and 13.56% in the 100 % flowering period. In our study, the crude protein ratios measured during the flowering stage were higher. This may be a result of differences in cultivar, ecological factors, and agronomic practices. Dvoracek et al. (2004) emphasized that the reason for the change in the protein ratio in the buckwheat plant is due to climatic factors rather than genotype.

Table 8. The means of dry matter yield and ADF measured at different sowing times and harvest stages in the field trial run with significant year x sowing times x harvest stage interaction.

Years	Sowing times	Dry matter yield (kg ha^{-1})			ADF (%)		
		Harvest stage			Harvest stage		
		Flowering	Milk	Dough	Flowering	Milk	Dough
2018	15 April	2940.7 e-g	3931.0 bc	4319.7 ab	27.58 e-g	33.63 a-c	28.32 d-f
	1 May	2435.0 f-ı	3791.1 bc	4285.5 ab	23.41 g-ı	36.47 a	33.70 a-c
	15 May	2122.7 ıj	2935.9 e-g	3578.4 cd	26.30 e-h	26.59 e-h	27.17 e-h
	1 June	1459.6 k	2308.3 hı	2041.8 ıj	25.92 e-h	27.11 e-h	26.87 e-h
2019	15 April	2950.3 e-g	4560.9 a	3942.3 bc	22.68 hı	30.30 b-e	32.57 a-d
	1 May	2976.1 e-g	2922.4 e-g	3958.9 bc	24.19 f-ı	28.12 d-g	30.41 b-e
	15 May	2413.5 g-ı	3062.5 de	3010.5 e	19.89 ı	28.52 d-f	29.64 c-e
	1 June	1581.1 jk	2978.8 ef	2827.3 e-h	20.03 ı	34.73 ab	28.13 d-g

Means followed by the same letters are not different for $P \leq 0.05$ according to LSD test.

Table 9. The means of crude protein (%) and crude protein yield (kg ha^{-1}) measured at different sowing times and harvest stages in the field trial run with significant sowing time x harvest stage interaction.

Sowing time	Crude protein (%)			Crude protein yield (kg ha^{-1})		
	Harvest stage			Harvest stage		
	Flowering	Milk	Dough	Flowering	Milk	Dough
15 April	21.61 a	12.80 e	11.40 e	638.3 a	546.1 b	464.6 cd
1 May	19.39 b	15.18 d	15.19 cd	524.4 bc	500.7 bc	621.3 a
15 May	22.66 a	15.97 cd	14.75 d	512.6 bc	479.0 b-d	482.5 b-d
1 June	19.45 b	16.17 cd	16.85 c	296.4 e	426.8 d	413.4 d

**: significant at $P \leq 0.01$. Means followed by the same letters are not different for $P \leq 0.05$ according to LSD test.

Crude protein yield

The effect of sowing time x harvest stage interaction on crude protein yield was found to be significant at the 1% probability level (Table 4). The highest crude protein yield (638.3 kg ha^{-1} and 621.3kg ha^{-1}) were detected in plots sown on 15 April and harvested during the flowering stage, and on 1 May and harvested during the dough stage. Especially, the high dry matter yield of buckwheat sown on 1 May and harvested during the dough stage caused the crude protein yield to be high. The lowest crude protein yield (296.4 kg ha^{-1}) was obtained from the plots sown on 1 June and harvested during the flowering stage (Table 9). Since crude protein yield is the product of dry matter yield and crude protein ratio, crude protein yield decreases in late sown and late harvested plants as a result of the decrease in both crude protein ratio and dry matter yield as Temel and Tan (2002) reported that the decrease in crude protein yield was due to the rapid decrease in hay yield resulting from the delay in sowing time. Alkay and Kokten (2020) reported that the crude protein yield of buckwheat grown in Bingol's conditions varied between 89.0-127.0 kg ha^{-1} and the crude protein yields obtained 25 April, 5 May, and 15 May sowing times were higher than crude protein yield obtained latest sowing time (1 June). Although the results obtained in our research are partially parallel with those of Alkay and Kokten (2020), the crude protein yields detected in our study were higher. This situation may be primarily explained by the variability of dry matter yields among different ecologies. Surmen and Kara (2017), who examined the effects of harvest stages in buckwheat-soybean mixtures, reported that the crude protein yield in buckwheat was 871.9 kg ha^{-1} at the 50% flowering stage and 689.3 kg ha^{-1} at the 100% flowering stage.

ADF

The effect of the interaction of year x sowing time x harvest stage on the ADF ratio was found to be significant at the 1% probability level (Table 4). The highest ADF ratio was found in plants sown on 1 May in 2018 and harvested during the milky stage, and the lowest in those sown on 15 May 2019 and harvested during the flowering stage. In some sowing times, there is a continuous increase trend depending on the harvest stage, while in some there is a fluctuating course. The fact that these trends are different between years has caused the triple interaction to be significant. For example, In the sowings made on April 15, 2018, depending on the harvest periods, the ADF ratio increased by 21.93% in the milky stage compared to the flowering stage, while this increase rate was 2.68% in the dough stage. On the other hand, at the same sowing time in 2019, these rates were 33.60% and 43.61%, respectively (Table 8). As the plant ages, the cell walls thicken, and the amount of structural substances in the plant increases. In this case, it leads to a decrease in the crude protein ratio, while the components representing the fibrous tissue such as ADF and NDF are increased. In the early phases of plant development, the stems contain nutrients in similar amounts to the leaves, and as the plant matures, the nutritional value of the stem decreases faster than that of the leaves (Gullap et al., 2021). Alkay and Kokten (2020) reported that the highest rate of ADF (42.04 %) in buckwheat was obtained by sowing on 15 May. The ADF values detected in our research were considerably lower than those of Alkay and Kokten (2020). Surmen and Kara (2017) reported that the ADF ratio increased from 28.04 % to 35.82 % in between 50% flowering and full bloom and Gullap et al. (2021) reported that it increased from 22.65 % to 29.66 % from the beginning of flowering to full bloom. Gullap et al. (2021) reported that the rate of ADF is higher in years when better plant growth is observed, structural substance production is increased, and accordingly, the dry matter yield is higher.

NDF

The effect of the interaction of year x sowing time x harvest stage on the NDF ratio of buckwheat was significant at the 5% probability level (Table 4). The highest NDF ratio was obtained from the plots sown on 1 May in 2018 and harvested during the milky stage, while the lowest was obtained from those sown on 15 May 2019 and harvested during the flowering stage. While the NDF ratio decreased from the first sowing time (15 April-42.08 %) to the last sowing time (1 June- 39.40 %) in 2018 in the harvests made during the milky stage, the NDF ratio increased in the same periods in 2019 (Table 10). In this case, the triple interactions were important. Alkay and Kokten (2020) reported that the highest rates of NDF (45.11% and 44.35%) were obtained by sowing on 5 May and 25 May, while the lowest NDF ratios resulted from sowing on 25 April and 15 May. In our research, the NDF rate of buckwheat increased gradually with the delay of the harvesting stage in all applications except for the 1 May and 15 May sowings in 2018, and 15 May and 1 June sowings in 2019. Surmen and Kara (2017) reported that the NDF ratio increased by approximately 28% from 50% flowering to 100% flowering. On the other hand, Gullap et al. (2021) reported that the highest NDF rate was obtained from harvesting at the full flowering and 50% flowering stages.

Table 10. The means of NDF and RFV measured at different sowing times and harvest stages in the field trial run with significant year x sowing times x harvest stage interaction.

Years	Sowing times	NDF (%)			RFV		
		Harvest stage			Harvest stage		
		Flowering	Milk	Dough	Flowering	Milk	Dough
2018	15 April	33.05 ıj	42.08 b-f	44.08 a-e	190.48 bc	139.61 g-k	143.48 g-k
	1 May	36.43 f-j	49.11 a	47.32 ab	185.62 b-d	115.39 k	124.79 jk
	15 May	40.14 c-h	36.60 f-j	37.92 e-ı	158.92 d-h	175.48 c-f	166.11 c-g
	1 June	34.74 h-j	39.40 d-ı	40.42 c-h	185.13 b-d	160.18 d-g	156.66 d-ı
2019	15 April	31.43 j	41.85 b-g	45.81 a-c	211.43 ab	146.42 f-j	128.99 h-k
	1 May	36.82f-j	38.26 e-ı	42.48 b-f	177.93 c-e	163.44 c-g	142.79 g-k
	15 May	30.86 j	41.38 b-g	40.85 c-h	221.40 a	150.24 e-j	150.23 e-j
	1 June	35.61 g-j	45.04 a-d	43.53 a-e	192.76 a-c	128.04 ı-k	143.22 g-k

Means followed by the same letters are not different for $P \leq 0.05$ according to LSD test.

RFV

The effect of triple interaction on RFV was found to be significant at the 1% probability level (Table 4). According to Table 10, the highest RFV was obtained from the plots sown on 15 May in 2019 and harvested during the flowering stage, and the lowest value was obtained from plants sown on 1 May in 2018 and harvested during the milky stage. In sowing times, there is a continuous increase trend depending on the harvest stage, while in some there is a fluctuating course. The fact that these trends are different between years has caused the triple interaction to be significant. For example, the RFV values detected in different harvest periods from the plants sown on April 15 in 2018 and 2019 clearly show this situation (Table10). The relative feed value (RFV) was developed in the USA to measure the nutritional value of alfalfa but is now used for other feeds as well. ADF and NDF values are used to calculate the relative feed value. The relative feed value is based on a value of 100 calculated from the 41% ADF and 53% NDF content of fully bloomed alfalfa hay. As the relative feed value falls below 100, the quality of the feed decreases; if it rises, the feed's quality increases. Accordingly, if NYD is below 75, it is considered to be of 5th quality, 75-86 of 4th quality, 87-102 of 3rd quality, 103-124 of 2nd quality, 125-150 of 1st quality, and above 150 is considered to be the best quality (Canbolat and Karaman, 2009). Therefore, low ADF and NDF values are desirable to achieve high RFV. In the study, the ADF and NDF ratios of plants sown on 15 May and harvested during the flowering stage in 2019 were lower than the plants with all other sowing times and harvesting stages. According to the two-year average results, the yield and quality characteristics of buckwheat planted for hay production are significantly affected by varying sowing and harvesting stages. Accordingly, to obtain high dry matter yield from buckwheat, sowing should be done during the early period (15 April) and the resulting plants should be harvested during the milky or dough stage. In terms of obtaining a high crude protein yield and RFV, sowing should be done during the early period (15 April) and harvesting should be done during the flowering stage. It has been determined that buckwheat that sown and harvested at the appropriate time provides very high yields and is of sufficient quality for hay production. For this reason, buckwheat is believed to be a potential alternative feed source to aid in closing the roughage shortage.

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ROW SEEDING CONFIGURATION REGULATES YIELD, QUALITY AND COMPETITION IN COMMON VETCH (*Vicia sativa* L.)-SUDANGRASS (*Sorghum sudanense* (Piper.) Stapf.) MIXTURE

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ABSTRACT

Intercropping used in modern agricultural systems is designed for growing more plant species in the same field for maximized growth parameters. This study was conducted to understand the effects of row seeding configuration (same, alternate and cross seeding row) and different Sudangrass ratios (25 and 50%) on yield, quality, and competitive ability of common vetch in second crop production. Among row seeding configuration, same row seeding maximized the aboveground biomass production but decreased belowground biomass, crude protein content and NDF (Neutral Detergent Fiber). Plant height, ratio, ADF (Acid Detergent Fiber) content, AGRNE (Aboveground Relative Neighbor Effect), and BGRNE (Belowground Relative Neighbor Effect) were not affected by row seeding configurations. In order to maximize the utilization from the row seeding configuration, the same row seeding can increase the aboveground biomass but mitigate belowground biomass and crude protein content. The row seeding configuration may represent a potential for yield, quality, and competitive ability when sown as a mixture using 25% Sudangrass ratio.

Keywords: Competition, intercropping, quality, second crop, yield

INTRODUCTION

Mixed cultivation has been arranged in modern agricultural systems for improving yield and soil properties, and for decreasing biotic and abiotic stress factors. It represents competition, complementarity, cooperation, and compensation among plant species during the cropping cycle (Justes et al., 2021). Yield, quality, and competitive ability of a mixture can exceed or regulate by choosing the best performing species (Acar et al., 2006). The positive effects of mixed species on the productivity of a stand are different depending on the plant species, plant properties, diversity, and climatic conditions (Ergon et al., 2016). The balance among facilitation, competition, and neutral effect can occur due to climatic, edaphic factors, plant density, physiology, life stage, invasion, and plant species (Manea and Leishman, 2011). Except for minor differences in the requirement of plant species, they compete for the same resources such as water, nutrient, light, or space during the growth period. The facilitative effect is more common under high abiotic stress conditions (Bertness and Callaway, 1994). Some species can facilitate by altering the soil microbial diversity and by increasing nitrogen assimilation due to nitrogen fixation and by ameliorating the local environment (Ehlers et al., 2014; Dang et al., 2020), but indirect facilitation is more common among plants. In addition to the facilitative effect, there is a negative impact among plant species depending on allelochemical contents and leachate. These compounds can inhibit the germination and growth performance of plant species (Cook et al., 2010; Ehlers et al., 2014).

In mixed cropping, row seeding is one of the most effective factors for regulating plant-plant interaction. How intercropping advantages or disadvantages in mixture alters with different row seeding?. Alternate row seeding is often suggested as a technique to improve the establishment and life forms in separate rows, which may reduces row competition, but increases the species diversity of a planting. Different row seeding regulates plant-plant interaction and increases the yield and the land equivalent ratio (Erkovan et al., 2008), but its advantage is unclear in the common vetch-Sudangrass mixture for second crop production. Same rows seeding configuration have the disadvantage of increasing competition especially space, but it has the advantage of preventing common vetch from lodging. Seeding different life forms in alternate and cross-seeding row may reduce competition, improve stand establishment, and increase yield and quality due to increasing land equivalent ratio and using resources such as light, nutrient, water etc. (Pokorny et al., 2020; Justes et al., 2021). Kilcher and Heinrichs (1958) suggested that seeding methods performed better or less than mixed cropping of alfalfa in terms of yield and competition. But Yolcu (2005) reported that same, alternate, and cross-seeding rows of alfalfa and smooth bromegrass mixture did not affect dry matter yield. However, there are no detailed studies on row seeding of Sudangrass-common vetch intercropping, especially on the second forage cropping.

In Turkey, especially Mediterranean Climate Zone, second crop production has an important role in improving forage production. Common vetch is the second most grown crop after alfalfa in Turkey, and has been successfully used in this zone. There is a disadvantage of common vetch cultivation as a cool season forage crop in the second crop growing period because of high temperature. This disadvantage of common vetch can be tolerated by growing in a mixture with warm-season plant species. Forage yield increases by choosing the best performing and sowing ratio of plant species (Ergon et al., 2016). The positive or negative effect of the sowing ratio on yield and yield components depends on the mutualistic interaction of species and environmental properties. For example, rapidly growing Sudangrass under high temperature and drought conditions creates a microclimate environment for other plant species due to temporal niche differentiation and increases their yield (Poorter and Navas, 2003; Justes et al., 2021), but may inhibit plant growth as it has allelopathic compounds (Cook et al., 2010). The positive effect of the sowing ratio on yield and yield components of mixtures occurs under various climatic conditions depending on plant species functional characteristics (Kavut and Geren, 2017; Ergon et al., 2016). Sudangrass is generally more competitive in mixtures with short plant species because of temporal niche differentiation, light conservation, water use efficiency, nutrient acquisition, etc. Hence, Sudangrass is sown later than other plants or using decreased seed rate for less competition. Decreased ratio in mixed cropping systems is known as an important strategy for plant-plant interactions (Koc et al., 2013).

Common vetch and oat, barley, wheat mixed cropping has been widely studied at different seeding ratios, harvest and sowing time to assess the performance yield, quality, and fertilizer use efficiency in the region (Dereli, 2015; Kavut et al., 2016; Ileri et al., 2020; Ileri et al., 2021). There are some problems, especially short growing season and wide temperature range conditions (Mazzafera et al., 2021). However, less attention has been given to sowing methods, row seeding, life form, plant height, canopy structure, and at the second crop production. The objectives of this study are to determine the effects of row seeding configurations in the common vetch-Sudangrass mixture for better yield, quality, and the less competitive effect between common vetch and Sudangrass, when the ratio of Sudangrass was reduced by 50 and 75% in the mixture.

MATERIALS AND METHODS

The field experiment was conducted during 2019 and 2020 after harvesting wheat in the second crop growing season at the experimental station of Eskisehir Osmangazi University, Faculty of Agriculture. The long-term average annual air temperature of experimental area was 12.8 ^{0}C, and nearly 180 days that were frost-free in a year. The monthly temperature of the experimental years in 2019 and 2020 were 12.3 and 13.0 ^{0}C, respectively (Table 1). The mean annual precipitation for the long term, 2019 and 2020 were 352.4 mm, 405.5 mm, and 299.2 mm, respectively (Table 1). The experimental soil characteristics were determined as described by Soil Survey Laboratory Staff (1992), and clay-loam in texture, slightly alkaline, lime, and organic matter contents were 14.6% and 1.62% (poor) respectively. Soil salinity level was quite low (0.07 %) and P_2O_5 and K_2O contents were 61.6 and 1688.0 kg ha^{-1} respectively.

Table 1. Climatic data for the study site, temperature (°C), monthly precipitation (mm) and humidity (%) during 2019 and 2020 and the long-term average (LTA; 1929–2020).

Months	Temperature (^{0}C)			Precipitation (mm)			Humidity (%)		
	2019	**2020**	**LTA**	**2019**	**2020**	**LTA**	**2019**	**2020**	**LTA**
January	4.3	0.3	0.3	60.2	52.7	38.7	91.0	78.7	98.2
February	3.4	4.1	4.7	50.1	43.3	32.5	79.6	70.8	92.6
March	6.3	8.2	9.3	13.4	20.0	33.4	64.5	63.5	81.6
April	9.5	10.9	13.1	26.7	13.0	35.0	69.3	57.2	67.8
May	16.5	16.3	16.5	42.2	38.9	44.8	65.1	58.0	86.1
June	20.9	19.5	20.4	45.7	74.3	30.6	67.9	63.5	83.3
July	**21.3**	**23.2**	**23.3**	**33.5**	**1.2**	**14.0**	**62.3**	**58.0**	**75.8**
August	**22.3**	**23.4**	**22.9**	**2.4**	**1.0**	**7.8**	**61.0**	**52.1**	**74.1**
September	**18.1**	**21.5**	**20.0**	**5.0**	**6.0**	**14.4**	**62.1**	**59.9**	**68.1**
October	**14.2**	**16.1**	**12.9**	**18.3**	**37.6**	**27.0**	**70.1**	**73.8**	**79.6**
November	7.9	6.3	7.5	33.9	1.4	29.2	76.2	72.7	80.3
December	2.9	5.7	3.6	74.1	9.8	45.1	89.9	77.2	93.6
Tot./Ave.	***12.3***	***13.0***	***12.8***	***405.5***	***299.2***	***352.4***	***71.6***	***65.5***	***81.8***

Bold lines: Growing season for the experiment

Common vetch (cv. Orakefe) and Sudangrass (cv. Gozde-80) were used as plant materials. The field trial was arranged in the Randomized Complete Block Design (RCBD) with 3 replications. Common vetch was sown at the 120 kg ha^{-1} (Acikgoz, 2001) and was mixed with Sudangrass by reducing the seeding rates as 50% (5 kg ha^{-1}) and 75% (2.5 kg ha^{-1}) considering the suggestions as 10 kg ha^{-1} for Sudangrass.

Plants were sown on 04 and 21 June in 2019 and 2020 years respectively depending on the wheat harvest in the region. All plots were 5 m of 1.5 m at different row seeding and Sudangrass sowing ratios. Common vetch was seeded as sole crop or as a mixture with Sudangrass. While common vetch and Sudangrass were seeded in 5 same rows 0.3 m spaced rows, they were separately seeded in alternate 0.15 m spaced rows between common vetch and Sudangrass, and in cross-seeding row 0.3 m spaced. Nitrogen (30 kg ha^{-1}) and phosphorus (70 kg ha^{-1} P_2O_5) were applied as Di-ammonium phosphate (DAP) and irrigation was applied every week using sprinkler and considering the requirement of the plants. Weed control was done by hand hoeing within the plots.

The harvest stage was determined considering in the lowest pods full set stage of common vetch as suggested by Kusvuran et al. (2014) and all plants were harvested together with the common vetch. The harvest date was 23 September and 14 October in 2019 and 2020 respectively. All plants were harvested in plots by cutting after taking out the 0.5 m from beginning and end of each row. Samples dried at 70 ºC until reached to constant weight at the oven to determine the aboveground biomass production. Root biomass was sampled in the same area of the plots after harvest. Excavated roots were firstly washed to separate from soil and dried at 70°C until reached to constant weight to determine the belowground biomass. Aboveground biomass samples were grounded to pass through a 2 mm sieve to determine crude protein (CP) content using the Kjeldahl method. Neutral detergent fiber (NDF) and acid detergent fiber (ADF) contents were determined due to the detergent method suggested by Van Soest et al. (1991). Relative neighbor effects of above-and belowground (ARNE and BRNE) among the mixed-species were determined using above-and belowground biomass production as in the formula that given by Oksanen et al. (2006).

$$RNE = (X_m – X_c) / Max (X_m \text{ or } X_c)$$

RNE; Relative neighbor effect, X_m; production in mixed sowing, X_c; production in sole

The analysis of variance was performed considering the General Linear Model (GLM). All data were tested for homogeneity and the variances were analyzed in terms of the completely randomized block design using SAS statistical software (SAS Institute, 2011). The means of treatments were compared by using the Bonferroni/Dunnett test.

RESULTS AND DISCUSSION

There was a significant difference in plant height among main factors and Sudangrass density x row seeding configuration interaction (Table 2). Common vetch sole sown height was higher at 2019 than in 2020 year (Table 2). Increasing the Sudangrass ratio from 25% to 50% in the mixture raised the plant height of common vetch from 48.38 cm to 52.38 cm ($p<0.0001$) (Table 2) (Figure 1). The plant height of sole growing common vetch was significantly higher than different row seeding configuration mixtures with Sudangrass ($p<0.001$), but there were no significant differences among the row seeding configurations (Table 2).

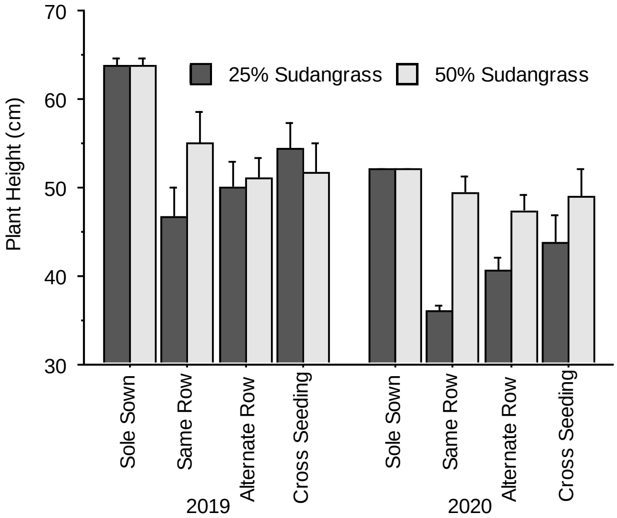

Figure 1. Plant height (± SE) in Sudangrass density and row seeding configuration during 2019 and 2020 sampling periods.

Overall common vetch ratio was 87.28% and it did not significantly vary between the years and among the Sudangrass densities ($p<ns$). The first and second-order interactions were not significant (Table 2). Common vetch ratio was higher in sole sowing than in other row seeding configurations mixture with different Sudangrass ratio ($p<0.0001$) (Table 2).

Aboveground biomass of common vetch and Sudangrass mixture was significantly affected by main factor year ($p<0.0001$), row seeding configuration ($p<0.0001$), and first and second-order interactions ($p<0.049$) except for main factor Sudangrass ratios (Table 2). Aboveground biomass was higher in the second year of the study (3956.4 kg ha^{-1}) and the highest value was determined in the same row seeding configuration (3902.8 kg ha^{-1}) while it was the lowest in the sole sowing of common vetch (2860.4 kg ha^{-1}) (Table 2) (Figure 2).

Except for the main factor Sudangrass ratio and second-order interaction ($p<ns$), years ($p<0.0001$), row seeding configuration ($p<0.0001$) and first order interaction ($p<0.049$) led to significant changes in belowground biomass production (Table 2) (Figure 3). Belowground biomass was found to be significantly higher in the second year (799.4 kg ha^{-1}) compared to the in the first year (658.3 kg ha^{-1}). On the other hand, cross-seeding and alternate row seeding configuration led to significantly higher belowground biomass than the other row seeding configurations (Table 2).

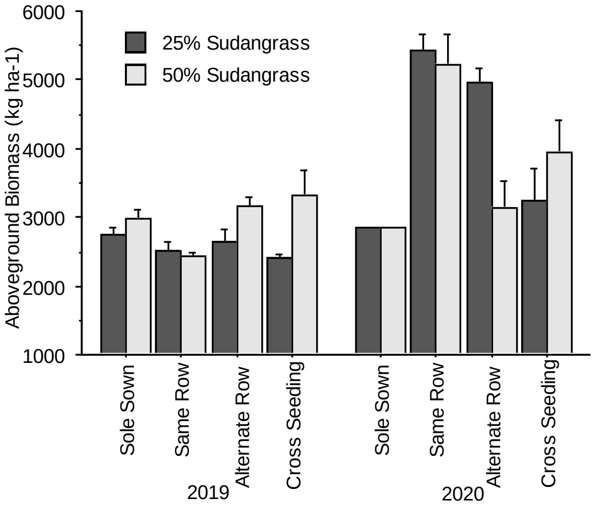

Figure 2. Aboveground biomass (± SE) in Sudangrass density and row seeding configuration during 2019 and 2020 sampling periods.

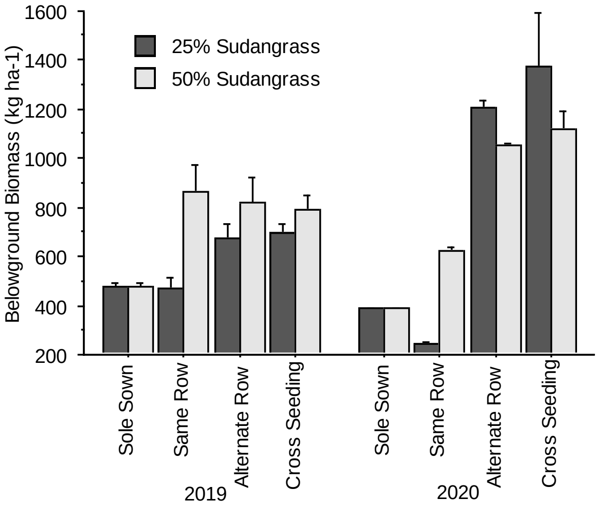

Figure 3. Belowground biomass (± SE) in Sudangrass density and row seeding configuration during 2019 and 2020 sampling periods.

Table 2. The results of analysis of variance (ANOVA) with main effects, first and second order interactions of Sudangrass ratio and row seeding configuration on plant height, common vetch ratio, above and belowground biomass, crude protein content, neutral detergent fibre (NDF), acid detergent fibre (ADF), above (AGRNE) and belowground (BGRNE) relative neighbour effect.

	Plant Height	Ratio	Aboveground Biomass (kg ha^{-1})	Belowground Biomass (kg ha^{-1})	Crude Protein (%)	NDF	ADF	AGRNE	BGRNE
Year (Y)									
2019	54.50 A	89.16	2781.0 B	658.3 B	14.04 B	41.89 A	33.18	-0.006 A	-0.185 A
2020	46.25 B	85.40	3956.4 A	799.4 A	16.93 A	34.97 B	32.91	-0.716 B	-0.876 B
Sudangrass Ratio (SR)									
25% Sudangrass	48.38 B	86.35	3353.3	691.2	15.47	38.94 a	33.25	-0.349	-0.537
50% Sudangrass	52.38 A	88.21	3384.1	766.6	15.49	37.92 b	32.83	-0.373	-0.524
Row Configuration (R)									
Sole	57.83 A	100.00 A	2860.4 C	434.5 C	14.66 B	38.63 A	33.12	-	-
Same Row	46.75 B	81.82 B	3902.8 A	549.1 B	14.45 B	39.06 A	33.16	-0.399	-0.416
Alternate Row	47.25 B	83.54 B	3478.8 B	938.3 A	16.10 A	39.15 A	32.98	-0.292	-0.582
Cross-seeding	49.67 B	83.76 B	3232.9 B	993.6 A	16.73 A	36.88 B	32.91	-0.391	-0.594
Mean	**48.40**	**87.28**	**3368.7**	**728.9**	**15.49**	**38.43**	**33.04**	**- 0.361**	**- 0.531**
Y	***	ns	***	***	***	***	ns	***	***
SR	**	ns	ns	ns	ns	*	ns	ns	ns
R	***	***	***	***	***	***	ns	ns	ns
Y x SR	ns	ns	**	***	ns	ns	ns	ns	ns
Y x R	ns	ns	***	*	***	**	ns	ns	ns
SR x R	*	ns	**	***	ns	**	ns	ns	ns
Y x SR x R	ns	ns	*	ns	*	***	ns	*	ns

Values followed by small and capital in a column shows significantly differences at P< 0.05 and P< 0.01 levels. ns: No statistical difference at P<0.05 and P<0.01. *Statistical difference at P< 0.05. **Statistical difference at P< 0.01. *** Statistical difference at P< 0.001.

Crude protein content was significantly affected by year (p<0.0001) and row seeding configuration (p<0.0001) (Table 2). Alternate and cross-seeding row seeding configuration positively affected crude protein content compared to sole and same row seeding configuration (Table 2). Crude protein content showed a variation depending on the year, Sudangrass ratio, and row seeding configuration. Hence, the second-order interaction was significant in the experiment (p<0.049) (Figure 4).

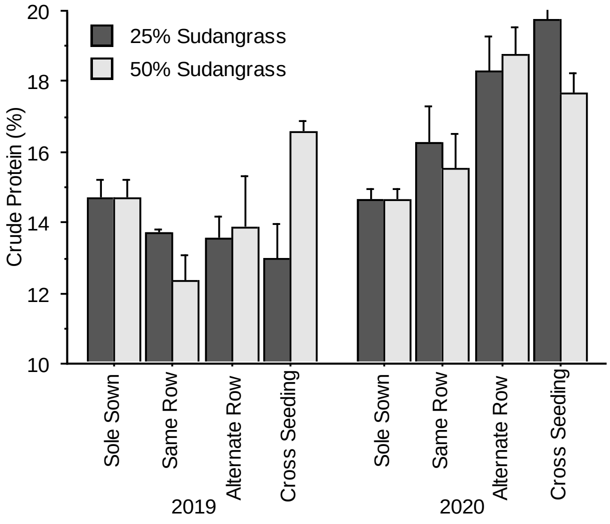

Figure 4. Crude protein content (± SE) in Sudangrass density and row seeding configuration during 2019 and 2020 sampling periods.

NDF content was significantly affected by main factors (p<0.049), first and second-order interactions (p<0.001) except for year x Sudangrass ratio interaction (Table 2). NDF content was higher in the first year (41.89%), and in the mixture with a 25% Sudangrass ratio (38.94%) (Table 2). Cross-seeding exhibited the significantly lowest NDF among row configurations (Table 2). There were significant variations in the NDF content. As a result of this, first and second-order interactions were significantly affected (Figure 5).

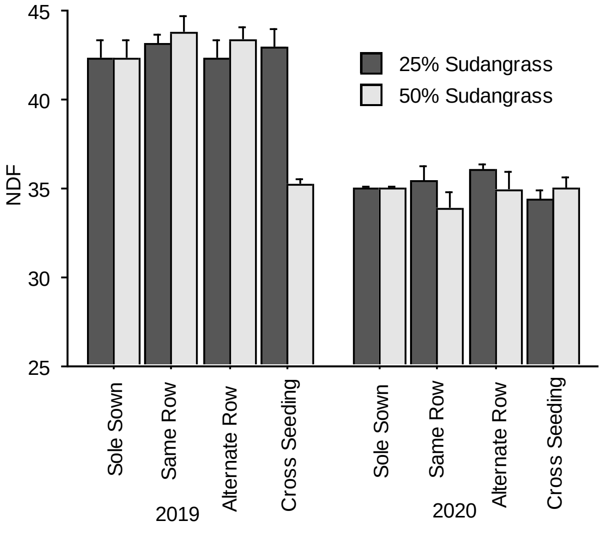

Figure 5. NDF content (± SE) in Sudangrass density and row seeding configuration during 2019 and 2020 sampling periods.

Overall ADF content was 33.04% and it did not significantly vary among the main factors, first and second-order interactions (Table 2).

The growth of the common vetch was significantly suppressed more than the second experiment year in competition with the Sudangrass diversity (p<0.0001) (Figure 6). There were no significant effects on the common vetch AGRNE first-order interactions; in contrast, there was significant effect of second-order interaction (Table 2). This was shown in the AGRNE, where Sudangrass ratio and row seeding configuration had a less positive effect in the first year, but more negative effect on the common vetch in the second year (Figure 6).

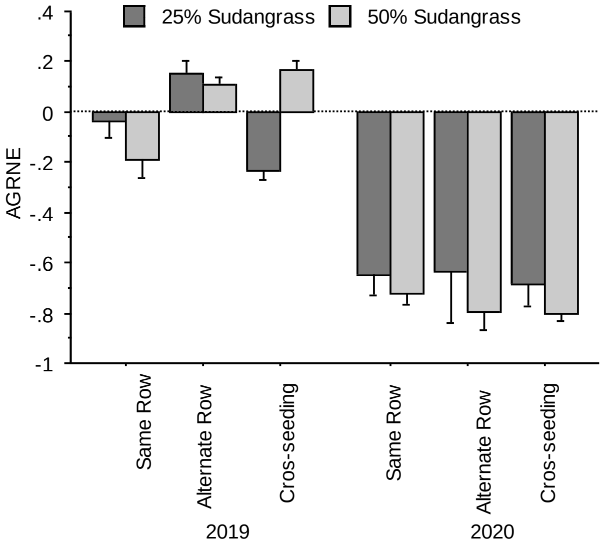

Figure 6. AGRNE (± SE) in Sudangrass density and row seeding configuration during 2019 and 2020 sampling periods.

Except for the year (p<0.0001), the main factors and the first and second-order interactions were not significant on BGRNE (Table 2). We found that common vetch BGRNE significantly suppressed more in 2020 than in 2019 (Table 2).

Crop production may be affected significantly by erratic weather conditions due to biotic and abiotic stress (Justes et al., 2021). The variability in weather conditions in the experiment years, especially temperature, played a deterministic role on plant height. It was higher in 2019 possibly due to cool-season characteristics of common vetch that the responses of climate conditions on growth properties have fully yet to be elucidated because of common phenomena of it (Lithourgidis et al., 2006; Baxevanos et al., 2021; Ileri et al 2021; Li et al., 2021). Intercropping resulted in a lower plant height in common vetch in comparison to the in sole grown, mainly due to the competition with Sudangrass. Similar results have been reported by Lavergne et al. (2021), Li et al. (2021). Common vetch was dominant within all mixtures and row seeding configuration and might have acted as different variations for each mixture.

Used plant species in the experiment have different sensitivities to climatic conditions. Common vetch (cool-season plant) grows better in cool conditions, while Sudangrass (warm-season plant) are more competitive in warmer conditions (Acikgoz, 2001). This shows that plant species had an advantage on interspecific competition when increasing temperature effects at second crops growing seasons. As a matter of fact, the plant height of common vetch was longer and both above and belowground less competed in 2019 that was cooler, but the above and belowground biomass has been lower because of the ratio and the growth performance of Sudangrass. In the second year of the experiment, the above and belowground biomass was higher because of increasing temperature and ratio of Sudangrass growth performance. Similar results have been reported by researchers (Ileri et al., 2021; Lavergne et al., 2021; Li et al., 2021). The sole stand of common vetch had lesser aboveground biomass about 1000 kg ha^{-1} compared to row seeding configurations, and the highest aboveground biomass was observed in the same row seeding. Alternate and cross-seeding rows did not increase the common vetch biomass production compared to same row seeding, but it is thought to reduce within row competition to improve stand establishment. Suitable mixtures due to niche complementarity of species increase the facilitative effect and aboveground biomass in the stand (Ergon et al., 2016). The helpful effects of symbiotic systems may facilitate increasing soil nutrient content, microbes, and radiation use efficiency in the mixture. Greater photosynthetic capacity, competition ratio, and radiation use efficiency increase aboveground biomass production in the row seeding configuration. Aboveground biomass production indicated a high magnitude variation in first and second-order interactions. This implies that the performance of plant species and row seeding configuration were less consistent in the mixture than two growing seasons (Baxevanos et al., 2021). Currently, the dominant plant of common vetch in the mixture was affected by the growth of Sudangrass, since the variation on aboveground biomass ranged within the year, Sudangrass ratio, and the row seeding configuration. The interaction taking place in mixtures is complex and occurs different during the growing seasons. The adoption of the mixture is challenging due to the short growing season and wide temperature range throughout the year. Consequently, these different responses of the common vetch to year, Sudangrass density, and row seeding configuration caused first and second-order interaction for aboveground biomass.

Although the proportion of Sudangrass in the composition was low in 2020, above and belowground biomass were more. The increase in belowground biomass in 2020 can be associated with substantial differences in temperature (Table 1). Belowground biomass mostly consisted of common vetch, whereas the ratio of Sudangrass belowground biomass was less. In 2019, lower belowground biomass can explain the fact that the growth of common vetch, which is a cool climate plant, was rapid and low productivity in the warmer period. Sudangrass presented a better growth performance in higher temperatures due to warm-season characteristics of the crop. As a result of this, competitive ability and productivity may change with the difference in temperature depending on the characteristics of the plant species (Dirks et al., 2021; Lavergne et al., 2021). Belowground biomass in the mixtures was more productive due to niche complementary and large contributions of plant roots. The sole grown common vetch produced the lowest belowground biomass compared to row seeding configurations, and also alternate and cross-seeding row configurations have greater belowground biomass than the same row seeding configuration. The greater belowground biomass could be explained by the contribution of mixtures. Because common vetch and Sudangrass roots properties and resource complementarity are different, hence complementarity increases when roots of common vetch or Sudangrass are away from zones of neighbors (Dirks et al., 2021). Common vetch and Sudangrass in alternate and cross-seeding row configurations may reduce competitive effects within the row due to decreasing competition for space, and their belowground performed increases (Koc et al., 2013), hence belowground biomass increases. The performance of common vetch belowground biomass production was less consistent over applications (years, density, and row configuration) than growing seasons. The differential responses of applications in the mixtures have not fully to be elucidated. Years and density-based variation of the interaction, which can be by the relative performance of different row seeding configurations. Likewise, the data of this study indicated the magnitude of interactions variation in comparison to the interactions for belowground biomass.

The crude protein content was affected significantly by years. The higher crude protein content was recorded the first year of the experiment. Generally, intercropping results in a variety of agronomic benefits along with improved crude protein content that in forage is the major criterion for forage quality appraisal. Legumes tend to improve the crude protein content of the mixture due to their higher protein content, also intercropping helps to a significant increase of crude protein content (Iqbal et al., 2019). The differences in climatic condition between the years affect growing condition, hence row seeding configuration effect on crude protein content changed depending on these factors (Table 1). The growing performance of the plant grown as sole or mixture had showed different performances depending on the differences between years (Carr et al., 2004; Pflueger et al., 2020). The crude protein content was significantly affected by row seeding configuration in the experiment. Row seeding configuration is often suggested as a technique to improve the establishment, yield, and quality (Tilley et al., 2008). Separate rows may decrease competition and may improve the establishment and the performance of plant species. Thus, the alternate and cross-seeding row had the highest crude protein content, whereas sole and same row showed the lowest crude protein content. The protein content of forage appears to be highly variable in genetically identical common vetch plants grown under the same conditions such as year, Sudangrass ratio, and row seeding configuration. Although there are some differences, mixtures showed partly consistent CP content among years, Sudangrass ratio, and row seeding configuration. This situation was the main reason for three-way interaction.

Year, Sudangrass ratio and row seeding configuration altered in the NDF content of the forage. The NDF contents were higher in the first year in the experiment. These might be related to increases in cellulosic component synthesis in the plant, because plant maturity plays a deterministic role in these contents (Erkovan et al., 2014; Kavut and Geren, 2017). Cross-seeding row conducted a significant decrease in NDF content compared to sole, same and alternate row, whereas the sole, same, and alternate row configuration did not show an interactive effect. Row seeding configuration is thought to reduce competition and heat stress to improve, warm weather causes generally an increase in cellulose content (Osman et al., 2010). Our findings implied that efficiency of year, Sudangrass ratio, and row seeding configuration are strongly related to climatic and competition condition that affects the availability of NDF content because the same treatment has a positive effect on NDF content due to increase cellulosic content.

In the summer, the high temperature could lead to greater competition between common vetch and Sudangrass due to several possible factors influencing the stability of plant species such as complementarity, resilience, and redundancy of plant species roles (Grant et al., 2014). Therefore, this could affect the above and belowground competition ability. Common vetch RNE performance decreased with increasing performance of Sudangrass. Because RNE can easily shift from competition to facilitation and vice versa that competitive intensity between common vetch and Sudangrass may be a key mechanism contributing to the stability of plant species under changing climate conditions. These results show the effect of climate and alterations in competitive ability including facilitation and competition among applications. Thus, competitive effects were occurred in the temperature due to increasing Sudangrass performance. As a result of this, AGRNE and BGRNE's competitive ability has increased in 2020 compared to 2019. These results showed the change of temperature, diversity, and induced alterations in competitive intensity including facilitation and competition in common vetch depending on the complexity of above and belowground relations. Consequently, these different responses of the common vetch to year, Sudangrass density, and row seeding configuration caused three-way interactions for AGRNE.

CONCLUSION

Our results showed a strong effect of the year on plant height, above and belowground biomass, crude protein, NDF, AGRNE, and BGRNE of common vetch as sole crop and mixture. The study shows that the seeding row seeding configuration of common vetch and Sudangrass mixture regulated some properties, but in the same row seeding aboveground biomass and NDF content was higher than other row seeding configurations. The same row seeding of common vetch with Sudangrass had an aboveground biomass production advantage for exploiting available resources, but alternate and cross-seeding row had an advantage for belowground biomass and crude protein content. This research indicates that row seeding configuration could have a significant effect on yield and quality when sown as a mixture using 25% Sudangrass ratio in the second crop conditions of semi-arid regions. These configurations of row seeding could provide economic and environmental benefits for sustainable production in the second crop season and further research is required to understand the net effect of root and canopy structure in various pedoclimatic conditions.

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ANALYSING GROWTH PATTERNS OF SELECTED TURKISH OAT CULTIVARS USING SIGMOIDAL MODELS

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ABSTRACT

Increasing popularity of oat was accompanied with the introductions of many new cultivars for the last few decades. The aim of this study was to characterize the growth and developments of Kahraman, Kucukyayla, Yeniceri, Sebat, Otag and Dirilis oat cultivars using sigmoidal growth models. Growth data comprised of weekly observations of dry weights and growth stages with three samplings for two consecutive years. Results indicated that the growing season were the determining factor for the dry matter accumulation until the stem elongation stage since genotype differences became apparent only in the later stages. Sigmoidal growth models were successfully fitted to the growth data, and allowed for further evaluations. Goodness of fit statistics implied that Logistic, Logistic Power and Ratkowsky models were the best fitting growth models to explain dry matter accumulations of oat cultivars. Analysis also showed that Otag, Yeniceri and Sebat cultivars reached the highest dry matter accumulations. Point of inflections on the Logistic models indicated that Kucukyayla and Kahraman were the earliest cultivars in the Marmara region. Comparison of cultivars by using the growth models proved to be informative in terms of understanding the genotypic variation.

Keywords: Dry matter accumulation, earliness, growth models, Marmara, oat, Turkey

INTRODUCTION

Oat (*Avena sativa* L.) is prominent for feed and grain for the Mediterranean environment. Oat is grown annually and can be sown in both autumn and spring, former being the usual for the temperate regions. In addition to its traditional usage as a fodder crop, oat grain also became an area of interest in the coming years. This is due to relatively recent discovery of its dietary benefits. Oat grain is reported to have a unique quality, often regarding to a high oil, micronutrient, souble fibre and beta glucan contents (Welch, 2012; Loskutov et al., 2021). Quality of oat grain, especially the high beta glucan contents are linked to reduce blood cholesterol levels and improve hemostatic factors, further improving the cardiovascular health (Tosh and Bordenave, 2020). Therefore, the role of oat grain as food are no longer restricted to oatmeals. Oat grain is utilized in the food industry in a variety of products including whole grain, flour, bakery products, food supplements and even oat milk as a coffee additive (Onning et al., 1998; Rasane et al., 2015).

