

e-ISSN: 2458-8377



SELCUK
UNIVERSITY
PRESS

SJAFS

SELCUK JOURNAL OF AGRICULTURE AND FOOD SCIENCES

VOL 38 NO 2 2024 AUGUST



<https://dergipark.org.tr/tr/pub/selcukjafsci>



Selçuk Journal of Agriculture and Food Sciences

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Product Information

Publisher	Selçuk University
Owner (On Behalf of SUAF)	Prof. Dr. Hüseyin YILMAZ Rector
Editor in Chief	Prof. Dr. Ercan Ceyhan, Selçuk University, Türkiye
Printing House	Selçuk University Press
Date of Publication	30.08.2024
Language	English
Frequency	Published three times a year
Type of Publication	Double-blind peer-reviewed, widely distributed periodical
Indexed and Abstracted	TR DİZİN GOOGLE SCHOLAR SCIENTIFIC INDEXING SERVICES (SIS) ARAŞTIRMAX CAB ABSTRACTS / GLOBAL HEALTH CAB ABSTRACTS/VETERINARY SCIENCE DATABASE CROSSREF CAB DIRECT EBSCO - Food Science Source EBSCO - Academic Search Main Edition EBSCO - Academic Search Alumni Edition EBSCO - Academic Search Premier EBSCO - Academic Search Complete EBSCO - Academic Search Elite EBSCO - Academic Search Ultimate MIAR SCILIT ESJİ Dimensions DOAJ OAJI ERIH PLUS ACARINDEX Cosmos
Aims and Scope	Selçuk Journal of Agriculture and Food Sciences aims to publish scientific research results from national and international priority areas that can be referenced in the future. It aims to publish articles in the fields of Horticulture, Plant Protection, Food Science and Technology, Agricultural Economics, Field Crops, Agricultural Structures and Irrigation, Agricultural Machinery, Soil Science, plant Nutrition, and Animal Husbandry. The journal aims to contribute to science by publishing high-quality publications of scientific importance. For this purpose, original research articles, reviews, case reports, and letters to the editor are published in all fields related to basic or extended field experiences in agricultural sciences.
Web Address	https://dergipark.org.tr/tr/pub/selcukjafsci
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Using Molecular Markers to Improve Potato Lines Resistant to Pathogens

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HIGHLIGHTS

- Approximately 130.000 potato hybrid seeds were used in the study.
- At the end of the study, it was concluded that marker-assisted selection could be used successfully in potato cyst nematode, wart, late blight and PVX resistance.

Abstract

Potato is a very important crop for human nutrition worldwide. Potato disease and pests can cause economic yield losses. In this study, approximately 130.000 potato seeds from different genetic backgrounds were obtained, evaluated, and selected from 2008 to 2016 for developing new cultivars. After several years, superior lines were tested in different locations to select new cultivars. At the end of the potato breeding program, eight superior potato lines (Nos 12-55-07, 12-55-16, 12-68-05, 12-69-39, 13-67-25, 13-66-23, 12-45-24 and 13-66-75) were submitted for registration as commercial cultivars. The use of resistance genes is the most effective method for controlling these diseases and pests, and DNA markers that are tightly linked to resistance genes are available. The aim of this research was to evaluate the use of closely linked molecular markers for combining *Gro1* and *H1* for cyst nematode resistance, *Sen1* for wart resistance or with *R1* for late blight or with *Rx1* for PVX in advanced breeding lines and commercial candidate cultivars. In the research, 61 advanced lines were checked for the presence of five markers. As a result, only two breeding lines (Nos 31-01-03 and 32-02-52) were determined to have positive results for four markers, and nine advanced lines were found to have positive marker results for *Sen1*, *H1*, and *R1* at the same time. In 6 advanced lines, positive results were acquired from both *Sen1*, *H1*, and *Rx1*, 2 lines *Sen1*, *Gro1*, and *H1*, and only one line No. 12-45-24 *Sen1*, *Gro1*, *H1*, and *Rx1* were found to have positive results for markers. Marker-assisted selection for cyst nematode, wart, late blight, and PVX will be performed using potato breeding programs.

Keywords: Cultivar cyst; *Globodera*; marker; potato

1. Introduction

Fungal, bacterial, and viral diseases and pests such as nematodes and potato beetles are the major threat to potato production worldwide. In potato breeding studies, cultivars resistant to bacterial, fungal, and viral diseases and pests are essential. Classic breeding for resistance to pathogens and pests includes the recognition of resistance resources, which are often found in local, wild and exotic genetic sources, the transfer of resistance agents into cultivars by backcrossing to potato advanced genotypes and phenotypic selection (Gebhardt et al. 2006). Pathologic tests for resistance in glasshouse / field conditions are basic, but space and time consuming are required. Alternatively, molecular markers-based DNA molecular markers could be used without specific facilities for different pathological tests (Babu et al. 2004; Xu and Crouch 2008; Bradshaw 2022).