Oat has always been a prominent cereal in Turkish agriculture. Oat breeding effords in Turkey for the last 20 years also seems to prioritize grain related traits over vegetative growth (Hısır et al. 2012; Hocaoglu and Akcura, 2020). In the literature of agronomic researches conducted in Turkey, Turkish oat cultivars are mainly compared by their grain yields and yield components when underlying biological mechanism remains unvisited. Comparing genotypes by the variation of their growth habits would provide insight about how genotypes differ from eachother (Karadavut, 2009). This comparison could also be useful from the standpoint of understanding the genotype environment interaction. Collection and statistical evaluation of a reliable growth data would not only reveal which genotypes are preferable for a given environment, but also potentially provide an insight about the underlying reasons.

A basic and effective way to evaluate plant growth is by implementing growth models. Using sigmoidal growth models to explain the biological growth has an impressively long history. Growth curves such as Logistic and Gompertz were known to be used in 19th century (Bollen and Curran, 2006) which is remarkable since their original formulas and variations are still in use today. Sigmoidal models produce curves resembling a streched “S” that corresponds well with the process of biological growth in general, whether it is the growth of a population or an individual organism. Therefore, these curves can be configured for a given data – a process called “the curve fitting” – to allow us to evaluate our data statistically. Today, many available softwares can perform this by fitting several models on the given data and ranking the most suitable models by their coefficients of determination (R^2), lower error statistics and other indicators such as Akaike's information criterion (AICC) or Bayesian information criteria (BIC). These parameters can vary according to the analysis and usually referred to as to "Goodness of fit" statistics. Curve fitting also yields shape parameters that defines the fitted curve on the given data. Some shape parametes can have biological meanings – allowing us to evaluate genotypes or environments by comparing the differences in between their curves. Although growth analysis has and will have a much wider use in the future, these approaches are seen as the main reason behind the growing popularity of the use of growth models in agronomy.

The objective of the study was to compare the dry matter growth of several oat cultivars under Marmara region condition by using the sigmoidal growth models to better understand the variation among their growth habits.

MATERIALS AND METHODS

Field Trials

Field trials were conducted in the Unit of Agricultural Production and Research of Canakkale Onsekiz Mart University Faculty of Agriculture in Canakkale (Turkey) for two consecutive growth seasons (2019-2020 and 2020-2021). Oat cultivars Kahraman, Dirilis, Kucukyayla, Otag, Sebat and Yeniceri were used as plant material, all of which were generally considered as suitable for the Marmara region. Cultivars Kahraman, Kucukyayla, Otag and Yeniceri were registered as early cultivars when Dirilis and Sebat were classified as mid-early. Field trials were sown in 6 November 2019 and 13 November 2020 in the first and the second year, respectively. Field trial was arranged in Randomized Complete Block Design (RCBD) in split-plots with three replications where genotype and sampling times were arranged as the main and sub plots, respectively. Agronomic applications of both trials were consistent including plot sizes, sowing densities (550 plants m^{-2}), fertilization and weed management. Each plot included six plant rows arranged with 0.2 m space apart, covering a total of 6 m^2 of the area. Phosphorus were applied before sowing as 6 kg da^{-1} P_2O_5 in the diammonium phosphate (DAP) form when nitrogen from DAP were complemented with an additional ammonium sulfate application in the beginning of stem elongation stage to a total of 8 kg da^{-1} N (which coincided with the 14th week of samplings in both years). Chlorsulfuron were used to control broad-leaved weeds while remaining weeds were controlled by hand.

Data Collection and Statistical Analysis

In order to identify the differences among varying growth patterns of oat cultivars, dry weights were monitored in a weekly base. Our aim was to assess the dry matter accumulations of oat cultivars with accuracy which required precision in the field measurements. In order to minimize the spatial variation within the plots, 300 plants were selected for sampling and marked from each plot after the emergence of the first leaf. This plant markings proved useful to guide the future plant samplings. In each sampling, growth stages of oats were assessed (Zadoks, 1974) for each plot, then 10 plants were randomly selected for plant height and number of tillers measurements. These measurements were used as the preliminary evaluation criteria reflecting the current sitation of the plot, after which the outliers were excluded from the evaluation. Finally, above-ground biomasses of randomly selected 3 plants were collected from the remaining plants. Fresh samples were dried in the drying oven for at least 48 hours in 105°C for dry weight measurements. Dry weight averages of each cultivar were used as the growth data which were recorded from the week when germinations were completed (Zadoks Scale 10) until the harvest maturity (Zadoks Scale 90). Total number of samplings varied between 26-28 weeks for the first year and 24-26 weeks in the second year.

Curve fitting on the field data identifies the growth patterns and yields several curve parameters that are biologically meaningful for us to use for comparison (Diel et al., 2020). In this study, growth data were fitted to the most commonly used sigmoidal models and results were evaluated with Curve Expert Professional v. 2.7.3 software (Hyams, 2010). Model efficiency were compared using standard error, R Square and corrected Akaike's information criterion (AICC). Random distribution of the residuals were tested by Wald-Wolfowitz runs test, results of which indicated that run patterns of the residuals were unlikely for all curves (<%5) meaning that the residuals were randomly distributed (Hyams, 2020). Since there was an excessive amount of output data, only three best fitting models were reported for each graph. Curve parameters *a*, *b* and *c* were derived from Curve Expert Professional v. 2.7.3 when point of inflection (PI) and weight at the point of inflection (WIP) were calculated manually according to the Wen et al. (2019).

RESULTS AND DISCUSSION

Growth curves of six oat cultivars for two growing seasons were presented in Figures 1- 12. Each growth data fitted to Gompertz, 3 Parameter Logistic, Logistic Power, MMF, Ratkowsky, 3 Parameter Richards and 3 Parameter Weibull models separately. In the Figures 1-12, weekly dry weight measurements of oat cultivars were presented with the standard error (Std Err), R Square and corrected Akaike's information criterion (AICC) of three best fitting models. Logistic, Gompertz, Logistic Power and Ratkowsky models displayed the overall best results with the highest R Square and lowest AICC values. First 10-15 weeks showed no significant increase of dry weight for any oat cultivar because of the winter dormancy since trials were sown in autumn. Stem elongation stage began in 17th week for all cultivars in the first year and 17-20th weeks in the second year, indicating that the date of stem elongation could be driven more by the environmental factors rather than genotypic variation. Rising temperatures and precipitation of the early weeks of the spring in the Mediterranean climate seemed to have triggered a rapid growth, which usually began during the last weeks of the tillering stage (Figures 1-12). Rapid increase of dry weight gain for all cultivars began in the 15th week in the first year and 15 – 18th weeks in the second year. Differences among the growth patterns of oat cultivars became evident in this period.

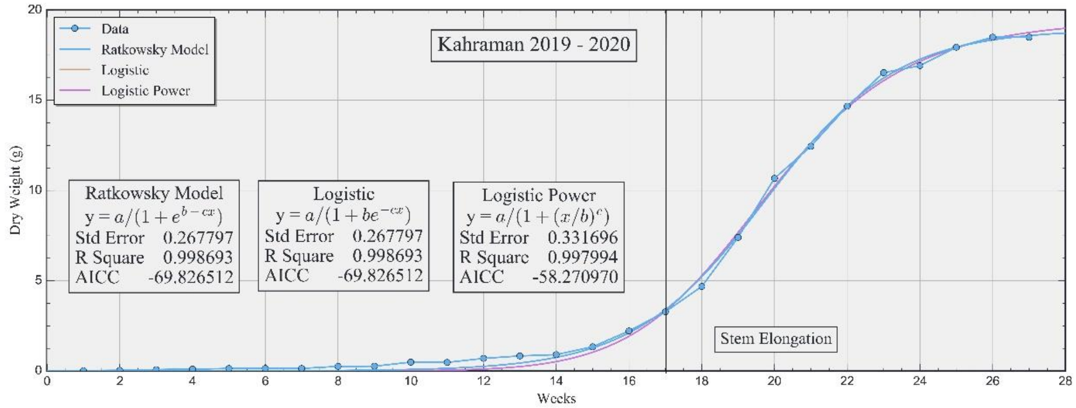

Figure 1. Identifying the dry weight increase of Kahraman oat cultivar with the sigmoidal curves (first year).

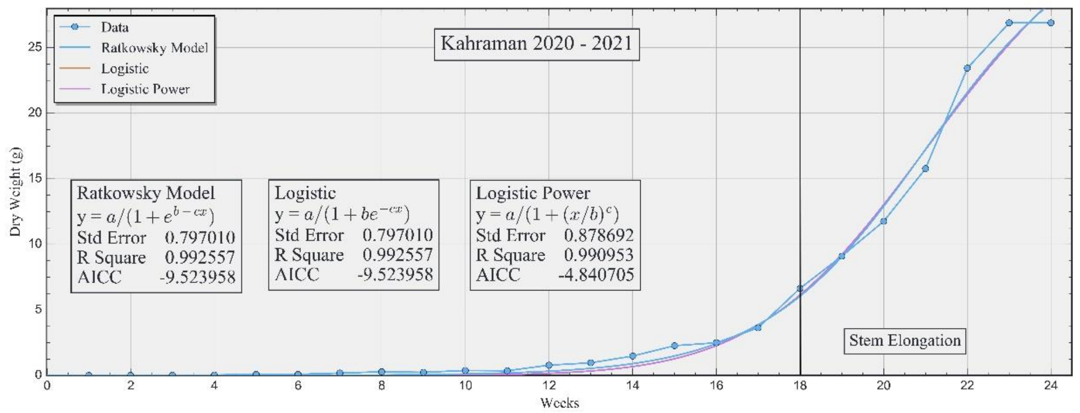

Figure 2. Identifying the dry weight increase of Kahraman oat cultivar with the sigmoidal curves (second year).

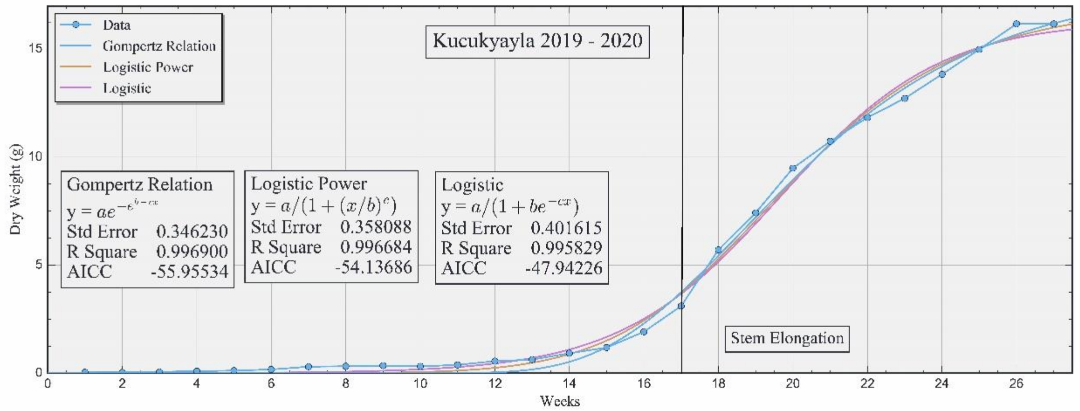

Figure 3. Identifying the dry weight increase of Kucukyayla oat cultivar with the sigmoidal curves (first year).

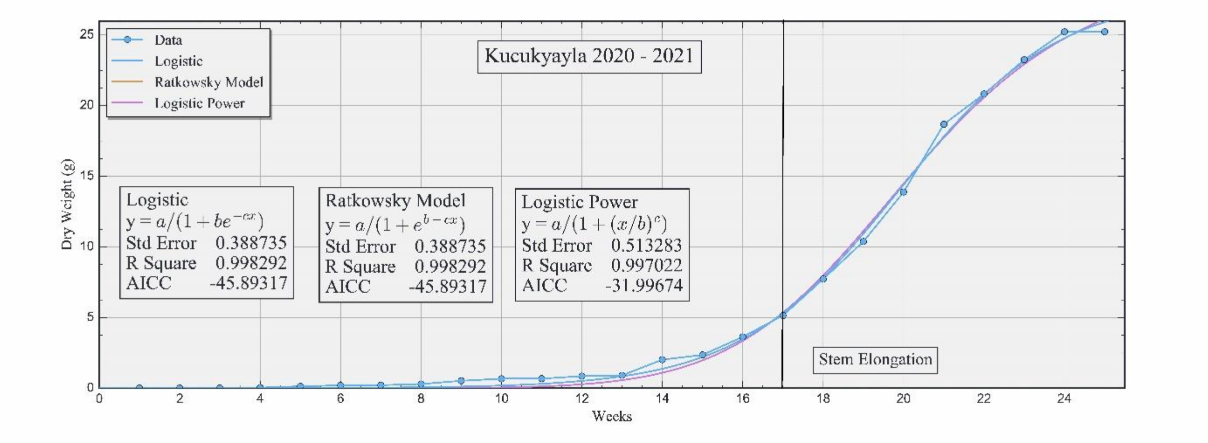

Figure 4. Identifying the dry weight increase of Kucukyayla oat cultivar with the sigmoidal curves (second year).

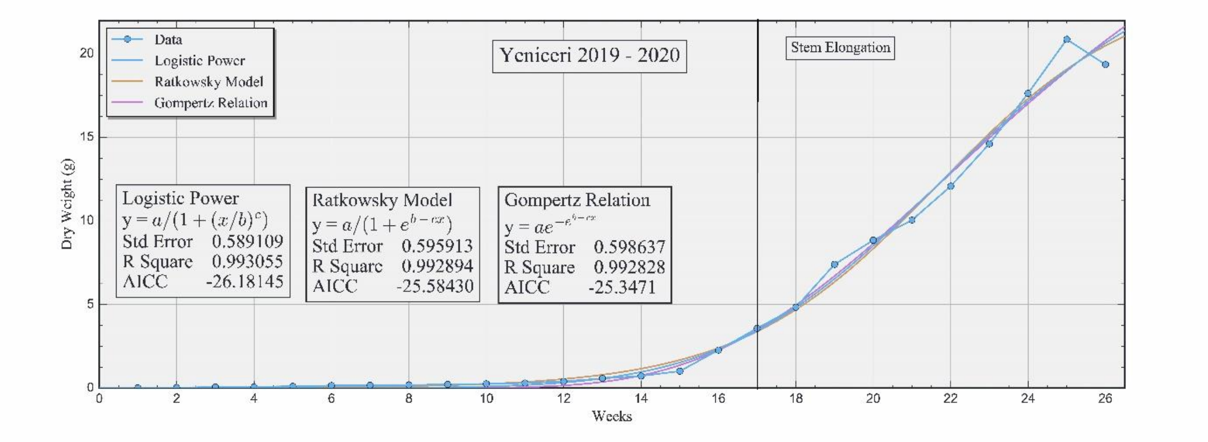

Figure 5. Identifying the dry weight increase of Yeniceri oat cultivar with the sigmoidal curves (first year).

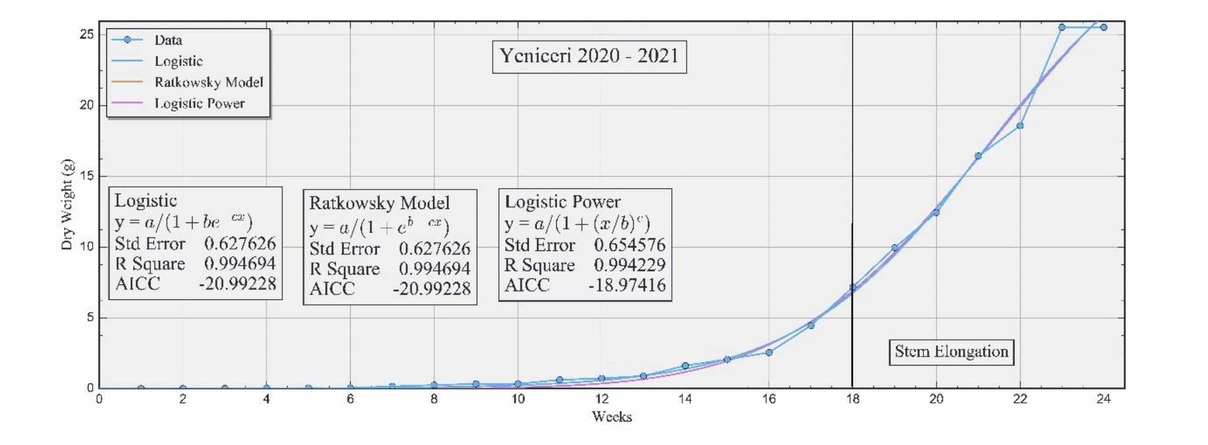

Figure 6. Identifying the dry weight increase of Yeniceri oat cultivar with the sigmoidal curves (second year).

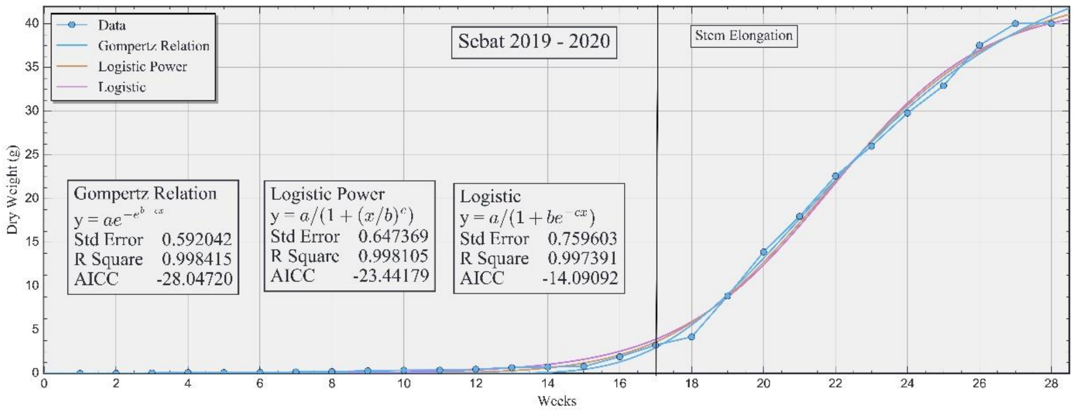

Figure 7. Identifying the dry weight increase of Sebat oat cultivar with the sigmoidal curves (first year).

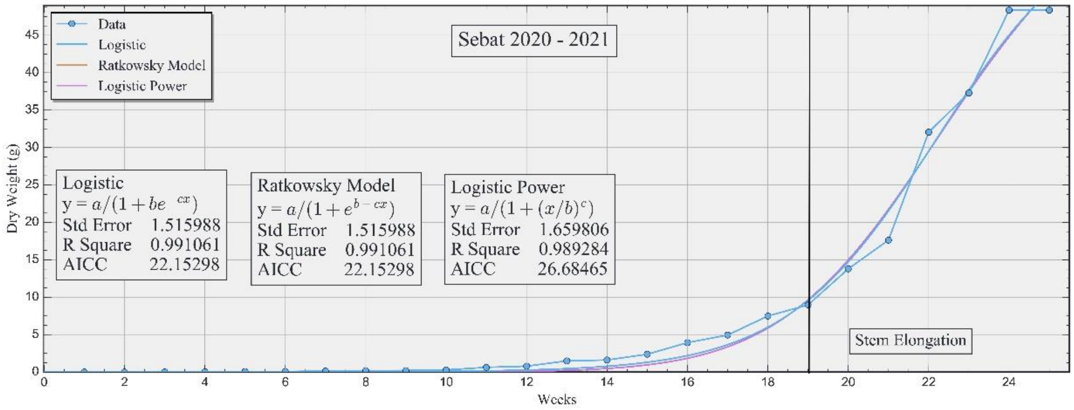

Figure 8. Identifying the dry weight increase of Sebat oat cultivar with the sigmoidal curves (second year).

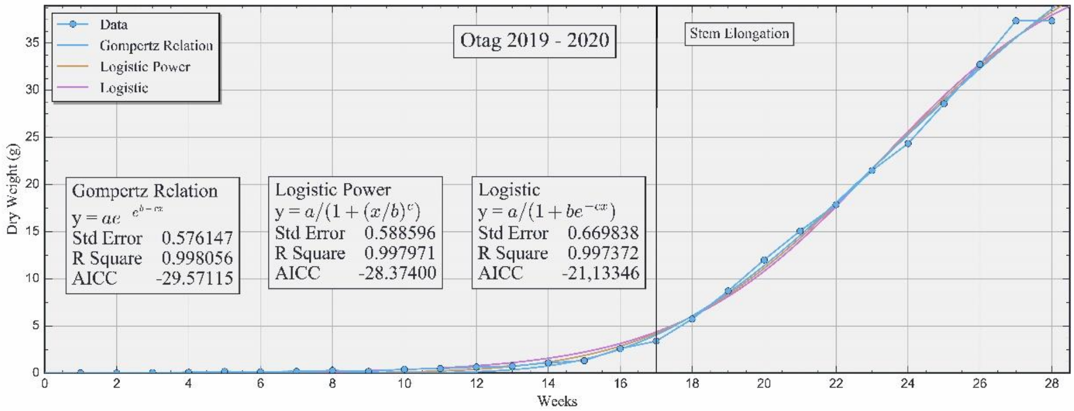

Figure 9. Identifying the dry weight increase of Otag oat cultivar with the sigmoidal curves (first year).

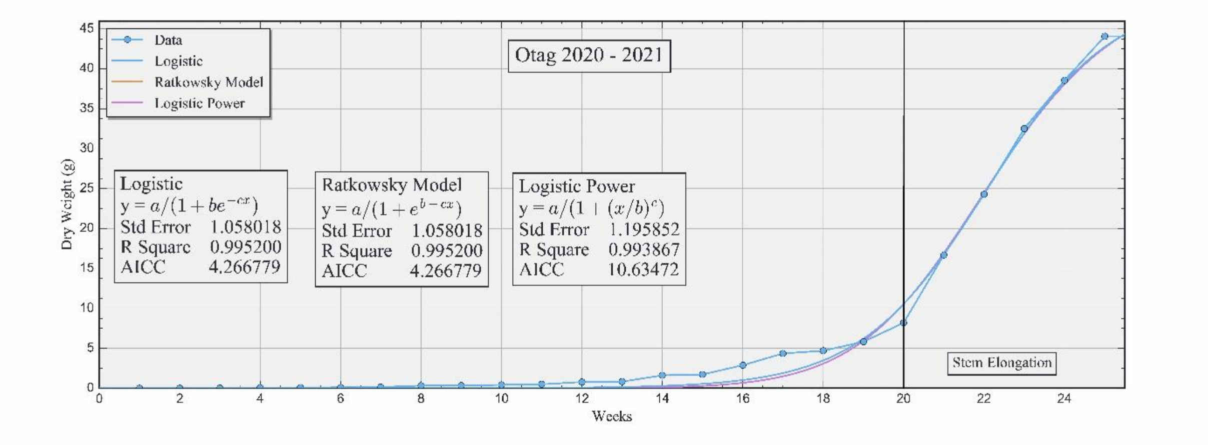

Figure 10. Identifying the dry weight increase of Otag oat cultivar with the sigmoidal curves (second year).

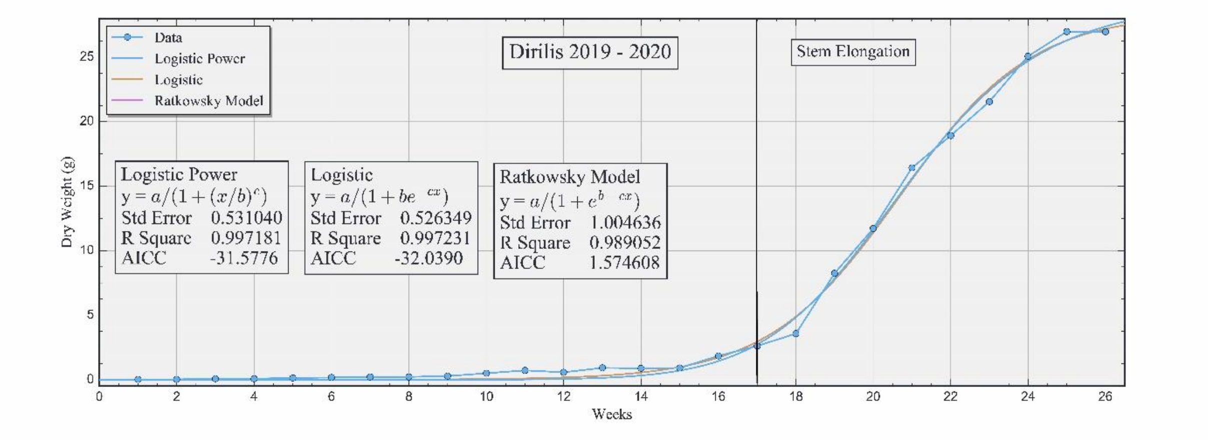

Figure 11. Identifying the dry weight increase of Dirilis oat cultivar with the sigmoidal curves (first year).

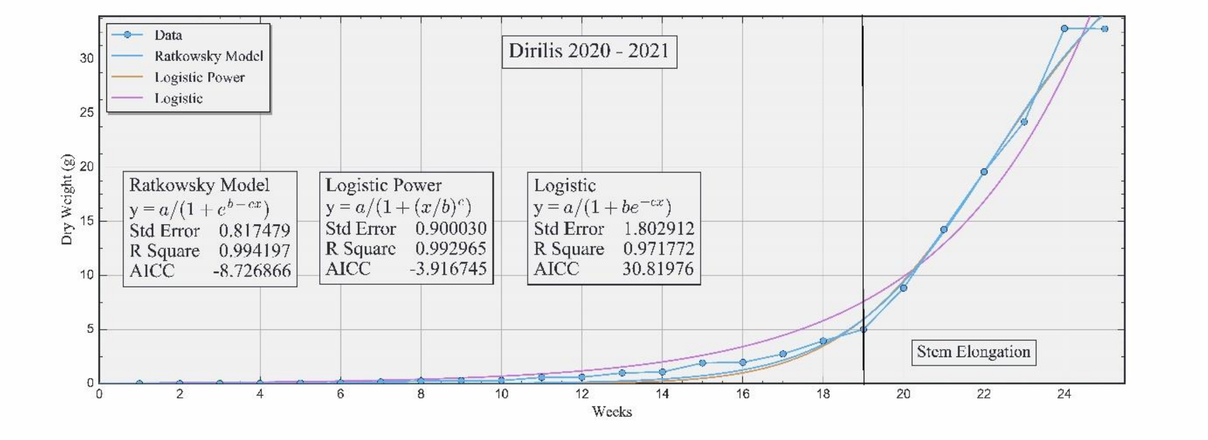

Figure 12. Identifying the dry weight increase of Dirilis oat cultivar with the sigmoidal curves (second year).

In order to evaluate these differences statistically, estimated curve parameters *a*, *b*, *c* were presented in Table 1. In growth analysis, parameter *b* is usually referred as a biological constant when parameters *a* and *c* are often attributed with biological meanings. Parameter *a* is associated with the maximum value of the curve when *c* is reported to reflect the growth rate in certain sigmoidal models (Tjørve, 2003; Keskin et al., 2009) According to the results, parameter *a* varied between 16.33 (Kucukyayla, Logistic) and 62.48 (Otag, Gompertz) in the first year and 27.86 (Kucukyayla, Logistic) and 63.34 (Sebat, Logistic Power, Table 1). Logistic model of Dirilis cultivar in the second year is excluded from this evaluation, parameters of which were outliers possibly due to reduced efficiency of the Logistic model as a result of unusually high growth rate observed in the latest weeks (Table 1, Figure 12). In this study, parameter *a* reflects the theoretical maximum dry weights of oat cultivars. Highest dry weight production were observed in Sebat and Otag cultivars in both years. It should be noted that maximum dry weights of oat cultivars were obtained individually from the tiller groups, therefore it is not an indication of the biological yield which reflects the dry weights collected from an area.

Table 1. Curve parameters of the sigmoidal models given in Figures 1-12

Genotype / Year / Model	a	b	c	PI*	WIP**	*Genotype / Year / Model*	a	b	c	PI	WIP
Kahraman						**Sebat**					
2019-2020						*2019-2020*					
Ratkowsky	18.923	10.920	0.553			Gompertz	48.548	5.356	0.255	6.593	17.860
Logistic	18.923	55284.724	0.553	19.748	9.461	Log Power	44.965	22.105	-9.294		
Log Power***	19.518	19.811	-10.351			Logistic	42.341	28611.734	0.469	21.873	21.170
2020-2021						*2020-2021*					
Ratkowsky	34.364	10.858	0.517			Logistic	62.072	143063.086	0.535	22.183	31.036
Logistic	34.364	51925.439	0.517	20.985	17.182	Ratkowsky	62.072	11.871	0.535		
Log Power	38.633	21.492	-9.395			Log Power	69.341	22.648	-10.376		
Kucukyayla						**Otag**					
2019-2020						*2019-2020*					
Gompertz	17.976	5.052	0.271	5.983	6.613	Gompertz	62.481	3.690	0.158	8.267	22.985
Log Power	17.145	5.444	0.296			Log Power	52.295	24.184	-6.890		
Logistic	16.329	9348.121	0.465	19.658	8.164	Logistic	44.624	4196.901	0.360	23.167	22.312
2020-2021						*2020-2021*					
Logistic	27.862	22440.710	0.504	19.865	13.931	Logistic	49.172	1291554.72	0.639	22.033	24.586
Ratkowsky	27.862	10.019	0.504			Ratkowsky	49.172	14.071	0.639		
Log Power	29.843	20.129	-9.002			Log Power	51.169	22.144	-13.266		
Yeniceri						**Dirilis**					
2019-2020						*2019-2020*					
Log Power	28.253	22.5	-7.008			Log Power	29.537	20.8	-11.346		
Ratkowsky	24.235	8.450	0.390			Logistic	28.397	180881.051	0.585	20.682	14.198
Gompertz	34.012	3.723	0.170	7.712	12.512	Ratkowsky	28.397	12.106	0.585		
2020-2021						*2020-2021*					
Logistic	33.935	11074.126	0.440	21.161	16.968	Ratkowsky	41.355	12.378	0.558		
Ratkowsky	33.935	9.312	0.440			Log Power	45.269	22.545	-11.049		
Log Power	40.856	22.150	-7.671			Logistic	$-1.5*10^{-9}$	$-3.1*10^{-10}$	0.2657		

*PI: Point of inflection, **WPI: Weight at the point of inflection, *** Log Power: Logistic Power

Other curve parameter, the parameter *c* can be associated with the growth rate in several models, although it is not often used to deduce biological meanings. Individual comparison of the curve parameters are not always meaningful since curve parameters often reflect the shape of the equations collectively. Parameter *a* in Logistic model, for example, reflects the maximum dry weight by being the only parameter related to the upper asymptote when other features of the curve such as overall shape or y-axis intersection are represented by parameters *b* and *c* together (Tjørve, 2003). Therefore, biological interpretations are restricted to several models and parameters.

Another biologically meaningful parameter that can be calculated from the equations of some models is the point of inflection (PI). Logistic and Gompertz models both have a fixed point of inflection (PI) where the rate of growth gets its maximum value (Goshu and Koya, 2013). PI of the Logistic curve is calculated with the equation "*a*/2", which remarks the week where plants reaches %50 of the maximum dry weight. PI of the Gompertz model is calculated with the formula "*a*/*e*" with *e* being the mathematical constant; therefore PI of Gompertz is located roughly around %37 of the total growth duration (Duan et al., 2015). Therefore, comparison PI can only be meaningful within the different curves of the same model.

In this case, PI should provide us an idea about how early oat cultivars reach to the maximum rate of dry matter accumulation (which is expected to coincide around late stem elongation stage) in Marmara region. Logistic model provided to be one of the best fitting models to our data which can be seen in high R square and low AICC values in Figures 1-12. Logistic model were constantly among the best fitting models with the only exception being the Dirilis cultivar on the second year. This allowed us to compare the PI and the dry weights at PI (WPI) values of Logistic models from each curve (Table 1). Cultivars that began rapid growing earlier than others are expected to have a lesser PI. In Table 1, PI varied between the weeks 19.66 (Kucukyayla) and 23.17 (Otag), both from the results of the first year (Table 1). Otag consistently had the highest PI for each year, making it the latest to develop rapid dry weight increase in both years. Cultivar Otag began rapid dry weight accumulation in later weeks when compared to other figures of the same year which can also be seen in Figures 11 and 12. Despite its late boom, Otag cultivar had the highest Parameter *a* value in the first year and second highest in the second year, indicating a faster growth in later periods. Other cultivars such as Yeniceri and Sebat also consistently had higher PI, thus were also late developing cultivars.

Cultivars with the lowest PI for both years were Kucukyayla and Kahraman with PI values ranging between 19.65 and 20.98 weeks (Table 1). PI of these cultivars were above 20 weeks for both years with the exception of Kahraman in second year. Variation of PI among cultivars may not seem significant at first, but in practice, each week can be critical and therefore may decisively affect the plant growth. Cultivar differences in terms of their ability to initiate rapid dry weight increase earlier is important for spring cultivation (Buerstmayr et al., 2007) but it may also gain interest for the Mediterranean climate. In a foreseeable future, growing season for winter crops are expected to be shortened (Saadi et al., 2015) and occurrences of heat waves to be increased (Kuglitsch et al., 2010) due to the climate change, which might restrict the growth of late-developing cultivars.

In conclusion, our results indicated that sigmoidal growth models explained oat dry weight increases with the R squares ranging from 0.971772 to 0.998693. This success of the curve fitting process on the growth data of oat makes way for improving our understanding of oat growth. We concluded that Logistic, Logistic Power and Ratkowsky models were the best fitting sigmoidal models for our data. Our comparison of oat cultivars implies that cultivars Otag, Yeniceri and Sebat generated higher maximum dry weights per plant samples. In addition, Otag and Sebat developed later for Marmara Region when Kucukyayla and Kahraman were the earliest cultivars in terms of dry matter accumulation. Although later developing cultivars seemed to accumulate higher dry weights, a larger set of genotypes would be needed to reach a definitive conclusion. In terms of expected consequences of global warming, earlier developing cultivars such as Kahraman and Kucukyayla may provide more consistent yields in the future.

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HETEROSIS AND COMBINING ABILITY VIA LINE × TESTER ANALYSIS FOR QUALITY AND SOME AGRONOMIC CHARACTERS IN SAFFLOWER

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ABSTRACT

This research was conducted in 2018 and 2019 at Tekirdag Namık Kemal University, Faculty of Agriculture, Field Crops Department, Research and Experimental area. It was aimed to determine general combining abilities (GCA) of parental lines, specific combining abilities (SCA) of hybrids, having F_1 hybrids from 5 female lines and 9 male testers. For this purpose, lines and testers were hybridized in all combinations using line x tester method in 2018. The field experiments for hybrids (F_1) were designed in a randomized complete block with three replications in 2019. The male parents Linas as high general combiners for developing increased seed and oil yields in safflower hybrids. The highest heterosis for oil content was calculated EC18 hybrid (14.281%). The highest heterosis was calculated in EC2 (34.079%) and EC4 (34.548%) for seed and oil yield, respectively. In the study, similar heterotic effect was observed between seed and oil yield as there is interaction between them. Hybrid EC2, EC4 and EC18 were determined as best combinations for high seed yield, oil content and oil yield according to the SCA and heterosis values. The promising hybrids will be grown in the next generations together with the other hybrids in order to ensure sufficient variation, and the selection will be started in the later generations such as F_3 and F_4.

Key words: ***Carthamus tinctorius*** **L., Combining ability, Heterosis, Line x tester, Safflower**

INTRODUCTION

Safflower (*Carthamus tinctorius* L.), which is from the *Asteraceae* family, started to be cultivated 3000 years ago and is one of the oldest cultivated plants. It contains 25-45% oil in its seeds, has two different types as linoleic (omega 6) and oleic (omega 9), has high quality edible oil, is suitable for biodiesel production, is cultivated in the form of residue and mixture and is considered as animal feed (Arslan et al., 2012; Culpan, 2015). On the other hand, drought tolerant and cultivation without irrigation enable especially availability of fallow areas (Arslan and Culpan, 2018). The oil content and seed yield of safflower are lower than other oil crops (soybean, sunflower, canola etc.), so safflower cultivation has not been able to develop both in Turkey and in the world. Therefore, various studies (selection and hybridization etc.) must be done in order to improve and increase seed yield and oil content especially and breeding studies must be conducted in order to develop new genotypes.

Suitable choice of parent based on their combining ability effects is crucial for the breeding studies. SCA (specific combining ability) of agronomic traits is an important and useful marker of the potential of inbred lines in generating successful hybrid combinations (Huang et al., 2010). General combining ability (GCA) is a measure of additive gene action, whereas SCA is related to dominance (Huang et al., 2010; Wang et al., 2010).

Line x tester method is widely used in quantitative trait inheritance studies. The method developed by Kempthorne in 1957 is one of the breeding strategies for estimates the GCA of parents and selecting of suitable parents and hybrid with high SCA (Kose, 2017). Heterosis or hybrid vigor describes the phenomenon in which hybrids formed between individuals of the same or closely related species are more vigorous than their parents (Timberlake, 2013).

Combining ability and heterosis are one of the most important characteristics for breeding improved new genotypes (Kose, 2017). Some researchers have studied the combining abilities and heterosis of safflower hybrid genotypes by using line x tester method for breeding characters (Ranga, 1982; Manjare and Jambhale, 1995; Golkar et al., 2012; Nai et al., 2014; Kose, 2017; Kanoje et al., 2018).

The aim of this study was to determine general combining abilities of parent lines, specific combining abilities and heterotic performance of hybrids. In addition, the main objectives of this research were to identify of suitable hybrids for the development of new lines and cultivars with high seed yield, oil content and oil quality.

MATERIALS AND METHODS

Location of experiment, soil and climate properties

The study was carried out between 2018 and 2019 at area of research and experiment, Field Crops Department, Faculty of Agriculture, Tekirdag Namik Kemal University (40°59'25.1"N 27°34'49.1"E) in Turkey. The soil of the experimental area was clayey (C), slightly alkaline (pH=7.10), and had sufficient phosphorus, rich in potassium but low in organic matter (1.33 %). The total rainfall at growing period of safflower (April to August) was 196 mm and mean monthly temperature 20.5 °C in experimental area in 2019. In this context, the soil and climate properties of the experimental area are sufficient and suitable for safflower.

Experimental design and measurements

Five selected lines [3 (PI 209287), 21 (PI 369842), 25 (PI 506427), 28 (PI 537601), Dincer] and nine testers [1 (PI 193473), 10 (PI 253520), 29 (PI 560161), 30 (PI 560177), 31 (PI 572432), 35 (PI 603208), Balci, Linas and Olas] were used for the important breeding objectives such as seed yield and oil content. Especially, safflower genotypes with high oil content; 29, 30, 31, 35, Balci, Linas and Olas were used as testers in the study. These lines and testers were obtained from Eskisehir Transitional Zone Agricultural Research Institute (Dincer and Balci), Trakya Agricultural Research Institute (Linas and Olas) and United States Department of Agriculture (USDA) Agricultural Research Service (PI coded). In this study, spininess was used as a morphological marker. Because spininess of safflower was controlled by single dominant gene and the spiny phenotype was wholly dominant to spineless (Pahlavani et al., 2004; Golkar et al., 2010). The lines (spineless) and testers (spiny) were used as female and male plant materials were hybridized through chemical male sterility (*ch-ms*) with gibberellic acid (GA_3) in June, 2018 (Baydar and Gokmen, 2003; Gokmen, 2004). Thus, 45 hybrid combinations were produced with the line x tester method (Table 1). The seeds obtained from each crossed head were harvested separately on August 1, 2018 and stored in the refrigerator at +4°C.

Table 1. Hybrid combinations of male and female genotypes

Parents		**Lines (♀)**				
		3	**21**	**25**	**28**	**Dincer**
Testers (♂)	**1**	3 x 1 (**BA1**)	21 x 1 (**BA7**)	25 x 1 (**BA13**)	28 x 1 (**BA19**)	D x 1 (**EC13**)
	10	3 x 10 (**BA2**)	21 x 10 (**BA8**)	25 x 10 (**BA14**)	28 x 10 (**BA20**)	D x 10 (**EC14**)
	29	3 x 29 (**BA3**)	21 x 29 (**BA9**)	25 x 29 (**BA15**)	28 x 29 (**BA21**)	D x 29 (**EC15**)
	30	3 x 30 (**BA4**)	21 x 30 (**BA10**)	25 x 30 (**BA16**)	28 x 30 (**BA22**)	D x 30 (**EC16**)
	31	3 x 31 (**BA5**)	21 x 31 (**BA11**)	25 x 31 (**BA17**)	28 x 31 (**BA23**)	D x 31 (**EC17**)
	35	3 x 35 (**BA6**)	21 x 35 (**BA12**)	25 x 35 (**BA18**)	28 x 35 (**BA24**)	D x 35 (**EC18**)
	Balci	3 x B (**EC1**)	21 x B (**EC4**)	25 x B (**EC7**)	28 x B (**EC10**)	D x B (**EC19**)
	Linas	3 x L (**EC2**)	21 x L (**EC5**)	25 x L (**EC8**)	28 x L (**EC11**)	D x L (**EC20**)
	Olas	3 x O (**EC3**)	21 x O (**EC6**)	25 x O (**EC9**)	28 x O (**EC12**)	D x O (**EC21**)

The seeds of hybrid (F_1) and their parents were sown by hand on April 19, 2019. The field experiments were designed in a randomized complete block with three replications. Each hybrid seeds were sown in plots with 2 rows; plots were 5 m long, with 50 cm between rows. Nitrogen and phosphorous were applied at the rate of 100 kg N (urea) and 60 kg P (DAP) per hectare, respectively. All phosphorus and half of the nitrogen were applied prior to seed sowing, and the remaining nitrogen was applied in stem elongation stage (about 1.5 months after sowing). Plants that were found to be spineless during the flowering period (non-hybrids) were cut off from the rows so that real hybrids remained in the rows. Ten plants per plot were selected randomly from each of the hybrids and parents for investigation yield components. These characters were plant height (cm), branch number per plant, head number per plant, head diameter (cm), number of seeds per head, 1000 seed weight (g), seed yield (kg ha^{-1}), hull content (%), oil content (%), oil yield (kg ha^{-1}), linoleic and oleic acid content (%). The all hybrids and parent plants were harvested by hand on August 5, 2019. The oil content analyzes of seeds were measured by reading in the Nuclear Magnetic Resonance (NMR) device as percentage. The seeds were dehumidified by keeping them in an oven at 70 ºC for 48 hours. The average oil content was calculated by making three readings in each plot on a 4 g seed NMR device (Erbas, 2012; Arslan and Culpan, 2020). The oil yield was calculated based on oil percentage and seed yield results (Goksoy et al., 2020). The fatty acid composition (linoleic and oleic acid) of hybrids and parents were analyzed in Gas Chromatography (Agilent 7820A GC) device.

Statistical analysis

The data of this study were analyzed with the TARPOPGEN software developed by Ozcan and Acikgoz (1999). The GCA variance effects of the parents and the SCA variance effects of the hybrids were calculated by the using of the line x tester method as described by Kempthorne (1957) and Singh and Chaudhary (1977):

$$g_i = y_{i.} - y_{..}$$

$$s_{ij} = y_{ij} - y_{..} - g_i - g_j$$

where g_i and g_j are the GCA effects for *i*-th and *j*-th lines, respectively; s_{ij} is the SCA effect for *ij*-th hybrid; y_{ij} is the trait value of *ij*-th hybrid; $y_{i.}$ is the average of the hybrids among *i*-th line crossed with a series of parents; $y_{i..}$ is the overall mean.

Heterosis was estimated based on two criteria, mid-parent heterosis (H_t) and high-parent heterosis (H_b), using the following formulae:

$$H_s\ (\%) = 100 \times (F_1 - MP)/MP$$

$$H_b\ (\%) = 100 \times (F_1 - HP)/HP$$

where F_1 is the mean performance of F_1 hybrids, MP is the parental mean, and HP is higher parent values for all tested traits. Heterotic effects were tested by the LSD test at the 0.05 and 0.01 levels. The *t*-test was used in order to determine the significance of GCA and SCA effects at 0.05 and 0.01 level.

RESULTS AND DISCUSSION

General combining ability (GCA)

The results of the variance analysis for line x tester population were given in Table 2a and Table 2b. The parents and hybrids showed significant differences according to the traits studied in the research. The lines used in the study showed a variation in terms of seed yield, oil content, oil yield and oleic acid content while the testers showed significant differences for seed yield and all quality traits. In addition, the interaction between lines and testers were indicated as significant for all traits except branch numbers (p<0.01).

Table 2a. Variance analysis of yield components for combining ability

SV		DF	Plant height	Branch number	Head number	Head diameter	Number of seeds per head	1000 seed weight
Replication	MS	2	0.789	0.159	3.637	0.010	28.350	13.3032
	F		2.962	1.459	2.433	1.395	11.617**	21.048**
Genotypes	MS	58	35.807	4.948	29.067	0.233	218.408	93.360
	F		134.333**	45.307**	19.443**	31.875**	89.495**	147.712**
Parents	MS	13	33.598	1.739	14.494	0.141	182.439	72.437
	F		126.047**	15.928**	9.695**	19.249**	74.756**	114.609**
Hybrids	MS	44	36.6780	5.669	26.700	0.266	233.813	97.916
	F		137.598**	51.919**	17.859**	36.264**	95.807**	154.922**
Parents vs Hybrids	MS	1	26.222	14.891	322.668	0.021	8.204	164.863
	F		98.375**	136.356**	215.825**	2.284	3.362	260.843**
Lines	MS	4	27.402	4.919	12.023	0.175	109.265	94.022
	F		0.748	1.069	0.446	0.611	0.383	0.878
Testers	MS	9	41.545	10.307	33.009	0.224	89.228	63.245
	F		1.134	2.239	1.224	0.781	0.313	0.591
Lines x testers	MS	32	36.620	4.604	26.958	0.287	285.527	107.071
	F		137.382**	42.160	18.032**	39.207**	116.998**	169.406**
Error		116	0.266	0.109	1.495	0.007	2.4405	0.6320

Table 2b. Variance analysis of seed yield and quality traits for combining ability

SV		DF	Seed yield	Hull content	Oil content	Oil yield	Linoleic acid content	Oleic acid content
Replication	MS	2	67.867	0.455	0.311	3.410	0.657	0.129
	F		2.610	1.011	3.005	0.999	1.403	1.386
Genotypes	MS	58	2913.227	21.865	13.593	315.182	738.736	741.136
	F		112.026**	48.493**	131.170**	92.336**	1576.516**	7923.343**
Parents	MS	13	3378.519	47.324	19.939	351.491	905.795	986.967
	F		129.918**	104.955**	192.400**	102.973**	1933.033**	10551.472**
Hybrids	MS	44	2835.481	14.825	11.997	310.436	664.482	657.463
	F		109.036**	32.880**	115.768**	90.945**	1418.055**	7028.808**
Parents vs Hybrids	MS	1	285.267	0.660	1.332	52.000	1834.103	1226.963
	F		10.970**	1.465	12.855**	15.234**	3914.111**	13117.213**
Lines	MS	4	7275.759	4.661	20.226	842.989	41.573	35.833
	F		9.919**	0.482	6.313**	9.229**	0.715	0.914
Testers	MS	9	9023.117	40.534	43.058	920.543	3401.307	3441.293
	F		12.301**	4.192**	13.439**	10.078**	58.502**	87.767**
Lines x testers	MS	32	733.537	9.668	3.204	91.340	58.140	39.209
	F		28.208**	21.443**	30.916**	26.759**	124.076**	419.182**
Error		116	26.004	0.450	0.103	3.413	0.468	0.093

SV: source of variation, DF: degree of freedom, MS: mean square, F: F value. **: significant at P<0.01

GCA effects (general combining ability) of the lines and testers in terms of the yield components were shown in Table 3a. Among the parents the highest positive GCA effect were obtained by the lines 21 and tester 31 in terms of plant height, and difference no significant statistically ($p>0.05$). The GCA effects of lines and testers on the branch number were insignificant, these values varied from -1.037 (testers 30) and 1.390 (testers 1). The general combining ability of the all parents in terms of head number, head diameter, number of seeds per head and 1000 seed weight were insignificant statistically (Table 3a). The present results confirm the findings of Golkar et al. (2012), Kose (2017), Kanoje et al. (2018) and Thorat and Gawande (2021).

Table 3a. GCA effects (general combining ability) of the parents for the yield components

Source of variation	Plant height	Branch number	Head number	Head diameter	Number of seeds per head	1000 seed weight
			Lines			
3 (♀)	-0.259	-0.436	-0.860	0.035	-2.104	-2.701
21 (♀)	1.685	-0.366	-0.353	0.110	1.111	-0.719
25 (♀)	-0.037	0.075	0.936	-0.091	-1.652	2.292
28 (♀)	-1.000	0.104	0.173	0.011	2.789	0.225
Dincer (♀)	-0.389	0.623	0.103	-0.128	-0.144	0.902
			Testers			
1 (♂)	-1.600	1.390	1.121	0.045	3.010	0.365
10 (♂)	-1.167	1.030	2.155	-0.105	-0.090	-0.478
29 (♂)	1.367	-0.990	-1.912	0.077	-2.303	-1.398
30 (♂)	-0.767	-1.037	1.761	-0.088	0.470	-1.937
31 (♂)	3.667	-0.544	-0.385	-0.092	-4.363	2.144
35 (♂)	-1.233	-0.224	-0.199	-0.045	-1.556	-1.325
Balci (♂)	-0.633	-0.064	-2.159	0.237	3.104	-0.655
Linas (♂)	-0.233	0.176	-0.305	0.100	1.317	-1.141
Olas (♂)	0.600	0.263	-0.079	0.077	0.410	4.425
SE (for lines)	0.099	0.064	0.235	0.016	0.301	0.153
SE (for testers)	0.133	0.085	0.316	0.022	0.403	0.205

GCA effects of the parents in terms of the seed yield and quality traits were given in Table 3b. Among the lines and testers while 1, 21, 28, Dincer, Balci, Linas and Olas had positive GCA effects and statistically significant ($p<0.01$), remaining other lines and testers had negative GCA effects for seed yield. For oil content among the parents significant and positive GCA effects were observed to 28, 30, 31, 35, Linas and Olas respectively. Therefore, these parents can be considered a great line and tester for a safflower breeding program. In addition, line 21, 28, Dincer and tester 1, 29, 35, Balci, Linas and Olas had significant and positive GCA effects in terms of oil yield among the parents. Particularly male parents Linas as high general combiners for developing increased seed and oil yields in safflower hybrids. Although GCA was not significant when the linoleic and oleic acid content was examined for the lines, testers were showed negative and positive highly significant GCA. Among the testers 35 and Olas had significant and negative GCA effects for linoleic acid content. On the contrary, same testers had significant and positive GCA effects for oleic acid content (13.122 and 35.623, respectively). This is expected in terms of linoleic and oleic acid composition. Golkar et al. (2011) reported that GCA effects for linoleic acid showed a range from -8.27 to 2.75 in F_1. Ratnaparkhi et al. (2015a) indicated that GCA values ranged from -2.357 to 1.327 in terms of oleic acid content. Because of the parents used by the researchers were linoleic type, their results are different from the results of our study. The parents with high GCA can be used to develop lines and cultivars with high seed yield, oil content and oil quality.

Specific combining ability (SCA)

SCA effect is an important characteristic for estimating and choosing suitable and superior hybrid combinations. In the study on SCA effects of 45 hybrids for all the traits were given in Table 4a and 4b. The SCA effects of hybrids were significant in terms of all traits (Table 2a and 2b, $p<0.01$). Among the hybrids the highest positive SCA effect were observed by the EC4 (5.115) in terms of plant height. Insignificant SCA effect for branch number was observed in 12 hybrids (Table 4a). BA1 hybrid had the highest positively and significant SCA in terms of head number (6.993). While EC2 hybrid (20.924) exhibited the highest positively and significant SCA effects for number of seeds per head, BA9 (-14.179) showed the lowest significantly negative SCA effects in terms of 1000 seed weight. Patel et al. (2018) reported that the highest SCA value for number of seeds per head was determined as 9.27 in the line (4) × tester (3) mating design. This difference may be due to the line (5) × tester (9) mating design in our study. The SCA effects of hybrids on the seed yield were significant, these values varied from -35.563 (EC5) and 28.236 (BA19). Nai et al. (2014) reported that SCA effects for seed yield showed a range from -1.05 to 23.50 in 45 F_1 genotypes, Kanoje et al. (2018) indicated that SCA values ranged from -38.19 to 43.93 in terms of seed yield. It can be said that the differences in our study are the safflower line and testers used by other researchers.

Table 3b. GCA effects (general combining ability) of the parents for the seed yield and quality

Source of variation	Seed yield	Hull content	Oil content	Oil yield	Linoleic acid content	Oleic acid content
			Lines			
3 (♀)	-9.572**	-0.071	-1.086**	-4.226**	0.309	-0.084
21 (♀)	16.010**	-0.044	-0.053	5.085**	1.799	-1.464
25 (♀)	-23.376**	0.682	0.023	-7.559**	-1.635	1.287
28 (♀)	3.991**	-0.453	1.330**	2.460**	-0.382	-0.726
Dincer (♀)	12.947**	-0.114	-0.214**	4.240**	-0.092	0.987
			Testers			
1 (♂)	10.824**	-0.083	-2.572**	0.956*	6.592**	-9.105**
10 (♂)	-15.376**	2.124**	-2.390**	-7.029**	8.066**	-8.632**
29 (♂)	-9.516**	-0.473**	0.136	2.864**	10.337**	-9.085**
30 (♂)	-25.296**	-0.046	0.717**	-7.466**	0.995**	-1.259**
31 (♂)	-39.463**	-1.576**	1.227**	-11.787**	2.889**	-4.281**
35 (♂)	-0.136	-3.206**	2.901**	2.956**	-12.608**	13.122**
Balci (♂)	25.310**	1.594**	-0.335**	7.810**	10.245**	-8.438**
Linas (♂)	33.570**	0.527**	0.263**	11.560**	8.807**	-7.945**
Olas (♂)	20.084**	1.137**	0.053	5.863**	-35.325**	35.623**
SE (for lines)	0.981	0.129	0.062	0.356	0.132	0.059
SE (for testers)	1.317	0.173	0.083	0.477	0.177	0.079

SE: standard error, *: significant at P<0.05, **: significant at P<0.01

The more than half of the hybrid combination had positive SCA effect for oil content and oil yield. While the highest positive and significant SCA effect was observed by the EC18 (2.014) in terms of oil content, the highest positive and significant SCA effect for oil yield was obtained from the EC4 hybrid (9.441). Similar results were reported earlier by Kose (2017) and Kanoje et al. (2018). In addition, Thorat and Gawande (2021) determined that the SCA values ranged from -2.145 to 2.145 in terms of oil content. It can be said that the reason for this difference is that the genotypes used belong to different geographical origins. While the highest positive and significant SCA effect was observed by the BA24 hybrid (12.616) in terms of linoleic acid content, the lowest negative and significant SCA effect for oleic acid content was obtained from the same hybrid (-13.668). Selection for high SCA effects in terms of seed yield, oil content and oil yield would be a suitable strategy for improvement new safflower cultivars and lines. In addition, choosing hybrids with low hull content will contribute to the development of lines and varieties with low and thin hull.

Heterosis and heterobeltiosis

Heterosis (Ht) and heterobeltiosis (Hb) values of hybrids for the selected characters in the study were presented in Table 5. Significant heterosis and heterobeltiosis were found for different hybrids for the selected characters (seed yield, oil content and oil yield). The highest heterosis and heterobeltiosis were calculated EC2 (34.079%) and EC4 (26.200%) hybrids for seed yield, respectively. The heterosis and heterobeltiosis values ranged from -6.567% to 14.281% and -12.571% to 5.597% for the oil content, respectively. The highest heterosis was calculated EC18 hybrid (14.281%). The highest heterosis was calculated in EC2 (34.079%) and EC4 (34.548%) for seed and oil yield, respectively. In the study, similar heterotic effect was observed between seed and oil yield as there is interaction between them. Ranga (1982) calculated the heterosis value as 55.0% in terms of seed yield in the F_1 hybrid. Ratnaparkhi et al. (2015b) calculated the heterosis values in terms of oil content -22.70% and -4.04%. Erbas (2012) calculated the heterosis value as 42.3% in terms of oil yield. The difference in the heterosis and heterobeltiosis results of different researchers studied in the present line and testers can be commented to the divergence of the material used in studies.

Heterosis touches on the phenomenon that lineage of diverse varieties of a species or crosses between species present more yield, development, and fertility than both parents (Birchler et al., 2010). Heterosis and heterobeltiosis values are crucial and considerable genetic parameters for plant breeding programs. These values are guidelines for selecting superior hybrids.

Table 4a. SCA effects (specific combining ability) of the hybrids for the yield components

Code	Hybrids	Plant height	Branch number	Head number	Head diameter	Number of Seeds per head	1000 seed weight
BA1	3 x 1	0.693*	-0.030	**6.993****	-0.211**	-5.270**	-6.603**
BA2	3 x 10	-3.074**	-1.837**	-0.273	-0.227**	-7.770**	2.330**
BA3	3 x 29	2.226**	-0.050	0.493	-0.300**	-9.656**	8.323**
BA4	3 x 30	-3.141**	0.130	**-4.647****	0.238**	5.270**	3.443**
BA5	3 x 31	1.093**	-0.564**	-3.200**	**0.527****	16.370**	0.445
BA6	3 x 35	3.659**	1.583**	-1.620*	-0.229**	-5.170**	-1.083*
EC1	3 x B	-4.107**	0.990**	0.640	-0.083	-9.996**	-2.949**
EC2	3 x L	-0.174	-1.150**	-0.547	0.308**	**20.924****	-6.120**
EC3	3 x O	2.826**	0.930**	2.160**	-0.022	-4.703**	2.214**
BA7	21 x 1	4.248**	-0.234	-1.214	-0.202**	-9.818**	0.198
BA8	21 x 10	-1.019**	1.326**	2.553**	0.245**	11.716**	9.598**
BA9	21 x 29	-3.885**	0.546**	-0.181	-0.008	-3.971**	**-14.179****
BA10	21 x 30	-7.585**	-0.641**	-4.254**	0.490**	11.022**	-4.229**
BA11	21 x 31	-1.185**	-0.301	0.693	0.042	6.889**	-5.637**
BA12	21 x 35	0.715*	0.713**	1.606*	-0.124*	-8.784**	6.515**
EC4	21 x B	**5.115****	-0.781**	0.166	-0.141**	-1.011	1.239**
EC5	21 x L	3.715**	-0.687**	-0.354	-0.273**	-11.124**	2.871**
EC6	21 x O	-0.119	0.059	0.986	-0.030	5.082**	3.625**
BA13	25 x 1	-0.030	0.725**	1.030	0.209**	1.312	5.777**
BA14	25 x 10	-0.130	0.785**	0.797	-0.077	-0.421	-5.093**
BA15	25 x 29	2.837**	0.805**	-0.703	0.230**	10.192**	-2.489**
BA16	25 x 30	4.637**	-0.681**	3.924**	-0.022	-0.881	0.233
BA17	25 x 31	2.037**	1.492**	1.370	-0.140**	-8.348**	-0.428
BA18	25 x 35	**-6.396****	-1.195**	-3.150**	0.381**	14.745**	0.221
EC7	25 x B	2.170**	0.079	1.810*	0.030	-0.615	2.385**
EC8	25 x L	-1.063**	-0.495**	-3.876**	-0.285**	-2.995**	-3.566**
EC9	25 x O	-4.063**	-1.515**	-1.203	-0.325**	**-12.988****	2.961**
BA19	28 x 1	-1.400**	**-2.071****	-4.107**	0.503**	6.038**	-1.972**
BA20	28 x 10	1.333**	-0.411**	-3.673**	-0.176**	-5.762**	-4.216**
BA21	28 x 29	-3.867**	-0.491**	-0.807	0.114*	5.384**	-1.256**
BA22	28 x 30	1.933**	0.822**	0.920	-0.245**	-11.222**	5.747**
BA23	28 x 31	-0.500	0.462**	0.033	-0.366**	-8.989**	5.199**
BA24	28 x 35	3.233**	0.942**	4.713**	0.228**	4.304**	-3.496**
EC10	28 x B	-0.700*	0.049	-2.260**	-0.326**	1.111	-0.632
EC11	28 x L	-0.100	**2.242****	5.153**	0.015	2.864**	-2.112**
EC12	28 x O	0.067	-1.544**	0.027	0.252**	6.271**	2.738**
EC13	D x 1	-3.511**	1.610**	-2.703**	-0.300**	7.738**	2.601**
EC14	D x 10	2.889**	0.137	0.597	0.234**	2.238**	-2.619**
EC15	D x 29	2.689**	-0.810**	1.197	-0.036	-1.949*	**9.601****
EC16	D x 30	4.156**	0.370	4.057**	**-0.461****	-4.189**	-5.193**
EC17	D x 31	-1.444**	-1.090**	1.104	-0.063	-5.922**	0.422
EC18	D x 35	-1.211**	-2.043**	-1.550*	-0.255**	-5.096**	-2.156**
EC19	D x B	-2.478**	-0.336	-0.356	0.521**	10.511**	-0.042
EC20	D x L	-2.378**	0.090	-0.376	0.235**	-9.669**	8.927**
EC21	D x O	1.289**	2.070**	-1.970**	0.125*	6.338**	-11.539**
SE		0.298	0.191	0.706	0.049	0.902	0.459

SE: standard error, *: significant at $P<0.05$, **: significant at $P<0.01$

Table 4b. SCA effects (specific combining ability) of the hybrids for the seed yield and quality

Code	Hybrids	Seed yield	Hull content	Oil content	Oil yield	Linoleic acid content	Oleic acid content
BA1	3 x 1	-18.268**	-0.579	-0.858**	-5.147**	0.885*	-0.286
BA2	3 x 10	-1.301	0.948*	0.420*	0.738	-1.079**	-1.070**
BA3	3 x 29	-9.028**	-1.172**	0.724**	-2.047	-0.561	1.284**
BA4	3 x 30	-14.515**	**3.568****	-1.351**	-5.009**	-1.668**	-2.902**
BA5	3 x 31	15.352**	-1.585**	0.206	5.526**	-2.053**	1.130**
BA6	3 x 35	1.125	0.861*	0.216	0.506	-1.275**	3.036**
EC1	3 x B	1.612	-1.105**	0.214	0.452	0.401	-0.084
EC2	3 x L	22.185**	-0.639	-0.103	5.965**	2.710**	-0.924**
EC3	3 x O	2.839	-0.299	0.533**	-0.985	2.641**	-0.184
BA7	21 x 1	16.117**	2.394**	0.685**	5.421**	-1.695**	0.934**
BA8	21 x 10	-3.350	2.120**	-0.500**	-1.950	0.031	2.021**
BA9	21 x 29	17.324**	-0.050	0.407*	6.001**	-0.010	-0.096
BA10	21 x 30	0.370	-0.310	-0.484*	-0.283	1.342**	-1.502**
BA11	21 x 31	-4.330	-1.413**	0.259	-1.035	0.378	0.450*
BA12	21 x 35	2.410	-0.816*	-0.794**	0.331	0.435	0.467**
EC4	21 x B	24.697**	-2.450**	1.158**	**9.441****	-1.422**	-0.183
EC5	21 x L	**-35.563****	2.367**	-0.196	**-12.179****	-3.480**	3.623**
EC6	21 x O	-17.676**	-1.843**	-0.537**	-5.746**	4.422**	-5.714**
BA13	25 x 1	-5.564	-1.216**	0.750**	-0.741	0.939*	-1.267**
BA14	25 x 10	7.936**	0.044	0.928**	3.721**	1.505**	-1.940**
BA15	25 x 29	-9.458**	0.841*	-0.671**	-3.401**	-0.210	-0.827**
BA16	25 x 30	7.089*	0.148	0.504**	2.171*	-3.404**	2.217**
BA17	25 x 31	6.989*	-0.256	-0.279	2.042	-2.692**	0.439*
BA18	25 x 35	-2.671	-2.559**	0.561**	-0.974	-3.591**	**6.546****
EC7	25 x B	-7.618*	2.008**	-1.471**	-3.848**	1.015*	-0.794**
EC8	25 x L	-12.378**	-0.376	-0.585**	-5.008**	-0.353	-2.678**
EC9	25 x O	15.676**	1.364**	0.265	6.038**	6.792**	-1.695**
BA19	28 x 1	**28.236****	0.420	-0.381*	7.877**	-4.254**	1.546**
BA20	28 x 10	2.302	-1.070**	1.211**	1.822	-2.658**	1.700**
BA21	28 x 29	-0.224	-0.974*	1.438**	1.470	0.131	0.616**
BA22	28 x 30	-13.944**	**-2.700****	0.370*	-4.528**	-4.473**	5.100**
BA23	28 x 31	-21.011**	0.430	-0.443*	-7.627**	2.319**	-1.068**
BA24	28 x 35	-3.138	0.860*	-1.997**	-3.150**	**12.616****	**-13.668****
EC10	28 x B	-4.118	0.676	0.139	-0.771	0.263	1.869**
EC11	28 x L	14.022**	-0.024	-0.052	4.846**	2.222**	0.066
EC12	28 x O	-2.124	2.383**	-0.286	0.059	-6.167**	3.838**
EC13	D x 1	-20.520**	-1.019**	-0.196	-7.410**	4.126**	-0.927**
EC14	D x 10	-5.587	-2.043**	**-2.058****	-4.331**	2.202**	-0.710**
EC15	D x 29	1.387	1.354**	-1.898**	-2.023	0.650	-0.977**
EC16	D x 30	21.000**	-0.706	0.961**	7.649**	8.203**	-2.913**
EC17	D x 31	3.000	2.824**	0.258	1.093	2.048**	-0.951**
EC18	D x 35	2.273	1.654**	**2.014****	3.287**	**-8.184****	3.619**
EC19	D x B	-14.573**	0.871*	-0.040	-5.274**	-0.258	-0.808**
EC20	D x L	11.733**	-1.329**	0.936**	6.376**	-1.099**	-0.088
EC21	D x O	1.287	-1.606**	0.025	0.633	-7.688**	3.755**
SE		2.944	0.388	0.186	1.067	0.395	0.177

SE: standard error, *: significant at P<0.05, **: significant at P<0.01

Table 5. Heterosis (H_t) and heterobeltiosis (H_b) values of hybrids for the selected characters

Code	Hybrids	Seed yield		Oil content		Oil yield	
		H_t	H_b	H_t	H_b	H_t	H_b
BA1	3 x 1	-11.576**	-24.723**	**-6.567****	-7.342**	-14.081**	-27.367**
BA2	3 x 10	1.362	-4.030	2.582**	-0.746	5.040	-3.598
BA3	3 x 29	-4.595	-6.287	0.105	-7.451**	-4.388	-10.143
BA4	3 x 30	-25.387**	-32.003**	-1.952*	-7.147**	-25.910**	-29.369**
BA5	3 x 31	14.650*	-13.018*	1.470	-6.539**	19.597*	-3.473
BA6	3 x 35	3.300	-7.733	4.191**	-5.948**	6.258	-13.236**
EC1	3 x B	15.380**	-5.736	2.405**	-0.626	17.344**	-6.338
EC2	3 x L	**34.079****	5.612	1.459	-3.202**	34.248**	2.176
EC3	3 x O	7.792*	-13.813**	-0.412	-7.656**	-3.143	-26.761**
BA7	21 x 1	17.119**	9.240**	-2.105*	-5.366**	16.083**	4.947
BA8	21 x 10	-1.953	-24.338**	-1.566	-8.540**	-5.589	-30.779**
BA9	21 x 29	19.975**	-4.828	-1.666*	-5.354**	19.185**	-2.590
BA10	21 x 30	-5.211**	-28.897**	0.005	-1.294	-4.278	-27.917**
BA11	21 x 31	-12.930**	-42.769**	0.771	-3.385**	-10.221	-39.720**
BA12	21 x 35	2.750	-8.897**	0.261	-5.883**	3.902	-2.241
EC4	21 x B	28.829**	**26.200****	4.477**	3.205**	**34.548****	**30.244****
EC5	21 x L	-14.466**	-16.767**	0.272	-0.260	-14.284**	-17.048**
EC6	21 x O	-9.475**	-10.142**	-4.468**	-7.764**	-13.490**	-17.046**
BA13	25 x 1	-2.488	-25.658**	-1.652	-4.934**	-3.023	-24.277*
BA14	25 x 10	10.270	1.217	3.546**	-3.795**	15.176*	13.634
BA15	25 x 29	-10.598	-20.685**	-4.721**	-8.290**	-15.260*	-27.324**
BA16	25 x 30	-1.335	-5.879**	3.319**	1.982*	0.783	-4.413
BA17	25 x 31	-4.798	-19.428**	-0.635	-4.730**	-4.555	-16.231*
BA18	25 x 35	-4.832	-24.469**	4.484**	-1.915**	-2.212	-25.936**
EC7	25 x B	3.979	-23.393**	-3.703**	-4.881**	0.596	-25.310**
EC8	25 x L	0.665	-28.038**	-0.718	-1.241	-0.269	-28.961**
EC9	25 x O	18.118**	-14.513**	-1.799*	-5.183**	14.525**	-18.882**
BA19	28 x 1	23.408**	20.638**	-2.362**	-6.754**	20.353**	12.509**
BA20	28 x 10	-2.062	-21.666**	7.483**	-1.285	3.017	-22.640**
BA21	28 x 29	-1.609	-18.961**	4.384**	1.706*	3.282	-13.093**
BA22	28 x 30	-26.420**	-42.899**	5.651**	**5.597****	-21.832**	-39.631**
BA23	28 x 31	**-40.809****	**-60.129****	1.584*	-1.412	**-38.941****	**-58.207****
BA24	28 x 35	-6.888	-13.713**	-0.399	-5.384**	-7.729*	-10.089**
EC10	28 x B	3.120	0.268	4.288**	1.747*	7.628*	7.241
EC11	28 x L	17.767**	9.369**	3.753**	3.001**	22.280**	14.291**
EC12	28 x O	-2.347	-7.582*	-0.713	-2.954**	-3.130	-10.256**
EC13	D x 1	-13.351**	-18.502**	0.510	-2.146*	-12.766**	-15.796**
EC14	D x 10	-5.920	-26.937**	-1.258	-2.695**	-7.535	-28.940**
EC15	D x 29	3.523	-17.330**	-3.804**	**-12.571****	2.156	-11.599**
EC16	D x 30	13.350**	-14.460**	10.325**	2.659**	28.868**	1.982
EC17	D x 31	-6.898	-38.540**	6.169**	-3.859**	4.455	-27.154**
EC18	D x 35	1.109	-9.644**	**14.281****	1.461	17.320**	16.437**
EC19	D x B	-2.057	-3.206	6.367**	1.372	4.233	0.472
EC20	D x L	18.180**	14.007**	9.649**	2.774**	34.067**	21.495**
EC21	D x O	3.037	1.376	2.482**	-6.586**	5.469	-5.231

* and **: significant at $P<0.05$ and $P<0.01$, respectively

CONCLUSIONS

Choice for lines and testers with high general combining ability effects and hybrids with high specific combining ability effects would be a suitable objective for high seed yield, oil content and oil quality in safflower. Especially hybrid EC2, EC4 and EC18 were determined as best combinations for high seed yield, oil content and oil yield according to the SCA and heterosis values. In addition, BA24 hybrid is promising for developing cultivars with high oleic acid content. The promising hybrids will be grown in the next generations together with the other hybrids in order to ensure sufficient variation, and the selection will be started in the later generations such as F_3 and F_4.

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EFFECTS OF DIFFERENT BORON APPLICATIONS ON SEED YIELD AND SOME AGRONOMICAL CHARACTERISTICS OF RED LENTIL

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ABSTRACT

The purpose of the study was to determine the effect of different boron application methods on grain yield and some agricultural properties of Sakar red lentil variety (*Lens culinaris* Medic.). The study was conducted in 2018-2019 and 2019-2020 growing years in the boron lack areas of Sanliurfa in Turkey. Experimental design was randomized complete block design with four replicates. Borax and Sakar lentil variety were used as Boron source and plant material, respectively. The methods of applying boron were control (0 kg B da^{-1}), to soil (0.20 kg B da^{-1}), to foliar spraying (when plants had 5-6 leaves 0.05 kg da^{-1} B and had pre-flowering period 0.05 kg da^{-1}B) and to soil+foliar application (to soil 0.10 kg B da^{-1} + to foliar when plants had 5-6 leaves 0.05 kg da^{-1} B and had pre-flowering period 0.05 kg da^{-1} B). According to average of two years; the highest seed yield was obtained from soil + foliar applications (198.57 kg da^{-1}). Also, the highest plant height (42.49 cm), thousand seed weight (41.80 g), biological yield (582.96 g m^{-2}) values were seen on soil + foliar applications. Protein content of seed (33.29 %) and the boron content of seed (19.09 mg kg^{-1}) values increased with boron applications. The most effective boron application method was determined as soil + foliar boron application but other application methods were more effective than control applications.

Keywords*: Boron, foliar application, grain yield, red lentil, soil application*

INTRODUCTION

Lentil is one of the best and the cheapest vegetable protein sources. Because it contains about 25% protein, 1.0% fat and 56% carbohydrate in seeds and also provides a significant amount of vitamin A and B (Grusak et al., 2016). On the other hand, lentil plants may adapt extreme climate conditions such as drought. For this reason, lentil is of great importance in arid climate regions due to obtaining acceptable yield. It is also being able to grow in soil types with poor fertility. Lentils have an important place in crop rotation to maintain soil fertility and root nodules. They can fix atmospheric nitrogen by symbiotic rhizobia.

In order to meet the world need of red lentils with an increasing population, it is necessary to increase the amount of yield per unit area. One way to rise the yield is to supply that the plant nutrition. Fertilizers and soil fertility play a major role for obtaining a higher yield (Rahman et al., 2013). It’s known that macro and micro nutrients play a large role in plant growing. Especially micro nutrient deficiencies can reduce the yield and quality of the crop. So, soil nutrient content must be known and macro and micro nutrients must be applied as to the soil, leaves or both of them for obtaining higher yield.

Micro-element deficiencies are much higher than estimated in the world and also in lentil grown areas. It is known that, even though the lack of micro elements in plants is not evident, micro element applications result in significant increases in productivity (Oktem et al., 2012).

Abid et al. (2017) specified that the treatment of micro nutrient fertilizer increases the lentil quality (protein %) and quantity of grain yield in arid region. Chakraborty (2009), emphasized that micronutrient application along with macronutrients could prove advantageous in increasing the grain yield of lentil in the weak soils of the dry areas.

Boron is a micronutrient that insufficiency in culture plants is more common than in other trace element deficiencies (Demirtas, 2005).

Lack of boron in the soil is one of the worldwide problem for plant growing and the boron defect has been explained in 132 countries in 80 plant varieties included lentil (Oktem, 2020). Many researchers have found that seed yield and also quality are positively affected appropriate level of boron applications. Boron intake was reduced under poor soil nutrient and drought stress conditions. Most of the boron in soil is found in organic matter. For this reason, soil boron deficiency encountered in low soil organic matter (Schulin et al., 2010). Soils with low organic matters content are more sensitive to boron deficiency because existent boron is released from organic matter (Dear and Weir, 2004). Boron affects generative growth more than vegetative growth in plants and boron accelerates flower formation and bud development. Tariq and Mott (2007) reported that dicotyledons plants require more boron than monocotyledon, because the roots of dicotyledons have a higher capacity to adsorb boron than roots of monocotyledons.

Yagmur and Kaydan (2005) reported that a 26% gain in seed yield was seen with foliar application of boron in lentils. In another study, Khurana and Arora (2012) reported that seed yield was increased by 21.4-23.3% with application of 0.75 B kg ha^{-1} boron applications. Quddus et al. (2014) explained that the highest seed yield obtained from 1.5 kg ha^{-1} boron application. Yang et al. (2009) emphasized that treatment of boron to soil rise seed yield as 46% compared to control in rapeseed. Kumar et al. (2018) said that grain yield of legumes was affected by positively 0.5 and 2.5 kg B da^{-1} of dosage. Halder et al. (2007) emphasized that the highest grain yield and 1000 kernel weight were obtained from 2 kg B ha^{-1}. Khan et al. (2019) explained that when nutrients applied on foliar, they get very quickly and directly to the leaf cells and because of that effect is very high. On the other hand, Nagula et al. (2015) stressed that the application of boron through soil or foliar application was found to be beneficial in stimulating plant growth and yield. Gupta and Solanki (2013) reported that the only way to accomplish the boron deficiency is its external application and it can be water soluable and sprayed on to the crop or the soil.

Boron deficiency appears in the vast majority of lentil growing areas in arid climate conditions such as the Southeastern Anatolia Region in Turkey where this study was conducted. Low organic matter content and boron deficiency at the soil are very common in Southeastern Anatolia Region. Oktem and Oktem (2006) reported that boron deficiency was found at 95% analyzed soil sample of Southeastern Anatolia Region. Boron levels of experiment area were 0.217 and 0.265 mg kg^{-1} in 2018 and 2019, respectively. These boron levels were below 0.5 mg kg^{-1} critical limit of boron in the soil (Vista et al., 2019).

This study, to determine the effect of different boron application methods to seed yield and some agricultural properties of Sakar red lentil cultivar. The study was carried out especially in areas with boron deficiency and in rain-fed conditions. Supplemental irrigation was not done during the growing period.

MATERIALS AND METHODS

Description of the Experimental Sites

The research was conducted in 2018-2019 and 2019-2020 for winter growth periods in Sanliurfa, Turkey.

Some of the chemical properties of the research area are shown in Table 1. The soil of the experiment area was clay, slightly alkaline and very low in salt content and organic matter. The research was set up in boron deficit soils of Sanliurfa Region. In both treatment years, soil samples were taken previous to seeding from the experiment land and some chemical characters of the soil were analyzed using the method that described by Jackson (2005). The soil characteristics of the trial area for 2018-19 and 2019-20 years were given in Table 2. It is clearly seen from table that boron levels of research area were below 0.5 mg kg^{-1} that was critical limit of boron in the soil.

Table 1. Some chemical properties of soil research area (0-20 cm)

Years	Saturation (%)	Total salt (%)	pH	Lime (%)	K_2O (kg da^{-1})	P_2O_5 (kg da^{-1})	Org. Mat. (%)	AvailableB (mg kg^{-1})
2018/19	59	0.80	7.79	26.3	133.4	4.23	0.48	0.217
2019/20	63	0.72	7.86	24.7	101.9	5.08	0.60	0.265

Table 2. Some meteorological data of Sanliurfa

Year	Parameters	**1**	**2**	**3**	**4**	**5**	**6**	**10**	**11**	**12**
2018	Max.Temp. (^{0}C)	17.8	18.9	26.8	32.1	36.3	43.1	34.2	27.5	18.2
	Min.Temp. (^{0}C)	2.0	4.1	6.1	9.3	12.2	16.2	9.3	5.4	0.5
	Av.Temp. (^{0}C)	8.1	10.4	15.5	19.9	23.0	28.6	21.6	13.0	8.6
	Av.Humidity (%)	67.0	68.2	52.9	38.4	50.1	36.6	45.6	72.5	84.9
	Rainfall (mm)	118.9	87.4	13.3	35.8	64.5	10.1	45.6	72.5	84.9
2019	Max.Temp. (^{0}C)	17.2	18.6	22.1	26.8	40.3	44.1	36.2	27.5	22.4
	Min Temp. (^{0}C)	-1.2	2.2	1.9	5.9	10.1	18.5	11.3	5.9	4.5
	Av.Temp. (^{0}C)	6.1	8.3	10.7	14.4	25.2	30.7	22.9	14.8	9.0
	Av. Humidity (%)	76.4	71.7	69.5	67.0	35.8	30.6	44.9	42.3	79.4
	Rainfall (mm)	113.8	83.8	156.7	97.4	7.3	9.9	45.1	6.7	277.7
2020	Max.Temp. (^{0}C)	14.0	19.4	24.7	29.4	38.0	41.6	34.2	26.1	18.7
	Min. Temp. (^{0}C)	0.2	-5.8	3.6	4.8	11.1	15.3	16.1	5.6	0.5
	Av.Temp. (^{0}C)	6.6	7.0	13.3	17.1	23.2	28.9	24.0	13.5	9.4
	Av. Humidity (%)	69.1	63.4	63.6	54.2	41.0	29.9	27.5	60.9	61.5
	Rainfall (mm)	76.9	24.1	90.8	68.3	39.1	0.4	0.0	84.3	17.9

Months; 1 January, 2 February, 3 March, 4 April, 5 May, 6 June, 10 October, 11 November, 12 December.
Data collected from Sanliurfa Meteorological Station (Anonymous, 2021)

Some climatic properties of the experiment area are shown in Table 2. It is seen from the detailed climatic data from Table that the weather was usually warm during winter months and rainfall were rare. During the time session for the research the most rainfall was seen on December of 2019.

Experimental Design and Treatments

Borax ($Na_2B_4O_710H_2O$, 11% B) was used as the boron source and Sakar red lentil variety also was used as plant substance in the research. The experimental design was the Randomized Complete Block Design (RCBD) with four replications. Boron applications consist of control (0 kg B ha^{-1}), for soil, on foliar and soil + foliar applications. Application methods and the amount of boron are given Table 3. Soil applications of boron were applied to soil before planting and mixed to soil. Foliar applications of boron were sprayed to leaves (Table 3).

Table 3. Boron application methods

Boron Applications	Application Time and Methods
Control	Without boron application
Soil	Before planting (0.2 kg da $^{-1}$B) sprayed to soil
Foliar	When plants has 5-6 leaves (0.1 kg da^{-1} B) + at pre-flowering period (0.1 kg da^{-1} B) sprayed to foliar
Soil + Foliar	0.1 kg da^{-1} B to soil + when plants had 5-6 leaves (0.05 kg da^{-1} B) + at pre-flowering period (0.05 kg da^{-1} B) sprayed soil and foliar

After the wheat harvest, which was the primary plant, the experiment area was plowed firstly and cultivated, then prepared for planting with a single pass of a disk-harrow. Planting dates were 30th November and 26th December for 2018 and 2019. Seed amount was 90 kg ha^{-1} for two years. Pneumatic seeder had been used in the planting. Plot sizes were 6 m x 1.6 m and each parcel had of 8 rows. Space between rows was 20 cm and an intra row space was 3-4 cm. Lentil seeds were planted at a 4-5 cm depth.

Nitrogen and phosphorus amounts of soil was identified before planting. Considering the amount of nitrogen and phosphorus in the soil, fertilization amount was completed to 6 kg da^{-1} N, 6 kg da^{-1} P_2O_5 (Oktem et al., 2011). Fertilizers of phosphorous and nitrogen were given with sowing as a banded. 20-20-0 fertilizer was used as phosphorous and nitrogen sources. The banded fertilizers were 50 mm to the side and 50 mm below the seed. Mechanical struggle was made against to weeds.

Lentil harvest was done by hand. Harvest dates were 28th May and 23rd May in 2019 and 2020, respectively. One row from right and left side and 0.5 m from the beginning and also end of the plot were not harvested because of plots' edge effect. After harvesting of plants from the plots, lentils yield was calculated per decar.

Statistical Analyses

Analyses of boron were extracted with hot water and defined by azomethine-H technique and also boron content in soil and seed was determined using same method (Kacar and Inal, 2008). The standard analysis of variance technics was used to analyzed the data and the combined analysis of variance (ANOVA) over two years was performed to obtain the interaction component. Least Significant Difference (LSD) test was made to determine differences between boron levels using the Mstat-C™ statistical software (Steel and Torrie, 1980).

RESULTS AND DISCUSSION

Thousand Seed Weight (g)

According to analysis of the variance, thousand seed weight value was statistically significant ($P \leq 0.01$) at year, boron treatments and year x boron treatment interaction (Table 4). The highest value of the thousand seed weight was observed at soil + foliar boron applications in both years (41.53 and 42.06 g) respectively. The lowest thousand seed weight was found in control applications at all treatment years (Table 4). At the average of two years, the highest thousand seed weight value was seen at soil + foliar boron application (41.80 g), while the lowest one was found at control applications (39.25 g).

Thousand seed weights depend on climatic factors and variety (Saracoglu and Oktem, 2021), but some agronomic activity such as fertilization can effect this value (Oktem and Oktem, 2020). In this research the thousand seed weight was affected positively from boron applications. This result may be due to better starch utilization resulting in increased grain set and grain development which improves grain size. Our findings are consistent with the findings of some researchers. Quddus et al. (2014) clarified that thousand seed weights were found significant variation with different levels of boron application and also the highest thousand seed weight was recorded at 1.5 kg B ha^{-1}. Hakkoymaz et al. (2006) reported that 3.75 kg da^{-1} B level in lentils gave the highest value of thousand seed weight. In a similar research the highest thousand seed weight values were obtained from the application of 0.2 kg B da^{-1} (Oktem et al., 2012).

Biological Yield (g m^{-2})

Biological yield value was found to be statistically significant ($P \leq 0.01$) at year, boron treatments and year x boron treatment interaction. As can be understood from

Table 4 the average values of biological yield ranged from 528.92 g m^{-2} to 582.96 g m^{-2} by boron applications. Biological yield generally depends on year and environmental conditions (Oktem and Oktem, 2019b). The lowest biological yield value was obtained from control while the highest value was found at soil + leaf applications.

Table 4. Thousand seed weight and biological yield values and LSD groups at the boron applications

Boron Applications	Thousand Seed Weight (g)			Biological Yield (g m^{-2})		
	2018/19	2019/20	Average	2018/19	2019/20	Average
Control	38.75 e[†]	39.76 d	39.25 C	456.46 g[†]	601.37 d	528.92 D
Soil	39.33 d	41.53 b	40.43 B	493.12 f	614.45 c	553.78 C
Foliar	40.94 c	42.02 ab	41.48 A	506.20 e	633.62 b	569.91 B
Soil + Foliar	41.53 b	42.06 a	41.80 A	507.90 e	658.01 a	582.96 A
Year Ave.	40.14 B	41.34 A	40.74	490.92 B	626.86 A	558.89
Year		**			**	
Boron (B)		**			**	
Year X B		**			**	

[†]: There is no significant difference between the averages entering the same letter group compared to the LSD test at 0.05 level.

[**]: Significant at the $P \leq 0.01$ probability levels

Some researchers have reported that treatment of B improves physiological processes in plant, resulting in enhanced growth and dry matter production (Asad and Rafique, 2000; Hussain et al., 2002). For this reason, it is thought that boron applications may have increased the biological yield of lentils. Boron application of soil + leaf was the most effective methods on biological yield.

Plant Height (cm)

The effect of different boron application methods on plant height was found to be statistically significant ($P \leq 0.01$). Plant height values varied between 37.51 cm (control) and 42.49 cm (soil + foliar applications) at the average of two years. The maximum values of the plant height were obtained from the addition of soil+ foliar while the lowest values were seen at control applications (Table 5).

Table 5. Plant height, seed yield, protein content values and LSD groups at the boron applications

Boron Applications	Plant Height** (cm)	Seed Yield** (kg da^{-1})	Protein Content ** (%)
Control	37.51 d	161.47 d	27.46 d†
Soil	39.75 c	179.32 c	29.98 c
Foliar	41.10 b	193.93 b	31.83 b
Soil + Foliar	42.49 a	198.57 a	33.29 a
Year Aver.	40.22	183.32	30.64

[**]: Significant at the $P \leq 0.01$ probability levels

Although the value of the plant height is characteristic of the variety, it can affect by the climate and growing conditions like fertilization (Khald and Oktem, 2021). In this study, the value of the plant height was positively affected by boron applications. Some researchers explained that boron applications positively affected the plant height. Dixit and Elamathi (2007) reported that boron application of 2 % significantly improved the plant height. Kaisher et al. (2010) emphasized that application of 0.5 kg da $^{-1}$ B significantly increased plant height. It was explained that significantly higher plant height with foliar application of boron (Praveena et al., 2018).

Seed Yield (kg da^{-1})

Boron application increased seed yield (Figure 1). The highest seed yield was obtained from soil + foliar in the first and second treat years, respectively (194.28 kg ha^{-1} and 201.85 kg ha^{-1}) while the lowest value was found without boron parcels in both experiment years (156.12 kg da^{-1} and 166.33 kg da^{-1}, respectively). Soil + foliar boron application (198.57 kg da^{-1}) gave the highest seed yield value, while the lowest value was obtained from control applications (161.47 kg da^{-1}) at the average of two years (Table 5).

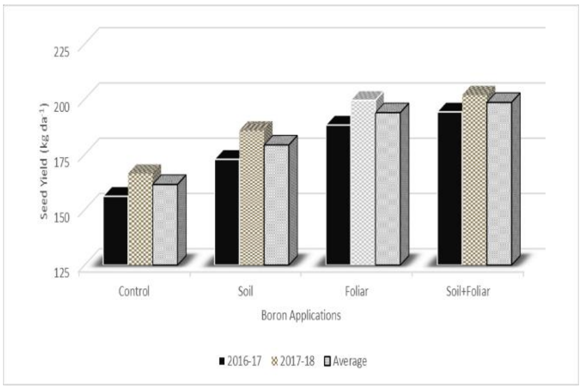

Figure 1. Seed yield values at different boron applications

Lentil cultivation is depending on rainfall in dry areas and seed yield increases in regular rainfall conditions. On the other hand, boron deficiency is often seen when the soil is calcareous and has low organic matter content. In addition, arid climatic conditions play a positive role occurring of boron deficiency. For this reason, when the sufficient amount of boron is applied to this type of soil, the seed yield increases. All these negative factors such as low rainfall, low organic matter and high calcareous are seen at our research area. Our findings are supported by Zengin (2012) who explained that boron deficiency in the soil can be eliminated by boron applications.

All boron application methods had a positive impact on seed yield and the most appropriate method was obtained from soil + foliar boron application. Our study was supported by some researchers' findings. Khurana and Arora (2012) observed that seed yield raised 25% over with the application of boron. It was reported that boron effected seed yield positively in lentil (Srivastava et al., 2000). In a similar study, Quddus et al. (2014) explained that the seed yield increased 13.8% with 1.5 kg ha^{-1} boron application according to control. Sakal et al. (2000) reported that lentils seed yield increased about 75 kg da^{-1} according the control applications with 1.5 kg ha^{-1} B applications in the calcareous soil. It was reported that 21 kg da^{-1} over seed yield obtained than control applications when 0.15 kg da^{-1} boron applied to lentils in calcareous soils (Kumar et al., 2018). It was explained that the highest wheat yield obtained from 2 kg ha^{-1} boron application which was 66% higher than control applications (Halder et al., 2007).