Citation: Özkaynak E (2023). Using molecular markers for improving of potato lines resistant to pathogens. *Selcuk Journal of Agriculture and Food Sciences*, 38(2), 193-204. <https://doi.org/10.15316/SJA.FS.2024.018>

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Received date: 13/05/2023

Accepted date: 21/05/2024

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Potato wart (*Synchytrium endobioticum*) is a significant quarantine disease in potato production areas. The development of resistant cultivars is needed for its management (Obidiegwu et al. 2014; Szajko et al. 2020). For resistance to *S. endobioticum* pathotype 1, *Sen1*, a single dominant gene was defined in diploid germplasm and mapped to potato chromosome XI (Hehl et al. 1999; Gebhardt et al. 2006). The potato cyst nematode *Globodera rostochiensis* is one of the most troublesome pests of the potato, and yield losses caused by potato cyst nematodes are estimated to be 30% worldwide (Milczarek et al. 2011; Milczarek 2012). The dominant gene *Gro1* was shown to offer resistance to the *G. rostochiensis* pathotype Ro1 (Paal et al. 2004). Another single dominant gene, *H1*, presents resistance to the pathotypes Ro1 and Ro 4 of *G. rostochiensis* (Milczarek et al. 2014). The high-resolution map of *H1* supplied firmly linked AFLP markers (Bakker et al. 2004), from which Ohbayashi et al. (2010) improved a sequence-tagged site marker PCN (Mori et al. 2011).

Potato late blight achieved by *P. infestans* is the most significant fungus disease of potato, especially in the rainy regions of the world. *R1* is located on potato chromosome V (Leonards-Schippers et al. 1992; Plich et al. 2016), which was cloned, and primers for *R1* have been pressed and used in breeding programs (Ballvora et al. 2002). PVX can infect potato seed production and account for 20%–30 % yield loss (Ahmadvand et al. 2013). Control of viral infections are generally the most effective method for cultivar genetic resistance. *Rx1* was the dominant resistance gene detected in *S. tuberosum* subsp. *andigena* (Ahmadvand et al. 2013). Ohbayashi et al. (2010) improved an STS marker linked to *Rx1*. The recombination frequency was found to be 1.3% in this marker (Mori et al. 2011). Molecular markers of PVX that are firmly linked to the genes have been improved for potato breeding programmes (Ahmadvand et al. 2013; Ohbayashi 2019).

In this study, we used molecular markers closely linked to the *Gro1* and *H1* genes for cyst nematode resistance, *Sen1* gene for wart resistance, *R1* gene for late blight, and *Rx1* gene for PVX in a potato breeding program to developed superior lines bearing multiple genes.

2. Materials and Methods

2.1. Plant material

Tetraploid potato populations, including European and local cultivars and genotypes, were used as parents for improving new cultivars resistant to potato cyst nematode, wart disease, and late blight in the potato advanced breeding program. Almost 130000 seeds from distinct genetic structures were ensured and evaluated between 2008 and 2016.

2.2. Plant growth

Potato breeding lines were planted with a 30 x 70 cm planting distance under field conditions. Fertilizer was administered at 40/50 kg ha⁻¹, P₂O₅/60/80 kg ha⁻¹ N and 80/100 kg ha⁻¹ K₂O in distinct trial fields and locations. Weeds were controlled by hand and herbicide after emergence. Disease, pest control, and irrigation were performed according to practice.

2.3. Selection of the advanced lines

The selection of the advanced breeding lines was applied during two periods (early season: January-May; medium-late season: May-October between 2008 and 2016) in the experimental fields at Yuksel Seed in Antalya. After the first two screenings, the field performances of selected potato lines were evaluated in the important potato production provinces Afyonkarahisar, Niğde, Adana, and Izmir in Turkey. Sixty-one improved breeding lines were chosen from these F1 populations because they contained resistance genes and other superior agronomic and tuber yield traits.

2.4. DNA isolation

Genomic DNA was isolated from young fresh leaves of potato lines using the Wizard Magnetic Kit (Promega) according to the manufacturer's instructions. The marker literature is listed in Table 1.

Table 1. Molecular markers used in the study.

Pathogen	Gene	Genetic control	Literature
<i>Globodera rostochiensis</i>	<i>Gro1</i>	dominant single gene	Paal et al. (2004); Gebhardt et al. (2006)
<i>Globodera rostochiensis</i>	<i>H1</i>	dominant single gene	Mori et al. (2011)
<i>Synchytrium endobioticum</i>	<i>Sen1</i>	dominant single gene	Bormann et al. (2004); Gebhardt et al. (2006)
<i>Phytophthora infestans</i>	<i>R1</i>	QTL	Ballvora et al. (2002)
PVX	<i>Rx1</i>	dominant single gene	Mori et al. (2011); Ohbayashi et al. (2010)

All PCR reactions were set up in a total volume of 25 µl containing 20 ng of genomic DNA, each forward and reverse primer at 0.4 µM, 1 PCR Buffer, 2 mM MgCl₂, 0.4 mM dNTPs, and 1 U of Taq DNA polymerase (Vivantis) and performed in the thermocycler PTC-200 (MJ Research, USA). *Rx1* gene resistance to potato virus X was screened using the RxSP-S3 and RxSP-A2 primer sets (Ohbayashi et al. 2010; Mori et al. 2011). PCR products were separated on a 2% agarose gel containing TAE buffer at 110 V for 2h and visualized under UV light after staining with ethidium bromide. Electrophoresis was performed on a 2.5% agarose gel.

3. Results

3.1. Breeding and selection

To develop new potato cultivars resistant to *Globodera rostochiensis*, *Synchytrium endobioticum*, *Phytophthora infestans*, and PVX, different genotypes with tuber flesh colors of red and purple were used in breeding. In the first two selection periods under field conditions, 680–840 potato lines were selected during those years. The performances of these lines were evaluated in two different potato production areas (Adana and Niğde Provinces) beginning in 2010. Later, in the 5th selection year, approximately 100-150 advanced potato lines were used using numerous tubers (50-60 tubers) in Turkey's different potato production areas (Afyonkarahisar, Niğde, Adana and İzmir). In the 6th selection year (utilizing 25-45 lines up to the years), minitubers were produced by tissue culture and evaluated in the target areas to develop cultivar candidates. Because of selection, 61 advanced potato lines with good agronomic properties and resistance genes were selected. Of these, 12 were selected as superior promising lines for the next 10 years (Table 2). Eight of 61 lines were selected as commercial candidate cultivars at the end of the large-scale trials conducted in 2012, 2013, and 2014. Moreover, they were evaluated in 2 locations for 4 replications with commercial control cultivars for cultivar registration in 2015 and 2016. Nos 12-55-07, 12-55-16, 12-68-05, 12-69-39, and 13-67-25 were registered as the names of Cevher, Demet, Asya, Maraton, and Soylu, respectively.

3.1. Molecular markers

61 advanced lines were analyzed for *Gro1*, *H1*, *Sen1*, *R*, and *Rx1* genes. PCR results of potato molecular markers linked to *Sen 1*, *H1*, and *R1* genes are shown in Figures 1-3. The *Sen-1* marker was used in 61 breeding lines, 42 of them were positive and 19 of them were negative. Four of them are commercial candidate lines because of their good agronomic and yield performance. The *H1* marker was evaluated in 61 advanced breeding lines. Of these, 43 yielded DNA bands, and the remaining did not produce DNA fragments. A total of 61 lines were checked for *Gro1* and *R1* genes using molecular markers. 12 breeding lines produced expected DNA fragments, and 49 lines have no bands for *Gro1*. 20 and 21 breeding lines produced DNA bands, and 41 and 40 lines did not presence for *R1* and *Rx1* markers, respectively.

Table 2. The presence/absence of markers and genes improved potato lines.

	Wart					Cist Nematode					LB					PVX				
<i>Female</i>	<i>Sen1</i>	<i>Gro1</i>	<i>H1</i>	<i>R1</i>	<i>Rx1</i>	<i>Male</i>	<i>Sen1</i>	<i>Gro1</i>	<i>H1</i>	<i>R1</i>	<i>Rx1</i>	Adv. Lines	<i>Sen1</i>	<i>Gro1</i>	<i>H1</i>	<i>R1</i>	<i>Rx1</i>			
YT-1*	+	+	+	-	+	YT-2	+	-	+	+	-	11-04-36	+	-	+	-	+			
YT-1	+	+	+	-	+	YT-2	+	-	+	+	-	11-04-39	+	-	+	+	-			
YT-3	-	-	+	-	+	YT-1	+	+	+	-	+	11-05-29	-	-	+	-	-			
YT-4	+	-	-	-	-	YT-5	-	-	+	-	-	12-03-85	-	-	-	-	-			
YT-4	+	-	-	-	-	YT-6	+	+	+	-	-	12-04-12	-	-	+	-	-			
YT-7	+	-	-	-	-	YT-1	+	+	+	-	+	12-16-79	-	-	+	-	-			
YT-7	+	-	-	-	-	YT-1	+	+	+	-	+	12-16-84	+	-	+	-	-			
YT-8	-	-	+	-	-	YT-1	+	+	+	-	+	12-44-12	-	-	-	+	-			
YT-8	+	-	+	-	-	YT-9	+	-	-	-	+	12-45-24	+	+	+	-	+			
YT-10	+	-	+	-	+	YT-11	+	+	+	-	-	12-52-100	-	+	+	-	+			
YT-10	+	-	+	-	+	YT-2	+	-	+	+	-	12-55-07	+	-	-	+	-			
YT-10	+	-	+	-	+	YT-2	+	-	+	+	-	12-55-16	+	-	+	+	-			
YT-10	+	-	+	-	+	YT-2	+	-	+	+	-	12-55-29	+	-	+	-	+			
YT-1	+	+	+	-	+	YT-8	+	-	+	-	-	12-68-05	-	-	+	-	+			
YT-1	+	+	+	-	+	YT-12	+	-	+	-	+	12-69-39	+	-	+	-	+			
YT-13	+	-	+	+	-	YT-1	+	+	+	-	+	12-123-03	+	-	-	+	-			
YT-1	+	+	+	-	+	YT-14	-	-	+	-	+	12-217-03	+	-	+	-	+			
YT-1	+	+	+	-	+	YT-10	+	-	+	-	+	12-200-02	+	+	-	-	-			
YT-11	+	+	+	-	-	YT-7	+	-	-	-	-	13-39-01	+	+	+	-	+			
YT-9	+	-	-	-	+	YT-1	+	+	+	-	+	13-46-25	+	-	-	-	-			
YT-1	+	+	+	-	+	YT-8	+	-	+	-	-	13-66-75	+	-	+	-	+			
YT-1	+	+	+	-	+	YT-8	+	-	+	-	-	13-66-23	+	-	+	-	+			
YT-1	+	+	+	-	+	YT-12	+	-	+	-	+	13-67-25	+	-	+	-	+			
YT-2	+	-	+	+	-	YT-8	+	-	+	-	-	13-80-16	-	-	+	+	-			
YT-2	+	-	+	+	-	YT-8	+	-	+	-	-	13-80-34	-	-	+	-	-			