Protein Content of Seed (%)

Boron application methods were significant for protein content of seed at the P≤ 0.01 (Table 5). The highest protein content of seed was found at soil + foliar application (32.63 % and 33.94 % respectively) while the lowest values were seen in the control applications during two years (Figure 2). According to average of two years, the highest protein content of seed value was obtained from soil + foliar applications (33.29%), whereas the lowest one was seen at control applications (27.46%). Protein content of seed was higher at the first year than second year. In the second treatment year, the protein content of the seed may have increased due to climatic factors. During this research rainfall was lower growing season of 2019-2020 than 2018-2019. In addition, all boron applications also affected protein content of seed positively. Oktem and Oktem (2019a) reported that the protein content of the kernel increased with water stress. As the water stress increases, the protein content of the grain also increases (Oktem, 2008a; Oktem, 2008b). Some researchers have reported that boron is essential for some physiological functions of the plant such as carbohydrate and protein metabolism and also it plays an important role in protein synthesis of legumes (Kulhary et al., 2017; Naqib and Jahan, 2017). In addition, Moshiul et al. (2018) observed that agronomic bio fortification with like B increased the seed protein content of legumes. When boron is applied to soils that are deficient in boron content, the seed protein content increases. This may be due to the positive effects of boron on nodule improvement and nitrogen fixation in the roots.

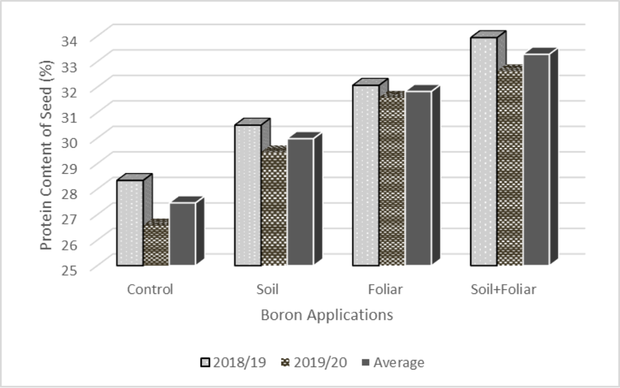

Figure 2. Protein content of seed values at different boron applications

Our results are in a good accord with some researchers' findings. Bayrak et al. (2005) said that boron fertilization to soil increased protein content of seed. Quddus et al. (2014) explained that B concentrations of seed for lentil were significant influenced by B treatment indicating that the B played a favorable role on protein synthesis. And also Singh and Singh (2014) reported that boron application with 1 kg ha^{-1} dosage to soil improved protein content in seeds.

Boron Content of Seed (mg kg^{-1})

According to the analysis of variance the boron content of seed was significant (P≤0.01) at year, boron application methods and boron x year interaction (Table 6). As can be seen from Table 6 that the boron content in the seed was the lowest in the control applications (11.79 and 13.80 mg kg^{-1}) in both years while the highest values were obtained from soil + foliar applications (18.37 mg kg^{-1} and 19.81 mg kg^{-1}). The highest boron content of seed was seen at soil + foliar applications (19.09 mg kg $^{-1}$) at the average of two years, while the lowest value was found at control applications (12.80 mg kg $^{-1}$).Boron content in the seed increased with all boron application methods. It has been reported that boron increased the boron content of lentil seed (Khurana and Arora, 2012). In another research it was stressed that B fertilization to soil increased boron content of lentil seed (Johnson et al., 2005).

Table 6. Boron content values and LSD groups at the boron applications.

Boron Applications	Boron content of seed (mg kg^{-1})		
	2018/19	2019/20	Average
Control	11.79 f	13.80 e	12.80 D
Soil	15.09 d	15.42 d	15.25 C
Leaf	16.47 c	17.76 b	17.12 B
Soil + Foliar	18.37 b	19.81 a	19.09 A
Year Aver.	15.43 B	16.70 A	16.07
Year		**	
Boron (B)		**	
Year X B		**	

†: There is no significant difference between the averages entering the same letter group compared to the LSD test at 0.05 level.
**: Significant at the P≤ 0.01 probability levels

CONCLUSION

All tested characteristics were positively affected by boron application methods. According to an average of two experiment years, the highest seed yield was found at soil + foliar applications (198.57 kg da^{-1}). Also the highest plant height (42.49 cm), thousand seed weight (41.80 g), biological yield (582.96 g/m^2) values were seen in soil + foliar applications. According to average of two years, protein content of seed (33.29%) and boron content of seed (19.09 mg kg^{-1}) values increased with boron applications. All applications of boron gave a higher seed yield than control applications. The most effective boron application method was determined as soil + foliar boron application but other application methods were more effective than control applications.

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COMPARISON OF OIL CONTENT AND FATTY ACIDS COMPOSITION OF SESAME (*Sesamum indicum* L.) VARIETIES GROWN AS MAIN AND DOUBLE CROP IN MEDITERRANEAN ENVIRONMENT IN TURKEY

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ABSTRACT

This study was conducted during 2018 and 2019 growing seasons at the Experimental area of Cukurova University, Faculty of Agriculture in Mediterranean region (Adana, Turkey). The objective of this study was to compare of oil content and fatty acids composition of sesame varieties grown as a main and double crop. The experimental design was a Randomized Complete Block with three replications. Arslanbey, Batem-Aksu, Batem-Uzun, Baydar-2001, Boydak, Cumhuriyet-99, Golmarmara, Hatipoglu, Kepsut-99, Muganli-57, Orhangazi-57, Osmanli-99, Ozberk-82, Sarisu, Tan-99, Tanas and Sari Susam genotypes were used as a plant material in this study. These varieties were registered by the different Research İnstitutes and Faculties in Turkey. Oil content and fatty acids composition (oleic acid, linoleic acid, stearic acid and palmitic acid) of sesame varieties were investigated and compared in main and double crop growing seasons.

The results showed that the considerable variation was found in oil content and fatty acids composition among the sesame varieties grown in main and double crop growing seasons. Oil content, palmitic and oleic acid percentage of the sesame varieties were higher in main crop than in double crop growing season whereas, stearic and linoleic acid percentage of the sesame varieties were found higher in double crop than in main crop growing season.

Keywords: Fatty acid composition, Growing season, Oil content, Sesame

INTRODUCTION

Sesame (*Sesamum indicum* L.) is one of the important oilseed crops widely grown in different part of the world (tropical and subtropical areas) and it has been cultivated mainly in Asia and Africa (Jeon et al., 2012). Sesame seeds contain approximately 35-60% oil, 18-25% protein and 16-18% carbohydrate (Uzun et al., 2008). İt is a good source of vitamin E and B1 and minerals such as calcium, phosphorous, potassium, magnesium, iron, copper, zinc and manganese. The seed contains all essential amino acids and fatty acids (Alyemeni et al., 2011). For these reason, sesame seed plays an important role in human health due to its high oil content and proper quality (low cholesterol and have some antioxidants), as well as a good protein content (Gharby et al., 2017). The sesame seed is commonly known as the “Queen of oil seeds”, probably due to its high nutritional and therapeutic value, and resistance to oxidation and rancidity (Sukumar et al., 2008).

Sesame seeds have high nutritional quality due to its high oil and protein content. Sesame seed is mostly used for edible purposes such as oil and confectionery. It is also used for various food products such as tahini, halva, rolls, crackers, cakes, cookies, buns, chips, and margarine, cosmetics, perfumery, soaps, paints, insecticides and pharmaceutical products (Ogbonna and Umar-Shaaba, 2011).

The sesame oil shows high degrees of stability and resistant to oxidative rancidity due to the presence of a number of endogenous antioxidants such as sesamin, sesamolin and sesamol (Alpaslan et al., 2001). Bedigian (2004) and Sankar et al. (2004) indicated that the sesame oil contains phytosterols associated with reduced blood cholesterol levels and hypertension in humans and reduces the incidences of certain cancers. The antioxidant compound called lignan such as sesamin in sesame oil, inhibits human lymphoid leukemia cells by inducing apoptosis (Miyahara et al., 2000). Sesamin and tocopherols have shown benefits in the prevention of hypertension and stroke (Noguchi et al., 2004).

The quality of sesame oil is depending on the fatty acid composition. Fatty acid composition in sesame seeds consists of unsaturated fatty acids such as oleic (35.9-42.3%) and linoleic (41.5-47.9%) acids and saturated fatty acids, mainly palmitic (7.9-12%) and stearic acids (4.8-6.1%) (Hwang, 2005). In sesame oil, oleic and linoleic acids are the predominant fatty acids and constitute more than 80% of the total fatty acid content (Teres et al., 2008). The high level of monounsaturated (oleic acid) and polyunsaturated (linoleic acid) fatty acids increases the quality of the oil for human consumption (Jones et al., 2008 and Mondal et al., 2010). Fatty acids are important diet for healthy living. They have several functions in the body including helping in transportation of oxygen in the bloodstream, aiding cell membranes development and function, keeping the skin healthy, preventing early aging, and more importantly, preventing cholesterols build up in the articles (Mzimbiri et al., 2014).

Alpaslan et al. (2001), Mohammed and Mekonnen (2010), Shilpi et al. (2014) and El Harfi et al. (2019) indicated that the oil content and fatty acid composition in oilseed crops is affected by genotype, location, environmental conditions (temperature, photoperiod and moisture content), growing conditions, planting date, plant density, fertilization and the interaction of these factors. Uzun et al. (2002) reported that oil content of sesame can be varied by climate conditions and it can decrease by delaying planting time. In additionally, seed oil content may vary considerably between genotype and seasons, and oil percentage tends to rise with increasing length of photoperiod (Weiss, 2000).

Were et al. (2006), Mondal et al. (2010), Yol et al. (2015), Kurt (2018) and El-Harfi et al. (2019) reported that the oleic and linoleic acids varied between 35.67-53.96% and 30.40-51.65% respectively in oils of different sesame varieties. El-Harfi et al. (2019) indicated that the total unsaturated fatty acids percentage varied between 79.5-82.34% in sesame oils.

Weiss (1971) indicated that the air temperature effects on growth and development of sesame plant and seed quality. The oil content and quality of seed decreased at the lower temperature, specially sesamin and sesamolin content. High temperature during the growing period increased oil content of sesame seed.

Sekhon and Bhatia (1972) and Gupta et al. (1998) reported that sowing date also influenced fatty acid composition of sesame by decreasing linoleic acid and increasing oleic and stearic acid content as sowing was delayed. Not only these conditions affect fatty acid composition but also genotypic factors play an important role in the process, resulting in the fact that each genotype shows different fatty acid composition. Gupta et al. (1998) indicated that he sowing date is also known to influence fatty acid composition of sesame by decreasing linoleic acid and increasing oleic acid contents as the sowing gets delayed. Ali et al. (2015) reported that the oleic acid percentage was increased from 38.2 to 42.6% and linoleic acid percentage was decreased from 40.0 to 37.0% with delaying the sowing date from 20th June to 30th July.

Kurt et al. (2016) reported that the palmitic and stearic acids are the predominant saturated fatty acids in sesame oil. Unal and Yalcin (2008), Uzun et al. (2008), Savant and Kothekar (2011), Jeon et al. (2012), Yol et al. (2015), Kurt (2018), Thakur et al. (2018) and El Harfi et al. (2019) indicated that the palmitic and stearic acids percentage in sesame genotypes varied between 7.42-12.00% and 2.10-8.90% respectively in different part of the world.

In according to the “Turkish Food Codex” given limit values for sesame seed, palmitic, stearic, oleic and linoleic acids must be changed 7.9-12.0%, 4.5-6.7%, 34.4-45.5% and 36.9-47.9%, respectively (Anonymous, 2009). Uzun et al. (2002) reported that in general, sesame oil contains about 47% oleic acid 39% linoleic acid 9.0% palmitic acid and 4.1% stearic acid.

The Mediterranean climate has suitable temperature regimes for growth and development of sesame plants. Sesame is an important annual oil crop in Turkey. Environmental conditions are suitable for sesame growing as a main and double crop in Mediterranean region in Turkey. The environmental factor effects on growth and development of sesame plants, especially day and night temperature and photoperiod are the most effective environmental factors for the seed yield and seed quality. İt is important to know the content of sesame fatty acids for better quality and safety of its product. Fatty acid composition of sesame oils is not constant. The oil content and fatty acid composition of sesame oil varies depending on environmental factors (temperature and photoperiod), varieties, sowing dates and cultural factors. The objective of the study was to comparing oil content and fatty acids composition of sesame varieties grown as a main and double crop in Mediterranean environment in Turkey.

MATERIALS AND METHODS

Material

Field experiments was conducted during 2018 and 2019 growing seasons at Experimental Farm of Agricultural Faculty, Cukurova University (Southern Turkey, 36^o59^l N, 35^o18^1 E and 23 m above sea level) as a main and double crop in Mediterranean region (Adana, Turkey). 16 different sesame cultivars such as Arslanbey, Batem-Aksu, Batem-Uzun, Baydar-2001, Boydak, Cumhuriyet-99, Golmarmara, Hatipoglu, Kepsut-99, Muganli-57, Orhangazi-57, Osmanli-99, Ozberk-82, Sarisu, Tan-99, Tanas and one local variety such as Sari Susam (Sari Susam was grown commonly in the Cukurova region) were used as a plant material in this study. These varieties were registered by the different Research İnstitutes and Faculties in Turkey. All of the varieties have indeterminate growth type and capsule dehiscence at ripening is completely shattering. Some of the seed and capsule characteristics of the sesame varieties were given at Table 1.

Table 1. Some seed and capsule characteristics of sesame varieties

Varieties	Seed Color	Capsule Type	Capsule number per leaf axil
Arslanbey	Dark brown	Small, long rectangle, bicarpellate	Tricapsulle
Batem-Aksu	Brown	Medium, long rectangle, bicarpellate	Monocapsulle
Batem-Uzun	Light brown	Wide, long rectangle, bicarpellate	Monocapsulle
Baydar-2001	Light brown	Wide, long rectangle, bicarpellate	Monocapsulle
Boydak	Brown	Wide, long rectangle, bicarpellate	Monocapsulle
Cumhuriyet-99	White	Small, long rectangle, bicarpellate	Monocapsulle
Golmarmara	White	Medium, long rectangle, bicarpellate	Monocapsulle
Hatipoglu	Dark brown	Small, long rectangle, bicarpellate	Monocapsulle
Kepsut-99	White	Small, long rectangle, bicarpellate	Monocapsulle
Muganli-57	Light brown	Wide, long rectangle, bicarpellate	Monocapsulle
Orhangazi-99	White	Medium, long rectangle, bicarpellate	Monocapsulle
Osmanli-99	White	Medium, long rectangle, bicarpellate	Monocapsulle
Ozberk-82	Light brown	Small, long rectangle, bicarpellate	Monocapsulle
Sari Susam	Yellow	Wide, long rectangle, bicarpellate	Monocapsulle
Sarisu	Yellow	Wide, long rectangle, bicarpellate	Monocapsulle
Tan-99	White	Medium, long rectangle, bicarpellate	Monocapsulle
Tanas	Yellow	Wide, long rectangle, bicarpellate	Monocapsulle

The soil texture was clay loam. The soil tests indicated that pH of 7.36-7.40 with high concentrations of K_2O (748-730 kg ha^{-1}) and low concentrations of P_2O_5 (23-25 kg ha^{-1}). İn addition, the organic matter and nitrogen content of the soil were very low. The lime content was varied between 28.4-27.3% (2018 and 2019) in the soil. The soil is suitable for the sesame growing.

This study was conducted in Adana province (Mediterranean environment) in Turkey and in this region, winters are mild and rainy, whereas summers are dry and warm, which is a typical of a Mediterranean climate. The average monthly air temperature during the research period (March-November) was varied between 16.8 and 29.7 ^{o}C in 2018, whereas it was 13.8 and 29.6 ^{o}C in 2019. The average air temperature was the higher during the research period in both years than long term average temperature. The total rainfall was 212.4 mm and 243.8 mm during the growing period in 2018 and 2019, respectively. The average relative humidity was ranged from 58.6% to 71.6% in 2018 and 57.6% to 69.3% in 2019. The differences between the years and long term for the climate data were not found very significant

Method

The field trial was arranged in a Randomized Complete Block Design (RCBD) with three replications. 200 kg ha^{-1} of Di-ammonium phosphate (364 kgha^{-1} N, 92 kg ha^{-1} P_2O_5) fertilizer was applied and incorporated to soil before planting. Urea (46%N) at the rates of 200 kg ha^{-1} was applied before first (beginning of flowering) irrigation in each two growing seasons. Plot size was 2.8 x 5.0 m (14.0 m^2) and spacing between row and plant was 70 and 10 cm, respectively. The seeds were sown by hand at the 6th May as a main crop and 22th June as a double crop in both years (Reserve seeds were used at the second years). During the growing period, recommended pesticides and fungicides were applied at proper time intervals to control insects and diseases. Furrow irrigation was applied at 15 days intervals to maintained soil moisture close to field capacity. The remaining cultural practices such as inter row cultivation and weed control were applied during the growing period. The plants were harvested by hand at the 6th September and 10th September for main crop and at the 22th October and 27th October for the double cropping in 2018 and 2019, respectively. After harvesting, the plants dried almost 15 days in the field and then they threshed by hand.

Data collection and analysis

The data belonging to oil content and oil quality characteristics such as fatty acids composition were recorded in each two growing seasons.

Determination of oil content; oil was extracted from sesame seeds using "Soxhlet", and oil percentage was estimated according to Association of Official Analytical Chemist (AOAC, 2010).

Determination of fatty acids composition: Fatty acid methyl esters were prepared according to AOAC (2010), method Ce 2-66 and analyzed with HP 6890 Series II Gas Chromatograph (GC) (Hewlett-Packard Company, Wilmington, DE, USA) equipped with a flame ionization detector and auto sampler.

The data were statistically analyzed by using JMP 8.1.0 package program with split plot design. The Least Significant Differences (LSD) test was used to compare the means of treatments at $p<0.05$ level (Caliskan et al., 2008; Steel and Torrie, 1980).

RESULTS AND DISCUSSION

According to two-year results, the data were statistical analyzed using repeated years split plot design by using variety and growing seasons factors. The variance analysis of the findings obtained from the study was shown in Table 2. İt can be seen in Table 2, only variety, season and season x variety factors were statistical significant for all the investigated characters, except oil content for the season.

Table 2. The F-values of the variance components of the investigated characteristics of sesame varieties grown in the field trial combined over two-years

Source of Variations	**df**	**Oil Content**	**Palmitic Acid**	**Stearic Acid**	**Oleic Acid**	**Linoleic Acid**
Year	1	ns	ns	ns	ns	ns
Season	1	ns	**	**	**	**
Year*Season	1	ns	ns	ns	ns	ns
Variety	16	**	**	**	**	**
Year*Variety	16	ns	ns	ns	ns	ns
Season*Variety	16	**	**	**	**	**
Year*Seasons*Variety	16	ns	ns	ns	ns	ns

df: Degree of freedom, ** $p < 0.01$, ns: not significant, O/L: Oleic acid/Linoleic Acid, İV: İodine value

Oil content

The data belonging to oil content of the sesame varieties at different growing seasons were shown in Table 3. According to two-year averages, the oil content of the sesame varieties varied between 42.9-51.1% in main crop and 43.7-50.8% in double crop growing season. The differences between the varieties for the oil content were statistically significant in both two growing seasons. The oil percentage was the highest in Cumhuriyet-99 (51.1% and 50.8%) and the lowest in Batem-Aksu (42.9% and 43.7%) among the sesame varieties in each two growing seasons. The oil content of the Boydak and Cumhuriyet-99 varieties were higher than Sari Susam (local variety) among the tested sesame varieties in each growing seasons. These differences between the genotypes for the oil content may have originated due to different genetic backgrounds and growing conditions (Baydar et al., 1999 and Uzun et al., 2002).

Table 3. The oil content, Palmitic (C16:0) and Stearic (C18:0) acids percentages of sesame varieties as average of two years (2018 and 2019)

Varieties	**Oil content** (%)		**Palmitic acid** (%)		**Stearic acid** (%)	
	Main crop	Double crop	Main crop	Double crop	Main crop	Double crop
Arslanbey	49.8	49.6	9.74	9.66	3.65	4.08
Batem-Aksu	42.9	43.7	9.52	9.13	2.70	2.89
Batem-Uzun	48.1	47.0	10.04	9.62	2.48	2.56
Baydar-2001	49.1	48.3	9.46	9.35	2.28	2.72
Boydak	50.3	50.2	9.78	9.43	3.23	4.02
Cumhuriyet-99	51.1	50.8	10.88	9.65	2.86	3.95
Golmarmara	46.5	46.0	10.03	9.68	2.74	3.12
Hatipoglu	48.9	48.4	9.62	9.51	3.06	3.73
Kepsut-99	48.9	48.4	9.66	9.59	2.87	3.65
Muganli-57	49.1	48.7	9.49	9.40	1.79	2.50
Orhangazi-99	49.2	48.4	9.41	9.41	2.08	2.59
Osmanli-99	50.0	48.1	9.89	9.89	2.72	3.60
Ozberk-82	49.6	48.3	10.08	9.39	2.67	3.00
Sari Susam*	50.1	50.1	10.03	9.61	2.58	3.20
Sarisu	45.8	45.3	10.12	9.77	3.03	3.77
Tan-99	49.0	48.5	9.73	9.60	1.93	2.59
Tanas	48.8	46.3	10.31	9.48	2.66	2.74
Average	48.6	48.0	9.87	9.44	2.67	3.22
LSD (%5 $_{\text{Variety-A}}$)	0.233	0.295	0.035	0.025	0.020	0.056
LSD (%5 $_{\text{Seasons-B}}$)	NS		0.023		0.065	
LSD (%5$_{\text{A x B}}$)	1.568		0.235		0.226	

*Control variety (Local variety)

According to a two year average, the average oil content of the sesame varieties was 48.6% in main crop and 48.0% in double crop growing seasons (Table 3). The average oil content of the varieties was found higher in main crop (early sowing date) than in late sowing date (double crop growing seasons). But, the differences between the growing seasons for the oil content were not significant.

Early planting dates may not be feasible in some seasons or under some soil conditions. Early planting resulted in the apparently unfavorable combination of cool vegetative-stage temperatures and warm seed-fill-stage temperatures. Furthermore, genotypic differences for oil content have been reported with oil content displaying more genetic variability among cultivars than the other oil traits (Kane et al., 1997; Baydar et al., 1999 and Ashri, 2007).

Palmitic and Stearic acids

According to a two year average, palmitic acid percentage of the sesame varieties varied between 9.41-10.88% in main crop and 9.13-9.89% in double crop growing season. The differences between the sesame varieties for the palmitic acid percentage were statistically significant in both growing seasons. The highest palmitic acid percentage was obtained from Cumhuriyet-99 (10.88%) in main crop and from Osmanli-99 (9.89%) variety in double crop growing season (Table 3). The differences between the varieties must be due to difference in genetic constitution of sesame cultivars.

The average palmitic acid percentage of the sesame varieties were 9.87% in main crop and 9.44% in double crop growing season. The differences between the growing seasons for the palmitic acid percentage were significant. Palmitic acid percentage was decreased from 9.87% to 9.44% with the sowing date delayed from 6th May (main crop) to 22th June (double crop). The average palmitic acid percentage of the sesame varieties was found higher in main crop than in double crop growing season. İnteraction between the variety and growing season was found significant and the highest palmitic acid percentage was obtained from Cumhuriyet-99 (10.88%) in main crop growing season (Figure 1.).

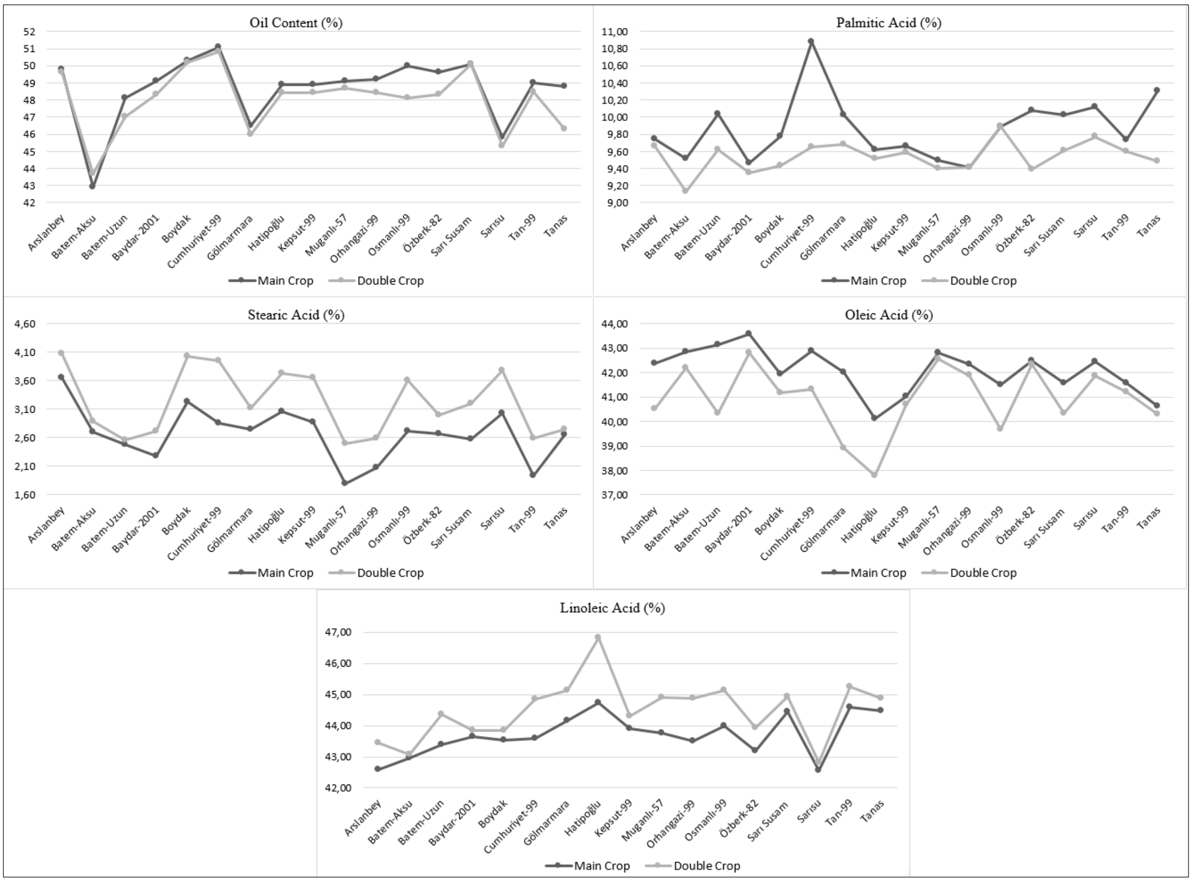

Figure 1. The comparison of average oil content, palmitic, stearic, oleic and linoleic acid of sesame varieties grown in main and double crop growing seasons

According to a two year average, stearic acid percentage of the sesame varieties varied between 1.79-3.65% in main crop and 2.50-4.08% in double crop growing season. The differences between the sesame varieties for the stearic acid percentage were statistically significant in both growing seasons. The differences between the varieties must be due to difference in genetic constitution of sesame cultivars. The highest stearic acid percentage was obtained from Arslanbey sesame variety (3.65% and 4.08%) in main and double crop growing season. The average stearic acid percentage of the sesame varieties were 2.67% in main crop and 3.22% in double crop growing season. The differences between the growing seasons for the stearic acid percentage were significant. Stearic acid percentage was increased from 2.67 to 3.22% with the sowing date delayed from 6th May (main crop) to 22th June (double crop). İnteraction between the variety and growing season was found significant and the highest stearic acid percentage was obtained from Arslanbey in both growing seasons (Table 3). These differences between the genotypes for the Palmitic and Stearic acids content may have originated due to different genetic backgrounds and growing conditions. The fatty acids composition in oilseed crops is affected by genotype, location, environmental conditions (temperature, photoperiod and moisture content), growing conditions, planting date, plant density, fertilization and the interaction of these factors (El Harfi et al., 2019). Similar results were reported by other researchers (Gupta et al., 1998; Uzun et al., 2008; Jeon et al., 2012; Ali et al., 2015; Yol et al., 2015; Kurt, 2016; Kurt, 2018; Vurarak et al., 2018 and El Harfi et al., 2019).

Oleic and Linoleic Acids

The data belonging to oleic and linoleic acids percentage of sesame varieties were shown in Table 4. According to a two year average, the oleic acid percentage of sesame varieties varied from 40.11 to 43.46% in main crop and from 37.78 to 42.81% in double crop growing seasons. The differences between the sesame varieties were statistically significant. The differences between the varieties must be due to difference in genetic constitution of sesame cultivars. The oleic acid percentage was the highest in Baydar-2001 (43.56% and 42.81%) and the lowest in Hatipoglu (40.11% and 37.78%) among the sesame varieties in each two growing seasons.

Table 4. The effect of growing season on oleic (C18:1) and linoleic (C18:2) acids percentage of sesame varieties in two years average (2018 and 2019)

Varieties	**Oleic acid** (%)		**Linoleic acid** (%)	
	Main Crop	Double Crop	Main Crop	Double Crop
Arslanbey	42.36	40.52	42.58	43.44
Batem-Aksu	42.85	42.20	42.97	43.07
Batem-Uzun	43.15	40.33	43.39	44.35
Baydar-2001	43.56	42.81	43.64	43.85
Boydak	41.93	41.16	43.54	43.85
Cumhuriyet-99	42.89	41.31	43.59	44.86
Golmarmara	41.99	38.90	44.16	45.14
Hatipoglu	40.11	37.78	44.73	46.83
Kepsut-99	41.03	40.70	43.89	44.29
Muganli-57	42.80	42.57	43.75	44.90
Orhangazi-99	42.35	41.88	43.50	44.88
Osmanli-99	41.49	39.66	43.98	45.14
Ozberk-82	42.49	42.35	43.19	43.93
Sari Susam*	41.56	40.33	44.46	44.92
Sarisu	42.45	41.87	42.57	42.82
Tan-99	41.57	41.19	44.59	45.26
Tanas	40.62	40.28	44.47	44.88
Average	42.07	40.99	43.71	44.49
LSD (%5 $_{\text{Variety-A}}$)	0.139	0.157	0.070	0.098
LSD (%5 $_{\text{Seasons-B}}$)	0.583		0.168	
LSD (%5$_{\text{A x B}}$)	1.246		0.813	

*Control variety (Local variety)

As it can be seen in Table 4, while the linoleic acid percentage of the sesame varieties were ranging between 42.57 and 44.73% in main crop growing season and it was ranged between 42.82 and 46.83% in double crop growing season. The differences between the varieties must be due to difference in genetic constitution of sesame cultivars. According to a two year average data, the differences between the varieties for the linoleic acid percentage were statistically significant. Linoleic acid percentage was the highest in Hatipoglu (44.73% and 46.83%) among the sesame varieties in both growing seasons. The lowest linoleic acid percentage was obtained from Sarisu (42.57% and 42.82%) in each two growing seasons. In according to the “Turkish Food Codex” given limit values for sesame seed oleic and linoleic acids must be changed 34.4-45.5% and 36.9-47.9%, respectively (Anonymous, 2009). As seen in Table 4, oleic and linoleic acids percentage of the sesame varieties grown in Turkey were found in the limits specified by “Turkish Food Codex” declaration in this study.

The average oleic acid percentage of the sesame varieties were 42.07% in main crop and 40.99% in double crop growing season. The differences between the growing seasons for the oleic acid percentage were significant. Oleic acid percentage was decreased from 42.07% to 40.99% with the sowing date delaying from 6th May (main crop) to 22th June (double crop). Comparing the main and double crop for the oleic acid percentage of sesame varieties, it was higher in main crop than in double crop growing season. İnteraction between the variety and growing season for the oleic acid was found significant and the highest oleic acid percentage was obtained from Baydar-2001 (43.56%) in main crop growing season (Figure 1.).

The average linoleic acid percentage of the sesame varieties were 43.71% in main crop and 44.49% in double crop growing season. The differences between the growing seasons for the linoleic acid percentage were significant. Linoleic acid percentage was increased from 43.71% to 44.49% with the sowing date delayed from 6th May (main crop) to 22th June (double crop). Comparing the main and double crop for the linoleic acid percentage of sesame varieties, it was lower in main crop than in double crop growing season (Table 4). İnteraction between the variety and growing season for the linoleic acid was found significant and the highest linoleic acid percentage was obtained from Hatipoglu (46.83%) in double crop growing season. According to a two year average data, linoleic acid percentage of the sesame varieties was found higher than oleic acid percentage in both growing seasons in this study (Table 4).

It has demonstrated that unsaturated fatty acids are influenced by the environmental conditions, mainly air temperature during seed filling and oil biosynthesis. Thus, under low temperature conditions, there is an increase of unsaturated acid of seed oil, which leads to a higher proportion of linoleic and linolenic acids. Contrarily, under high temperature conditions, there is a low proportion of these acids and a high proportion of oleic acid in seed oil. İn addition to that, amplitude of maximum and minimum temperature as well as duration of plant exposure these temperature during seed filling effect significantly fatty acids compositions (Deng and Scarth, 1998). The climatic conditions are the main factors influencing the fatty acids composition such as oleic and linoleic acids, especially the variations in temperatures during seed filling stage (Andersen and Gorbet, 2002). The genetic components play an important role in the process, resulting in the fact that each genotype shows different fatty acid composition (Sekhon and Bhatia, 1972). Vurarak et al. (2018) reported that the oleic and linoleic acids percentage was varied between 41.09% and 40.38% in main crop and between 40.32% and 40.49%, respectively in double crop growing seasons. They also indicated that the oleic acid percentage was higher in main crop than in double crop, contrarily the linoleic acid percentage was higher in double crop than in main crop growing season.

As a result, the differences between the sesame varieties for the oleic and linoleic acids percentage were found statistically significant in both growing seasons. As it can be seen in Table 4, while the sowing date was delaying, the oleic acid percentage was decreased. On the other hand the linoleic acid percentage was increased. These results were in agreement with the findings of Were et al. (2006), Arslan et al. (2007), Uzun et al. (2007), Unal and Yalcin (2008), Mondal et al. (2010), Yol et al. (2015), Ali et al. (2015), Kurt (2018), Vurarak et al. (2018) and El-Harfi et al. (2019).

CONCLUSIONS

The results showed that the sesame varieties were registered by the different Research İnstitutes and Faculties in Turkey can be grown successfully as a main and double crop in Mediterranean region in Turkey. İn this study, considerable variation was found in oil content and fatty acids composition among the sesame varieties grown in main and double crop growing seasons. Oil content, palmitic acid and oleic acid of the sesame varieties were higher in main crop than in double crop growing season. On the other hand, stearic acid and linoleic acid of the sesame varieties were found higher in double crop than in main crop growing season.

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INVESTIGATION OF THE EFFECT OF PGPR ON YIELD AND SOME YIELD COMPONENTS IN WINTER WHEAT (*Triticum aestivum* L.)

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ABSTRACT

The plant growth-promoting rhizobacteria (PGPR) that live actively in plant roots and rhizosphere and support plant growth has gained widespread importance in agriculture. This study was carried out to obtain and identify the PGPR isolates from wheat soil and determine their ability and capacity on plant growth and yield. So, they were obtained from soil, and they were identified as *Bacillus* spp. (*B. simplex* and *B. pumilus*) by biochemical tests and MALDI-TOF MS (Matriks assisted lazer desorption ionization time of flight massspectrometry). After the wheat seeds (Flamura-85) were treated for PGPR, the field experiment was conducted with inoculated and non-inoculated seeds at the area of the Field Crops Department, Tekirdag Namık Kemal University in 2016-2018. The experiment was arranged in a split-plot design with three replicates for each treatment. In the experiment, some parameters such as plant height (PH), spike length (SL), number of grain per spike (NGPS), grain weight per spike (GWPS), and grain yield (GY) were evaluated and compared between treatments. The study has shown that PGPR treatments support plant growth and significantly increase yield between 9.6% and 29.29%. Especially, W3 and W4 strains (*B. simplex*) were showed a significant effect on the GY. According to the results, we can mention that using the PGPR promotes wheat growth and lead to increasing yield in the wheat. The use of PGPR can give promising results for sustainable and eco-friendly agricultural practices.

Keywords: PGPR, plant growth, seed treatment, winter wheat (*Triticum aestivum* l.), yield

INTRODUCTION

The plant growth-promoting rhizobacteria (PGPR) is the soil bacteria and one of the most important and agronomically useful soil microbiota (Kloepper and Schroth,1978; Bhattacharya and Jha, 2012). It can help plant growth indirectly by reducing plant pathogens, or directly by supporting the uptake of nutrients such as P, N, K also they can affect the biomass of the plants (Cakmakci et al., 2006; Ibiene et al., 2012; Khan et al., 2013). The PGPR can colonize and grow fastly onto the surface of the seed or the root, it can support plant growth, the germination rate of the seed, improve transplant emergence, and respond to stress conditions. For all those, it is an excellent alternative and tool to improve the agricultural system eco-friendly. Increasing crop production, the PGPR is given a good opportunity to prevent the use of a large number of chemical fertilizers and pesticides, which are generally overused in soil (Kumar et al., 2017). Many studies have demonstrated the abilities of plant growth-promoting microorganisms to increase plant nutritional status and reduce the use of pesticides (Meena et al., 2017; Aloo et al., 2019).

The PGPR can reduce the input of chemical fertilizers and pesticides without reducing yield. So, nowadays it is used as a popular biopesticide, photostimulation, and biofertilizer for sustainable agriculture (Rodrigurez et al., 2006; Santos et al., 2020). The PGPR can be applied to some different parts of the plant such as seed, root, and foliar. Even though the most common application of PGPR is known as a seed treatment, the root and foliar treatments also are used (Podile and Kishore, 2006). The application of PGPR supported most of the plants such as canola, grasses, maize, rice, and wheat. Recently studies indicated that PGPR is more effective on wheat yield. Some researchers obtained that PGPR providing P to wheat plants over the growing season, P-solubilizing rhizosphere bacteria at associated with wheat growth stages as vegetative biomass production and generative biomass production (Khan et al., 2013; Schadler et al., 2019; Asık and Arıoglu, 2020). All those results and documents try to understand the importance of PGPR for sustainable agriculture. The PGPR which is a productive and new alternative in the agricultural system also contains strains from genera such as *Pseudomonas*, *Serratia*, *Azospirillium*, *Azotobacter*, *Bacillus*, *Burkholderia*, *Enterobacter*, *Rhizobium*, *Erwinia*, *Acinetobacter*, *Alcaligenes*, *Arthrobacter*, and *Flavobacterium* (Bashan and de-Bashan, 2005; Esitken et al., 2010) well recognized. However, *Bacillus* is an important genus within the PGPR group.

More studies referred to *Bacillus* spp. produce endospores that can tolerate extremes of the condition such as temperature, pH, and exposure to pesticides and fertilizers. Also, *Bacillus* spp. have been shown to improve root growth and morphology total root area in crops (Backman et al., 1997; Adesemoye and Kloepper, 2009; Duca et al., 2014; Shen et al., 2016; Nguyen et al., 2019).

This study aimed to collect and identify the PGPR strains in the wheat fields and also obtain their ability and capacity on plant growth and yield under the field conditions.

MATERIALS AND METHODS

Isolation of the PGPR from soil

Soil materials were collected from Tekirdag provinces during the 2015 and 2016 growing seasons. Nutrient agar (NA) media was used for isolation at $10^{-5} – 10^{-6}$ serial dilutions. The agar plates were incubated at 28 °C for 48 h. Each colony was used as an isolate in NA plates and followed by purification on the new NA plate with a repeated plating method (Schmidt and Belser, 1982). A total of 53 candidate PGPR isolates were isolated and used for further study.

Selection of the PGPR

Some tests such as hypersensitivity reaction test, soft rot test, and growth 37 °C were performed to select the candidate of PGPR isolates.

A hypersensitivity reaction test of the candidate PGPR isolates was carried out by inoculating tobacco leaves, using a highly concentrated bacterial suspension (~10^8 cfu/ml). The effectiveness of the bacterial isolates was evaluated by the absence of disease symptoms and hypersensitivity reaction. Candidate PGPR isolates were further checked for potato (*Solanum tuberosum* L.) soft rot test on potato slices. A loopful bacterial colony was taken from pure fresh culture and spread on surface-sterilized potato slices and incubated to obtain the occurrence of any soft rot on potato tissues. The existence of soft rot on potato slices showed the positive effect of pectolytic activities of the tested PGPR isolates. Lastly, the growth at 37 °C on the NA test was done. So human pathogenic isolates were eliminated with this test.

Identification of PGPR

Physiological and biochemical characteristics of the candidate PGPR isolates were determined with Gram reaction, oxidase, and production fluorescent pigmentation (Lelliot and Stead, 1987). After that, all isolates were analyzed with MALDI-TOF MS to be identified (Pavlovic et al., 2012). So, the raw spectra of the unknown PGPR isolates were used for pattern matching against the reference spectra of the database. The results of the pattern-matching process were expressed as proposed by the manufacturer, with log (scores) values ranging from 0 (no similarity) to 3 (absolute identity) (Carolis et al., 2012; Ziegler et al., 2012; Uysal et al., 2019)

Seed inoculation

The Flamura-85 wheat cultivar was used as experimental material in this study. Seeds of the cultivar were done surface-sterilization using 0.1% NaClO for 2 min and rinsed five times with sterilized water. Candidate PGPR isolates were grown on NA for experiments. A single colony from each isolate was transferred into a 50 ml flask, containing nutrient broth (NB) and kept in flasks overnight on a rotating shaker at 200 rpm. Bacteria grown on NB were diluted with sterile NB. to a final concentration was arranged as 10^8 CFU (colony forming unit) ml^{-1} in a spectrophotometer. For treatments, wheat seeds were inoculated with the candidate PGPR suspension of 10^8 CFU ml^{-1} along for 30 min before sowing. For control, non-inoculated seeds were used.

Field experiment

The experimental site and growing conditions: This study was carried out in the experimental area of Field Crops Department of Agricultural Faculty of Tekirdag Namik Kemal University, Tekirdag, Turkey during the 2016-2017 and 2017-2018 wheat growing seasons. Tekirdag district locates at latitude 40° 36’-40° 31’ and longitude 26° 43’-28° 08’and altitude is 10 m. The climate data during the 2016-2017 and 2017-2018 wheat growing seasons and long-term average were given in Table 1. In the first year of the study, the total precipitation and the average temperatures were 360.3 mm and 9.8 °C, respectively. (Table 1). In the second year of the study, the total precipitation and the average temperatures were 612.7 mm and 12.1 °C respectively (Table 1).

Table 1. The monthly climatical data measured during the winter wheat-growing seasons in 2016-2017 and 2017-2018*

Months	Precipitation (mm)			Average temperature (°C)		
	2016-17	2017-18	Long term	2016-17	2017-18	Long term
November	43.1	85.2	55.2	3.8	11.7	11.4
December	43.1	94.8	86.2	3.8	9.5	7.2
January	107.0	76.5	69.9	1.9	6.6	4.4
February	38.8	95.3	54.7	6.5	7.2	5.3
March	32.2	63.7	55.6	9.1	10.2	6.8
April	51.6	10.6	42.9	11.2	14.0	11.5
May	16.7	114.2	37.6	16.8	15.2	16.6
June	36.8	75.4	37.8	22.0	22.4	22.5
Total	369.3	612.7	439.9	-	-	-
Average	-	-	-	9.38	12.1	28.9

*Source: Tekirdag meteorology station

Under field conditions, inoculated and non-inoculated wheat seeds were sown in Tekirdag on November 10, 2016, for first-year experiments and November 15, 2017, for second-year experiments with a density of 500 grains m^{-2}. The plot size was 5.10 m^{2} (5 m x 1.02 m). The experiments were arranged in randomized complete block design with 3 replicates. For two years the experiment was fertilized at different times along season like sowing (20.20.0 composed fertilizer), tillering (urea, %46 N), and stem elongation (calcium ammonium nitrate %26N). The experiment was harvested in early July and yield and yield components were studied.

Statistical analysis

The JMP version 5.0 package statistical software was used for all data involving calculations and the comparison of each isolate for all measurements. The data were statistically analyzed using ANOVA. Duncan's multiple range test at a probability level of 0.05 was used to separate the means.

RESULTS

Isolation and selection of the candidate PGPR strains

A total of 53 candidate PGPR were isolated from wheat fields in Tekirdag. All isolates were subjected to growth at 37°C, tobacco (*Nicotiana tabacum*) HR, and potato rotting. Following the tests, none of the strains grown at 37 C, 46 isolates were found positive for tobacco HR. Any isolates weren't shown pectolytic activity on potato slices (Table 2). So, seven-candidate PGPR strains were retained for using further study.

Table 2. Identification of candidate PGPR strains

Strain code	Bacterial species	Gram reaction	Florescent	Oxidase	HR	Pectolytic activity	Growth at 37 °C
W1	*B. pumilus*	+	-	-	-	-	-
W2	*B. simplex*	+	-	+	-	-	-
W3	*B. simplex*	+	-	+	-	-	-
W4	*B. simplex*	+	-	+	-	-	-
W5	*B. simplex*	+	-	+	-	-	-
W6	*B. simplex*	+	-	+	-	-	-
W7	*B. pumilis*	+	-	-	-	-	-

W1: *B. pumilus*, W2: *B. simplex*, W3: *B. simplex*, W4: *B. simplex*, W5: *B. simplex*, W6: *B. simplex*, W7: *B. pumilus* They are code or name of bacterial strain that are identified as *B. pumilus* and *B. simplex*

Identification of candidate PGPR strains

According to the biochemical characterization of strains were identified *Bacillus* genera including *B. simplex* and *B. pumilus* (Table 2).