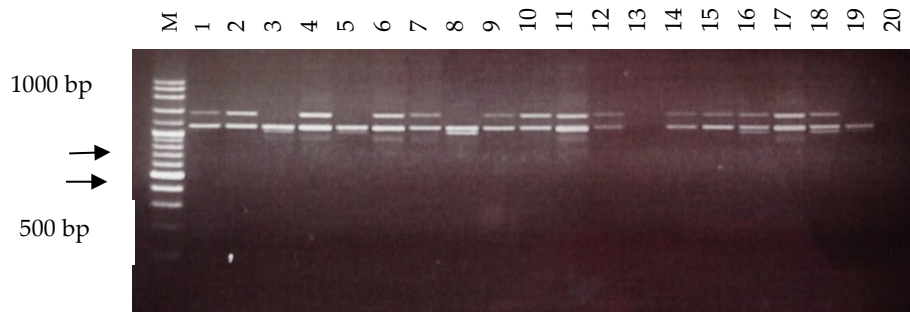
YT-15	+	-	+	-	-	YT-16	+	-	-	+	-	22-06-44	+	-	-	+	-
YT-4	+	-	-	-	-	YT-1	+	+	+	-	+	22-19-29	-	-	-	+	-
YT-1	+	+	+	-	+	YT-16	+	-	-	+	-	22-22-20	+	-	-	+	+
YT-10	+	-	+	-	+	YT-16	+	-	-	+	-	22-26-25	+	-	-	+	-
YT-17	+	-	+	-	+	YT-11	+	+	+	-	-	22-27-03	+	-	+	-	+
YT-17	+	-	+	-	+	YT-9	+	-	-	-	+	22-28-34	+	+	+	-	-
YT-9	+	-	-	-	+	YT-16	+	-	-	+	-	22-32-01	+	-	-	+	+
YT-18	+	-	+	-	-	YT-11	+	+	+	-	-	22-96-09	+	+	-	-	-
YT-18	+	-	+	-	-	YT-17	+	-	+	-	+	22-99-02	+	+	-	-	+
YT-18	+	-	+	-	-	YT-17	+	-	+	-	+	22-99-33	+	-	+	-	+
YT-19	+	-	+	-	-	YT-20	+	-	+	-	+	22-102-71	+	-	+	-	+
YT-19	+	-	+	-	-	YT-20	+	-	+	-	+	22-107-29	-	-	-	-	-
YT-21	+	-	+	+	-	YT-1	+	+	+	-	+	22-128-07	+	-	+	+	-
YT-1	+	+	+	-	+	YT-12	+	-	+	-	+	31-23-44	+	-	+	-	-
YT-2	+	-	+	+	-	YT-12	+	-	+	-	+	31-28-03	+	-	-	-	+
YT-1	+	+	+	-	+	YT-14	-	-	+	-	+	31-40-30	+	-	+	-	-
YT-15	+	-	+	-	-	YT-16	+	-	-	+	-	31-58-21	+	-	-	+	-
YT-1	+	+	+	-	+	YT-2	+	-	+	+	-	31-69-01	+	-	-	-	-
YT-1	+	+	+	-	+	YT-2	+	-	+	+	-	32-A-322	+	-	+	-	-
YT-1	+	+	+	-	+	YT-9	+	-	-	-	+	32-01-03	+	+	+	+	-
YT-1	+	+	+	-	+	YT-16	+	-	-	+	-	32-02-52	+	+	+	+	-
YT-11	+	+	+	-	-	YT-9	+	-	-	-	+	32-13-05	+	+	+	-	-
YT-1	+	+	+	-	+	YT-9	+	-	-	-	+	32-35-23	-	+	+	-	-
YT-1	+	+	+	-	+	YT-13	+	-	+	+	-	32-71-33	+	-	+	+	-
YT-16	+	-	-	+	-	YT-9	+	-	-	-	-	41-34-33	-	+	+	-	-
YT-22	+	-	+	+	-	YT-23	+	-	+	+	-	41-90-11	+	-	+	+	-
YT-24	+	-	-	-	+	YT-25	-	-	-	+	-	41-119-86	-	-	+	-	+
YT-1	+	+	+	-	+	YT-23	+	-	+	+	-	41-125-16	-	-	+	-	-

YT-1	+	+	+	-	+	YT-25	-	-	-	+	-	41-129-16	+	-	+	+	-
YT-1	+	+	+	-	+	YT-25	-	-	-	+	-	41-129-96	-	+	+	-	+
YT-1	+	+	+	-	+	YT-25	-	-	-	+	-	41-129-102	+	-	+	-	-
YT-1	+	+	+	-	+	YT-10	+	-	+	-	+	41-132-104	+	-	+	+	-
YT-8	+	-	+	-	-	YT-10	+	-	+	-	+	41-133-33	-	-	+	-	-
YT-2	+	-	+	+	-	YT-10	+	-	+	-	+	41-141-14	+	-	+	+	-
YT-4	+	-	-	-	-	YT-25	-	-	-	+	-	41-154-15	-	-	-	-	-
YT-10	+	-	+	-	+	YT-11	+	+	+	-	-	41-164-28	-	-	+	-	+
												Total	+: 42	+: 12	+: 43	+: 20	+: 21
													-: 19	-: 49	-: 18	-: 41	-: 40

	Wart	Cist Nematode			LB	PVX
	<i>Sen1</i>	<i>Gro1</i>	<i>H1</i>	<i>R1</i>	<i>Rx1</i>	
32-01-03 and 32-02-52 potato lines	+	+	+	+	-	
9 potato line	+	-	+	+	-	
6 potato line	+	-	+	-	+	
22-28-34 and 32-13-05 potato lines	+	+	+	-	-	
12-45-24 potato line	+	+	+	-	+	

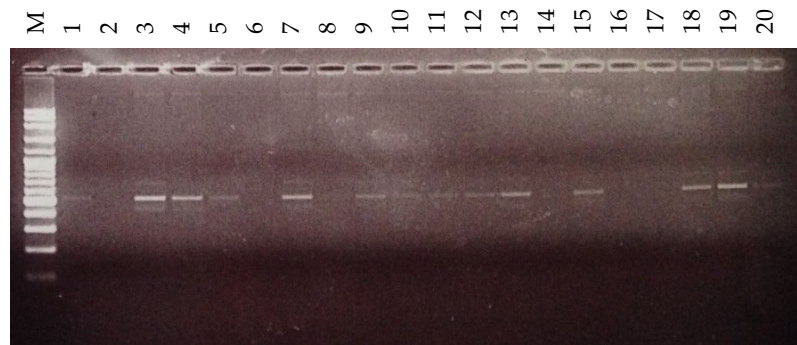
+: presence of molecular marker, -: absence of molecular marker. LB: Late Blight

*: YT-1 to YT-25; European cultivars, exotic cultivars, and local genotypes.