Two out of seven strains were selected according to their biochemical test results. Then these two strains were analyzed by MALDI-TOF MS. The strains were similarly identified like biochemical test results as *B. simplex* and *B. pumilus*.

Effect of plant growth and yield under field conditions

Results of variance analyses are presented in Table 3. The results indicated that grain yield (GY), spike length (SL), plant height (PH), number of grain per spike (NGPS), and grain weight per spike (GWPS) had some difference at 1% probability level (Table 3).

Table 3. ANOVA results of yield and yield components

S.O.V	GY	SL	PH	GWPS	NGPS
Replication	1732.646ns	0.175ns	15.484 ns	0.051 ns	0.776 ns
Year (Y)	403333.333**	0.630ns	48.682ns	0.745**	58.963**
Strain (S)	6725.131**	0.158ns	7.850ns	0.029ns	5.389ns
YxS	10476.762**	0.233ns	48.510**	0.189**	27.661**
Error	1636.268	0.181	13.930	0.034	3.925
General	12261.700	0.194	18.980	0.072	8.715

*: significant at α level %5, **: significant at α level %1, ns: non-significant,
S.O.V: source of variation, GY: grain yield, SL: spike length, PH: plant height, GWPS: spike length, NGPS: grain weight per spike

The morphological properties such as PH, SL, NGPS, GWPS, and GY were observed and evaluated for each of the PGPR strains treatment and compared with the non-inoculated control. The statical analysis indicated that interaction between grain yield and years was found significantly important (Table 3). Therefore, the years were evaluated separately (Table 4).

Table 4. Means of yield and yield components of wheat at different PGPR straints in the field trial run in the 2016 -17 and 2017-18 periods

2016-2017 field trails					
Treatment	PH (cm)	SL (cm)	NGPS	GWPS (g)	GY (kg ha^{-1})
W1	92.63	9.67	41.40b	2.08c	7236.6ab
W2	90.06	9.36	40.33b	2.03c	6963.3cd
W3	89.70	9.53	41.20ab	2.10abc	7443.3a
W4	88.53	9.57	44.26ab	2.44a	6790.0d
W5	86.76	9.74	46.20a	2.42ab	6566.6e
W6	94.20	9.77	41.93ab	2.19abc	6973.3bcd
W7	93.30	9.47	39.80ab	2.08bc	7016.6bc
Control	93.96	9.61	40.00ab	2.02c	6946.6cd
MSE	-	-	3.585	0.025	38.506
2017-2018 field trails					
W1	87.80	9.33a	42.16bc	2.00ab	4326.6ab
W2	85.36	9.50ab	44.40abc	1.93ab	4130.0b
W3	88.93	8.96b	43.43abc	1.61b	5163.3ab
W4	90.86	9.10b	41.76bc	1.73b	6090.0a
W5	94.46	8.90b	40.66c	1.63b	5403.3ab
W6	86.56	9.33ab	43.00bc	1.87b	5660.0ab
W7	90.73	9.43ab	46.56ab	2.43ab	5786.6ab
Control	85.93	10.00a	47.63a	2.33a	4710.0ab
MSE	-	-	4.362	0.040	3255.619

MSE: mean squared error , W1: *B. pumilus*, W2: *B. simplex*, W3: *B. simplex*, W4: *B. simplex*, W5: *B. simplex*, W6: *B. simplex*, W7: *B. pumilus*
W1: *B. pumilus*, W2: *B. simplex*, W3: *B. simplex*, W4: *B. simplex*, W5: *B. simplex*, W6: *B. simplex*, W7: *B. pumilus* They are code or name of bacterial strain that are identified as *B. pumilus* and *B. simplex*

The number of grain per spike

In the first year, the application of PGPR was measured between 39.80 to 46.20 and 40.66 to 46.56 in the second year. Especially, W5 was increased number of grain pers pike compared to control. W5 strain showed highly effective at 15.5% (Table 4). Also, the mean values of NGPS in 2017-18 were higher than in 2016-17. This may have been caused by longer spikes in 2018 when the high rainfall was received. It can be said that high precipitation affects NGPS.

Spike length

Results of ANOVA indicated a statistically significant difference in spike length was not affected by any PGPR treatments (Table 3) when both years' experiments were evaluated. It varied between 9.47 and 9.77 cm in the first year and 8.90 to 9.43 cm in the second year (Table 4). Also, the first-year experiment showed that the W5 strain was enhanced SL at the rate of 1.35 (Table 4).

Plant height

All treatments were in the same group as statistically, W5 PGPR treatment (86.76 cm) in the first year and W2 PGPR treatment (85.36 cm) in the second year decreased the plant height (Table 4). The mean value of PH in 2016-17 was higher than in 2017-18 years. However, the mean values of SL in the 2016-17 year were lower than the 2017-18 year. These two results were parallel each together. The higher amount of precipitation in the first year was enhanced to PH. But it may get lower the mean value of SL (Table 4).

Grain weight per spike

It ranged from 2.03 g to 2.44 g in the first year and 1.63 g to 2.43 g in the second year (Table 4). Grain weight per spike was not affected by PGPR treatments (Table 3). However, in the first year, the W4 strain increased grain weight per spike at the rate of 20.79% (Table 4).

Grain yield

It was markedly influenced by PGPR treatments. The application of PGPR except in the first-year results indicated that W3 increased the value (7443.3 kg ha^{-1}) of GY at the rate of 7.15%. Also, W1 (7236.6 kg ha^{-1}) and W7 (7016.6 kg ha^{-1}) led to more grain yield when compared to non-inoculated control. However, second-year experiments, W3, W4, W5, W6, and W7 increased the GY between 9.62% and 29.29% (Table 4). According to results, strains were found the effects on plant growth during 2017-2018. The PGPR has supported wheat growth. So, our results indicated that as expected results, mean values of GY in 2016-2017 were higher than 2017-2018.

DISCUSSION

Wheat is one of the world's most important crops. Over 760 million tons of wheat were produced in 2020 worldwide (FAO,2021). The potential of wheat yield represents the yield of a cultivar grown in environments which is an adaptation of nutrients and water regime, pesticide application influences wheat yield and quality (Kovacevic, 2007). Recently, some studies that include a way to protect plants and soil against chemical damage, maintained to rise yield and quality of wheat. The use of PGPR-based products has given the opportunities that they are considered safer than many of the chemicals now in use and not considered harmful to ecological processes or the environment (Lucas Garcia et al., 2004).

Here, the PGPR isolates were collected and identified as *Bacillus* spp. including *B. pumilus* and *B. simplex* strains by biochemical tests and MALDI-TOF MS assay. Following studies to obtain the PGPR strains ability on wheat growth and yield in field conditions. Previous studies indicated that *Bacillus* spp. associated with rhizosphere is one of the important PGPR. The first commercial bacterial fertilizer called Alinit was developed from *Bacillus* spp. They could support the plants in many ways such as biofilm could develop biofilms, solubilization phosphate, N fixing, or uptake nutrients. However, *Bacillus* spp. increased crop yield by 40% (Kilian et al., 2000; Haas and Defago, 2005; Beauregard et al., 2013; Lyngwi and Joshi, 2014; Berendsen et al., 2016).

In the present study, to determine whether PGPR interaction ability could make enhance plant performance, a field experiment was carried out with PGPR (*B. simplex* and *B. pumilus*) inoculated seeds and non-inoculated seeds. In the first-year experiment, PGPR showed increasing all plant parameters such as PH, SL, NGPS, GWPS, and GY. On the other hand, the second-year experiment indicated that PGPR just increased PH and GY. Also, both PH and GY values have been higher percentages than in the first-year experiment. Up to this point, when the results for both years were compared with control, the weather condition such as temperature and rainfall might have played a key role. PGPR strains were risen the GY by over 29% in the second year according to control. However, GY was increased over by 7% in the first year. Nguya et al. (2019) mentioned that the response of PGPR strains seems to correlate with air and soil temperate during winter. The low temperature might make a negative effect on the colonization and survival of PGPR when seeds are inoculated and sown in autumn. As a parallel with our results, the temperature for the first year in autumn was lower (3.8 °C) than the second year (11.7 °C). So, PGPR might not survive in the condition. Also, higher colonization at early stages helps PGPR to be more competitive. In addition, PGPR cannot better-adapted local microflora and soil microfauna (Bashan et al., 2014). Therefore, *Bacillus* spp. strains referred to in this study could not be successful to colonize in the first year because of weather and other microorganism population in the soil. So, in the second year, the temperature was suitable for *Bacillus* strains also they might have better-adapted ability and extent their population. For all those, *Bacillus* strains led to a rising rate of GY from 7% to 29%. In the present research, a significant increase in GY was recorded in treatments where all these PGPR species were inoculated. This increase in yield might be the result of N fixing and phosphate solubilization capacity of these inoculated strains (Krey et al., 2013; Puri et al., 2016). Grain yield is one of the significant parameters between yield and yield components. It is usually characterized by biotic and abiotic environmental factors. The present study mentions that PGPR led to having more grain yield. The results were in agreement with the finding of Dobbelaere et al. (2001) and Saber et al. (2012). Moreover, *Bacillus* strains were increased PH for two years. Similarly, Kumar et al. (2014) were reported that *B. megaterium* has a positive effect on the PH. Interestingly, Akbar et al. (2019) referred to species of *Bacillus* MCR-7, *B. subtilis* did not show promising results on GWPS. Conversely, our result indicated that *B. simplex* strain W4 increased GWPS in the first-year field experiment. It also pointed out variable significant differences in the bioactivity of different types of similar PGPR species (Shaharooa et al., 2008). However, Alsaady et al. (2020) point out the number of grains per spike. They indicated those treatments of PGPR such as *Pseudomonas fluorescens*, *Streptomyces*, *Azotobacter*, *Azospirillum*, *B. subtilis*, *B. pumilis*, and bacterial mixture found effective on NGPS. Similarly, the treatment of the W5 strain led to a significant increase in NGPS in this study.

The value of yield independence of yield components such as plant height, spike length, leaf area, or stem high were also found associated with the vegetative period. These components are in direct connection with productivity in wheat (Knezevic et al., 2007). So, the study results were given a similar outcome with previous studies. For two years field experiments were given evidence to see the effect of PGPR on plants growth and yield. Also, *Bacillus* strains referred to as *B. simplex* and *B. pumilus* increased the yield like as most studies (Turner and Backman 1991; Garcia et al., 2003; Kokolis-Burella et al., 2003; Adesomoye et al., 2008; Mısra et al., 2010; Yildirim et al., 2011; Ibiene et al., 2012). Besides, for each year, observed the yield parameters showed some changes. We have thought that the climate and environmental factors are caused the changes (Bouaziz and Hicks, 1990; Acevedo et al., 1991; de Freitas and Germida, 1992; Dobbelaere et al., 2001).

CONCLUSIONS

In conclusion, the PGPR has some ability such as the use of biofertilizer to grow plants and pesticides for plant diseases. Therefore, it increases agricultural productivity and has an important role in the sustainable agricultural industry. The present study indicates the beneficial effects of *B. simplex* and *B. pumilus* strains inoculation on wheat growth in field conditions. The results of grain yield indicated that *B. simplex* and *B. pumilus* have an advantage especially strain W3 and W4 led to a significant increase in grain yield. It is concluded that the application of PGPR performed better than non-inoculated application. Therefore, PGPR seems a good alternative instead of using synthetic chemical fertilizers. This approach could be reduced the application of synthetic chemical fertilizers. According to our results, PGPR has promising and eco- and environmentally-friendly, and economical outcomes for winter wheat growth and production. It should be developed for future studies as multidisciplinary for the sustainability of winter wheat production.

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INHERITANCE OF YIELD AND SOME FIBER PROPERTIES OF LINE X TESTER HYBRIDS IN COTTON (*Gossypium hirsutum* L.)

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ABSTRACT

This study was determined the general combining ability (GCA) of parents and specific combining abilities of their hybrid combinations. Furthermore, hybrid vigor of F_1 was evaluated by investigating genetic structure of the lines and testers. A total of 28 F_1 hybrids (4 lines and 7 testers) were used as experimental material, which were developed from line × tester method. Investigated traits, except fiber length were influenced by non-additive gene effect in the created populations. Heterosis values for seed cotton weight per boll, earliness ratio, ginning percentage, fiber length and micronaire were positive, while seed cotton yield was negative. Gloria, TMN 199 and ZN 1018 proved the best parents for seed cotton yield, while TMN 199, GW 2357 and TMN 170 were best for earliness ratio. Similarly, GW 2357 and Gloria were best for seed cotton weight per boll, whereas Ipek 607 proved best for seed index. Likewise, TMN 199, TMD 139 ZN 1018 were best for ginning percentage, while Gloria, Flash, Ipek 607 and UA 48 proved best for fiber length. In the same way, Ipek 607 and TMD 139 were best parents for fiber fineness, whereas Gloria and UA 48 proved best for fiber strength. These parents had the highest overall combining abilities for respective traits. The hybrid combinations, i.e., TMN 199 × Ipek 607, TMN 199 × UA 48, Flash × GW 2357, Gloria × Ipek 607, Gloria × UA 48 and ST 468 × ADN 712 had the highest combining abilities for seed cotton yield. Similarly, TMN 199 × TMN 170 exhibited higher combining ability for earliness ratio, whereas Flash × GW 2357 and Gloria × Ipek 607 had better combining ability for seed cotton weight per boll. Likewise, TMN 199 × GW 2357 and Gloria × ADN 712 resulted in higher combining ability for seed index, while Gloria × TMD 139, TMN 199 × TMN 170, Flash × ZN 1018 and ST-468 × TMN 139 proved better for ginning percentage. Nonetheless, Flash × ADN 712 and ST-468 × Ipek 607 had better combining ability for fiber length, whereas ST-468 × Ipek 607 exhibited better combining ability for fiber micronaire. In the same way, TMN 199 × UA 48, Flash × Ipek 607, ST-468 × GW 2357 and ST-468 × UA 48 had better combine ability for fiber strength. The study identified the most promising hybrids for respective traits and overall combining abilities.

Keywords: Combining ability, fiber quality, ginning percentage, line-tester, seed cotton yield

INTRODUCTION

Cotton is the most important fiber source for Turkey and other countries in the world. It provides the most important material for textile, ginning, oil, feed, and paper industries. Community awareness has rapidly increased on cotton products, and many features of the fiber are used to assess the quality. For instance, fiber fineness affects string quality by 20-35%, fiber length by 35%, and fiber strength by 30-35% (Guvercin et al., 2019). These circumstance force breeders to develop new cultivars with more productive and high quality fibers and explore the gene effects of both parents and hybrids on the desired traits. Line × Tester is a statistical method that can fulfill this necessity. It was first used by Kempthorne in 1957. Hybrid potency is a method that compares the traits of hybrids with their parents in the early generation. It is known as heterosis and heterobeltiosis. While heterosis is a method of determining that a hybrid is closer to which of the parents, heterobeltiosis is a method of determining the status of the hybrid versus superior parent

Several earlier studies have reported that many traits are managed by non-additive gene effects. Boyaci (2011) reported that fiber fineness, boll seed weight and earliness characteristics showed negative heterosis, while both positive and negative heterosis could be seen in other characteristics. In addition, many researchers determined that non-additive genes, in other words, dominant, epistatic or dominant × epistatic genes were effective in the management of yield, ginning percentage, earliness, single boll seed weight, 100 seed weight, fiber fineness and fiber strength traits in Line × Tester populations (Ashokkumar et al., 2010; Saravanan et al., 2010; Kalpande et al., 2014; Sawarkar et al., 2015; Sajjad et al., 2016; Guvercin, 2016; Patil et al., 2018; Ullah et al., 2020; Unay et al., 2020).

Basal et al. (2009) and Karademir et al. (2009) reported that fiber length is managed by additive genes. In addition, Babar et al. (2001), Lakho et al. (2001) and Karademir (2004) indicated high heterosis in ginning percentage, whereas Karademir (2004) and Boyaci (2011) reported high heterosis for 100-seed weight.

The aim of this study was to determine the general combining abilities (GCA) of parents and specific combining abilities (SCA) of hybrids, genetic influence affecting traits in populations as well as the hybrid powers of the crossed lines (Heterosis and Heterobeltiosis) in accordance with the Line × Tester analysis method among the cotton varieties developed recently in Turkey

MATERIALS AND METHODS

The study was carried out in Nazilli-Aydin province (37° 54′ 45″ N, 28° 19′ 14″ E, altitude: 64 m) during 2015 and 2016. Gloria, TMN 199, TMN 170 and ST 468 cotton varieties were used as homozygote lines, whereas GW 2357, ZN 1018, GW 2357, UA 48, ADN 712, Ipek 607 and TMN 139 were used as testers.

The crosses were made in accordance with the Line × Tester analysis method and 28 hybrid combinations were obtained during 2015. The hybrid combinations and their 11 parents were planted at Nazilli Cotton Research Institute on 11 May 2016. The experiment was laid out according to Randomized Complete Block Design (RCBD) with 3 replications. Parents and hybrids were planted in plots of replications as 1 row with length of 12 m, inter-row spacing of 0.70 m and intra-row spacing of 0.10 m. The soil of the experimental area is rich in phosphorus (3.5 ppm) and potassium (350 ppm), poor in organic matter (1.4%), medium calcareous (20.98%), alkaline (pH: 7.95) and saline (0.036%) (Anonymous, 2018). During 2016, 90 kg ha^{-1} of pure nitrogen (46% urea) was applied, followed by the first irrigation (4 furrow irrigations in total). Harvesting was performed manually twice on 8 October 2016 and 1 November 2016.

The highest (38.2 ºC) and the lowest (7.1 ºC) temperature in the region was recorded during August and November, respectively. In long-term data, the highest average temperature was observed in July (28.4 ºC), the lowest in December (8.2 ºC), and maximum in July (36.1 ºC).

Data relating to seed cotton yield (SCY; kg ha^{-1}), earliness (ER; %), fiber fineness (FF; mic.), fiber length (FL; mm), fiber strength (FS; g tex^{-1}), ginning percentage (GP; %), 100 seed weight (g) and single boll weight (g) were collected during the study. Fiber properties were determined by USTER HVI 1000.

Values obtained were analyzed according to the Line × Tester method with the statistical package program (TARPOPGEN) developed by Ozcan and Acikgoz (1999) in accordance with Kempthorne (1957) and Sing et al. (1982), and the means were compared with the $LSD_{(0.05)}$ (least significant difference) test. Heterosis (Chiang and Smith, 1967) and heterobeltiosis (Fonseca and Patterson, 1968) values were determined with the following formulas.

Heterosis (%); $Ht = (F_1\text{-}MP)/MP) \times 100$, Mean Parents = (Line parent + Tester parent)/2 (1)

Heterobeltiosis (%); $Htb = (F_1\text{-Better Parent})/\text{Better Parent}) \times 100$ (2)

RESULTS AND DISCUSSION

The differences between genotypes were significant (p>0.01). Both parents and crosses contributed significantly to variation; however, the contribution of parents was more significant than hybrids (Table 1). These results stem from the significance of parent's GCA and the hybrid's SCA values. However, parents showed negative GCA for seed cotton yield, which indicated that GCA/SCA ratio was negative. This pointed out that dominant genes were effective in seed cotton yield.

Table 1. Variance analysis according to line × tester analysis

Source of variation	DF	SCY	GP	ER	BW	SW	FF	FL	FS
Replication	2	13104.45	0.925	55.466	0.036	0.017	1.991	0.059	5.559
Genotypes	38	42967.24**	18.249**	138.650**	0.954**	4.593**	8.213**	0.240**	16.476**
Parents	10	20213.75**	27.727**	148.103**	0.786**	5.287**	12.375**	0.431**	31.915**
Parents vs Hybrids	1	40318.73**	15.628**	1191.412**	5.526**	70.577**	0.209	0.215**	4.182
Hybrids	27	51492.56**	14.835**	96.157**	0.846**	1.893**	6.968**	0.170*	11.214**
Lines	3	21500.17**	4.019**	422.811**	1.434**	5.066**	6.645**	0.508**	8.787**
Tester	6	18637.41**	59.965**	146.384**	2.582**	1.391*'*	25.291**	0.377**	36.581**
Line × Tester	18	67443.01**	1.595**	24.973**	0.170**	1.531**	0.913**	0.045**	3.163**
Error	76	5269.67	0.424	40.513	0.067	0.041**	0.512	0.032	2.213
δ^2 GCA		-354.454	0.294	1.582	0.015	0.008	0.135	0.003	0.179
$^{\delta2}$ SCA		20.724.446	0.390	-5.180	0.034	0.496	0.134	0.004	0.519
δ^2GCA/δ^2SCA		-0.017	0.750	-0,3	0.440	0.170	1.007	0.750	0.340

SCY: Seed cotton yield (kg ha $^{-1}$), GP: Ginning percentage (%), ER (%), FL: Fiber length (mm), FF: Fiber fineness (Micronaire), FS: Fiber strength (g/tex), BW:Boll weight (g), SW: seed weight (g)

In addition, it is clearly understood from GCA/SCA ratio (<1) that dominant genes are effective in other traits except for fiber fineness (1.007). Basal et al. (2009), Karademir et al. (2009), and Ekinci and Gencer (2014) reported that additive gene effect was important in fiber length, and dominant gene effect in other traits were compatible with the results of Ali et al. (2016) and Sajjad et al. (2016).

General Combining Ability of Parents (GCA)

According to parental GCA, TMN 199 was positive to ginning percentage, earliness rate, 100 seed weight, fiber fineness, but negative to seed cotton yield, boll weight, fiber length and fiber strength. Flash variety was positive to seed cotton yield, 100 seed weight and fiber length. Gloria variety was positive to seed cotton yield, boll weight, 100 seed weight, fiber length and fiber strength. The ST 468 was negatively correlated to the traits for which Gloria had possitive correlations, except for fiber strength (Table 2).

On the other hand, ADN 712 variety from the tester contributed positively towards ginning percentage, boll weight, and fiber fineness, and negatively to earliness rate, fiber length and fiber strength. The GW 2357 variety was positive to seed cotton yield, earliness rate, boll weight and fiber fineness, and negative to ginning percentage and fiber strenght (Table 2). Similarly, Ipek 607 variety was positive to seed cotton yield, boll weight, 100 seed weight, fiber length and fiber fineness and negative to ginning percentage, earliness rate and fiber fineness. Likewise, TMD 139 variety was positive to ginning percentage, whereas negative to other traits. The TMN 170 variety was positive to ginning percentage, earliness rate, boll seed weight and 100 seed weight, while negative to seed cotton yield and fiber length. The variety UA 48 was positive to boll weight, 100 seed weight, fiber length and fiber strength, whereas negative to ginnig percentage. The ZN 1018 variety was positive to seed cotton yield, ginning percentage and fiber fineness, while negative to boll weight, 100 seed weight, fiber length and fiber strength (Table 2). These results indicate that crucial new genotypes can be developed if ZN 1018, GW 2357 and Ipek 607 varieties are crossed with a high ginning percentage cultivar.

Table 2. GCA effects of parents for investigated properties

Lines and Testers	SCY	GP	ER	BW	100 SW	FF	FL	FS
Lines								
TMN 199	-14.747	0.53**	4.564*	-0.096**	0.537**	0.186**	-0.520**	-0.683**
Flash	35.11**	0.12	-2.004*	0.053	0.070**	0.038	0.093**	-0.421
Gloria	16.40**	-0.50**	-5.297*	0.331**	0.047**	-0.188	0.752**	0.569**
ST 468	-36.77**	-0.15**	2.737*	-0.288	-0.654**	-0.036	-0.324**	0.536**
Testers								
ADN 712	-2.11	0.21**	-4.542*	0.066*	-0.027	0.056**	-0.439**	-0.785**
GW 2357	36.64**	-0.40**	5.729*	0.293**	-0.013	0.124**	-0.877	-0.935**
Ipek 607	26.94**	-4.06**	-3.032*	0.588**	0.260**	-0.340**	2.664**	1.774**
TMD 139	-16.65	3.38**	-1.658	-0.766**	-0.286**	-0.101**	-0.905**	-1.168**
TMN 170	-39.86**	1.05**	2.654*	0.120**	0.352**	0.017	-0.663**	0.215
UA 48	-4.847	-0.66**	-0.016	0.173**	0.294**	0.036	1.390**	2.882**
ZN 1018	73.17**	0.48**	0.864	-0.473**	-0.579**	0.207**	-1.199**	-1.985**

Specific Combining Ability of Hybrids (SCA)

A total of 12 hybrids displayed negative SCA for seed cotton yield, 13 for ginning percentage and fiber length, 15 for earliness rate and fiber fineness, 14 for boll seed weight, 16 for 100 seed weight, 15 for fiber fineness, 13 for fiber length and fiber strength unlike other combinations (Table 3).

It has been reported that fiber length is managed by both additive (Ekinci and Gencer, 2014) and non-additive (dominant and epistatic) (Basal et al., 2009) gene effects. Fiber fineness and strength is managed by additive (Karademir and Gencer, 2010), and fiber fineness by both additive and non-additive (Ekinci and Gencer, 2014) gene effects. Basal et al. (2009) reported that fiber strength is governed by non-additive genes. Among studied traits, TMN 199 × Ipek 607, TMN 199 × UA 48, TMN 199 × ZN 1018, Flash × GW 2357, Flash × TMN 170, Gloria × Ipek 607, Gloria × UA 48, ST 468 × ADN 712, ST 468 × TMD 139 and ST 468 × TMN 170 were significant for seed cotton yield. Similarly, Flash × ADN 712 and Flash × ZN 1018 remained significant for ginning percentage, whereas Flash × ADN 712 and Gloria × TMD 139 for boll weight. Likewise, TMN 199 × GW 2357, TMN 199 × Ipek 607, Flash × TMN 170, Flash × UA 48, Gloria × ADN 712, ST 468 × GW 2357, ST 468 × Ipek 607 and ST 468 × UA 48 proved significant for 100 seed weight, while Flash × TMN 170, Gloria × GW 2357 and Gloria × Ipek 607 were significant for fiber fineness. In the same way, Flash × ADN 712 and ST 468 × GW 2357 remained significant for fiber length, whereas Flash × Ipek 607 and ST 468 × GW 2357 hybrids were significant for fiber strength. The hybrids with the highest special adaptability for seed cotton yield in the current study were Gloria × Ipek 607, whereas ST 468 × GW 2357 had the highest special adaptability for fiber length and strength (Table 3).

Table 3. SCA effects of hybrids for investigated characteristics

F_1 Hybrids	SCY (kg ha^{-1})	GP (%)	ER (%)	BW(g)	100 SW(g)	FF (mic)	FL (mm)	FS (g tex^{-1})
TMN 199 × ADN 712	55.71	-0.016	-0.26	-0.039	-0.516**	-0.025	0.25	0.408
TMN 199 × GW 2357	-119.28**	0.256	-1.214	0.094	1.253**	-0.052	0.002	-0.642
TMN 199 × Ipek 607	97.88**	0.0272	1.847	-0.118	0.568**	0.074	-0.39	-0.617
TMN 199 × TMD 139	3.38	0.355	-2.102	-0.044	-0.690**	-0.067	-0.006	0.592
TMN 199 × TMN 170	-26.37	-0.269	3.594	-0.168	0.046	-0.002	-0.279	-0.092
TMN 199 × UA 48	160.60**	-0.063	0.31	0.168	-0.718**	-0.025	0.042	0.008
TMN 199 × ZN 1018	149.27**	-0.335	-2.175	0.106	0.057	0.098	0.23	0.342
Flash × ADN 712	5.694	0.643**	0.553	0.253**	-0.306**	0.059	0.781**	0.246
Flash × GW 2357	124.51**	-0.353	1.288	0.045	-0.224**	-0.058	-0.838**	-1.53**
Flash × Ipek 607	-207.92**	-0.207	-0.001	-0.18	-0.535**	0.005	0.318	1.665**
Flash × TMD 139	-12.44	-0.652**	-1.238	-0.143	0.047	-0.026	-0.136	0.163
Flash × TMN 170	86.66**	-0.378	2.176	0.316	0.483**	0.129**	0.305	-0.487
Flash × UA 48	-22.56	0.238	1.17	-0.024	0.691**	0.133	-0.421	-0.12
Flash × ZN 1018	-20.03	0.690**	-3.949	-0.267**	-0.156	-0.241	-0.009	0.08
Gloria × ADN 712	-236.34**	0.602	0.272	-0.141	1.398**	-0.054	-0.968**	0.456
Gloria × GW 2357	-41.1	0.685	2.033	-0.003	-1.254**	0.085**	-0.273	0.039
Gloria × Ipek 607	260.38**	0.447	-3.423	0.133	-0.175	0.098**	-0.131	-1.202
Gloria × TMD 139	-66.85	-2.015	2.851	0.516**	0.670**	0.07	0.402	-0.927
Gloria × TMN 170	-130.74**	0.174	-1.303	-0.259**	-0.383**	-0.232**	0.536	0.623
Gloria × UA 48	174.32**	-0.248	-4.173	-0.233**	-0.356**	-0.094	0.14	-0.044
Gloria × ZN 1018	40.34	0.356	3.744	-0.013	0.1	1.121	0.295	1.056
ST 468 × ADN 712	128.93**	0.057	-0.564	-0.073	-0.576**	0.02	-0.063	-1.111
ST 468 × GW 2357	35.87	-0.588	-2.107	-0.137	0.226**	0.026	1.109**	2.139**
ST 468 × Ipek 607	-150.33	-0.312	1.577	0.164	0.142**	-0.178	0.052	0.164
ST 468 × TMD 139	70.80**	1.007	0.49	-0.329	-0.027	0.017	-0.259	0.173
ST 468 × TMN 170	70.45**	0.473	-4.467	0.111	-0.17**	0.106	-0.562	-0.044
ST 468 × UA 48	8.84	0.074	2.692	0.089	0.383**	-0.013	0.239	0.156
ST 468 × ZN 1018	-169.57	-0.71	2.379	0.175	-0.001	0.022	-0.516	-1.477

Mean performance of parents and F_1 crosses

The values of the studied traits of the parents (line and tester) are given in Table 4. Seed cotton yield of the lines ranged from 7035.7 to 5496.0 kg ha^{-1}, ginning percentage from 42.03 to 40.25%, earliness rate from 78.86 to 66.50%, boll seed weight from 6.23 to 5.44 g, 100 seed weight from 10.43 to 9.90 g, fiber fineness from 5.17 to 5.57 micronaire, fiber length from 32.01 to 28.96 mm, and fiber strength from 37.33 to 30.80 g tex^{-1}. Seed cotton yield of the testers ranged from 621.82 to 462.30. kg da^{-1}, ginning percentage from 42.10 to 32.19%, earliness rate from 87.90 to 70.31%, boll seed weight from 6.40 to 4.79 g, 100 seed weight from 13.86 to 8.90 g, fiber fineness from 4.47 to 5.62 micronaire, fiber length from 35.26 to 28.43 mm, and fiber strength from 40.13 to 29.46 g tex^{-1} (Table 4).

On the other hand, seed cotton yield of hybrids of these parents ranged from 8261.9 kg ha^{-1} (Gloria × Ipek 607) to 3003.39 kg ha^{-1} (Gloria × ADN 712), ginning percentage from 44.99% (TMN 199 × TMD 139) to 36.19% (ST 468 × Ipek 607), earliness rate from 91.93% (TMN 199 × TMN 170) to 69.36% (Gloria × Ipek 607), boll seed weight from 7.38 g (Gloria × Ipek 607) to 4.94 g (ST 468 × TMD 139), 100 seed weight from 13.59 g (TMN 199 × GW 2357) to 10.58 g (ST 468 × ZN 1018), fiber fineness from 4.78 micronaire (ST 468 × Ipek 607) to 5.82 micronaire (TMN 199 × ZN 1018), fiber length from 34.09 mm (Flash × Ipek 607) to 28.90 mm (ST 468 × ZN 1018) and fiber strength from 37.30 g tex^{-1} (ST 468 × UA 48) to 30.80 g tex^{-1} (ST 468 × ZN 1018). The ST 468 × ZN 1018 had low 100 seed weight, fiber length and fiber strength; however, low 100 seed weight had not affected the seed cotton yield and ginning percentage.

Among the lines, TMN 199 had the highest ginning percentage (42.03%) and earliness rate (78.86%), whereas Gloria had the highest seed cotton yield (7035.7 kg ha^{-1}), boll seed weight (6.23 g), 100 seed weight (10.30 g), fiber length (32.01 mm) and fiber strength (37.33 g tex^{-1}). Among testers, ZN 1018 had the highest seed cotton yield (6218.82 kg ha^{-1}), whereas TMD 139 resulted in the highest ginning percentage (42.97%). Likewise, GW 2357 had the highest earliness rate (79.89%) and single boll seed weight (6.50 g), Ipek 607 had the highest fiber length (35.26 mm), fiber strength (34.73 g tex^{-1}) and fiber fineness (4.47 micronaire) (Table 4).

Among hybrids, Gloria × Ipek 607 produced the highest seed cotton yield (8261.9 kg ha^{-1}), while TMN 199 × TMD 139 and Flash × TMD 199 resulted in the highest ginning percentage (44.99% and. 44.89%). Similarly, TMN 199 × TMN 170 recorded the highest earliness rate (91.93%), Gloria × Ipek 607 had the highest boll weight (7.38 g), TMN 199 × Ipek 607 observed the highest 100 seed weight, Flash × Ipek 607 resulted in the highest fiber length and strength, and the thinnest fibers were recorded for ST 468 × Ipek 607 (Table 4).

Table 4. Mean values of observed characters

Parents and F_1 Hybrids	SCY (kg ha^{-1})	GP (%)	ER (%)	BW (g)	SW (g)	FF (mic)	FL (mm)	FS (g tex^{-1})
Lines								
TMN 199	6992.0	42.03	78.86	5.61	9.90	5.57	30.84	32.66
Flash	5496.0	40.25	66.50	5.99	10.43	5.42	30.30	34.10
Gloria	7035.7	40.41	67.36	6.23	10.30	5.17	32.01	37.33
ST 468	5698.4	41.74	71.03	5.44	9.93	5.42	28.96	30.80
Testers								
ADN 712	4623.0	40.04	70.31	5.59	9.23	5.14	30.09	31.86
GW 2357	4726.1	39.21	79.89	6.50	9.86	5.48	29.93	29.30
Ipek 607	5757.9	32.19	66.90	6.24	13.86	4.47	35.26	34.73
TMD139	5123.0	42.97	68.80	4.79	8.90	4.60	30.76	32.23
TMN 170	5103.1	42.10	87.90	6.40	9.46	5.27	29.85	33.73
UA 48	5234.1	36.99	77.02	6.02	9.56	5.42	33.74	40.13
ZN 1018	6218.2	41.09	79.72	5.48	9.56	5.62	28.43	29.46
Mean$_{(\text{Parents})}$	5638.0	39.80	75.70	5.70	10.60	5.10	30.80	33.20
LSD$_{(0.05)}$	1178.0	1.28	15.80	0.40	0.36	0.30	1.01	2.28
Crosses								
TMN 199 × ADN 712	5613.1	41.45	80.83	6.26	11.81	5.55	30.30	32.66
TMN 199 × GW 2357	3517.8	41.11	90.20	6.62	13.59	5.59	29.60	31.46
TMN 199 × Ipek 607	6325.3	37.26	84.50	6.70	13.18	5.25	32.90	34.20
TMN 199 × TMD 139	4944.4	44.99	81.92	5.42	11.38	5.35	29.50	32.46
TMN 199 × TMN 170	4414.6	42.04	91.93	6.18	12.75	5.53	29.50	33.16
TMN 199 × UA 48	3422.6	40.53	85.98	6.57	11.93	5.53	31.90	35.93
TMN 199 × ZN 1018	7301.5	41.40	84.37	5.86	11.83	5.82	29.50	31.40
Flash × ADN 712	6071.4	40.41	75.12	6.70	11.55	5.48	31.40	32.76
Flash × GW 2357	6454.3	40.10	86.13	6.72	11.65	5.43	29.30	30.83
Flash × Ipek 607	3765.8	36.58	76.08	6.79	11.61	5.03	34.09	36.73
Flash × TMD 139	5285.7	44.89	76.22	5.47	11.65	5.24	30.07	32.30
Flash × TMN 170	6043.6	41.53	83.94	6.82	12.72	5.51	30.70	33.03
Flash × UA 48	5301.5	40.43	80.27	6.53	12.87	5.54	32.08	36.06
Flash × ZN 1018	6107.1	42.02	76.03	5.40	11.15	5.33	29.90	31.40
Gloria × ADN 712	3003.9	41.03	71.55	6.58	13.10	5.14	30.30	33.96
Gloria × GW 2357	4611.1	40.50	83.58	6.95	10.60	5.35	30.60	33.40
Gloria × Ipek 607	8261.9	36.60	69.36	7.38	11.95	4.90	34.30	34.86
Gloria × TMD 139	4553.5	41.59	77.01	6.41	12.25	5.12	31.20	32.20
Gloria × TMN 170	3682.5	41.45	77.17	6.52	11.83	4.93	31.60	35.13
Gloria × UA 48	7083.3	39.31	71.63	6.60	11.80	5.08	33.30	37.13
Gloria × ZN 1018	6523.5	41.05	80.43	6.17	11.38	5.47	30.80	33.36
ST 468 × ADN 712	6125.0	40.84	78.75	6.03	10.56	5.37	30.10	32.36
ST 468 × GW 2357	4849.2	39.59	87.48	6.19	11.37	5.44	30.90	35.46
ST 468 × Ipek 607	3623.0	36.19	82.40	6.79	11.35	4.78	33.40	36.20
ST 468 × TMD 139	5448.4	44.96	82.69	4.94	10.85	5.21	29.50	33.26
ST 468 × TMN 170	5162.6	42.10	82.04	6.27	11.37	5.42	29.50	34.43
ST 468 × UA 48	4896.8	39.99	86.53	6.30	11.84	5.32	32.30	37.30
ST 468 × ZN 1018	3892.8	40.34	87.10	5.74	10.58	5.52	28.90	30.80
Mean$_{(\text{crosses})}$	5220.0	40.10	81.00	6.20	11.77	5.20	31.03	33.70
LSD$_{(0.05)}$	1200	0.92	7.00	0.80	1.00	0.28	1.24	2.36

Hybrid vigor (Heterosis and Heterobeltiosis)

Heterosis values of hybrids for seed cotton yield ranged between -48.46% (Gloria × ADN 712) and 29.15% (Gloria × Ipek 607), whereas the values for ginning percentage varied between 6.16% (ST 468 × TMD 139) and -2.59% (ST 468 × ZN 1018). Likewise, heterosis values for earliness rate ranged between 19.48% (ST 468 × Ipek 607) and -0.77% (Gloria × UA 48), whereas the values for boll seed weight varied between 18.4% (Gloria × Ipek 607) and -3.4% (ST 468 × TMD 139). Similarly, heterosis values for 100 seed weight ranged from 37.6% (TMN 99 × GW 2357) to -4.6% (ST 468 × Ipek 607), whereas the values for fiber fineness varied between 5.21% (TMN 199 × TMD 139) and -5.65% (Gloria × TMN 170). In the same way, heterosis values for fiber length were in the range of 5.03% (ST 468 × GW 2357 and 3.94% (TMN 199 × TMD 199), whereas the values for fiber strength differed from 18.02% (ST 468 × GW 2357) to -8.38% (TMN 99 × ADN 712) (Table 5).

Gloria × Ipek 607 and Flash × GW 2357 hybrids showed significant positive heterosis for boll weight and 100 seed weight, and contributed to seed cotton yield, while TMN 199 × TMN 170 hybrid exhibited high positive and significant heterosis for seed cotton yield and 100 seed weight. The hybrids TMN 199 × UA 48, Flash × Ipek 607, Gloria × ADN 712, Gloria × GW 2357 and ST 468 × ZN 1018 exhibited high and positive heterosis for boll weight and 100 seed weight, and negative heterosis for seed cotton yield, possibly due to insufficient number of bolls (Table 5).

Table 5. Heterosis values (%) of cross combinations for investigated traits

Hybrids	SCY	GP	ER	BW	SW	FF	FL	FS
TMN 199 × ADN 712	-3.34	1.01	8.37	11.7**	23.5**	3.64	-0.51	1.23
TMN 199 × GW 2357	-39.95**	5.37**	13.63**	9.3**	37.6**	2.44	-2.52**	-8.34*
TMN 199 × Ipek 607	-0.77	0.4	15.94**	13.1**	11.0**	4.65	-0.39	1.48
TMN 199 × TMD 139	-18.37*	5.86**	10.96*	4.3	21.1**	5.21**	-3.94*	0.051
TMN 199 × TMN 170	27.01**	-0.05	10.25*	2.9	31.7**	2.06	-2.51	-0.1
TMN 199 × UA 48	-44.01**	2.57*	10.31*	13.0**	22.6**	0.64	-1.11	-1.28
TMN 199 × ZN 1018	10.54	-0.39**	6.41	5.7*	21.6**	4.05	-0.36	1.07
Flash × ADN 712	20	0.66*	9.82	15.7**	17.5**	3.85	4.17*	-0.65
Flash × GW 2357	26.28*	0.91	17.67**	7.6*	14.8**	-0.37	-2.39	-2.73
Flash × Ipek 607	-33.07**	0.97	14.06**	11.1**	-4.4**	1.79	4.00*	6.73*
Flash × TMD 139	-0.48	7.86**	12.66**	1.5	20.5**	4.59	-1.51	-2.61
Flash × TMN 170	14.03	0.85	8.73	10.0*	27.9**	3.12	2.35	-2.6
Flash × UA 48	-1.18	4.66**	11.86*	8.7**	28.7**	2.15	0.19	-2.82
Flash × ZN 1018	4.26	3.30**	3.99	-1.6	11.6**	-3.41	1.83	-1.2
Gloria × ADN 712	-48.46**	2.00**	3.94	11.3**	34.2**	-0.23	-2.21	-1.83
Gloria × GW 2357	-21.59*	1.73**	13.53**	9.2**	5.1**	0.41	-1.13	0.25
Gloria × Ipek 607	29.15**	0.81	3.33	18.4**	-1.1**	1.66	1.99	-3.23
Gloria × TMD 139	-25.09**	-0.24	13.12**	16.4**	27.6**	4.74	-0.37	-7.42*
Gloria × TMN 170	-39.32**	0.46	-0.59	3.2	19.8**	-5.65**	2.4	-1.12
Gloria × UA 48	15.45	1.56**	-0.77	7.8**	18.8**	-4	1.3	-4.13
Gloria × ZN 1018	-1.56	0.73	9.37	5.4	14.7**	1.36	2.15	-0.09
ST 468 × ADN 712	18.68	-0.01	11.43*	9.3**	10.2**	1.7	2.27	3.29
ST 468 × GW 2357	-6.966	-2.2	15.93**	3.7	14.9**	-0.18	5.03*	18.02**
ST 468 × Ipek 607	-36.75**	-2.08	19.48**	16.2	-4.6**	-3.4	4.05*	10.47
ST 468 × TMD 139	0.69	6.16**	18.27**	-3.4	15.2**	3.99	-1.1	5.55
ST 468 × TMN 170	-4.4	0.43	3.24	5.8*	17.2**	1.31	0.32	6.71
ST 468 × UA 48	-10.41	1.57**	16.90**	9.9**	21.5**	-1.91	3.12	5.16
ST 468 × ZN 1018	-34.66**	-2.59**	15.55**	5.1	8.6**	0.03	1	2.21
Mean	-3.7	10.62	8.2	5.2	17.41	1.3	0.15	0.6

According to populations' average, positive hybrid vigor (heterosis) for boll weight and 100 seed weight were not reflected in yield due to earliness; thus, the hybrids produced less number of bolls and seed cotton yield. However, heterosis in ginning percentage (10.62%) partially reduced the reflection and decreased fiber production. While hybrid powers in fiber properties caused coarseness in fiber fineness, heterosis in fiber length and strength found weak. Our findings differed with Boyaci (2011) in terms of seed cotton yield and 100 seed weight, and exhibit similarities with Karademir (2004) in terms of ginning percentage, with Basbag et al. (2007) in terms of earliness, with Ali et al. (2016) in terms of boll seed weight, with Ashokkumar et al. (2013) in terms of fiber fineness and strength, and with Coban and Unay (2013) in terms of fiber length.

The heterobeltiosis values of the hybrids differed between 17.43% (Flash × GW 2753) and -57.30% (Gloria × ADN 712) for seed cotton yield, 4.70% (TMN 199 × TMD 139) and -13.28% (ST 468 × Ipek 607) for ginning percentage, 16.42% (ST 468 × TMD 139) and -12.21% (Gloria × TMN 170) for earliness rate, 18.3% (Gloria × Ipek 607) and -9.2% (ST 468 × TMD 139) for boll seed weight, 37.3% (TMN 99 × GW 2357) and -18.1% (ST 468 × Ipek 607) for 100 seed weight, 3.56% (TMN 199 × ZN 1018) and -11.92% (ST 468 × Ipek 607) for fiber fineness, 5.26% (ST 468 × Ipek 607) and 4.91% (Flash × UA 48) for fiber length, and 5.75% (Flash × Ipek 607) and -15.15% (ST 468 × GW 2357) for fiber strength (Table 6).