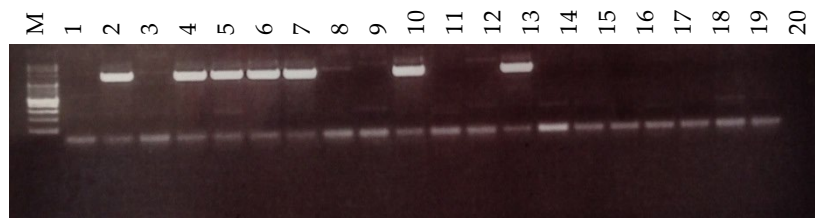
M, molecular marker; resistant lines: 1) 11-04-36, 2) 12-45-24, 4) 12-55-07, 6) 12-55-16, 7) 12-69-39, 9) 13-67-25, 10) 22-27-03, 11) 22-99-33, 12) 22-102-71, 13) 31-23-44, 14) 31-58-21, 15) 31-69-01, 16) 32-A-322, 17) 32-28-03, 18) 41-129-102; susceptible lines: 3) 12-03-85, 5) 41-125-16, 8) 12-68-05, 19) 32-35-23

Figure 1. 1400-bp PCR marker of the dominant allele *Sen1* for resistance to pathotype 1 of *Synchytrium endobioticum*.



M – molecular marker; resistant lines: 1) 11-04-36, 3) 12-55-16, 4) 12-68-05, 5) 12-69-39, 7) 13-66-75, 9) 13-67-25, 10) 22-27-03, 11) 22-99-33, 12) 31-23-44, 13) 32-A-322, 15) 41-119-86, 18) 41-125-16, 19) 41-129-96, 20) 41-164-28; susceptible lines: 2) 12-03-85, 6) 12-55-07, 8) 22-99-02, 14) 31-28-03, 16) 31-58-21, 17) 41-154-15

Figure 2. 506-bp PCR marker of dominant allele *H1* for resistance to the Ro1 pathotype of *Globodera rostochiensis*.



M – molecular marker; resistant lines: 2) 12-55-07, 4) 12-55-16, 5) 22-26-25, 6) 31-58-21, 7) 41-129-16, 10) 41-132-104, 13) 41-141-14; susceptible lines: 1) 12-03-85, 3) 12-68-05, 8) 12-69-39, 9) 13-67-25, 11) 22-27-03, 12) 22-96-09, 14) 22-99-33, 15) 31-23-44, 16) 31-28-03, 17) 41-119-86, 18) 41-125-16, 19) 41-129-102, 20) 41-133-33

Figure 3. CAPS marker SPUD237 (digestion of *AluI*) for detection of resistance to the late blight *R1* allele

Five genetic cultivars (YT-1, YT-2, YT-8, YT-9 and YT-10) were used more as male and female parents compared with other lines and cultivars (Table 3).

Table 3. Summary results of the parents and marker tests.

F/M	Presence / absence of molecular markers in parents						Advanced lines in the presence of the gene <i>Sen1</i>			Advanced lines in the presence of the gene <i>Gro1</i>			Advanced lines with the presence of gene <i>R1</i>			Advanced lines in the presence of the gene <i>Rx1</i>					
	Total	<i>Sen1</i>	<i>Gro1</i>	<i>H1</i>	<i>R1</i>	<i>Rx1</i>	+	-	%	+	-	%	+	-	%	+	-	%			
YT-1	32	+	+	+	-	+	22	10	69	3	29	9	25	7	78	11	21	34	9	23	28
YT-2	11	+	-	+	+	-	9	2	82	0	11	0	8	3	73	5	6	45	3	8	27
YT-10	9	+	-	+	-	+	7	2	78	2	7	22	6	3	67	5	4	56	3	6	33
YT-9	8	+	-	-	-	+	6	2	75	6	2	75	7	1	88	2	6	25	1	7	12
YT-8	8	+	-	+	-	+	2	6	25	1	7	13	7	1	88	2	6	25	4	4	50

Note. F/M: female or male; YT-1 was used 32 advanced lines female or male.

The most used cultivar is YT-1 as a parent. This cultivar was used in 32 advanced line's female or male line. These five cultivars were found to be positive for the *Sen1* marker. YT-1 was found to be positive for *Gro1* and YT-2 positive for *R1* marker. In advanced lines of five mostly used parents, high percentage positive results for *H1* (67-88%) and *Sen1* (except 25 % in YT-8) were determined (Table 3). Advanced lines with the presence of *Gro1* (75%), *R1* (56%), and *Rx1* (50%) were found to be over 50% in Y-9, YT-10, and YT 8, respectively.

4. Discussion

4.1. Breeding and selection

To select new potato cultivars resistant to some important pathogens, 25 different parents were used as mother and father lines. In the mother and father lines, the *Gro1*, *H1*, *R1*, *Sen1*, and *Rx1* genes were analyzed using potato molecular markers linked to the related genes. In addition, the 61 advanced potato lines developed because of breeding studies were analysed using the same markers. In this study, molecular markers were used first at the beginning of the breeding program in mother and father lines and second after the 4th selection year. On the other hand, having good agronomic, plant, and tuber traits, advanced lines could begin to be selected after the 4th selection year. After 5th selection year, advanced potato lines were tested for molecular markers. Eight of the 61 lines were selected as commercial candidate cultivars: 12-45-24, 12-55-07 (Cevher), 12-55-16 (Demet), 12-68-05 (Asya), 12-69-39 (Maraton), 13-66-23, 13-66-75, and 13-67-25 (Soylu); they were submitted for cultivar registration, and some of them were registered.

4.2. Molecular markers

In the research, only two breeding lines (32-01-03 and 32-02-52) obtained positive results for all molecular markers, except *Rx1*. In nine advanced lines, positive marker results were found for *Sen1*, *H1* and *R1*. In six advanced lines, positive results were acquired from both *Sen1*, *H1*, and *Rx1*, and positive results were found for the *H1* and *R1* markers. Milczarek et al. (2011) used *H1* and *Gro1* markers, and in their research, in some cultivars, the markers linked to *Gro1* and *H1* were determined as in our research. They found it in 50 potato lines from 67 that were tested and resistant, and only one *H1*-positive potato breeding line was susceptible, which was an encouraging result. Similar to Milczarek et al. (2011) and Milczarek (2012), in our study we found 43 advanced lines out of 61 that were positive for the *H1* marker. As a result of Milczarek et al. (2011) and Milczarek (2012) studies, markers *H1* and *Gro1* were used to determine resistant breeding potato genotypes, and their presence was set against the conclusion of resistance tests.

Ortega and Lopez-Vizcon (2012) found that the existence of the *Gro1-4* locus of *G. rostochiensis* Ro1 resistance was assessed in 43 breeding clones, with 15 of them being positive (34.9%). In other words, cultivars that could have the *H1* gene and six breeding genotypes were controlled for the presence of the *H1* marker, with all cultivars and two of the breeding genotypes being positive (33.3%). In our study, we found similar results to Ortega and Lopez-Vizcon (2012) for *Gro1* and *H1* markers.

Antonova et al. (2017) used a subset of 113 potato cultivars. All the analyzed cultivars elicited the diagnostic marker of the *Sen1* gene, whereas several susceptible cultivars lost this diagnostic fragment. The tested markers of *Gro1-4* and *H1* which present resistance to the *G. rostochiensis pathotype* Ro1, revealed dissimilar presumability. In the molecular screening of potato cultivars, it is better to use a few markers of these genes. Saynakova et al. (2018) used the multiplex PCR technique for genes for resistance to potato wart disease and *G. rostochiensis*. 40 samples were tested using genetic markers to recognize genes for resistance to wart disease (*Sen1*) and *G. rostochiensis* (*H1*, *Gro1*) in the genome. The sample contained two cultivars, three populations produced by self-pollination of the cultivar 'Ideal', and 35 individually selected potato hybrids. As a result of Saynakova et al. (2018) researchers identified markers for *Sen1* in 19 samples, *H1* in 12 samples, and *Gro1* in 6 samples.

Twenty breeding lines were found to be positive for the *R1* molecular markers test. Sharma et al. (2013) reported similar results as in this study. They tested potato breeding materials identified by the *R1* gene. The results of their molecular marker screening showed that 17 lines possessed the *R1* gene. Further, these lines

were tested for *P. infestans* resistance in the laboratory using the detached leaf method. *R1* resistant lines were partitioned as highly resistant, resistant, and moderately resistant. Potato late blight resistance breeding will greatly benefit the use of R genes. Potato cultivars/genotypes in which the durability of resistance has been previously demonstrated are superb breeding components for broad-spectrum R genes (Plich et al. 2015).

Twenty-one lines were positive for *Rx1*. Shaikhaldein et al. (2018) were 25 genotypes of which three contained the *Rx1* genes. They reported that *Rx1*, including genotypes/clones, should be regarded as a support for potato crop development. Genotypes/clones that demonstrate the presence of molecular markers are violently suggested to be used by breeders to improve new PVX extreme resistance potato cultivars (Özkaynak 2020).

In this study, to determine the prevalence of resistance genes in potato breeding lines, we tested 5 resistance genes: *Sen1*, *Gro1*, *H1*, *R1*, and *Rx1*. We revealed that many breeding lines and commercial candidate cultivars have not only *H1*, *Gro1*, but also *Sen1*, *R1*, and *Rx1*, which are potentially resistant to potato cyst nematode, potato wart disease, late blight and PVX. The comparison of molecular marker test expenses with the costs of phenotypic evaluation of cyst nematode, late blight, potato wart resistance, and PVX in advanced potato breeding programs in Turkey presented here clearly demonstrates that the use of molecular markers is cheaper. Similar conclusions were drawn by Mori et al. (2011), Ortega and Lopez-Vizcon (2012), Slater et al. (2013), and Milczarek et al. (2014). 12 of these advanced lines (Nos 12-03-85, 13-39-01, 13-66-81, 22-06-44, 22-27-03, 22-32-01, 31-58-21, 32-A-322, 32-02-52, 41-125-16, 41-129-96 and 41-154-15) were selected as superior promising lines over the next 10 years.