Table 6. Heterobeltiosis values (%) of cross combinations for yield and fiber properties

Variations	SCY	GP	ER	BW	SW	FF	FL	FS
TMN 199 × ADN 712	-19.72**	-1.38**	2.5	11.5**	19.3**	-0.36	-1.73	0.00
TMN 199 × GW 2357	-49.68**	-2.18**	12.90**	1.9	37.3**	0.36	-3.96*	-3.67
TMN 199 × Ipek 607	-9.53	-11.30**	7.15	7.4*	-4.9**	-5.69	-.6.64**	-1.53
TMN 199 × TMD 139	-29.28**	4.70**	3.88	-3.4	15.0**	-3.95	-4.07*	-0.61
TMN 199 × TMN 170	-36.86**	-0.13	4.58	-3.4	28.8*	-0.66	-4.07*	-1.67
TMN 199 × UA 48	-51.04**	-3.56**	9.02	9.2**	20.5**	-0.72	-5.36**	-10.46**
TMN 199 × ZN 1018	4.42	-1.50**	5.83	4.5	19.5**	3.56	-4.26*	-3.87
Flash × ADN 712	10.46	0.39	6.85	11.9**	10.8**	1.11	3.81*	-3.91
Flash × GW 2357	17.43	-0.39	7.81	3.4	11.7**	-0.91	-2.98	-9.57**
Flash × Ipek 607	-34.59**	-9.13**	13.72**	8.8**	-16.2*	-7.19	-3.32*	5.75
Flash × TMD 139	-3.82	4.4**	10.78*	-8.6**	11.7**	-3.38	-2.24	-5.27
Flash × TMN 170	9.96	-1.3**	-4.5	6.4**	22.0**	1.66	1.6	-3.12
Flash × UA 48	-3.53	0.42	4.22	8.4**	23.4**	2.09	-4.91**	-10.13**
Flash × ZN 1018	-1.78	2.24**	-4.63	-5.8	6.9**	-5.1	-1.31*	-7.91*
Gloria × ADN 712	-57.30**	1.52**	1.76	5.6	27.2**	-0.58	-5.13**	-9.01**
Gloria × GW 2357	-34.46**	0.21	4.62	6.9**	2.9*	-2.43	-4.33*	-10.53**
Gloria × Ipek 607	17.42**	-9.44**	2.98	18.3**	-13.8*	-5.28	-2.72	-6.60*
Gloria × TMD 139	-35.27**	-3.21**	11.94*	2.9	18.9**	-1.1	-2.31	-13.75**
Gloria × TMN 170	-47.65**	-1.54**	-12.21**	1.8	14.9**	-6.51	-1.04	-5.89
Gloria × UA 48	0.67	-2.73**	-6.99	6.0*	14.6**	-6.15	-1.29	-7.47**
Gloria × ZN 1018	-7.27	-0.098	0.89	-0.9	10.6**	-2.67	-3.56	-10.62**
ST 468 × ADN 712	7.48	-2.055	10.87*	7.9*	6.3**	-0.98	0.34	1.56
ST 468 × GW 2357	-14.9	-5.16**	9.5	-4.7	14.5*	-0.73	3.32	15.15**
ST 468 × Ipek 607	-37.77**	-13.28**	16.01**	8.9**	-18.1**	-11.92	-5.26**	4.22
ST 468 × TMD 139	-4.38	4.64**	16.42**	-9.2**	9.3**	-3.93	-4.00*	3.2
ST 468 × TMN 170	-9.4	0.009	-6.67	-2.1	14.5**	-0.12	-1.18**	2.07
ST 468 × UA 48	-14.06	-4.2**	12.35*	4.6	19.2**	-1.97	-4.19*	-7.05
ST 468 × ZN 1018	-37.39**	-3.34**	9.25	4.7	6.6	-1.72	0.08	0
Mean	-16	-2	5.3	7.1	11.9	-2.3	-2	-3.5

Except Gloria × Ipek 607, positive but non-significant heterobeltiosis values for seed cotton yield of other hybrids indicated that the trait is managed by dominant gene instead of additive genes and environment effect. The Flash × ADN 712 hybrid showed positive heterobeltiosis for all traits except fiber fineness and strength, Flash × GW 2357 for seed cotton yield and 100 seed weight, Flash × TMN 170 for seed cotton yield, boll seed weight, 100 seed weight and fiber length, Gloria × Ipek 607 for seed cotton yield, earliness rate, boll seed weight and 100 seed weight. In addition, positive heterobeltiosis values were observed for boll seed weight and 100 seed weight from the population averages. The positive heterobeltiosis values for earliness trait were not reflected positively on the number of bolls and seed cotton yield. Moreover, due to negative heterobeltiosis (% -2.0) recorded for ginning percentage, negative effect on fiber yield was appeared (Table 6).

CONCLUSION

The parents and hybrids significantly differed for the studied traits. In F_1 generation, dominant genes were more effective in the management of all studied traits, except fiber length. This indicated that plant selections should be started at F_3 and F_4. In addition, parents proved superior to hybrids for the studied traits, except fiber strength necessitates more care in plant selection due to low variation.

The hybrids TMN 199 × Ipek 607, TMN 199 × UA 48, TMN 199 × ZN 1018, Flash × GW 2357, Flash × TMN 170, Gloria × Ipek 607, Gloria × UA 48, ST 468 × ADN 712 proved significant for seed cotton yield. Similarly, Flash × ADN 712, Flash × ZN 1018 were significant for ginning percentage, whereas Flash × ADN 712, Gloria × TMD 139, Gloria × TMN 170 and Gloria × UA 48 remained significant for boll weight. Likewise, TMN 199 × GW 2357, TMN 199 × Ipek 607, Flash × TMN 170, Flash × UA 48, Gloria × ADN 712, ST 468 × GW 2357, ST 468 × Ipek 607 and ST 468 × UA 48100 were significant for 100 seed weight, while Gloria × TMN 170 proved significant for fiber fineness. In the same way, Flash × ADN 712, Gloria × ADN 712, ST 468 × GW 2357 were significant for fiber length, while Flash × Ipek 607 and ST 468 × GW 235 remained significant for fiber strength. Gloria × Ipek 607 proved the most promising hybrid with its F_1 combination in terms of high seed cotton yield, economical ginning percentage and earliness rate as well as superior fiber properties.

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DETERMINATION OF OIL QUALITY AND FATTY ACID COMPOSITIONS OF SOME PEANUT (*Arachis hypogaea* L.) GENOTYPES GROWN IN MEDITERRANEAN REGION

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ABSTRACT

This study was conducted to determine the performance of fifteen peanut lines from India and four peanut varieties (NC-7, Halisbey, Sultan and BATEM-5025) registered in Turkey. The present study was carried out in Osmaniye, which is under Mediterranean climate conditions as main crop season in 2020 and 2021. The experimental design was randomized complete block design (RCBD) with three replications. The highest oil content of peanut was observed in ICGV 10193 (52.16%±0.34), followed by ICGV 10179 (51.58%±0.35) and ICGV 16013 (51.47%±0.35). However, Oleic Acid / Linoleic Acid (O/L) ratio and iodine value are both indicators of peanut shelf life and oil stability. ICGV 15074 and ICGV 16013 came to forefront with high O/L ratio (9.46±1.46, 5.44±0.61) and low iodine value (72.68±0.60, 76.41±0.69), respectively. As a result of the study, it was concluded that some Indian peanut lines, mentioned above, can be proper to include breeding program due to their higher oil and oleic acid contents.

Keywords: *Arachis hypogea* L., fatty acid composition, iodine value, oil content.

INTRODUCTION

Peanut, also known as groundnut, is a valuable and highly nutritional product all over the world. Its seed contains 43-55% oil and 25-28% protein depending on market types and years along with essential mineral elements such as Na, Cu, Zn, Fe, Ca, Mg and K. In addition, it is a good source of E, K and B group vitamins. Due to these properties, peanut (*Arachis hypogea* L.) is important source of nutrition for both humans and animals (Bakal and Arioglu, 2019; Ergun and Zarifikhosroshahi, 2020; Yasli et al., 2020; Yilmaz et al., 2022).

In 2020, the World produced 53.7 million tonnes peanut with shells in 31.6 million hectares. Asia and Africa contributed to this production by about 90% of total production, and the rest was produced by Americas. The most important producer countries, accounting for more than half of the total production, were China (~18 million tonnes) and India (~10 million tonnes). In the same year, Turkey contributed to the total production by 215 927 tonnes in about 54 775 hectares. Even if Turkey had low contribution on production, Turkey doubled average yield compared to the World (FAO, 2022; TUIK, 2022).

Peanut oil includes seven major fatty acids which are oleic (C18:1), linoleic (C18:2), arachidic (C20:0), palmitic (C16:0), stearic (C18:0), behenic (C22:0) and lignoceric (C24:0) acids. Linoleic and oleic acids, two important acids, explain about 80% of the total fatty acid composition (Onemli, 2012; Yol and Uzun, 2018). The fatty acid composition plays an important role in determining nutrition, shelf life, and flavor of peanut. High oleic acid content, monounsaturated fatty acid, supplies an extended shelf life for peanut-derived products, and reduces cardiovascular disease risk and decreases low-density lipoprotein cholesterol levels. Also, linoleic acid is the most effective polyunsaturated fatty acid for lowering serum cholesterol (Mora-Escobedo et al., 2015; Yol and Uzun, 2018; Bakal and Arioglu, 2019).

The high oleic acid to linoleic acid (O/L) ratio and low iodine value (IV) provide a long shelf life and good stability. The stability of peanut seed oil and the degree of unsaturated fatty acid can be determined by using iodine value. A high O/L ratio (>10:1) in peanut results in an increased shelf life up to ten times and improved flavor compared to a normal O/L ratio (1.5:1) (Yol et al., 2017; Bakal and Arioglu, 2019). Peanut can be a valuable alternative crop for the irrigated areas of the Mediterranean basin which has suitable temperature regimes for both vegetative and reproductive growth of peanut (Caliskan et al., 2008a; Yol and Uzun, 2018).

Cil et al. (2016) carried out a field research on genotypes originated from India (3) and varieties registered in Turkey (9). It was reported that Indian genotypes came forefront by their oil content and yield, and could be used as a breeding material. Yol et al. (2017) conducted a field experiment in Western Mediterranean with 256 peanut genotypes, 186 of them Indian lines, and reported that the mean of oil content was 48.8% and unsaturated fatty acid was 78.7%. Asik et al. (2018), Bakal and Arioglu (2019), and Ergun and Zarifikhosroshahi (2020) reported that oil content and fatty acid compositions of peanut were affected by genotypes, years, seed maturity and environmental conditions such as temperature, precipitation, etc. It was found the oil content of peanut varied between 49.01-53.78%, 43.91-49.48% and 56.62-50.30%, respectively. Besides, it was indicated that unsaturated fatty acid composition was composed of about 80% of total fatty acid compositions.

The aims of the present study, the first research to use these Indian lines, were to; *i*. determine the performance of Indian lines in comparison with the varieties registered in Turkey, and *ii*. select the proper Indian lines for the breeding program.

MATERIALS AND METHODS

Materials

Fifteen lines (ICGV 10176, 10178, 10179, 10193, 10207, 10208, 10209, 10220, 15074, 16013, 16017, 06040 and 06099) originated from India and four varieties (NC-7, Halisbey, Sultan and BATEM-5025) registered in Turkey were used as plant materials in the study. Indian lines were provided from ICRASAT (International Crops Research Institute For The Semi-Arid Tropics) and other varieties, used as control, from Osmaniye Oil Seed Research. Experiments were carried out in the experimental fields of Osmaniye Oil Seed Research (37°03′41”N, 36°06′79”E; 50 m) in Eastern Mediterranean of Turkey during the main growing seasons of 2020 and 2021.

The pH of the clay soil used in the study was slightly alkaline (pH ~8). Lime content (~3%) and organic matter (~2%) of the soil were optimum. Total precipitation and average temperature during 2020 and 2021 growing period and long year were shown in Figure 1. The total precipitation was 237.3 mm in 2020 and 88.0 mm in 2021. Although long year (266.5 mm) was similar with 2020, it was different from 2021. This difference resulted from April and May, 2021. The average temperatures in the studied years and long year had no significant differences. The average temperatures were 24.8°C, 25.0°C and 24.3°C in 2018, 2019 and long year, respectively.

Methods

Experiments were conducted in a Randomized Complete Block Design (RCBD) with three replications. Each plot had 5 m long four rows with 70 × 15 cm spacing. Di-ammonium phosphate (18% N, 46% P_2O_5) fertilizer was used at the rates of 25 kg da^{-1} before sowing. Sowing was performed on April 12, 2020 in the first year and on April 29, 2021 in the second year. Hand weeding was performed with the emergence of the plants. The first irrigation was made when the plants flowered adequately and the drought was seen. The drip irrigation was done six times and for ten hours during the period from the beginning of the gynophore to the formation of pod in the studied years. Manuel harvests were performed on September 17, 2020 in the first year and on September 27, 2021 in the second year. Harvests were performed from two inner rows by taking into consideration side effects.

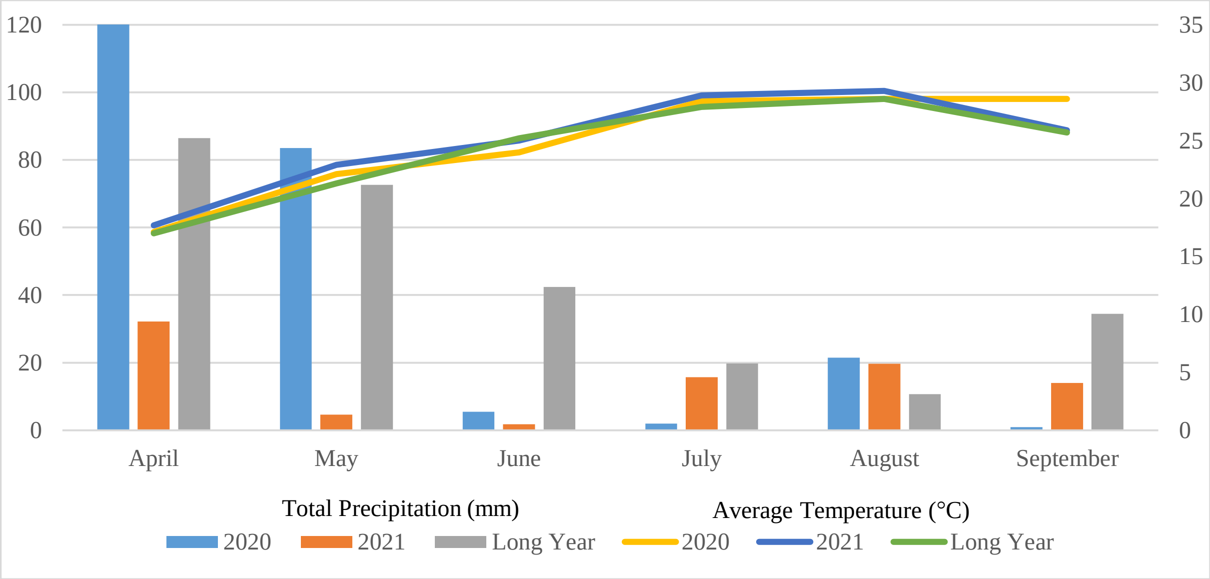

Figure 1. Climate parameters of the research field (2020, 2021 and long-year average)

Conventional soxhlet apparatus which consisted of a distillation flask, sample holder (thimble), siphon and condenser was used for extracting the seed oil, and solvent material was diethyl ether. The percent of peanut oil content was determined using the formula below:

$$\text{Oil content (\%)} = \frac{\text{Weight of oil extracted (g)}}{\text{Weight of the seed sample (g)}} \times 100$$

Fatty acid compositions of peanut seeds were analyzed by Thermo Scientific ISQ Single Quadrupole TR-Fame Gas Chromatography–Mass Spectrometry (GC-MS) system. Column had these properties; 5% Phenyl Polysilphenylene-silohexane, 0.25 mm inner diameter x 60 m length, 0.25 µm film thickness. Helium (99.9%) was used as a carrier gas which had 1 mL min^{-1} flow rate. The ionization energy was set at 70 eV and the mass range m / z 1.2-1200 amu. Scan Mode was used for data collection. The structure of each compound was identified with the Xcalibur software using mass spectra (Wiley 9).

Temperature program involved the following steps. The temperature of machine was warmed up to 120°C and waited for 1 min, increased by 10°C per minute until 175°C and waited for 10 min, increased by 5°C per minute until 210°C and waited for 5 min, and increased by 5°C per minute until 230°C and waited for 6 min. Split flow rate was 20 mL min^{-1}.

Iodine values (Chowdhury et al., 2015) and Oleic acid/Linoleic acid ratio were calculated with the help of the following formula:

$$\begin{aligned}\text{Iodine Values (IV)} \\ &= [(\text{oleic acid} \times 0.8601) \\ &+ (\%\ \text{linoleic acid} \times 1.7321)]\end{aligned}$$

$$\begin{aligned}\text{Oleic Acid/Linoleic Acid (O/L) Ratio} \\ &= \frac{\%\ \text{oleic acid (18:1)}}{\%\ \text{linoleic acid (18:2)}}\end{aligned}$$

Statistical Analysis

Experimental data were subjected to analysis of variance in accordance with Randomized Complete Block Design (RCBD) joined years with the aid of MSTAT-C and SPSS v22. Means were compared with the aid of Duncan's multiple range test (Steel and Torrie, 1980).

RESULTS AND DISCUSSION

Oil Content and Unsaturated Fatty Acid Compositions

Peanut oil is one of the most valuable protein and oil sources with its high oil and unsaturated fatty acid contents. The high oil content is an important quality parameter even if not by itself. The oil content was affected significantly ($p < 0.01$) by genotypes according to ANOVA. However, year and genotype × year interaction was no significant ($p > 0.05$) for oil content (Table 1). The oil contents obtained from genotypes in current study varied between 41.26-52.16% (Table 2). The highest oil contents were obtained from ICGV 10193, ICGV 10179 and ICGV 16013 with the value of 52.16%, 51.58% and 51.47%, respectively. This trio, was in same statistical group, had higher oil content compared to control varieties (NC-7, Halisbey, Sultan, BATEM-5025). There were no significant differences between years, and the oil contents of years were 46.97% in 2020 and 46.78% in 2021. Cil et al. (2016) conducted a field experiment with 9 varieties registered in Turkey and 3 Indian lines (ICGV 88365, ICGV 99085 and ICGV 00391), and observed variation in oil content in different genotypes of peanut, which was mainly driven by environmental conditions. It was reported that oil contents of Indian lines were about 50.6%. Yol et al. (2017) used 186 Indian lines as plant material to identify the oil and fatty acid profile of peanut and reported the average oil content as 48.8%. Ergun and Zarifikhosroshahi (2020) found an increase in oil content with change in genotype and growing conditions such as climatic conditions, environment and growing season. These findings were supported by most of researchers like Caliskan et al. (2008b), Cil et al. (2016), Onat et al. (2017), Yol et al. (2017), Asik et al. (2018) and Ergun and Zarifikhosroshahi (2020), who conducted their study in similar regions.

Table 1. Results of the analysis of variance for characteristics studied in the study.

Source of Variation	df	Oil Content	Oleic Acid	Linoleic Acid	Palmitic Acid	Stearic Acid	Behenic Acid	Arachidic Acid	Lignoceric Acid	O/L Ratio	Iodine Value
Year	1	ns	**	**	**	**	**	**	**	**	**
Block	4	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
Genotypes	18	**	**	**	**	**	**	**	**	**	**
Y x G	18	ns	**	**	**	**	**	**	**	**	**
CV (%)		2.65	4.01	6.54	5.35	3.97	4.29	6.55	19.06	9.86	3.35

df: Degree of freedom; ns: Non-significant; CV: Coefficient of variation; ** $p < 0.01$

The peanut seed is rich in monounsaturated (oleic acid) and polyunsaturated (linoleic acid) fatty acids which were affected significantly ($p < 0.01$) by genotype, year and their interaction (Table 1). Oleic acid (C18:1) varied between 33.57-68.30% with an average of 46.89% whereas linoleic acid (C18:2) ranged from 8.05 to 45.46% with a mean of 30.82%. Oleic and linoleic acid ratios were found as 47.48% and 31.66% in 2020, and 46.31% and 29.97% in 2021, respectively. It was found that the unsaturated fatty acid ratios decreased due to the insufficient precipitation in 2021 compared to that in the first year of the study (2020). Hassan et al. (2005) stated that fatty acid compositions were affected by environmental conditions such as temperature, moisture, etc. The highest and lowest oleic acid content was observed in ICGV 15074 and ICGV 10178, respectively. The genotypes, ICGV 16013 and ICGV 16017, from India had >50% oleic acid while ICGV 10217, ICGV 10218, ICGV 10220 and ICGV 10208 had >40% oleic acid. The maximum and minimum linoleic acid contents were detected in the lines ICGV 06040 (45.46%) and ICGV 15074 (8.05%), respectively. Considering all the genotypes together, the total averages of major unsaturated fatty acid compositions of peanut were about 77.71%. As can be seen in Figure 2, the genotype with the highest oleic acid content also had the lowest linoleic acid content. There was a negative correlation between oleic and linoleic acids. These findings were similar with those of Mora-Escobedo et al. (2015), Yol et al. (2017), Asik et al. (2018), Yol and Uzun (2018), Bakal and Arioglu (2019), Uckun et al. (2019) and Ergun and Zarifikhosroshahi (2020).

Table 2. Mean values of oil content, unsaturated fatty acids and oil quality parameters.

	Oil Content (%)	Oleic Acid (C18:1) (%)	Linoleic Acid (C18:2) (%)	O/L Ratio	IV
Varieties					
ICGV 10176	49.40±0.47 bc	34.80±0.85 jk	41.30±0.71 b	0.85±0.03 lm	101.47±0.74 bc
ICGV 10178	46.49±0.67 e	33.57±0.52 k	42.83±0.98 b	0.79±0.02 m	103.07±1.92 b
ICGV 10179	51.58±0.35 a	35.98±1.14 j	40.57±1.24 bc	0.90±0.05 km	101.23±1.67 bc
ICGV 10193	52.16±0.34 a	38.81±1.12 i	36.56±1.82 de	1.08±0.09 jl	96.71±2.36 d
ICGV 10207	43.26±0.64 f	39.47±1.55 i	36.16±2.83 e	1.15±0.13 ik	96.59±3.64 d
ICGV 10208	43.06±0.44 f	42.33±0.56 h	34.92±1.62 ef	1.23±0.07 hj	96.90±2.46 d
ICGV 10209	42.71±0.45 fg	39.50±1.31 i	35.72±1.05 ef	1.11±0.06 il	95.84±1.85 d
ICGV 10217	43.33±0.43 f	48.34±1.12 f	31.51±0.72 gh	1.54±0.07 fg	96.16±0.70 d
ICGV 10218	41.26±0.41 g	45.56±0.97 g	33.46±0.81 fg	1.37±0.05 gi	97.14±1.13 d
ICGV 10220	43.78±0.38 f	45.52±0.96 g	31.79±1.56 gh	1.45±0.06 gh	94.21±3.29 de
ICGV 15074	46.68±0.45 e	68.30±1.57 a	8.05±1.07 m	9.46±1.46 a	72.68±0.60 j
ICGV 16013	51.47±0.35 a	63.94±1.83 b	12.36±1.10 l	5.44±0.61 b	76.41±0.69 i
ICGV 16017	41.51±0.37 g	59.32±1.20 c	18.75±0.85 k	3.19±0.13 c	83.49±2.21 h
ICGV 06040	47.58±0.62 de	34.59±0.73 jk	45.46±0.62 a	0.76±0.02 m	108.48±0.94 a
ICGV 06099	48.96±1.09 cd	35.66±0.73 jk	38.74±1.97 cd	0.94±0.06 km	97.78±3.28 cd
NC-7	49.37±0.39 bc	59.84±1.19 c	21.60±0.67 j	2.79±0.14 d	88.89±1.04 fg
Halisbey	47.73±0.61 de	51.12±1.01 e	29.89±1.06 h	1.73±0.08 f	95.73±1.56 d
Sultan	49.55±0.47 bc	53.93±0.79 d	25.90±0.53 i	2.09±0.04 e	91.24±1.28 ef
BATEM-5025	50.75±0.24 ab	60.40±0.58 c	19.92±0.32 jk	3.04±0.05 c	86.44±0.82 gh
Years					
2020	46.97±0.47	47.48±1.59 A	31.66±1.52 A	2.35±0.37 A	95.67±1.33 A
2021	46.78±0.53	46.31±1.38 B	29.97±1.29 B	1.95±0.19 B	91.74±1.20 B
Mean	46.87±0.35	46.89±1.05	30.82±0.99	2.15±0.21	93.71±0.91

SEM: Standard Error of the Mean. Letters show different groups in each column.

Oil Quality Parameters

The ANOVA results of the genotypes showed a significant ($p < 0.01$) effect on oleic acid to linoleic acid (O/L) ratio and Iodine Value (IV) for all independent variables (Table 1). The low IV and high O/L ratio provide a long shelf life and good stability for peanut. Besides, high oleic acid content of peanut oil is valuable nutrient for augmented thermos-oxidative stability and human health (Ergun and Zarifikhosroshahi, 2020). The varieties of peanut have long shelf life when O/L ratio is equal or above 10, whereas most of varieties have normal O/L ratio as 1.5. The O/L ratios varied between 0.76-9.46 with the average of 2.15. As can be seen in Table 2, the maximum and minimum O/L ratios were observed in ICGV 15074 and ICGV 06040, respectively. Two lines among all genotypes, ICGV 16013 and ICGV 16017, had higher O/L values than 3 which were 5.44 and 3.19, respectively. Similarly, IV was affected significantly ($p < 0.01$) by genotypes, years and genotype × year interaction. IV of peanut genotypes ranged from 72.68 to 108.48 with an average of 93.71. The highest and lowest IV was detected in ICGV 06040 and ICGV 15074, respectively. Most of Indian lines' iodine values varied between 90-100. The desirable Iodine Values was observed in genotypes ICGV 15074 (72.68), ICGV 16013 (76.41) and ICGV 16017 (83.49). These three peanut lines came to forefront with their O/L ratio and IV. It has been reported that O/L ratio of peanut genotypes depends on the genetic structure of genotype and environmental factors (Chowdhury et al., 2015; Mora-Escobedo et al., 2015; Gulluoglu et al., 2016; Yol et al., 2017; Ergun and Zarifikhosroshahi, 2020; Yilmaz, 2022). These findings were similar with the results of these researchers.

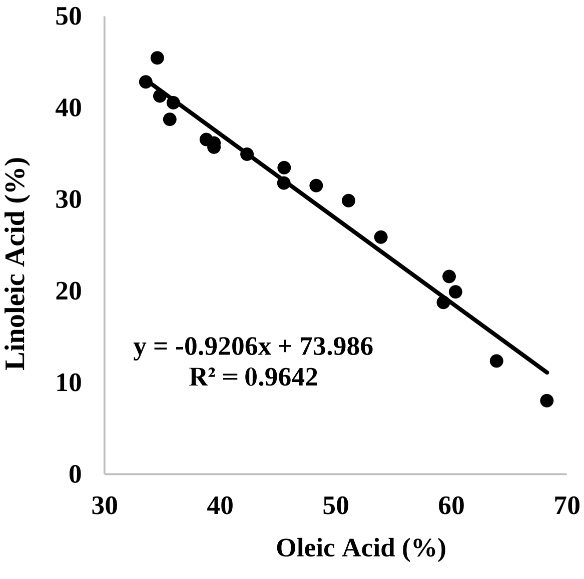

Figure 2. Relationship between linoleic and oleic acid.

Saturated Fatty Acid Compositions

Saturated fatty acids were affected significantly ($p < 0.01$) by genotype, year and their interaction (Table 1). Peanut oil is comprised of five major saturated fatty acids which are palmitic (C16:0), stearic (C18:0), behenic

(C22:0), arachidic (C20:0) and lignoceric (C24:0) acids (Table 3). Palmitic acid is more important among saturated acids due to its amount. The highest and lowest palmitic acid ranged from 9.73 to 12.59% with the mean of 11.22%. Only two within all the studied genotypes were below 10% and they were BATEM-5025 (9.73%) and NC-7 (9.98%), respectively. The lowest palmitic acid content was observed in ICGV 15074 (10.16%) among Indian lines. Stearic acid contents of peanut oil varied between 1.45-2.58% with an average of 2.02%. The maximum and minimum values were detected in ICGV 10208 and ICGV 06040, respectively. As regards to behenic acid, the values ranged from 1.60 to 4.05% and the mean was 2.84%. The lowest behenic acid was observed in control variety Sultan whereas ICGV 06040 had the minimum value of behenic acid (1.96%) among Indian lines. The highest and lowest arachidic acids were observed in ICGV 10207 with 2.30% and Halisbey with 1.24%, respectively. The average of arachidic value was 1.82%. Similar with results of stearic and behenic acids, ICGV 06040 had the lowest arachidic acid content with 1.30% within Indian lines. ICGV 10193 and NC-7 had the maximum and minimum lignoceric acid contents with 2.05 and 0.57%, respectively. The mean of the lignoceric acid was 1.09%. The lowest content was observed in ICGV 10217 with 0.61% among Indian lines. It was observed that saturated fatty acid ratios in 2021 were higher than those in the first year of the study (2020) due to the lower precipitation (Hassan et al., 2005). Saturated fatty acids are used in production of soaps, agricultural chemical and fatty alcohols. However, peanut oil is not adequate for commercial application compared to palm oil (Yol et al., 2017). These findings were in agreement with the results of Hassan et al. (2005), Hassan and Ahmad (2012), Mora-Escobedo et al. (2015), Yol and Uzun (2018), Uckun et al. (2019) and Ergun and Zarifikhosroshahi (2020). Isleib et al. (2008) and Onemli (2012) reported that fatty acid compositions are related to the plant growth habit. In the present study, fatty acid compositions of peanut genotypes were oleic acid (~47%), linoleic acid (~31%), palmitic acid (~11%), behenic acid (~3%), stearic acid (~2%), arachidic acid (~2%), and lignoceric acid (~1%) according to the mean of 2-year results (Figure 3).

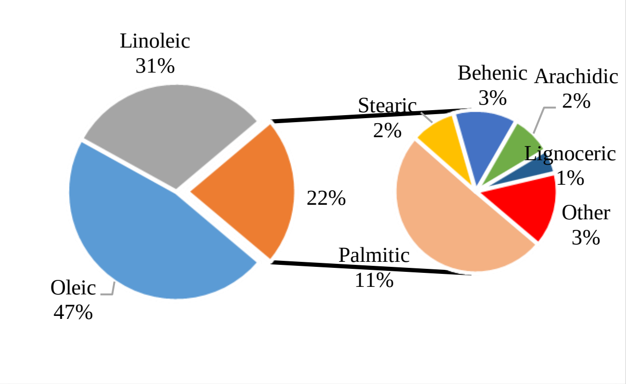

Figure 3. Fatty acid composition of peanut seed oil according to 2-year obtained data.

Table 3. Means of saturated fatty acid composition of varieties.

	Palmitic Acid (C16:0) (%)	Stearic Acid (C18:0) (%)	Behenic Acid (C22:0) (%)	Arachidic Acid (C20:0) (%)	Lignoceric Acid (C24:0) (%)
Varieties					
ICGV 10176	12.01±0.44 ab	1.55±0.04 h	3.03±0.09 f	1.92±0.09 bd	1.07±0.05 eg
ICGV 10178	12.10±0.40 ab	1.50±0.04 h	2.92±0.05 fg	1.87±0.14 cd	1.37±0.01 cd
ICGV 10179	11.91±0.44 ac	1.78±0.16 g	3.72±0.04 bc	1.88±0.11 cd	1.58±0.12 bc
ICGV 10193	11.65±0.49 bd	2.07±0.30 e	3.63±0.23 cd	1.77±0.12 df	2.05±0.42 a
ICGV 10207	12.29±0.71 ab	2.26±0.14 d	3.56±0.20 d	2.30±0.20 a	0.98±0.10 fg
ICGV 10208	12.22±0.22 ab	2.58±0.24 a	2.55±0.43 j	1.83±0.25 cde	1.27±0.36 de
ICGV 10209	11.20±0.43 ce	2.29±0.21 cd	3.79±0.24 b	2.25±0.15 a	1.58±0.28 bc
ICGV 10217	10.97±0.22 df	1.80±0.03 fg	2.18±0.37 k	1.32±0.19 g	0.61±0.14 h
ICGV 10218	10.91±0.13 dg	1.85±0.04 fg	2.85±0.23 gh	1.72±0.11 ef	1.22±0.24 ef
ICGV 10220	10.37±0.64 fi	1.89±0.06 f	3.19±0.21 e	1.97±0.12 bc	1.09±0.09 ef
ICGV 15074	10.16±0.40 gi	1.75±0.07 g	4.05±0.22 a	2.30±0.13 a	1.66±0.10 b
ICGV 16013	10.50±0.94 ei	2.37±0.06 c	3.53±0.08 d	2.21±0.03 a	1.02±0.05 eg
ICGV 16017	10.68±0.60 eh	2.37±0.27 c	2.74±0.51 hi	1.65±0.25 f	1.01±0.19 eg
ICGV 06040	12.36±0.98 ab	1.45±0.04 h	1.96±0.38 l	1.30±0.24 g	0.66±0.11 h
ICGV 06099	12.59±0.43 a	2.46±0.23 b	2.69±0.16 i	1.96±0.11 bc	0.80±0.07 gh
NC-7	9.98±0.42 hi	2.08±0.08 e	2.01±0.16 l	1.64±0.11 f	0.57±0.05 h
Halisbey	10.48±0.17 ei	1.81±0.02 fg	1.70±0.16 m	1.24±0.14 g	0.71±0.03 h
Sultan	11.19±0.21 ce	2.04±0.04 e	1.60±0.04 m	1.39±0.04 g	0.69±0.09 h
BATEM-5025	9.73±0.13 i	2.45±0.02 ab	2.20±0.07 k	2.04±0.04 b	0.98±0.05 fg
Years					
2020	10.49±0.16 B	1.80±0.4 B	2.63±0.09 B	1.66±0.04 B	0.91±0.04 B
2021	11.96±0.17 A	2.23±0.07 A	3.05±0.14 A	1.97±0.08 A	1.29±0.09 A
Mean	11.22±0.13	2.02±0.04	2.84±0.09	1.82±0.04	1.10±0.05

SEM: Standard Error of the Mean. Letters show different groups in each column.

Correlation

The Pearson correlation matrix is provided in Table 4. A significant negative correlation (r = -0.951) was observed between oleic and linoleic acids, as expected. This relationship was also reported by Yol et al. (2017) and Bakal and Arioglu (2019). Behenic acid had a strong positive correlation with arachidic acid (r = 0.835) and lignoceric acid (r = 0.737) whereas oleic acid and palmitic acid had a negative correlation (r = -0.558). There was a significant and positive correlation between lignoceric and arachidic acid (r = 0.547) even not as strong as the other relationships. Yol et al. (2017) also observed that palmitic acid had positive correlation with low values with all saturated fatty acids.

Table 4. Correlation coefficients among the fatty acid compositions based on average of two years.

	Oleic Acid	Linoleic Acid	Palmitic Acid	Stearic Acid	Behenic Acid	Arachidic Acid
Oleic Acid	1					
Linoleic Acid	**-.951**	1				
Palmitic Acid	**-.558**	**.421**	1			
Stearic Acid	**.217**	**-.360**	**.191**	1		
Behenic Acid	-.109	-.101	.173	**.330**	1	
Arachidic Acid	.028	**-.225**	.156	**.531**	**.835**	1
Lignoceric Acid	-.136	-.030	**.191**	**.349**	**.737**	**.547**

$p < 0.05$ in bold.

CONCLUSION

O/L ratio and IV are both indicators of peanut shelf life and oil stability. High O/L ratio and low IV provide extensive shelf life, better stability and high quality of oil. ICGV 15074 and ICGV 16013 came to forefront with high O/L ratio (9.46±1.46, 5.44±0.61) and low IV (72.68±0.60, 76.41±0.69), respectively. Besides, ICGV 10193 had higher oil content (52.16%±0.34) compared to ICGV 15074 (46.68%±0.45). In addition to these peanut lines, ICGV 10179 (51.58%±0.35) and ICGV 16013 (51.47%±0.35) may be recommended for peanut breeding program due to their high oil content.

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EVALUATION OF WHEAT GENOTYPES: GENOTYPE × ENVIRONMENT INTERACTION AND GGE BIPLOT ANALYSIS

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ABSTRACT

This research was carried out to evaluate the grain yield, yield traits and some quality traits of 18 bread wheat genotypes at seven environments in Thrace region using principal component analysis and genotype (G) + genotype × environment interaction biplot analysis to determine the genotypes with high yield and desired quality characteristics during the 2016-2017 and 2017-2018 growing seasons. The experiments were arranged in a randomized complete block design with four replications. Genotype, environment and genotype × environment interactions (GE) were found statistically significant at $p \leq 0.01$ level for all investigated traits. Mean values of the cultivars varied from 4841-6807 kg ha^{-1} for grain yield, 118.6-131.6 days for heading date, 80.4-104.7 cm for plant height, 7.7-10.4 cm for spike length, 16.4-20.3 for number of spikelets per spike, 34.2-59.6 number of grains per spike, 1.49-2.41 g grain weight per spike, 72.0-77.8 kg hl^{-1} for test weight and 36.6-45.3 g for thousand kernel weight. Principal component biplot analyses explained the relationships between the investigated traits and genotypes at a ratio of 60.9%. According to the principal component (PC) biplot analysis, it was observed that there was a positive and significant relationship between grain yield and test weight, a negative relationship with grain yield and spike length and grain weight per spike. GGE biplot analysis explained 82.65% of the relationship of G + GE for grain yield. According to the GGE biplot analysis, two mega environments were determined and Lucilla and Glosa genotypes took place in the biggest mega environment consisted of four environments as superior genotypes.

Keywords: Environment, GGE-biplot, grain yield, wheat

INTRODUCTION

Due to its wide adaptation ability, high nutritional value, cultivation area, production and yield potential, the bread wheat *(Triticum aestivum* L.*)* has an important role both the world and the Turkey (Kaydan and Yagmur, 2008). The issue of adequate and balanced nutrition is becoming more and more important with each passing day (Dogan and Kendal, 2013; Kilic et al., 2014). Due to the rapid increase in population, the increase in the need for food and the decrease in planting areas require much more effort to develop high yielding and quality cultivars. Wheat yield and quality differ from year to year due to genetic structure of varieties, climatic conditions, soil structure of production areas, abiotic and biotic stresses and cultural practices (Gungor and Dumlupinar, 2019). It is important to determine wheat varieties with high yield and quality, suitable for different ecological conditions and to expand their production in order to reduce the effect of environmental factors (Oral et al., 2018; Aydogan et al., 2020).

According to the recent official data (Anonymous, 2021), during the year 2020, 20.5 million tons of wheat was produced from an area of 6.9 million hectares in Turkey. Four million tons of durum wheat (3180 kg ha^{-1}) was produced from 1.2 million ha, and 16.5 million tons of bread wheat was produced from 5.6 million ha (2910 kg ha^{-1}). Wheat cultivation is carried out on 48% (449.523 ha) of the agricultural lands of the Thrace region (935.259 ha), where includes the provinces of Tekirdag, Edirne and Kirklareli. One million and seven hundred thousand tons of wheat was produced in the Thrace region in 2020, and the average grain yield (3810 kg ha^{-1}) is higher than the average of Turkey (2910 kg ha^{-1}).

Due to the importance of wheat farming in the region, suitable conditions for wheat farming in the Thrace region and the increase in the efficiency of high-yielding, high-quality and stable genotypes that are well adapted to the region will help increase production.

Since Thrace region has suitable conditions for growing wheat and the determination of high yield, quality and stable varieties that are well adapted to the region will benefit to increase the production. Climate and ecological characteristics, which are the main factors that make up the environment, affect the quality of genotype performance positively or negatively.

For this reason, it is desired to develop stable genotypes that perform across the environments. It is important to determine the GE interactions by testing the genotypes in different environments before their production in large areas. One of the most effective methods to determine GE interactions is the GGE (genotype × genotype environment) biplot analysis (Yan et al., 2000; Yan, 2001; Yan and Kang, 2003). The most important reasons that the method preferred by the researchers may be shown as the interactions of genotypes with different environments and might be presented in a simple and understandable way. Many features of genotypes can be displayed graphically, and it allows comparing the relationships between genotypes and features visually (Acikgoz et al., 2007; Ilker et al., 2009; Akcura, 2011; Sahin et al., 2011; Kilic et al., 2012; Basbag et al., 2021; Sayar et al., 2022). In recent years, many researchers working on different plant species have used the graphs created by this analysis (Sayar et al., 2013; Kilic et al., 2014; Kendal and Sayar, 2016; Sayar and Han, 2016; Oral et al., 2018; Wardofa et al., 2019; Tulu and Wondimu, 2019; Aktas, 2020).

In this study, it was aimed to evaluate the grain yield, yield components and some quality characteristics of some bread wheat cultivars at different locations in the Thrace region by using PCA and GGE biplot analysis and to determine the genotypes with high yield and adaptability characteristics in different environments.

MATERIALS AND METHODS

In the research, 18 commercial bread wheat cultivars, which are commonly produced in the Thrace region, were used as materials (Table 1). The trials were conducted in seven different environments, at Kirklareli, Tekirdag and Edirne locations, representing the entire Thrace region, during the 2016-2017 and 2017-2018 plant growing seasons (Table 2).

Table 1. Bread wheat cultivars used in the this study.

Cultivars	Owner Company/Institute	Date of registration	Growth type
Masaccio	ProGen Seed Company	2014	Alternative
Lucilla	ProGen Seed Company	2017	Alternative
Kate A-1	Trakya Agricultural Research Institute	1988	Alternative
Gelibolu	Trakya Agricultural Research Institute	2005	Winter
Tekirdag	Trakya Agricultural Research Institute	2005	Winter
Kopru	Trakya Agricultural Research Institute	2015	Winter
Rumeli	Trakya Agriculture Seed Company	2012	Winter
Pehlivan	Trakya Agricultural Research Institute	1998	Winter
Asli	ProGen Seed Company	2017	Winter
Flamura-85	Tareks Seed Company	1999	Winter
Glosa	Tareks Seed Company	2014	Winter
Selimiye	Trakya Agricultural Research Institute	2009	Winter
Esperia	Tasaco Seed Company	2011	Winter
Midas	ProGen Seed Company	2014	Winter
Saban	Trakya Agricultural Research Institute	2014	Winter
Krasunia odes’ka	Yildiz Seed Company	2008	Winter
Aldane	Trakya Agricultural Research Institute	2009	Alternative
Bereket	Trakya Agricultural Research Institute	2010	Winter

Table 2. Growing seasons, code, soil type and amounts of precipitation of environments.

Growing seasons	Code	Environment	Soil type	Precipitation (mm)
2016-2017	E1	Luleburgaz	Clayey loamy	366.3
2016-2017	E2	Edirne	Clayey loamy	408.0
2016-2017	E3	Babaeski	Clayey	366.3
2016-2017	E4	Hayrabolu	Clayey	451.7
2017-2018	E5	Luleburgaz	Clayey loamy	696.3
2017-2018	E6	Babaeski	Clayey loamy	696.3
2017-2018	E7	Kesan	Clayey	799.6

The research was arranged in a Randomized Complete Block Design (RCBD) with four replications. Sowing was done between the end of October and the first week of November in both growing seasons, and it was done by hand in 5 m long plots with 20 cm row spacing and 6 rows with 500 seeds per m^2. In the research, the plot sizes were 6 m^2 both at planting and at harvest (6 m x 1 m). Weed control was done by hand in the trial plots and no application was made for diseases and pests.

Fifty kg ha^{-1} nitrogen and 50 kg ha^{-1} phosphorus were applied at the sowing, and the top dressing was divided into two parts: 90 kg ha^{-1} nitrogen during tillering and 60 kg ha^{-1} nitrogen during jointing stage. Harvest was done in the first week of July in both growing seasons.

Plant height, heading date (the number of days between January 1st and the day when the plants are 50% spike in each plot), spike length, number of spikelets per spike, number of grains per spike, grain weight per spike, thousand kernel weight, test weight (Vasiljevic et al., 1980), and grain yield were all evaluated in the study (Dokuyucu et al., 2004; Karaman, 2017)

Statistical Analyses

The data obtained for two years were subjected to combined analysis of variance, analyses each year and location were considered as environments. A two-way fixed effect model was applied to determine the influence of the main effects of variation (GE) and their interaction (GE) on each trait, and the Duncan test was performed to compare the means. Principal component analysis was calculated over the average data and evaluated with the biplot approach (SAS Institute Inc. JMP 15.1, 2020). GGE Biplot analyzes were calculated with Genstat 14th (VSN International Ltd., 2011) software over seven environments using average data (Yan, 2001).

RESULTS AND DISCUSSION

Genotype by Environment Interaction (GEI)

The study conducted with 18 bread wheat cultivars, the effects of genotype, environment and GE interaction were found to be statistically significant at the $p \leq 0.01$ level in terms of all traits examined (Table 3).

The effect of G, E and GE interaction on grain yield were 28.37%, 47.91% and 23.72% respectively (Table 3). It has been determined that the effect of environmental variation on grain yield is higher than other variation sources. In previous studies conducted by different researchers, it was stated that the effect of genotype on grain yield ranged from 1.3 to 33.46%, environment 35.28-90.76%, and GE interaction between 7.12-31.45% (Kaya et al., 2002, Kaya et al., 2006, Mohammed, 2009, Oral et al., 2018; Tulu and Wondimu, 2019; Wardofa et al., 2019; Aktas, 2020). While the cultivars' average grain yield was 5991 kg ha^{-1}, the cultivar Lucilla had the highest grain yield (6807 kg ha^{-1}) and the lowest grain yield was obtained from the cultivar Aldane (4841 kg ha^{-1}) (Table 4). The lowest grain yield was found at the E1 (5469 kg ha^{-1}) location, and the highest grain yield was obtained at the E2 (7251 kg ha^{-1}) location (Table 6). Grain yield is a multi-gene controlled trait and is affected by many factors such as the genetic potential of the genotype, the ecology of the region and the applied cultural practices (Kaydan and Yagmur, 2008; Sahin et al., 2016; Aktas et al., 2017; Tekdal et al., 2017; Gungor and Dumlupinar, 2019; Aydogan et al., 2020; Takač et al., 2021).

Table 3. Mean square values for the investigated traits

Sources of variations	**Genotype (G)**	**Environment (E)**	**G x E**	**Error**	**C.V. (%)**
Degrees of Freedom	17	6	102	357	
Grain Yield/	63540.8**	303984.7**	8852.3**	4296.1	10.9
Variability (%)	28.37	47.91	23.72		
Heading Date/	235.38**	308.39**	7.66**	0.38	0.51
Variability (%)	60.33	27.90	11.78		
Plant Height/	1108.68**	1716.59**	85.22**	12.11	3.75
Variability (%)	49.81	27.22	22.97		
Spike Length/	11.18**	19.38**	1.25**	0.41	6.95
Variability (%)	43.76	26.78	29.46		
No of Spikelet /Spike/	24.67**	51.23**	4.61**	1.66	6.77
Variability (%)	35.04	25.68	39.28		
No of Grain/Spike/	950.87**	4380.40**	200.36**	42.31	13.60
Variability (%)	25.71	41.79	32.50		
Grain Weight/Spike/	1.52**	16.33**	0.49**	0.12	16.75
Variability (%)	14.90	56.30	28.81		
Test Weight/	63.36**	827.50**	12.83**	0.32	0.75
Variability (%)	14.65	67.55	17.80		
Tohusand Kernel Weight/	199.16**	1186.38**	21.64**	0.37	1.51
Variability (%)	26.64	56.00	17.37		

** Significant at the $P < 0.01$ probability level, * Significant at the $P < 0.05$ probability levels.

The effect of genotype on the heading date was determined as 60.33%, the effect of the environment as 27.90%, and the effect of GE interaction as 11.78% (Table 3). The mean heading date of the cultivars was 122.2 days and the earliest one was 118.6 days (Glosa), and the latest cultivar was Midas with 131.6 days (Table 5). According to the environmental averages, the date of heading varied from 119.4-125.8 days (Table 6). The latest heading date was obtained at the E3 (125.8 days) location and the shortest at the E6 (119.4 days) location. According to previous researchers, it has been stated that the duration of heading date varies between 103-171 days, which were conducted in different ecological conditions, and genotypes and environments were effective together in the formation of significant differences in terms of the duration of heading date. (Kilic et al., 2012; Dogan and Kendal, 2013; Sakin et al., 2017; Gungor and Dumlupinar, 2019; Akan et al., 2021).

Table 4. Mean grain yield (kg ha^{-1}) of cultivars across test environments

Cultivars	**E1**	**E2**	**E3**	**E4**	**E5**	**E6**	**E7**	**Mean**
Masaccio	6044	9004	6504	6269	5942	6346	7274	6769 a
Lucilla	5871	8344	6102	6702	6325	6621	7687	6807 a
Kate A-1	5867	6452	4965	5911	5854	5173	6952	5882 efg
Gelibolu	5794	6882	5269	5961	5858	5100	6835	5957 def
Tekirdag	5717	5588	4717	6396	5604	5559	5449	5576 gh
Kopru	5613	7413	5419	5900	5384	5875	6843	6064 c-f
Rumeli	5611	7954	6361	5938	5692	5638	7143	6334 bc
Pehlivan	5600	6506	5073	6238	5417	5138	5257	5604 gh
Asli	5459	7581	6162	5892	5717	5946	6432	6170 cde
Flamura-85	5450	7211	5669	5602	5817	5254	6297	5900 d-g
Glosa	5423	8217	6140	6788	6058	5996	7556	6597 ab
Selimiye	5398	6952	5488	5933	5571	6058	6035	5919 d-g
Esperia	5340	6371	5133	5483	5804	5792	6126	5721 fgh
Midas	5244	8598	5619	5971	5913	5745	6525	6230 cd
Saban	5223	7455	6340	6256	4546	5525	6441	5969 def
Krasunia odes’ka	5038	7856	5473	5963	6025	5767	5985	6015 c-f
Aldane	4890	5340	4169	5290	3958	5633	4610	4841 ı
Bereket	4867	6792	4944	5229	4246	6004	6266	5478 h
Mean	5469 e	7251 a	5530 de	5984 c	5541 de	5732 d	6428 b	5991

The effect of genotype on plant height was 49.81%, the effect of environment was 27.22%, and the effect of GE interaction was determined as 22.97% (Table 3). The average plant height of the genotypes varied from 80.4-104.7 cm (Table 5). The longest plant height was obtained from the cultivar Midas (104.7 cm), and the shortest plant height was obtained from the cultivar Esperia (80.4 cm). According to the environmental averages, the highest plant height was found at the E3 (99.6 cm) location, and the shortest plant height was determined at the E4 (87.4 cm) location (Table 6). Plant height is a crucial vegetative factor for the genotypes’s adaptation to the area, and it can affect yield and quality indirectly (Dogan and Kendal, 2013). Plant height in wheat can vary depending on genetic structure, climate and soil characteristics and cultural practices applied, and it has been determined that it varies between 71-125 cm in previous works (Dogan and Kendal, 2012; Dogan and Kendal, 2013; Sakin et al., 2017; Gungor and Dumlupinar, 2019; Akan et al., 2021).

The effect of genotype on spike length was determined as 43.76%, the effect of environment as 26.78% and the effect of GE interaction as 29.46% (Table 3). The spike length of the genotypes varied from 7.7-10.4 cm. The highest spike length was measured in the cultivar Kate A-1 (10.4 cm) and the lowest spike length was measured in the cultivar Massacio (7.7 cm) (Table 5). When the environemental averages were examined, they had the highest spike length at the E7 (10 cm) location and the lowest spike length at the E2 and E4 (8.7 cm) locations (Table 6). In the studies carried out in different ecological conditions, the spike length varied from 7.3 to 10.35 cm. (Sakin et al., 2017; Gungor and Dumlupinar, 2019).

The effect of genotype on the number of spikelets per spike was determined as 35.04%, the effect of environment as 25.68%, and the effect of GE interaction as 39.28% (Table 3). The average number of spikelets per spike of genotypes ranged from 16.4 to 20.03. Cultivar Gelibolu (20.3) had the highest number of spikelets per spike, and cultivar Massacio (16.4) had the lowest (Table 5). According to the environmental averages, the highest number of spikelets per spike was found at the E7 (20.3) location and the lowest at the E2 (18.0) location (Table 6). Kurt and Yagdi (2013) reported a 17.3 to 19.5, Gungor and Dumlupinar (2019) showed a 16.5-21.2, while, Akan et al. (2021) indicated an 18.15-22.13 number of spikelets per spike.

The difference in the number of fertile spikelets and florets in the spike according to the genotypes is the source of the difference in the number of grains in the spike (Bayram et al., 2017). The effect of genotype on the number of grains per spike was 25.71%, the effect of environment was 41.79%, and the effect of GE interaction was 32.50% (Table 3). According to the genotype averages, the number of grains per spike varied from 34.2 and 59.6, and the highest number of grains per spike was found in the cultivar Midas (59.6) while the lowest in the cultivar Massacio (34.2) (Table 5). According to the environmental averages, E7 location had the highest value (57.5) in terms of the number of grains per spike, while E2 location had the lowest value (40.4) (Table 6). In other studies, Bayram et al. (2017), 13.7-26.6; Gungor and Dumlupinar (2019), 27.2-49.7; Kara et al. (2016), 34.4-54, Ozen and Akman (2015), 22-46, Aktas et al. (2017) reported that it ranked from 42.21 to 52.34. The difference in the number of grains in the spike varies according to the genetic structure of the genotypes and climatic characteristics. (Ozen and Akman, 2015; Kara et al., 2016; Aktas et al., 2017; Bayram et al., 2017; Gungor and Dumlupinar, 2019).

Table 5. Means of yield components and quality traits of 18 bread wheat cultivars

Cultivars	HD	PH	SL	SNS	GNS	GWS	TW	TKW
Masaccio	121.8 fg	87.0 h	7.7 e	16.4 ı	34.2 ı	1.49 f	75.6 f	38.8 g
Lucilla	121.5 gh	90.3 g	9.5 b	18.9 d-g	50.9 cd	1.77 e	77.0 bc	38.3 h
Kate A-1	121.4 ghı	101.6 b	10.4 a	18.8 e-h	50.8 cd	2.09 cd	76.4 d	38.6 gh
Gelibolu	119.9 j	90.5 g	8.9 c	18.7 e-h	50.0 cd	2.19 bc	77.3 b	42.2de
Tekirdag	118.6 k	85.7 h	9.5 b	19.5 bcd	46.2 ef	2.11 cd	74.9 g	41.9 e
Kopru	123.2 d	94.1 e	9.4 b	20.3 a	55.1 b	2.41 a	73.9 ı	42.2 de
Rumeli	121.6 gh	96.3 d	8.9 c	19.8 abc	47.5 de	1.94 de	77.3 b	37.1 j
Pehlivan	122.4 e	101.1 b	9.4 b	18.9 d-g	39.3 h	1.99 d	76.9 c	45.3 a
Asli	125.6 b	99.1 c	9.0 c	19.0 def	45.6 ef	1.81 e	75.6 f	37.0 j
Flamura-85	122.9 d	91.2 g	9.7 b	18.3 gh	45.8 ef	2.10 cd	77.8 a	43.0 bc
Glosa	118.8 k	86.5 h	8.5 d	18.4 fgh	47.9 de	2.00 d	76.0 e	38.9 g
Selimiye	122.1 ef	90.4 g	9.0 c	18.3 gh	41.5 gh	1.79 e	76.5 d	42.3 d
Esperia	121.6 gh	80.4 ı	8.8 cd	19.3 cde	50.8 cd	2.12 cd	74.6 gh	36.6 k
Midas	131.6 a	104.7 a	8.7 cd	20.1 ab	59.6 a	2.37 ab	76.5 d	37.2 j
Saban	119.9 j	90.6 g	9.6 b	19.1 de	48.9 cde	2.20 bc	75.4 f	42.8 c
Krasunia odes'ka	124.1 c	91.4 fg	10.3 a	20.0 ab	52.3 bc	2.35 ab	74.4 h	38.0 ı
Aldane	121.4 hı	93.2 ef	9.4 b	18.1 h	43.8 fg	2.00 d	74.1 ı	43.2 b
Bereket	121.1 ı	98.2 c	9.6 b	19.9 abc	50.2 cd	2.18 bc	72.0 j	40.3 f
Mean	122.2	93.3	9.3	19.1	48.6	2.1	75.7	40.3

HD: Heading date, PH: Plant height, SL: Spike length, SNS: Number of spikelets per spike, GNS: Number of grains per spike, GWS: Grain weight per spike, TW: Test weight, TKW: Thousand kernel weight

The effect of genotype on grain weight per spike was 14.90%, the effect of environment was 56.30%, and the effect of GE interaction was determined as 28.81% (Table 3). The cultivar Kopru had the highest grain weight per spike of 2.41 g, while the cultivar Masaccio had the lowest grain weight per spike of 1.49 g. (Table 5). According to environmental averages, grain weight per spike ranged from 1.58 to 2.81 g, with the maximum value at the E7 (2.81 g) location and the lowest value at the E1 (1.58 g) location (Table 6). In studies conducted in different environments, the grain weight per spike was determined as 1-2 g, (Ozen and Akman, 2015), 1.4-1.9 g (Kara et al., 2016), 1.9-2.6 g (Aktas et al., 2017) and 0.93-2.25 g (Gungor and Dumlupinar, 2019).

Table 6. Average of yield component and quality traits in seven environments

Environments	HD	PH	SL	SNS	GNS	GWS	TW	TKW
E1	122.6 c	95.6 c	9.0 d	19.0 c	41.9 c	1.58 d	75.5 e	35.8 g
E2	122.3 d	92.3 d	8.7 e	18.0 d	40.4 c	1.94 c	80.8 a	45.3 b
E3	125.8 a	99.6 a	9.1 d	18.3 d	42.1 c	1.66 d	77.7 b	39.3 d
E4	123.4 b	87.4 f	8.7 e	18.4 d	42.2 c	1.61 d	76.0 d	37.1 f
E5	120.4 f	89.7 e	9.5 c	19.5 b	53.9 b	2.34 b	73.4 f	37.6 e
E6	119.4 g	87.9 f	9.8 b	19.7 b	56.7 a	2.41 b	69.9 g	40.1 c
E7	121.4 e	97.9 b	10.0 a	20.3 a	57.5 a	2.81 a	76.4 c	46.2 a
Mean	122.1	91.4	9.2	18.8	47.1	2.0	75.6	39.9

HD: Heading date, PH: Plant height, SL: Spike length, SNS: Number of spikelets per spike, GNS: Number of grains per spike, GWS: Grain weight per spike, TW: Test weight, TKW: Thousand kernel weight

The effect of genotype on test weight was found to be 14.65%, the effect of environment was 67.55%, and the effect of GE interaction was determined as 17.80% (Table 3). The cultivar Flamura-85 (77.8 kg hl^{-1}) had the highest test weight, while the cultivar Bereket (72.0 kg hl^{-1}) had the lowest (Table 5). According to the environmental averages, the lowest test weight was determined at the E6 location (69.9 kg hl^{-1}), and the highest at the E2 (80.8 kg hl^{-1}) location (Table 6). In studies conducted by different researchers (Schuler et al., 1994, Diepenbrock et al., 2005; Ozen and Akman, 2015; Kara et al., 2016; Mut et al., 2017; Gungor and Dumlupinar, 2019), it has been determined that the test weight varied from 69.3-82 kg hl^{-1}. In terms of test weight, values above 82 kg hl^{-1} are considered perfect, but at least 72 kg hl^{-1} should be preferred. Test weight is a quality parameter that determines the flour yield in the flour industry has commercial importance and is desired to be high (Mut et al., 2017). They reported that test weight is especially influenced by the environment and can be affected depending on factors such as genotype and agronomic practices.

Environmental factors and climatic conditions affect the thousand kernel weight significantly. The effect of genotype on thousand kernel weight was determined as 26.64%, the effect of environment as 56.00%, and the effect of GE interaction as 17.37% (Table 3). The highest thousand kernel weight was obtained in cultivar Pehlivan (45.3 g), and the lowest value was obtained in cultivar Esperia (36.6 g) (Table 5). According to the environmental averages, the thousand kernel weight varied from 37.6-46.2 g, the lowest thousand kernel weight was obtained at the E1 (35.8 g) location, and the highest thousand kernel weight was obtained at the E7 (46.2 g) location (Table 6). In other studies, it has been reported that the thousand kernel weight varied from 29.2-47.2 g. (Ozen and Akman, 2015; Mut et al., 2017; Tekdal et al., 2017; Gungor and Dumlupinar, 2019; Aydogan et al., 2020).

Principal Component (PCA) and GGE-Biplot Analysis

Principal component analysis resulted in a two-dimensional PCA score accounted for 60.9% of the total variation (Figure 1). Principal component 1 had a value of 23.5%, which indicates the genotype effect that was low in this case, and PC2 was 37.4%, which demonstrates the environment effect that was high in the study. Many researchers reported that higher total variation value (PC1+PC2) (≥50%) ensures to more reliable interpretation of a biplot graph (Sayar and Han, 2015; Kendal et al., 2016; Basbag et al., 2021; Sayar et al., 2022). Additionally; it was reported that when the angle between the vectors representing the features from 0° to 90°, there is a positive relationship. (Ilker et al., 2009; Sayar and Han, 2015; Dogan et al., 2016; Karaman, 2020). Grain yield was found to have a negative relationship with spike length, thousand kernel weight and grain weight per spike, but a positive relationship with test weight. It was determined that there was a positive relationship between the number of grains per spike, the number of spikelets per spike, plant height and heading date. Since the plant height and test weight traits had a short vector, their effects within the variation were determined to be lower than the other traits. Karaman (2020) reported that grain yield had a positive relationship with test weight and thousand kernel weight traits. Kahraman et al. (2021) stated that there was a positive relationship between grain yield and thousand kernel weight, and a negative relationship between grain yield and test weight.

Yan et al. (2000) stated that according to the GGE biplot analysis method, the genotypes located at the corners of the polygon were the genotypes with the highest value or the ideal characteristics for the related characters. The cultivars evaluated in the research were found superior; Midas for heading date, Lucilla, Masaccio Rumeli and Asli cultivars for grain yield, Aldane for thousand kernel weight, Bereket for spike length. In addition, Esperia and Kate A-1 cultivars were more stable than other genotypes.

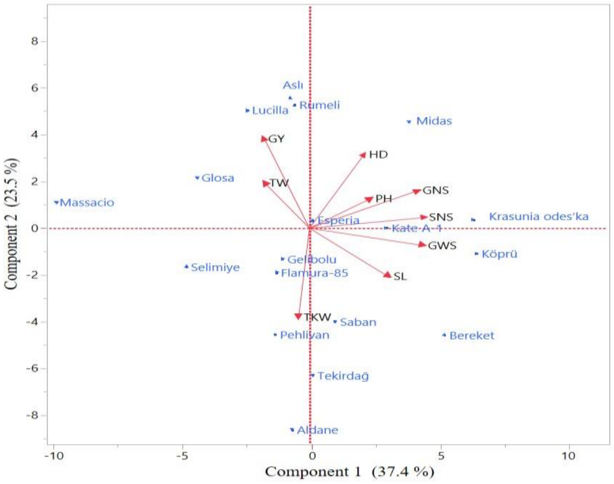

Figure 1. Relationship among genotypes and investigated for grain yield

GGE Biplot is a software program that analyzes graphs. With PC1 70.53%, PC2 12.12%, and total PC1+PC2 82.65%, a scatter plot graph demonstrated the relationship between genotype + GE. E5, E1, E4 and E7 locations developed a similar environment, as did E3, E2, E6 locations, resulting in two different mega-environments. Cultivars Rumeli, Asli and Masaccio were located in the mega environment formed by E2, E3, E6 locations, while cultivars Lucilla, Glosa and Gelibolu were located in the same zone with the mega environment formed by E1, E4, E5 and E7 locations and became superior genotypes for those environments. Cultivars Aldane, Bereket and Tekirdag were the farthest from the origin and mega-circles, had low stability, while cultivars Lucilla and Glosa were determined as the most ideal and stable cultivars. Cultivars Kopru (6064 kg ha^{-1}) and Krasunia odes'ka (6015 kg ha^{-1}) were determined to be more stable with a grain yield above the experiment mean (5991 kg ha^{-1}) which were on the right quadrant and close to the origin. GGE biplot analyses are used by many researchers as a selection tool in the evaluation of different plant species in terms of many characteristics (Figure 2).

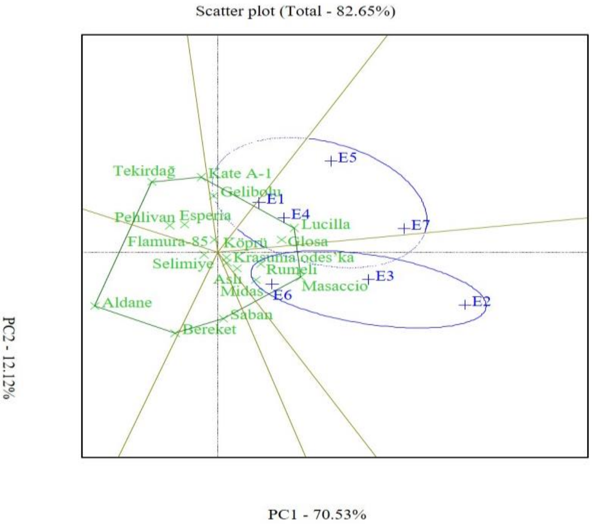

Figure 2. Scatter plot graph of GGE biplot Analysis

Genotypes in the first circle in the GGE biplot analysis are considered as ideal genotypes. While cultivar Lucilla was closest to the center circle, it was followed by cultivars Glosa, Masaccio and Rumeli, respectively. Cultivars Aldane, Bereket and Tekirdag were determined as the furthest genotypes from the first circle (Figure 3).

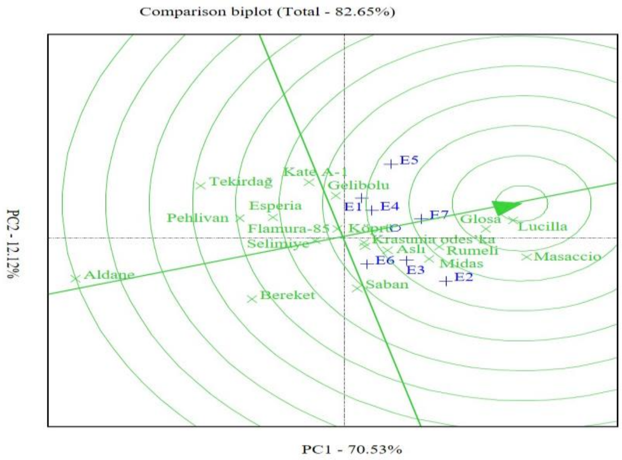

Figure 3. Comparison biplot "ideal genotype" using GGE biplot with scaling focused on genotypes

In GGE Biplot analyses, environments closer to the center circle, same as genotypes, are considered ideal environments. While E2 environment had the highest yield average (7251 kg ha^{-1}), E7 stood out as the ideal environment by taking place in the center circle which may be due to lack of separation power among the cultivars of the E2 environment. Aktas (2020) also reported the same situation, which is consistent with our findings. The E2 and E3 circles were determined as the circles close to the central circle after the E7 location, respectively. E6 was determined as the furthest environment from the central circle (Figure 4).

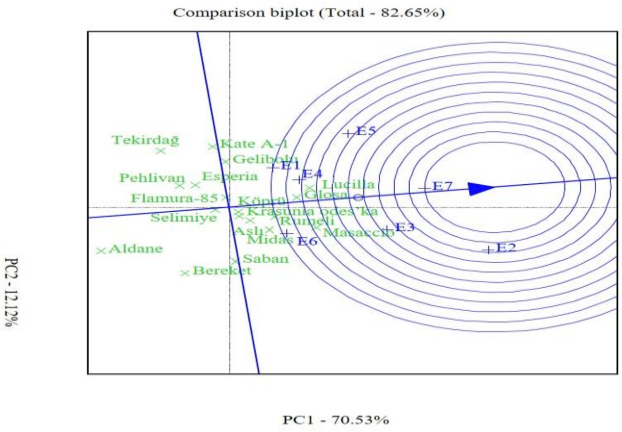

Figure 4. Comparison biplot of "ideal environment" for grain yield using GGE biplot

CONCLUSION

This research was carried out with 18 commercial bread wheat genotypes in seven environments in the Thrace region. The effect of environment was detected for grain yield, number of grains per spike, test weight and thousand kernel weight. In addition genotype effect was found significant on heading date, plant height and spike length traits, and number of spikelets per spike trait was affected by GE interaction. According to PC biplot analysis, grain yield was positively correlated with test weight, while negatively correlated with grain weight per spike and spike length. According to the results of the GGE biplot analysis, cultivars Rumeli and Masaccio were located at Edirne (E2), Babaeski (E3) and Babaeski (E6) environments. cultivars Lucilla, Glosa and Gelibolu were located in the same zone with the Luleburgaz (E1), Hayrabolu (E4), Luleburgaz (E5) and Kesan (E7) locations and became the superior genotypes for those environments. In this study, which was carried out at different environments for two years in the Thrace region, cultivars Lucilla, Glosa, Masaccio and Rumeli were found outstanding genotypes in terms of both high yielding and stable.

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DETERMINING THE GRAIN YIELD AND NUTRITIONAL COMPOSITION OF MAIZE CULTIVARS IN DIFFERENT GROWING GROUPS

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ABSTRACT

Due to its adaptability to different climates, different growing times, high photosynthetic capacity, nutrient value, and yield, maize is an important crop that is widely grown all over the world. This study was conducted to determine the grain yield and some nutrition traits of 18 maize cultivars under the ecological conditions prevalent in Bilecik in the years 2019 and 2020. Experiments were carried out in randomized complete block design with three replications. There were significant ($P<0.01$) differences among the cultivars in terms of grain yield and all quality traits. In addition, there were significant differences between the two years in terms of the investigated traits except for potassium, neutral detergent fiber, and total phenolic and ash content. According to the two year-average; grain yield (TV), test weight (TW), thousand grain weight (TGW), ash (AC), fat (FC), protein (PC), starch (SC), acid detergent fiber (ADF), neutral detergent fiber (NDF), and total phenolic (TP) contents of the maize cultivars were 13.00 ton ha^{-1}, 73.85 kg, 317.76 g, 1.59%, 5.95%, 11.22%, 65.69%, 4.25%, 15.17%, and 7.16 mg GA/g, respectively. The condensed tannin (CT), free radical scavenging activity (DPPH), total flavonoid (TF), potassium (K), phosphorus (P), magnesium (Mg), zinc (Zn), iron (Fe), manganese (Mg), and copper (Cu) contents were 0.39%, 7.97%, 0.48 mg QE/g, 4.24 g kg^{-1}, 3.58 g kg^{-1}, 1.24 g kg^{-1}, 2.36 g 100g^{-1}, 0.96 g 100g^{-1}, 0.67 g 100g^{-1}, and 0.20 g 100g^{-1}, respectively. The highest of grain yield were determined of the maize cultivars of ADA-9510 Larigal, Kerbanis, Keravnos, SY-Inove, Dracma, Kilowatt, ADA-9516 and SY-Gladius. According to the Biplot graph, the cultivars ADA-9510, SY-Inova, SY-Gladius, Kalideas, Dracma, Kerbanis, and Keravnos were prominent in terms of TV, SC, DPPH, TP, NDF, CP. The cultivars Arifiye, SY-Antex, Kolessaus, and Sakarya were prominent in terms of many quality traits such as PC, FC, AC, TF, TW, ADF, K, P, Mg, Fe, Zn, and Mn.

Keywords: Bilecik, corn, protein, total phenolic, quality, yield

INTRODUCTION

Cereals are rich sources of carbohydrates, proteins, lipids, minerals, and vitamins. Therefore, they play an important role in solving malnutrition problems all over the world (Nawaz et al., 2013). Maize (*Zea mays* L.) is an important crop that is cultivated widely across the globe. With a greater yielding potential than the other cereals, it ranks first in grain production with an annual yield of 1.1 billion tons globally (FAO, 2020). Maize, which is a C4 plant with different growing times and high yield, is a species with a high photosynthetic capacity. Its adaptability to different climates has allowed its agriculture to spread over large areas (Ozdemir and Sade, 2019). In addition to being used as human food and animal feed, maize is a resource for many unique industrial and commercial products such as breakfast foods, popcorn, alcohol, starch, glucose, spirits, oil, semolina, paint, soap, glue, insecticides, shaving cream, toothpaste, rubber tires, rayon, and molded plastics, etc. (Balconi et al., 2007).

Of the world corn production is used 60% for animal feed, 20% for human food (direct consumption), 10% for food processing and 10% for other purposes and as seeds. Maize serves as a staple food for a large proportion of the world’s population and in certain countries. The chemical content of the maize grain is very important for human and animal diets. Maize grains have high levels of starch, protein, different sugar derivatives, fiber, and fat content, as well as significant amounts of iron, magnesium, potassium, vitamin A, B1, B3, B9, and C (White and Johnson, 2003). Additionally, maize flour is considered to be superior to wheat, rice, and oats in terms of nutritional and antioxidant properties (Nawaz et al., 2013). While maize stands out as a significant source of human food in developing countries, it is used more as animal feed and industrial raw material in developed countries. Especially in emerging countries, maize accounts for up to 60% of the daily intake of protein. It also supplies different vitamins and minerals that are important for the human diet, particularly for children, the elderly, and pregnant women (Ozcan, 2009).

A significant number of metabolic disorders and diseases result from malnutrition. Considering that the vast majority of the global population consumes maize as the main bread grain, more detailed knowledge of the nutritional properties of maize cultivars will be beneficial in the production of maize with improved nutritional quality (Ndukwe et al., 2015).

Worldwide research shows that the global agricultural crop production must be doubled by 2050 to meet the growing demand driven by population growth, dietary changes, and biofuel consumption (Akgun et al., 2019). The two ways generally adopted to meet these increasing demands are expanding the cultivation areas and increasing the yield to be obtained per unit area. As with many cultivated plants in the world, it is not possible to expand maize cultivation areas any further. Because the final limit has already been reached in agricultural fields that can be cultivated. Thus, it is evident that corn production can be enhanced only by increasing the yield per unit area. To this end, in addition to determining the appropriate cultivation techniques, it is necessary to develop high-yielding and high-quality cultivars that can adapt to the conditions of the region they are grown (Uysal, 2019).

The purpose of this study was to investigate the grain yield and nutritional content per grain in 18 maize cultivars of different maturity groups.

MATERIALS AND METHODS

Plant Materials and Field Experiments

In this study, a total of eighteen different maize cultivars registered in various institutions and organizations in Turkey were used (Table 1). This study was carried out in the field of the Agricultural Research and Application Center of the Faculty of Agriculture and Natural Sciences, Bilecik Seyh Edebali University (30° 10′ N, 40° 11′ E and 500 m) in the years 2019 and 2020.

Table 1. FAO groups and institutions/organizations of the cultivars used in the trials

Cultivar name	FAO group	Registration date	Name of institution and organization
AGA	720	2015	Sakarya Maize Research Institute, Turkey
Samada-07	700	2009	Sakarya Maize Research Institute, Turkey
Keravnos	700	2020	KWS, Turkey
Kilowatt	700	2013	KWS, Turkey
Kolessous	680	2012	KWS, Turkey
Sakarya	650	2005	Sakarya Maize Research Institute, Turkey
Arifiye	650	1972	Sakarya Maize Research Institute, Turkey
Ada-523	650	2000	Sakarya Maize Research Institute, Turkey
Ada-9516	650	2000	Sakarya Maize Research Institute, Turkey
Ada-9510	650	2000	Sakarya Maize Research Institute, Turkey
Larigal	600	2006	Sakarya Maize Research Institute, Turkey
SY-Gladius	600	2019	Sygenta, Turkey
Kerbanis	550	2013	KWS, Turkey
SY-Inove	450	2015	Sygenta, Turkey
Dracma	450	1998	Sygenta, Turkey
SY-Antex	400	2018	Sygenta, Turkey
Kalideas	250	2016	KWS, Turkey
Simpatico	200	2021	KWS, Turkey

The climate data for 2019 and 2020 were obtained from the Turkish State Meteorological Service and are given in Table 2. While the average temperature for long years was 21.0 °C, it was determined to be 20.8 °C and 20.7 °C for 2019 and 2020, respectively. The total rainfall for 2019, 2020, and long years were determined to be 182.4 mm, 229.0 mm, and 206.9 mm, respectively. In addition, the average relative humidity in 2019, 2020, and long years was 64.0%, 63.8%, and 62.8%, respectively.

The soil at the trial site had similar physical and chemical properties in both 2019 and 2020. The physical soil characteristics showed that the soil texture was loamy sand with low organic matter and clay. The trials were performed in the soil with the following characteristics: 2.18 dSm^{-1} slightly salty, 7.72 pH, 1617 kg ha^{-1} exchangeable potassium (K_2O) content, 254 kg ha^{-1} available phosphorus content (P_2O_5), and 1.32% organic matter.

In the trials, sowing was done on May 3, 2019 in the first year, and on May 6, 2020 in the second year in randomized blocks design with three replications. Each plot consisted of 4 rows that were 5 m in length and 70 cm apart from each other. In the trials, the distance between the plots was 1 meter and the blocks were 2 meters apart from each other. The number of seeds was calculated as 80000 seeds per hectare. Based on the soil analysis results, all the plots in the trials were fertilized with 39.1 kg N and 100 kg P_2O_5 per ha^{-1} (di-ammonium phosphate) during sowing. When the plants are 40-50 cm (V4- V6 leaf stage), urea (% 46 N) fertilizer was divided into two parts and given to the soil during the first hoeing and throat filling. The total rate of N applied was 200 kg ha^{-1}. In the experiment, drip irrigation system was used, and irrigation was applied at weekly intervals or as needed. All cultivars were given the same amount of water at the same time. Pest and weed controls were performed according to general local practices and recommendations. Ten plants were randomly selected per plot to determine the yield and quality traits. Harvesting was done by hand (on Oct 5 2019 in the first year and on Oct 7 2020 in the second year) after the sheath of the cob had dried completely.

Table 2. Meteorological data for the experiment areas

Months	**Temperature (˚C)**			**Rainfall (mm)**			**Relative humidity (%)**		
	LY	2019	2020	LY	2019	2020	LY	2019	2020
May	16.1	16.7	17.5	47.7	55.2	35.0	64.5	62.0	60.1
June	19.9	19.8	19.0	39.3	139.1	62.4	62.0	59.7	68.0
July	21.7	22.9	23.8	30.9	1.2	35.4	61.0	63.0	60.3
August	23.5	22.4	23.3	11.2	9.9	6.5	62.0	60.9	56.7
September	19.3	19.1	21.4	31.2	4.7	8.0	64.3	67.0	65.2
October	14.2	16.0	17.1	46.6	18.9	35.1	70.4	69.9	66.6
Average	**19.1**	**19.5**	**20.4**				**64.0**	**63.8**	**62.8**
Total				**206.9**	**229.0**	**182.4**			

Grain yield, physical and chemical analyses

The cobs of the ten randomly selected plants were blended. The grain moisture was determined by drying the collected grains in an oven. The grains from each plot were weighed and the value obtained was corrected for 14% humidity. Subsequently, the resulting values were converted into grain yield as tons per hectare. Test Weight (TW) was determined using the special apparatus according to the 55-10 Approved Methods (AACC, 2010) and expressed in kilograms per hectoliter (kg/hL). Thousand grain weight (TGW) was determined by weighing 1000 seeds counted with a seed counting device (Chopin technologies-Numigral).

Maize grains that were separated for chemical analyses were ground in a hammer mill to pass through a 0.5 mm sieve. The ground samples were stored at ±4 C in a cold storage unit until they were analyzed. In the ash analyse, the temperature of the furnace was gradually increased to 500 ºC and the samples were burned for 8 hours at this temperature until completely ashed. Fat content was determined with the Soxhlet method (Welch, 1977). Protein content was determined with the use of the Micro Kjeldhal method of Concon and Soltess (1973). Starch contents of the samples were determined with the aid of enzymatic test kits (Megazyme) according to the AACC Approved Methods 76-13.01 (AACC, 2000). ADF and NDF values were determined in an ANKOM 220 Fiber Analyzer device (Van Soest et al., 1991) in maize grains. The total flavonoid content was determined according to Arvouet-Grand et al. (1994) with some modifications. Each sample (200 µL) was mixed with 100 µL of aluminum nitrate (10%) and 100 µL of potassium acetate (1 M). The total volume of the solution was adjusted to 5mL with ethanol. Similarly, a blank was prepared by adding methanol in place of the sample. Absorbance measurements were read at a spectrophotometer at the absorbance value of 417 nm after 40 min incubation at room temperature in dark conditions. Total flavonoid content was expressed as mg equivalents of quercetin (QE) g-1 DW according to the equation obtained from the standard quercetin graph and calculated from the calibration curve (R2= 0.9994) (Yavuz and Gulumser, 2022). The effect of each sample on the 2,2-diphenyl-1-picryl-hydrazylhydrate (DPPH) radical was identified according to Gezer et al. (2006). The total condensed tannin content was determined by adding 6 ml of tannin solution to 0.01 g of ground seed, then placing it in a tube and mixing on a vortex. The tubes were tightly capped and kept at 100 ° C for 1 hour and the samples were allowed to cool (Yildirim et al., 2021; Yildiz et al., 2021). Then, they were read at a spectrophotometer at the absorbance value of 550 nm (Bate Smith, 1975). Condensed tannins were calculated by the following formula: absorbance (550 nm x 156.5 x dilution factor) / Dry weight (%). Mineral matter analysis was performed with the ash samples obtained to determine the ash content. Subsequently, 1 N HCl was added to the ash samples and left to sit for 30 minutes. Subsequently, the samples were filtered with filter paper and diluted up to 50 mL with ultrapure water. The potassium, magnesium, zinc, iron, manganese, and copper contents were determined through inductively coupled plasma mass spectrometry (ICP-MS) using a Thermo Scientific - iCAPQc (Bremen, Germany). The phosphorus content was determined by the "Olsen" method (Olsen and Sommers, 1982).

Statistical Analysis

The combined data of the two years was subjected to an analysis of variance utilizing a randomized complete block design. Statistical analysis was performed using the Minitab 19 package program. Means were compared with LSD test ($p<0.05$). The principal component analysis (PCA) was performed based on all the investigated traits and the relationship between the genotypes. A Biplot graph was created by using the JMP 13 statistical package program (JMP, 2013).

RESULTS AND DISCUSSION

Table 3 shows the mean, SEM, SD, minimum, and maximum values of the investigated traits of the maize cultivars included in our study. Tables 4, 5, and 6 indicate that the effect of the year was highly significant on investigated traits, except for DM, AC, NDF, TF, and K. For all of the traits investigated (except for DM), there were very significant (P>0.01) differences among genotypes (Table 4, 5, and 6). These results indicate that the year and cultivars significantly impacted on investigated traits that were expressed by the maize cultivars.

Table 3. The mean values and ranges of the grain yield and 19 quality traits of the 18 maize cultivars in the combined data of the years.

Traits					**Range**	
	Mean	**SEM**	**SD**	**CV**	**Minimum**	**Maximum**
Grain yield (ton ha^{-1})	13.00	0.45	1.91	8.71	9.58	16.44
Test weight (kg)	73.85	0.43	1.80	2.44	70.06	76.86
Thousand grain weight (g)	317.76	9.77	5.46	7.05	200.29	380.10
Ash (%)	1.59	0.02	0.07	4.26	1.49	1.74
Fat (%)	5.95	0.17	0.73	4.19	5.01	7.44
Protein (%)	11.22	0.19	0.82	7.26	10.12	13.24
Starch (%)	65.69	0.42	1.80	2.73	62.39	68.45
Acid detergent fiber (%)	4.25	0.11	0.46	6.86	3.65	5.22
Neutral detergent fiber (%)	15.17	0.15	0.65	4.30	13.96	16.41
Total phenolic (mg GA/g)	7.16	0.22	0.94	6.08	5.88	9.53
Condensed tannin (%)	0.39	0.00	0.01	3.66	0.36	0.42
Free radical scavenging activity (%)	7.97	0.22	0.91	5.42	6.21	9.93
Total flavonoid (mg QE/g)	0.48	0.02	0.08	5.92	0.31	0.61
Potassium (g kg^{-1})	4.24	0.06	0.24	5.61	3.77	4.72
Phosphorus (g kg^{-1})	3.58	0.06	0.26	7.38	3.18	4.24
Magnesium (g kg^{-1})	1.24	0.04	0.15	3.03	1.00	1.55
Zinc (g 100g^{-1})	2.36	0.07	0.28	3.06	1.92	2.79
Iron (g 100g^{-1})	0.96	0.04	0.15	4.52	0.72	1.27
Manganese (g 100g^{-1})	0.67	0.02	0.07	5.16	0.51	0.79
Copper (g 100g^{-1})	0.20	0.01	0.03	3.92	0.16	0.30

SEM: Standard error of mean; SD: Standard deviation; CV: Coefficient of Variation

The grain yield of the maize cultivars ranged from 9.58 t ha^{-1} (SY-Antex) to 16.44 t ha^{-1} (ADA-9510). In terms of grain yield, the cultivars Keravnos, Kilowatt, SY-Gladius, Kerbanis, SY-Inove, Dracma, ADA-9510, Larigal, and ADA-9516 were placed in the same statistical group. The mean test weight ranged from 70.06 kg (Dracma) to 76.86 kg (Simpatico) with a mean value of 73.85 kg. It is thought that the difference between cultivars in terms of yield may be due to the difference in the FAO group, genetic structure and environmental factors. Thousand grain weights ranged from 200.29 g (Samada-07) to 380.10 g (SY-Gladius) with a mean value of 317.76 g (Tables 3 and 4). Although there is not much difference in rainfall between years, it is thought that very little rainfall in July in 2019 may have resulted in low yields. The rainfall during the 2019 growing season (229.0 mm) was more favorable for the expression of GY, TW, and TGW among the cultivars than the rainfall in 2020 (182.4 mm) (Table 2). Videnovic et al. (2013) stated that the grain yield in maize varies from year to year. Climatic factors such as temperature, sunshine hours, and precipitation besides genetic factors were the primary factors affecting maize growth and yield (Zhou et al., 2016). Ilker et al. (2009) reported that genotypes were affected by the environment and that some genotypes had higher yields due to high levels of environmental adaptation. Sahin and Kara (2021) reported that cultivars with a high grain yield had high thousand grain and test weights while cultivars with low grain yield had the lowest values of thousand grain and test weights due to having a higher number of grains per ear. It was reported by the researchers that the differences in grain yield, test weight, and thousand grain weight may be caused by ecological factors and agricultural practices, particularly by cultivar traits (Bayisa et al., 2022). Erawati et al. (2021) reported that each cultivar has a specific level of resistance and different genetic potential in responding to its environment. They also suggested that cultivars have the potential to provide high yields, but when environmental conditions are not favorable, then the variety cannot realize its full potential. Kalkan and Sade (2009) reported that grain yield may vary according to the maturity groups in a study they conducted on hybrid maize cultivars with different FAO groups in Konya. In the study, cultivars in the FAO 400, 250 and 200 groups showed low yields when compared to other cultivars.

Macronutrients such as fat, protein, and carbohydrates provide energy and materials that are important to ensure body composition while micronutrients such as vitamins and minerals ensure the metabolic pathways and the role of macronutrients (Biesalski and Tinz, 2018). The average ash, fat, protein and starch contents of the maize grains were 1.59% (range: 1.49-1.74%), 5.95% (range: 5.01-7.44%), 11.22% (range: 10.12-13.24%), and 65.69% (range: 62.39-68.45%), respectively (Table 3). The ash and protein was higher in the first year (1.60% and 11.36%) than in the second year (1.58% and 11.09%). The fat and starch contents of the cultivars were higher in the second year (6.28% and 66.93%) than in the first year (5.63% and 64.45%) (Table 4). Ash content is defined as the total amount of minerals accumulated in the seeds. Ali et al. (2010) indicated that the ash content of maize grains was 1.07 - 1.16%. Lipids are a diverse group of compounds with different functions such as energy storage and a structural component of cell membranes (Fahy et al., 2011). It is known that maize has a high concentration of lipids among cereals, following oat. In previous studies on maize, a wide range of fat content has been reported, namely 3.21 – 7.71% (Ullah et al., 2010). The protein content of maize grains is an important quality parameter. It was reported in previous studies that the protein contents were influenced by genotypes, location, and cultural practices (Seebauer et al., 2010). The protein content of maize cultivars was reported to be between 8.50% and 10.49% by Irinkoyenikan et al. (2016). Starch is the primary digestible carbohydrate of the plants, thus offering an important source of energy in human nutrition and animal feeding. Beckles and Thitisaksakul (2014) reported that cultivars, rainfall, temperature, soil type, and growth conditions could be more effective on the starch content in grains than genetic conditions. Previous studies have reported that the starch content of maize cultivars ranges from 38.83% to 67.09% (Thakur et al., 2021). Hegyi et al. (2007) reported that different maturity groups had significant effects on yield and some quality traits.