5. Conclusions

Molecular markers linked to the loci of interest could be used in advanced potato breeding to select resistant lines/clones/genotypes (Bradshaw 2022). The phenotypic assessment of resistance to *G. rostochinensis*, potato wart, late blight, and PVX is costly and time consuming. Using molecular markers facilitates the selection of resistant lines at the early and preliminary stages of potato breeding which ensures a rapid decrease in the number of individuals under selection in further steps. To be applicable and suitable for molecular marker-assisted selection, the marker should be cheap, practical in use, reproducible, and special for the character. Main conclusions;

1. For a successful potato breeding program, approximately 20.000 F1 seeds were used at a minimum.
2. The most effective method to control diseases and pests were used resistance genes tightly linked to DNA markers.
3. For potato wart, cyst nematode, *Globodera rostochinensis*, late blight, and PVX, molecular markers were effectively used.
4. The results showed that the genetic background is determinative and that it is important when using potato molecular markers.

The genomic information generated as a result of this research will facilitate the estimation of phenotypic outcomes by minimizing large-scale screening of lines in future breeding programs for each generation.

Author Contributions: The authors have read and agreed to the published version of the manuscript.

Funding: This research was funded by the Scientific and Technological Research Council of Turkey and its TEYDEP (grant numbers 3110172 and 1140133).

Conflicts of Interest: The authors declare no conflicts of interest.

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Evaluation of The Use of Different Foot Types in Pneumatic Precision Single Grain Stubble Seeder

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HIGHLIGHTS

- Measuring the fuel consumption of different types of embedding feet
- Determining the degree of sprout emergence of different types of burying feet in the field
- Determination of soil surface roughness to different types of burying feet
- Conducting laboratory trials of the pneumatic precision seeder

Abstract

In this study, the effects of a pneumatic precision stubble planter with different planter feets on fuel consumption, post-sowing surface profile unevenness and field germination emergence values in second-crop corn production were investigated. Field trials were carried out on the adhesive tape system at Selçuk University Faculty of Agriculture Sarıcalar Research and Production Farm, and laboratory experiments were carried out on the adhesive tape system at Selçuk University Faculty of Agriculture, Department of Agricultural Machinery and Technologies Engineering. Field trials were carried out at a working speed of 5.4 km h⁻¹, and adhesive tape experiments were carried out at a tape speed corresponding to a working speed of 5.4 km h⁻¹. According to laboratory and field conditions; Acceptable seed spacing values of 0.5-1.5Z were found to be over 80%, and 0.5<Z and 1.5Z were found to be below 10%. After the stubble sowing process, soil surface roughness values were determined as 14.28% on the axe-type burying foot and 16.35% on the double-disc burying foot. Fuel consumption was determined as 10.90 l ha⁻¹ for the pneumatic precision stubble seeder with ax cultivator foots, and 9.81 l ha⁻¹ for the pneumatic precision stubble seeder with disc cultivator. The FGE (field germination emergence rate) rate of the pneumatic seeder with an ax-type planter foot was determined as 87.30%, and the FGE rate of the pneumatic precision seeder with a double disc-type planter foot was determined as 85.50%. Ax-type planter foot gave better results than disc foot in terms of surface profile unevenness, field germination emergence rate and sowing depth in both applications.

Keywords: Stubble sowing; corn; fuel consumption; field germination emergence; surface profile roughness

Citation: Çıtıl E, Marakoğlu T, Çarman K (2024). Evaluation of the use of different foot types in pneumatic precision single grain stubble seeder. *Selcuk Journal of Agriculture and Food Sciences*, 38(2), 205-215. <https://doi.org/10.15316/SJAfS.2024.019>

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Received date: 23/10/2023

Accepted date: 04/06/2024

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1. Introduction

The introduction should briefly place the study in a broad context and highlight why it is important. It is increasing fuel prices in parallel with the decrease in oil resources in the world has necessitated the search for cost-reducing alternatives in agriculture, as in many areas. In addition, rapidly developing agriculture in parallel with the increasing population has aimed to increase productivity without considering sustainability, and as a result, intensive use of pesticides, fertilizers and excessive soil cultivation has come to the fore. The soil loses its fertility as a result of intensive use of pesticides, fertilizers and excessive tillage. In addition to the negative effects of increasing fuel prices on production costs, protective tillage and direct sowing techniques have been developed in order to improve the soil structure that has deteriorated as a result of the intensive use of agricultural lands day by day. Conservation tillage and direct sowing methods, which save time and fuel, prevent erosion and allow efficient use of agricultural areas for a long time (Çakır et al. 2007).

It is a soil cultivation system in which at least 30% of the soil surface remains covered with plant residues after seed sowing, in order to ensure the continuity of soil fertility by reducing soil and water erosion in protective soil tillage methods. In the majority of studies on conservation tillage, it has been stated that conservation tillage and stubble cultivation increase energy efficiency by 25% to 100% and reduce energy need by 15% to 50% (Karayel and Özmerzi 2007).

Conservation tillage is an agricultural practice in which sufficient vegetation and plant residues are left on the soil surface in order to protect water and soil, minimizing energy use and costs. In stubble sowing, profitable production is achieved by minimizing soil erosion that may occur due to various reasons. Protection of soil moisture, energy consumed, labor force used and even agricultural machinery used are additional gains that should be emphasized as much as the protection of the soil (Köller 2003).

No-till agriculture; It is the direct sowing of seeds into undisturbed stubble soil with any soil tillage machine. In the direct cropping system, the soil is left undisturbed from sowing to harvest and from harvest to sowing. The burying feet of the direct sowing machine open a narrow incision in the soil and sowing is done. Therefore, direct sowing machines do not disturb the soil outside of the sowing process. The planters used to open a furrow at the depth where the seed will be deposited must be able to cut through plant residues and sink deep enough into undisturbed soil to provide sufficient depth. The solution to the weed problem is provided by herbicides applied directly before or after sowing. The application method and time of herbicide are selected according to the density of the weed and climatic conditions (Çıkman et al. 2017).