Table 4. Two-year average values for grain yield and some quality traits of the 18 maize cultivars

	GY		TW		TGW		AC		FC		PC		SC	
Cultivar	**		**		**		**		**		**		**	
AGA	13.00	b-f	73.10	abc	359.34	abc	1.54	cde	6.67	ab	10.12	f	66.11	a-e
Samada-07	12.94	b-g	74.21	ab	200.29	i	1.54	cde	6.26	bcd	11.97	bcd	65.91	a-e
Keravnos	14.93	abc	73.12	abc	313.65	efg	1.55	c-e	6.12	b-e	10.60	ef	66.89	a-d
Kilowatt	14.25	a-d	73.55	abc	369.50	ab	1.69	ab	5.34	efg	11.45	cde	64.11	b-e
Kolessous	11.81	c-g	76.42	a	310.80	efg	1.57	b-e	5.76	d-g	11.42	cde	66.49	a-d
Sakarya	11.06	d-g	74.83	ab	340.00	b-e	1.51	de	6.00	b-e	10.77	ef	65.36	a-e
Arifiye	10.49	efg	75.51	ab	300.99	fg	1.58	b-e	7.22	a	13.24	a	62.39	e
Ada-523	11.97	b-g	74.71	ab	329.10	c-f	1.55	cde	6.28	bcd	11.08	def	65.57	a-e
Ada-9516	13.77	a-e	74.40	ab	274.17	h	1.49	e	6.64	abc	11.49	cde	65.21	a-e
Ada-9510	16.44	a	72.49	bc	287.79	a-d	1.57	b-e	5.15	g	10.60	ef	67.40	ab
Larigal	15.31	ab	71.93	bc	324.51	def	1.70	ab	6.32	bcd	11.05	def	64.27	b-e
SY-Gladius	13.32	a-f	74.24	ab	380.10	a	1.58	b-e	5.50	d-g	10.59	ef	67.00	abc
Kerbanis	13.46	a-e	73.26	abc	339.80	b-e	1.62	a-d	5.22	fg	11.16	c-f	65.27	a-e
SY-Inove	14.90	abc	71.82	bc	350.70	gh	1.57	b-e	5.01	g	10.51	ef	68.45	a
Dracma	14.28	a-d	70.06	c	311.54	efg	1.58	b-e	5.25	fg	10.70	ef	67.48	ab
SY-Antex	9.58	g	76.42	a	336.10	cde	1.65	abc	7.44	a	12.13	abc	63.08	cde
Kalideas	12.52	b-g	72.32	bc	281.75	gh	1.62	a-e	5.19	fg	10.59	ef	68.36	a
Simpatico	10.00	fg	76.86	a	309.56	efg	1.74	a	5.83	c-g	12.56	ab	63.02	de
Year	*		**		**		ns		**		*		**	
2019	12.66	b	71.47	b	310.35	b	1.60		5.63	b	11.36	a	64.45	b
2020	13.34	a	76.22	a	325.18	a	1.58		6.28	a	11.09	b	66.93	a

*: Significant at the $p<0.05$ probability level, **: Significant at the $p<0.01$ probability level, ns: non-significant, GY: Grain yield (ton ha-1), TW: Test weight (kg), TGW: Thousand grain weight (g), AC: Ash content (%), FC: Fat content (%), PC: Protein content (%), SC: Starch content (%)

The average ADF and NDF contents of maize cultivar ranged from 3.65% (Arifiye) to 5.22% (Larigal) and from 13.96% (Arifiye) to 16.41% (Kilowatt), respectively. ADF and NDF values were higher in the second year. However, statistical differences were observed in ADF between the years, but not in NDF. ADF indicates the amount of cellulose, lignin, and insoluble protein in the plant cell wall structure. It is also a good indicator of feed digestibility and energy intake of the animal. It has been reported that the digestibility and energy value of feeds containing high ADF are low (Mut et al., 2017). NDF expresses the amount of cellulose, hemicellulose, lignin, cutin, and insoluble protein in the plant cell wall structure. Since the NDF value directly affects the feed intake of the animals, the lower this value is the higher the animal's feed intake in the terms of feed (Van Soest et al., 1991). These values are very important in maximizing feed quality. Radosavljevic et al. (2012) reported that the ADF and NDF values ranged from 3.89% to 4.88% and from 17.59% to 29.84%, respectively.

Measuring the levels of various antioxidants in different maize cultivars provides key information for efforts to improve the antioxidant levels in corn. In this study, the average TP, CT, DPPH, and TF contents of the maize grain were 7.16 mg GA/g (range: 5.88 mg GA/g– 9.53 mg GA/g), 0.39% (range: 0.36% - 0.42%), 7.97% (range: 6.21% - 9.93%), and 0.48 mg QE/g (range: 0.31 mg QE/g - 0.61 mg QE/g), respectively (Tables 3 and 5). Natural antioxidants, especially phenolics and flavonoids, are safe and bioactive. Flavonoids show antioxidant activity and have considerable effects on human nutrition and health. In recent decades, phenolic and flavonoid-rich natural diets with antioxidant activity have fostered an interest in nutrition and food science (Aryal et al., 2019). DPPH is one of the most important methods adopted for evaluating the antioxidant properties of plants. Condensed tannins contribute to astringency in foods (Dykes and Rooney, 2007). These compounds are abundantly present in maize, especially in bran (Shah et al., 2016). Biofortification to produce corn with improved antioxidant activities could be a solution for some health problems. The researches have suggested that the phytochemicals in grains demonstrate significant beneficial contribution in reducing the risk of many diseases due to their potent antioxidant activities (Shahidi, 2009). Maize grains contain nutritionally valuable antioxidants that benefit human health by reducing age-related disorders such as cardiovascular disease, diabetes, obesity, neurodegenerative disorders, and cancer (Bae et al., 2021). Therefore, maize is considered to be a functional food. In addition, previous studies showed that the flavonoids and phenolic compounds have positive effect on the productivity and health of animals, as well as rumen fermentation and the control of nutritional stress such as bloat and acidosis (Lee et al., 2017). These compounds also have indirect effects on the environment. Lascano and Cardenas (2010) reported that ¼ of the methane gas released into the atmosphere is produced in the digestive system of ruminants. Condensed tannins inhibit certain hydrogen-producing protozoans and methane-producing organisms that use hydrogen directly in the rumen, and reduce greenhouse gas emissions (Martin et al., 2016). But tannin binds to proteins, carbohydrates, and minerals, which decrease the digestibility of these nutrients and reduce the feed efficiency of ruminants and monogastrics during feeding. Martinez-Martinez et al. (2019) reported that the total phenolic, flavonoid, and DPPH contents of the maize grain ranged between 69.46 mg GAE 100g^{-1} and 137.39 mg GAE 100g^{-1}, 0.02 mg QE g^{-1} and 0.19 mg QE g^{-1} and 3.17% and 6.75%, respectively. Nawaz et al. (2013) reported that the condensed tannins content of the maize grain ranged between 0.494 g/100 g and 0.556 g/100 g.

Table 5. Two-years average values for some quality traits of 18 maize cultivars

	ADF	NDF	TP	CT	DPPH	TF
Cultivar	**	**	**	**	**	**
AGA	4.95 ab	14.54 def	7.45 b	0.39 abc	7.90 b-e	0.49 cde
Samada-07	4.14 b-f	14.83 c-f	7.08 bcd	0.36 c	8.81 abc	0.45 def
Keravnos	4.25 b-f	14.79 c-f	7.32 bc	0.38 bc	8.54 abc	0.41 efg
Kilowatt	4.81 abc	16.41 a	7.51 b	0.40 abc	7.73 b-e	0.60 ab
Kolessous	4.44 a-f	14.95 b-f	6.73 bcd	0.39 abc	7.27 b-e	0.56 abc
Sakarya	4.06 c-f	14.73 c-f	5.90 c	0.39 abc	6.26 de	0.61 a
Arifiye	3.65 f	13.96 f	7.11 bcd	0.39 abc	8.03 bc	0.56 abc
Ada-523	4.75 a-d	15.54 a-d	7.41 b	0.37 bc	7.81 b-e	0.39 fg
Ada-9516	3.69 f	14.32 ef	6.50 bcd	0.39 abc	8.60 abc	0.52 a-d
Ada-9510	4.03 c-f	15.33 a-e	7.06 bcd	0.40 abc	8.56 abc	0.31 g
Larigal	5.22 a	16.11 ab	9.53 a	0.39 abc	9.93 a	0.45 def
SY-Gladius	4.03 c-f	15.29 a-e	7.11 bcd	0.39 abc	8.17 bc	0.50 b-e
Kerbanis	4.25 b-f	15.85 abc	9.13 a	0.42 a	8.95 ab	0.49 cde
SY-Inove	3.85 ef	15.36 a-e	6.65 bcd	0.38 abc	8.01 bcd	0.39 fg
Dracma	3.73 f	15.44 a-e	7.52 b	0.40 ab	7.10 cde	0.51 b-e
SY-Antex	4.69 a-e	14.45 def	5.88 d	0.37 bc	6.21 e	0.46 def
Kalideas	3.96 def	15.38 a-e	6.46 bcd	0.40 ab	7.60 b-e	0.48 c-f
Simpatico	4.05 c-f	15.81 abc	6.56 bcd	0.40 abc	8.03 bc	0.48 c-f
Year	**	ns	ns	**	**	**
2019	3.76 b	15.08	7.18	0.40 a	7.16 b	0.31 b
2020	4.75 a	15.26	7.14	0.38 b	8.78 a	0.65 a

**: Significant at the $p<0.01$ probability level, ns: non-significant, ADF: Acid detergent fiber (%), NDF: Neutral detergent fiber (%), TP: Total phenolic (mg GA/g), CT: Condensed tannin (%), DPPH: Free radical scavenging activity (%), TF: Total flavonoid (mg QE/g)

The mineral composition of the maize grain is an important parameter that needs to be considered in human nutrition and feed animal. The results of the mineral content grains of different maize cultivars are shown in Table 6. The analysis shows the level of K (3.77 g kg^{-1}- 4.72 g kg^{-1}), P (3.18 g kg^{-1}- 4.25 g kg^{-1}), Mg (1.00 g kg^{-1} - 1.55 g kg^{-1}), Zn (2.79 g 100g^{-1} - 1.92 g 100g^{-1}), Fe (0.72 g 100g^{-1} - 1.27 g 100g^{-1}), Mn (0.51 g 100g^{-1} - 0.78 g 100g^{-1}), and Cu (0.16 g 100g^{-1} - 0.30 g 100g^{-1}) (Tables 3 and 6). The K, Mg, Zn, Fe, and Cu contents of the genotypes were higher in the first year while the P, Mg, and Mn contents were higher in the second (Table 6). In previous studies, it was reported that magnesium content varies depending on the cultivar (Ullah et al., 2010), years (Ferreira et al., 2012), environments (Gu et al., 2015), and agricultural practices (Kresovic et al., 2018). Mineral deficiencies influence billions of people around the world. Mineral insufficiency causes decreased working efficiency, cardiovascular diseases, cancer, autoimmune diseases, high healthcare costs, and increased rates of premature death. Therefore, mineral elements such as Cu, Ca, Mn, Zn, and Fe are important from the viewpoint of malnutrition (Welch and Graham, 2004). The maize grain is an excellent and relatively inexpensive source of certain minerals, especially in underdeveloped countries. In recent years, one of the aims of plant breeding programs has been to increase mineral accumulation in cereal grains. This approach is a sustainable strategy to increase the use of micronutrients in diets as there are no further costs once new cultivars are developed (Neeraja et al., 2017). In a previous study on the mineral content of maize grains in different cultivars, Ullah et al. (2010) determined that the maize grain contained K (2915 ppm - 3471 ppm), Na (540.30 ppm - 620.41 ppm), Ca (410 ppm -5 90 ppm), Fe (38.02 ppm - 56.14 ppm), Zn (37.05 ppm - 52.04 ppm), Mg (985.2 ppm - 1125.3 ppm), and Cu (11.02 ppm - 14.25 ppm).

Table 6. Two-year average values for mineral matter contents of the 18 maize cultivars

	K	P	Mg	Zn	Fe	Mn	Cu
Cultivar	**	**	**	**	**	**	**
AGA	3.97 cd	3.68 bcd	1.28 a-e	2.05 ef	0.90 c-f	0.73 abc	0.23 bc
Samada-07	4.15 bcd	3.47 c-f	1.23 a-e	2.03 ef	0.83 ef	0.70 a-f	0.30 a
Keravnos	4.02 cd	3.45 c-f	1.13 b-e	1.92 f	0.92 cde	0.65 c-g	0.19 bc
Kilowatt	4.72 a	3.63 b-e	1.32 a-e	2.27 b-f	1.02 bcd	0.76 ab	0.17 c
Kolessous	4.14 cd	3.56 b-e	1.27 a-e	2.58 abc	0.94 b-e	0.78 a	0.19 bc
Sakarya	4.12 cd	3.76 bc	1.27 a-e	2.73 a	1.13 ab	0.70 a-f	0.21 bc
Arifiye	4.42 abc	4.25 a	1.55 a	2.64 ab	1.25 a	0.71 a-e	0.19 bc
Ada-523	3.96 cd	3.74 bc	1.32 a-e	2.04 ef	0.88 c-f	0.72 a-d	0.24 ab
Ada-9516	3.77 d	3.90 ab	1.38 abc	2.19 def	0.78 ef	0.60 e-h	0.18 bc
Ada-9510	4.35 abc	3.28 ef	1.05 cde	2.00 ef	0.72 f	0.51 h	0.18 bc
Larigal	4.32 abc	3.60 b-e	1.32 a-e	2.79 a	1.27 a	0.66 b-g	0.18 bc
SY-Gladius	4.17 bcd	3.49 c-f	1.11 b-e	2.23 c-f	0.93 cde	0.58 gh	0.20 bc
Kerbanis	4.35 abc	3.50 c-f	1.20 b-e	2.34 b-e	0.85 def	0.61 e-h	0.17 c
SY-Inove	4.24 a-d	3.19 f	1.00 e	2.43 a-d	0.92 cde	0.60 fgh	0.18 bc
Dracma	4.35 abc	3.33 def	1.09 b-e	2.35 b-e	0.89 c-f	0.62 d-h	0.18 bc
SY-Antex	4.30 a-d	3.82 bc	1.42 ab	2.47 a-d	1.03 bcd	0.65 c-g	0.16 c
Kalideas	4.28 a-d	3.18 f	1.01 de	2.61 abc	0.86 def	0.65 c-g	0.20 bc
Simpatico	4.68 ab	3.68 b-e	1.34 a-d	2.76 a	1.06 bc	0.79 a	0.19 bc
Year	ns	**	**	**	**	**	*
2019	4.23	3.47 b	1.12 b	2.41 a	0.99 a	0.65 b	0.21 a
2020	4.25	3.70 a	1.35 a	2.30 b	0.93 b	0.68 a	0.19 b

*: Significant at the p<0.05 probability level, **: Significant at the p<0.01 probability level, ns: non-significant, K: Potassium (g kg^{-1}), P: Phosphorus (g kg^{-1}), Mg: Magnesium (g kg^{-1}), Zn: Zinc (g 100g^{-1}), Fe: Iron (g 100g^{-1}), Mn: Manganese (g 100g^{-1}), Cu: Copper (g 100g^{-1})

Biplot Analysis

A genotype × trait Biplot was used to simultaneously demonstrate the relationships between traits and genotypes. The Biplot also helped identify the cultivars with superior traits. The two-year average values of the investigated traits of the 18 maize cultivars are displayed in Tables 4, 5, and 6. The Biplot graph explained 54.7% of the total variation (PCA1 33.9% and PCA2 20.8%) (Figure 1).

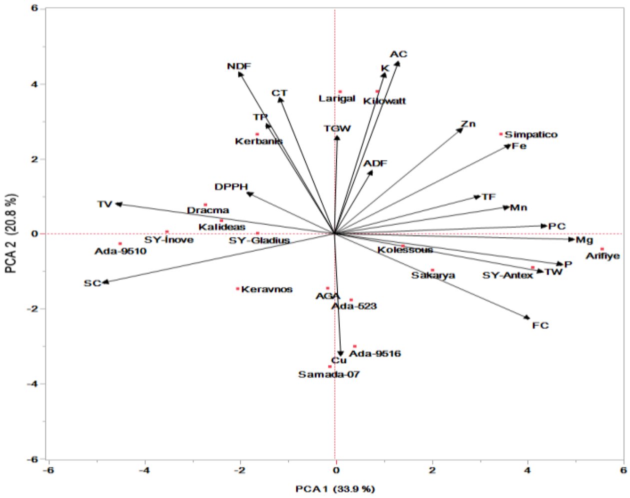

Figure 1. Genotype × Trait biplot graph for the investigated traits of the maize cultivars

In the Biplot graph, vector angles below 90° indicate that the traits are positively correlated; vector angles above 90° indicate a negative correlation, and finally, vector angles equal to 90° indicate that the traits are not related (Yan and Tinker, 2006). The vectors in Figure 1 were drawn individually from the biplot origin to each of the traits. The TV showed a highly significant and positive correlation with SC, DPPH, TP, NDF, CT, and TGW (<90˚). The cultivars ADA-9510, SY-Inova, SY-Gladius, Kalideas, Dracma, Kerbanis, and Keravnos were prominent in terms of grain yield. The cultivars Simpatico, Arifiye, SY-Antex, Kolessaus, and Sakarya were prominent in terms of many quality traits such as PC, FC, AC, TF, TW, ADF, K, P, Mg, Fe, Zn, and Mn. This result is partly consistent with a previous report (Amegbor et al., 2022). Yousaf et al. (2021) showed the negative association between starch and fat content.

CONCLUSIONS

Increasing the nutrient contents in cereals, which are the main food crops worldwide, is a sustainable approach to improving nutritional well-being. There are many types of maize in the market both in the world and in Turkey. However, although the yield and quality characteristics of these cultivars are different from each other, their responses to the ecology of the region may vary. Bilecik province is located in a region that is very suitable for grain mazie cultivation with its ecological conditions and irrigation facilities. As a matter of fact, the agricultural and quality traits determined in the cultivars used in the study were superior to the studies conducted in most regions of Turkey. The results of this study indicate that the grain yield and quality traits of many maize cultivars have significant differences. The highest of grain yield were determined of the maize cultivars of ADA-9510 Larigal, Kerbanis, Keravnos, SY-Inove, Dracma, Kilowatt, ADA-9516 and SY-Gladius. According to the Biplot graph, the cultivars ADA-9510, SY-Inova, SY-Gladius, Kalideas, Dracma, Kerbanis, and Keravnos were prominent in terms of grain yield. The cultivars Simpatico, Arifiye, SY-Antex, Kolessaus, and Sakarya were prominent in terms of many quality traits such as PC, FC, AC, TF, TW, ADF, K, P, Mg, Fe, Zn, and Mn. Knowing the change rates of nutritient values in maize cultivars will contribute to the farmer, user and industry field.

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EFFECTS OF DIFFERENT NITROGEN SOURCES ON TURF QUALITY AND PLANTS GROWTH OF SOME WARM-SEASON TURFGRASSES

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ABSTRACT

This research was conducted to determine the effects of different nitrogen sources and rates on some warm-season turfgrasses under Mediterranean-type climate conditions in 2017-2018. The experiment was arranged in a randomized complete block design with split-split plot arrangement and having three replications. In the arrangement, turfgrass species as the main plot, nitrogen sources as the subplots, and nitrogen doses as the sub subplots. The main plots consist of four nitrogen sources: two slow-release fertilizers, one organomineral fertilizer, and one sewage sludge. Subplots consist of three warm-season turfgrass species; zoysiagrass (*Zoysia japonica* Steud.), hybrid Bermudagrass (*Cynodon transvaalensis x Cynodon dactylon*), seashore paspalum (*Paspalum vaginatum* Sw.), and one cool-season turfgrass species, tall fescue (*Festuca arundinacea* Schreb.). The nitrogen doses were as follows; 0.0, 2.0, 3.0 and 4.0 g m^{-2}. Turf color and quality were evaluated visually monthly. Also, clipping weight is determined. According to the result, slow-release and organomineral fertilizers can be considerable as N sources that will meet the nutritional needs of the turfgrasses. Zoysiagrass and seashore paspalum showed almost equivalent scores and gave sufficiently dark turf color and quality. Turfgrass should be fertilized at least with 3.0 g m^{-2} N to provide acceptable turf color and quality.

Keywords: Nitrogen sources, nitrogen rate, turf color, turf quality, warm-season turfgrasses

INTRODUCTION

Warm-season turfgrasses belong to the Poaceae family and have a C_4 photosynthetic system. Due to the C_4 mechanism, warm-season turfgrasses have better resistance to drought stress and exhibits improved nitrogen and water use efficiency nearly two times higher than that of C_3 grasses (Braun et al., 2020). Due to global warming and the associated climatic change, the adoption of warm-season plants for decorative and sports turfs has recently been pushed in Mediterranean nations (Minelli et al., 2014; Giola et al., 2019; Kir et al., 2019).

Excessive nitrogen fertilizer application is a typical technique to guarantee that plants do not become N-deficient, especially in urban environments. Excess nitrogen that is not used by the plant adds to a waste of natural resources as well as money (Trenkel, 2010; Hopkins, 2020). Nitrogen fertilizer may result in nitrogen loss to the environment via ammonia volatilization, nitrate leaching, and denitrification/nitrification byproducts. Nitrogen loss, in its different mobile forms, adds to problems in the atmosphere and hydrosphere, which eventually impair human and animal health (Mulvaney et al., 2009; Olson et al., 2009; Hopkins, 2020). On a wide range of species, research has been conducted to improve nitrogen usage efficiency by following optimally management techniques such as applying the proper source at the right rate, positioning, and time (Stevens et al., 2007; LeMonte et al., 2018; Hopkins, 2020). Using slow-release fertilizers combined with improved irrigation techniques may reduce N_2O emissions so reducing N_2O emissions from turf may help mitigate climate change and atmospheric ozone destruction (Braun and Bremer, 2018). Besides slow-release; organomineral and sewage sludge fertilizers can offer many potential benefits for the fertilization of turfgrasses such as decreasing the leaching losses of nutrients (Granlund et al., 2000).

Slow-release fertilizers have lower nitrogen loss by leaching or volatilization, fewer chances of fertilizer burn, fewer applications at higher rates. Also, slow-release fertilizers provide economic savings and consistent release of nitrogen over a long period. Besides, slow-release nitrogen sources provide for more uniform growth compared to synthetic sources (Hummel and Waddington, 1981; Simonne and Hutchinson, 2005). Organomineral fertilizer is defined as organic fertilizers or soil improvers that are produced from organic ruins such as poultry litter, turf, or sewage sludge, with at least one inorganic source (Antille et al., 2013). These fertilizers contain vary in macro-nutrients, micro-nutrients, and organic matter content, but they provide nutrients via gradual solubilization (Eghball and Power, 1994; Carvalhoa et al., 2014). The use of organomineral and slow-release fertilizers is one of the backbones of sustainable agricultural practices to eliminate the negative effects of the unbalanced use of chemical fertilizers on humans and the environment. Andiru et al. (2015) stated that slow-release fertilizer applications are as effective on plant growth as standard fertilizers. Sewage sludge is the product of sewage treatment in wastewater treatment plants and includes high levels (10% to> 20%) of organic matter, nitrogen, phosphorus, potassium, and some micronutrient elements such as copper, zinc, iron (Brady and Weil, 1999). The use of sewage sludge compost and other processed sludge products for turfgrass establishment and maintenance is an environmentally sound and cost-effective way to use sewage sludge-derived products. In addition to inorganic fertilizers, compost can be applied to the surface of turfgrass. Thus adding macro and micronutrients to the soil (Angle, 1994).

It is a necessity to investigate the use of alternative nitrogen sources to reduce environmental pollution. This research was conducted to the determination of the effects of nitrogen sources and nitrogen doses on turf color, quality, and clipping yield for some warm-season turfgrasses.

MATERIALS AND METHODS

This study was carried out in the turfgrass experimental plots of Research and Training Centre of Faculty of Agriculture, Bursa Uludag University, in Bursa, Turkey in 2017 and 2018. The research area has a Mediterranean-type climate. Table 1 shows the temperature, precipitation, relative humidity in 2017 and 2018 growing season, and long-term average records. The long-term average temperature was 19.5 °C, the average relative humidity was 61.7%, and seasonal precipitation was 281.3 mm in the region. Relative humidity of the 2017-2018 growing seasons were higher (8% and 11%, respectively) than the long-term average records. Likewise, average temperatures of the 2017-2018 growing seasons were higher (0.2 and 0.9°C, respectively) than the long-term average. The 2017-2018 growing season drier than the long-term average.

Table 1. Climate data of the research area.

Months	**Temperature (°C)**			**Precipitation (mm)**			**Relative Humidity (%)**		
	2017	**2018**	**LT***	**2017**	**2018**	**LT**	**2017**	**2018**	**LT**
April	12.2	15.8	12.8	38.1	14.2	63.4	68.8	70.8	66.1
May	17.2	19.9	17.6	33.3	89.8	44.3	71.5	76.5	62.0
June	22.1	23.5	22.1	56.4	59.2	34.3	70.0	70.1	57.8
July	24.6	26.1	24.6	18.9	9.6	15.3	63.0	63.5	56.2
August	24.5	26.4	24.3	6.3	1.8	15.7	66.4	59.6	57.3
September	22.9	21.8	20.1	0.1	29.6	39.5	56.4	67.8	63.8
October	14.4	16.9	15.2	57.6	60.6	68.8	73.2	76.7	68.7
Tot./Ave.	*19.7*	*21.4*	*19.5*	*210.7*	*264.8*	*281.3*	*67.0*	*69.3*	*61.7*

* LT: Long term (1950-2015)

The experiment was carried out on previously established turfgrass plots. The treatments were set up in a randomized complete block design with a split-split plot arrangement, having three replications. The trial was conducted on plots established in 2013. Turfgrass species were designated as the main plot, nitrogen sources as the subplots, and nitrogen doses as the sub subplots. The main plot size 6 m × 4 m= 24 m^{-2}, sub subplot size was 1 m × 2 m = 2 m^{-2}, total area 24 m^{-2} × 16 = 384 m^{-2}. The main plots consist of four nitrogen sources; Floranid® (16-7-15 + 2 MgO + minor elements), Biosmart® (23-5-7 + minor elements), Sewage sludge (4,7-1,9-0,6), and Hexaferm® (12-15-5 + 10SO_3 + minor elements). Types of fertilizers used in the research; Floranid and Biosmart were slow-release fertilizers, sewage sludge was obtained by collecting and processing solid wastes of vegetable from the wastewater treatment system of Penguen Food Company. The sludge was dried to evaporate the water and reduce the moisture content. The dried muds have been ground and turned into granules for ease of application. Sewage sludge was analyzed at Department of Soil Science and Plant Nutrition, Faculty of Agriculture, Bursa Uludag University before the beginning the experiment. Analysis results are presented in Tables 2 and 3. Hexaferm was organomineral fertilizer. The organomineral fertilizer contains at least 20% organic matter, 7% humic-fulvic acid, 5% elemental sulfur-derived $SO_{3,}$ and zinc, as well as N, P, K.

Subplots consist of three warm-season turfgrass species, zoysiagrass (*Zoysia japonica* Steud. cv. Zenith), seashore paspalum (*Paspalum vaginatum* Sw. cv. Seaspray), hybrid Bermudagrass (*Cynodon transvaalensis x Cynodon dactylon* cv. Tifdwarf), and one cool-season turfgrass species, tall fescue (*Festuca arundinacea* Schreb. cv. Jaguar 4G). The nitrogen doses were grouped into sub-subplots. Nitrogen was applied monthly at rates of NS1: 0.0 (control) g N m^{-2}; NS2: 2.0 g N m^{-2}; NS3: 3.0 g N m^{-2} and NS4 4.0 g N m^{-2}. Irrigation was provided at 3-day intervals via a rotary sprinkler system. Nitrogen fertilizer treatments began in mid-April 2017 and lasted for seven months. Turfgrass color and quality were rated monthly. During the growing seasons, turfgrass color ratings were assessed visually on a scale of 1-9 (1 = completely yellow; 5 = unacceptable; 6 = acceptable; 9 = dark green). Turf quality was evaluated visually using a scale of 1-9 (1 = poorest; 5 = unacceptable; 6 = minimally acceptable; 7 = good; 9 = excellent) on each plot based on turfgrass uniformity, density, and color (Krans and Morris, 2007). When the plants reached a height of 6 to 8 cm, the subplots were mowed to a height of 4 cm. A 0.5 m 1.0 m strip across the center of each plot was cut at each clipping date, dried at 70 °C for 48 hours, and then weighed (Candogan et al., 2014).

Color, quality, and clipping yields data were based on two years average results. Except for visual turf color and quality, only clipping yield data were statistically analyzed. The differences between treatment means were evaluated by the least significant difference (LSD) at P= 0.05 probability level. (Steel and Torrie, 1980; Acikgoz et al., 2004; Kir et al., 2019).

Table 2. Analysis results of sewage sludge and soil.

Parameters (on a dry matter basis)	**Soil**	**Sewage Sludge (Penguen)**
pH (1:2.5, soil:water and 1:10, sludge:water)	8.48	6.73
Electrical conductivity (mmhos/cm)	468	6780
% N	0.106	4.66
Ammonium-N (mg.kg^{-1})		4.66
Total-P %		1.87
Available-P (mg.kg^{-1})	30.95	785.9
Organic C (%)	2.091	42.15
C/N (%)		5.24
Total Na (mg.kg^{-1})	675	1262.5
Total K (mg.kg^{-1})	5180	6050
Total Ca (%)	17415	2.51
Total Mg (mg.kg^{-1})	15090	7768.8
Total Mn (mg.kg^{-1})	788.3	625.6
Total Fe (mg.kg^{-1})	36210	9211.3

Table 3. Analysis result of some heavy metal standards and contents of sewage sludge and soil (mg kg^{-1}).

	Standards*			
Parameters	**Soil**	**Sewage Sludge**	**Research Area Soil**	**Sewage Sludge (Penguen)**
Total Cd	1-3	10	0.148	1.4
Total Zn	50-300	2500	54.95	578.9
Total Cr	100	1000	134.0	176.4
Total Ni	30-75	300	97.08	93.1
Total Cu	50-140	1000	33.48	115.3
Total Pb	150-300	750	10.42	30.3

RESULTS AND DISCUSSION

Results of visual turf color and quality average values present in Table 4, and clipping yield average values present in Table 5. Since warm-season turfgrasses were yellowish-brown throughout the winter months, only data from the spring-summer-autumn seasons were statistically analyzed for clipping yield (Kir et al., 2019).

During the experiment, nitrogen sources (NS), turfgrass species (TS), nitrogen doses (ND) substantially impacted visual turfgrass color, quality, and clipping yield. NS x TS interaction had a significant effect on turf color, quality, and clipping yields, except for the NS x TS interaction of the quality in spring season. NS x ND interaction color, quality, and clipping yields values were found significant for all season observations. TS x ND interactions turfgrass color, quality, and clipping were significant for all observations (Table 4). In the present study, generally the sewage sludge fertilizer produced minimum color and quality values among nitrogen sources, while the highest values were obtained from the application slow-release fertilizer. Biosmart, Hexaferm and Floranid gave high turf color and quality (Table 5). The highest clipping yield was obtained from Hexaferm slow-release fertilizer, and the lowest clipping yield was obtained from sewage sludge fertilizer during the trial (Table 6).

Differences were observed between the effects of nitrogen sources used in our experiment. Higher color and quality values were obtained in one of the slow-release fertilizers compared to the other. This situation may have developed due to many different reasons, from minor element differences in the content of fertilizers to coating material. Some researchers state that slow-release fertilizers show different properties according to the coating material (Azeem et al., 2014; Mehmood et al., 2019). Slow-release fertilizers nitrogen releasing rate is affected by coating thickness, soil temperature, soil moisture, and soil N concentration gradient, which also affects plant growth (Braun and Bremer, 2018). Ucgun et al. (2020) reported that in the studies conducted with slow-release fertilizers, there were differences between fertilizers belonging to different companies, as well as differences between fertilizers belonging to the same company and using the same coating material, so emphasizes that the effect of factors such as the thickness of the coating material on the release time of the fertilizers. The nutrient release of slow-release fertilizers over time cannot be predicted exactly and the amount of release depends on soil and climatic conditions (Azeem et al., 2014).

Biosmart x zoysiagrass, Biosmart x seashore paspalum, and Hexaferm x zoysiagrass interactions gave the highest turfgrass color and quality values. Sewage sludge x hybrid Bermudagrass, sewage sludge x tall fescue interactions gave the lowest color and quality ratings.

Table 4. Result of variance analysis of color, quality, and clipping yield under nitrogen sources (NS), turfgrass species (TS), and nitrogen doses (ND).

Factor	**Color**			**Quality**			**Clipping yield**		
Sources of variation	**Spr.*****	**Sum.**	**Aut.**	**Spr.**	**Sum.**	**Aut.**	**Spr.**	**Sum.**	**Aut.**
NS	*	**	*	*	*	**	*	**	*
TS	**	**	**	**	**	**	**	**	**
ND	**	**	**	**	**	**	**	**	**
NS x TS	*	**	*	ns	*	*	*	**	**
NS x ND	**	**	**	**	*	*	**	*	*
TS x ND	**	**	**	**	**	**	**	*	**
NS x TS x ND	ns	*	*	*	*	*	**	**	*

*, **: F-test significant at $p \leq 0.05$ and $p \leq 0.01$, respectively. ns: not significant.
***: Spr: Spring, Sum.: Summer, Aut.: Autumn

Table 5. Turf color and quality of some turfgrass species (TS) under different nitrogen sources (NS), and nitrogen doses (ND).

			Color					**Quality**		
NS	**Spr.**	**Sum.**	**Aut.**	**Win.**	**Mean**	**Spr.**	**Sum.**	**Aut.**	**Win.**	**Mean**
NS_1	5.6	6.2	5.9	-	5.9	5.5	5.9	5.7	-	5.7
NS_2	5.9	6.4	6.0	-	6.1	5.8	6.2	5.8	-	5.9
NS_3	5.3	5.8	5.4	-	5.5	5.3	5.7	5.2	-	5.4
NS_4	5.7	6.2	5.9	-	5.9	5.7	6.1	5.9	-	5.9
Mean	5.6	6.1	5.8	-	5.8	5.6	6.0	5.7	-	5.7
TS										
1	5.2	6.0	5.4	-	5.5	5.1	5.8	5.2	-	5.4
2	5.9	6.5	6.0	-	6.1	5.7	6.2	5.9	-	5.9
3	6.0	6.7	5.9	-	6.2	5.9	6.4	5.7	-	6.0
4	5.5	5.6	6.0	-	5.7	5.5	5.5	5.9	-	5.6
Mean	5.6	6.2	5.8	-	5.8	5.6	6.0	5.7	-	5.7
ND										
0.0	3.1	3.7	3.4	-	3.4	3.0	3.4	3.3	-	3.2
2.0	5.5	6.1	5.7	-	5.7	5.4	5.9	5.7	-	5.7
3.0	6.6	7.1	6.7	-	6.8	6.5	6.9	6.5	-	6.6
4.0	7.4	7.9	7.4	-	7.5	7.3	7.7	7.2	-	7.4
Mean	5.6	6.2	5.8	-	5.8	5.6	6.0	5.7	-	5.7

*: NS1: Floranid® (16-7-15 + 2 MgO + minor elements), NS2: Biosmart® (23-5-7 + minor elements), NS3: Sewage sludge (4,7-1,9-0,6), and NS4: Hexaferm® (12-15-5 + $10SO_3$ + minor elements)
**: 1: Tifdwarf (*Cynodon transvaalensis x Cynodon dactylon*), 2: Seaspray (*Paspalum vaginatum* Sw), 3: Zenith (*Zoysia japonica* Steud.), 4: Jaguar 4G (*Festuca arundinacea* Schreb.)
***: g N m^{-2}

The sewage sludge fertilizer exhibited values above 6 with respect to color and quality, which is acceptable for some observations. However, compared to other nitrogen sources, it exhibited lowest turf color, quality, and clipping yield values (Table 5). In our previous study, which we carried out using three different sewage sludge, it was determined that the application of sewage sludge at rates as low as 4.0 g N m^{-2} was beneficial as at least 6.0 g N m^{-2} nitrogen dose. Although sewage sludge was less effective than chemical fertilizers, it at least resulted in acceptable turfgrass color and quality, so it was stated that sewage sludge may be an alternative to chemical fertilizers (Zere and Bilgili, 2016). In a study conducted by applying sewage sludge to Bermudagrass, researchers reported positive results for turf growth. The use of sewage sludge is an alternative to waste disposal as it meets environmental and economic requirements (Nobile et al., 2014). However, heavy, or toxic metals in sewage sludge cause heavy metal accumulation and threaten long-term soil quality (Dai et al., 2007; Lin et al., 2017). The sewage sludge used in the study was collected from the activated sludge system of a food processing and canning factory, not from industrial production, so the heavy metal concentration of the sewage sludge was low. Therefore, they did not pose environmental damage or a potential health risk. Heavy metal contents of the sewage sludge were all lower than the recommended limit values in sewage sludge as stated by USEPA 40 CFR Part 503 regulations (USEPA, 1994).

Table 6. Clipping yield of some turfgrass species (TS) under different nitrogen sources (NS), and nitrogen doses (ND).

NS	Spring	Summer	Autumn	Winter	Mean
NS_1	116.9 b	119.5 c	99.5 c	-	111.9
NS_2	134.3 a	154.1 a	134.4 a	-	140.9
NS_3	110.5 b	118.7 c	97.2 c	-	108.8
NS_4	126.2 a	134.9 b	120.2 b	-	127.1
Mean	121.9	131.8	112.8		122.2
$LSD_{0.05}$	8.08	4.55	10.90	-	
TS					
1	69.0 d	86.2 c	77.7 d	-	77.6
2	149.3 b	164.1 b	127.7 b	-	147.0
3	182.4 a	200.3 a	154.1 a	-	178.9
4	87.2 c	76.5 d	92.2 c	-	85.3
Mean	121.9	131.7	112.9		122.2
$LSD_{0.05}$	8.50	7.04	8.19	-	
ND					
0.0	37.8 d	47.4 d	40.9 d	-	42.0
2.0	102.3 c	111.4 c	95.3 c	-	103
3.0	140.1 b	156.4 b	130.7 b	-	142.4
4.0	207.8 a	211.8 a	184.6 a	-	201.4
Mean	122	131.7	112.8		122.2
$LSD_{0.05}$	6.82	8.63	8.23	-	

*: F-test significant at $P < 0.05$, **: F-test significant at $P \leq 0.01$, ns: not significant.
NS: Nitrogen Sources, TS: Turfgrass species, ND: Nitrogen Doses.

The highest color and quality values were obtained from Biosmart x 4.0 g N m^{-2} interaction for the spring season, and from the Biosmart x 4.0 g N m^{-2} and hexaferm x 4.0 g N m^{-2} interaction for the summer and autumn seasons. Also, Biosmart x 4.0 g N m^{-2} interaction gave the highest clipping yield, and Floranid x 0.0 g N m^{-2}, sewage sludge x 0.0 g N m^{-2} interactions gave the lowest clipping yield values.

In our study, zoysiagrass and seashore paspalum showed high color and quality values for summer season. Likewise, the highest clipping yields were obtained from zoysiagrass and seashore paspalum (Table 5, 6). Due to improved morphological uniformity seashore paspalum and zoysiagrass are high turf quality (Hanna and Anderson, 2008). Hybrid Bermudagrass color, quality, and clipping yield values found lower than other warm-season turfgrasses (Table 5, 6). Rezende et al. (2020) determined that Bermudagrass has slow growth, and consequently a low need for maintenance. During our study, tall fescue, which is a cool-season turfgrass, has low color and quality scores in the summer season when compared to warm-season turfgrasses. The highest color and quality values of tall fescue were obtained in the spring and autumn season when the temperature is lower than summer months. As a result, higher turfgrass color and quality values were obtained from the warm-season turfgrasses at low nitrogen doses in the summer season than the cool-season turfgrasses. Temperature is the major factor limiting the growth of cool-season turfgrasses during the summer months (Jiang and Huang, 2001). Researchers indicated that warm-season turfgrass optimum growth rate occurs at lower nitrogen concentrations than cool-season grasses also, nitrogen requirements are less than cool-season grasses. Warm-season grasses grow faster than cool-season turfgrass across a wide range of nitrogen concentrations (Wilson and Brown, 1983; Brown, 1985).

As a rule, warm season turfgrasses perform well in the summer on average, whereas cool-season turfgrasses do well during the coldest months (Richardson et al., 2008; Charif, 2021). Some researchers have observed that warm-season grass species use less water to generate the same dry matter weight due to their distinct physiology, therefore they are more suitable to Mediterranean climates and have greater recovery when compared to cool-season turf species (Croce et al., 2004; Turgeon, 2012; Volterrani and De Bertoldi, 2012; Lulli et al., 2012; Fontanelli et al., 2017). When warm-season turfgrasses begin active development during an optimal spring transition, cool-season turfgrasses lose vigor and density (Horgan and Yelverton, 2001).

The amount of nitrogen applied was the most important factor in determining turfgrass quality (Trenholm and Unruh, 2005). Increasing the dose of N application enhanced the color and quality for both years in our research. The 4.0 g N m^{-2} dose provided the highest turf color and quality. The second highest turf color and quality obtained was 3.0 g N m^{-2}. Except for the summer season of color, 2.0 g N m^{-2} nitrogen dose exhibited unacceptable turf color and quality. In all seasons, the control N rate (0.0 g N m^{-2}) ranked unsatisfactory quality (rating < 6). Clipping yield values increased with increasing nitrogen doses (Table 6). Turf quality is a multifaceted feature that plays a critical role in turfgrass assessment (Russi et al., 2004). The visual ratings of turfgrass quality, which is based on a mix of color, density, uniformity, disease or environmental stress, and other factors, is generally done monthly or seasonally on a scale of 1 to 9 (Morris and Sherman, 2000). In the present study, an acceptable turfgrass color and quality were attained under the 3.0 g N m^{-2} dose treatment throughout all months of the trial season. (Table 5). Our previous study applied the doses of chemical fertilizers (0, 2, 4, 6 g N m^{-2}) on three different warm-season turfgrass species of the hybrid Bermudagrass (Tifdwarf: *Cynodon dactylon x Cynodon transvaalensis*; Gobi, Sydney: *Cynodon dactylon* L. Pers.) was determined a nitrogen dose of 4.0 g N m^{-2} for acceptable turfgrass color and quality in the Mediterranean-type climate (Bilgili et al., 2017). It can be said that the application of microbial fertilizer in lower doses compared to chemical fertilizer improves color and quality.

Zoysiagrass x 4.0 g N m^{-2} and seashore paspalum x 4.0 g N m^{-2} interactions gave the highest turf color and quality values. While zoysiagrass x 4.0 g N m^{-2} interactions gave the highest clipping yields, hybrid Bernudagrass x 0.0 g N m^{-2} gave the lowest clipping yields.

Warm-season turfgrasses develop best in the temperature range of 25-35°C. These species thrive well in the summer when the temperature is high. In autumn and winter, when the temperature drops below 10°C, their growth ceases and their color turns yellow-brown (Christians, 2004; Bilgili et al., 2016). "Dormancy", which appears in the form of yellowing in warm-season turfgrass plants in winter, occurs as a result growth arrest and fragmentation of chlorophyll molecules, only the living growth points remaining in the knuckles of stolons and rhizomes survive the winter (Avcioglu, 1997). According to the recorded data of both years, which revealed similar results; the earliest zoysiagrass entered the dormant period in autumn, followed by seashore paspalum, and hybrid Bermudagrass varieties, respectively. The period of dormancy for both years showed substantial similarity to the previous year for zoysiagrass (80 and 83 days), seashore paspalum (133 and 123 days), and hybrid Bermudagrass (108 and 110 days) (Table 7). Some researchers declare that zoysiagrass enters the dormant period at the earliest in autumn, and they report results consistent with our findings. Also, researchers working on zoysiagrass report that this turfgrass plant has a shorter dormant period than other warm-season turfgrasses (Croce et al., 2004; Aaron et al., 2007; Salman, 2008). Certain components of turfgrass care, such as fertilizer, mowing, and irrigation type, can also impact quality, color, and dormancy and should be considered when evaluating turfgrasses for dormancy length.

Table 7. Dates of the beginning and the exit from dormancy during the years of trial.

Species	Beginning of dormancy	Cessation of dormancy	Vegetation (days)	Dormancy (days)
Tifdwarf		31.03.2017		108
Seaspray		22.03.2017		133
Zenith		08.03.2017		80
Tifdwarf	04.12.2017	24.03.2018	248	110
Seaspray	10.12.2017	12.04.2018	263	123
Zenith	10.12.2017	03.03.2018	277	83
Tifdwarf	03.12.2018		254	
Seaspray	08.12.2018		271	
Zenith	08.12.2018		280	

CONCLUSION

In this research, two slow-release fertilizers, one organomineral fertilizer, and one sewage sludge were used. Results of this study showed that effects of the slow-release fertilizers used in the study were different. Among the slow-release fertilizers, slow-release fertilizer (NS2) gave better turf color and quality than slow-release fertilizer (NS1) showed a lower performance. The slow-release fertilizer (NS2), organomineral fertilizer gave high turf color and quality. The lowest performance was obtained sewage sludge fertilizer. Zoysiagrass and seashore paspalum showed almost equivalent scores in terms of color and quality parameters. We can say that zoysiagrass and paspalum turfgrass species are more successful than other turfgrass species. Our results suggest that to provide acceptable turf color and quality at least throughout the growing season should be fertilized with 3.0 g m^{-2} N monthly. To eliminate the negative environmental effects caused by chemical fertilizers, a sustainable understanding, and program that envisages the effective application of microbial fertilizers in agriculture should be put into effect. In this context, slow-release and organomineral fertilizers can be assessable as sources that will meet the nutritional needs of the turfgrasses.

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