In evaluating the performance of the tillage machine, the surface smoothness of the processed soil is an important feature in the preparation of the seedbed and in the control of water flow and soil erosion caused by rainfall (Römkens and Wang 1987).

Soil surface smoothness is a property affected by the processes performed on the soil. It is the soil surface structure formed by the random preparation of clods on the soil. Surface roughness occurs due to deterioration of the soil surface due to natural reasons or cultural processes (Guillobez and Arnaud 1998; Hauer et al. 2001).

Bayhan et al. (2001) in their study titled Reduced tillage and direct sowing applications in second crop silage corn agriculture, found that the lowest fuel consumption was in the direct sowing application among the applications and the highest efficiency was obtained from the application using a soil tillage combination, and similarly, Yalçın and Çakır (2006) in their study, they obtained fuel consumption values as 60 l ha⁻¹ in the traditional method and 7.5 l ha⁻¹ in the direct sowing method.

Especially in the single grain precision sowing method, where it is desired to plant as many seeds as the number of plants to be grown, the importance of seed distribution increases as each plant that does not germinate or does not continue its development well will directly affect the yield. In precision sowing, planter layouts have the primary effect of ensuring the uniformity of seed distribution on the row. However, it should not be forgotten that the problems that may occur in the proper placement of the seeds thrown from the planter into the soil have an important effect on the quality and success of the sowing process. Even if the precision

sowing machine has the most modern, highest quality sowing arrangement, sowing failures may occur due to the inability of the burying feet to perform their duties well (Önal 1995).

In this study, the effects of the pneumatic precision stubble seeder (P.P.S.S.) with different burying feet on fuel consumption, surface surface roughness, field sprout emergence and row distances in second-crop corn production were investigated.

2. Materials and Methods

Field trials were carried out in Selçuk University Faculty of Agriculture Sarıcalar Research and Production Farm, and laboratory trials were carried out with 3 replications on the adhesive tape system in the application workshop of Selçuk University Faculty of Agriculture, Department of Agricultural Machinery and Technologies Engineering. Some soil properties of the trial area are given in Table 1.

Table 1. Some soil properties of the in trial area

	% Sand	20.4
Tekstüre Analysis	% Silt	24.7
	% Clay	54.9
Tekstüre Class	Clay-Loam	
Applications		
Volume Weight (g.cm ⁻³)	(0-20 cm)	1.51
Moisture content (%)	(0-20 cm)	17.85
Penetration resistance (Mpa)	(0-20 cm)	1.41
Porosity (%)		43.01
pH		7.5

Before applying stubble sowing, the amount of stubble per unit area of the application field was determined. To determine the amount of stubble, the average amount of stubble was determined by counting the amount of stubble in a 1x1 m² frame created from different points of the field (Figure 1).



Figure 1. Determination of stubble amount

In the trials, two different vacuum-type pneumatic precision single grain stubble planters (P.P.S.S.) with 4-row axes and double-disc burying feet were used (Figure 2 and Figure 5).

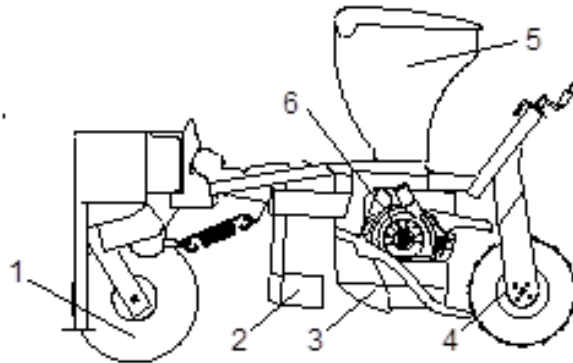


Figure 2. P.P.S.S. with axle burying foot

(1. Stubble cutting disc, 2. Stubble cleaner, 3. Axle burying foot, 4. Pressing wheel, 5. Seed storage, 6. Planter layout)



Figure 3. Burying foot with axle

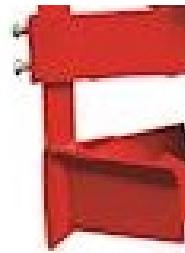


Figure 4. Stubble cleaner

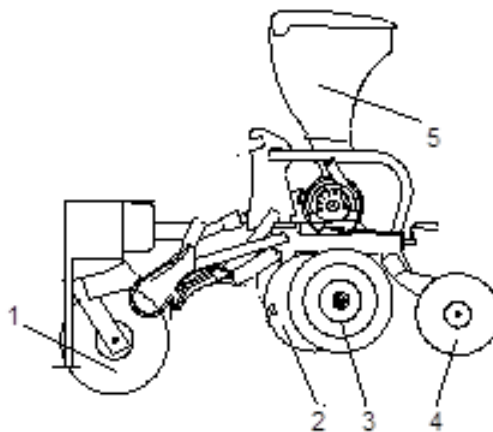


Figure 5. P.P.S.S. with Double disc burying foot

(1. Stubble cutting disk, 2. Double disc burying foot, 3. Depth adjustment wheel, 4. Pressing wheel, 5. Seed tank)

The diameter of the double-disc burying foot is 372 mm and the angle between them is 10°.



Figure 6. Double disc burying foot

Stubble-cutting discs are placed at the front of each unit of both machines to enable stubble cultivation in stubble fields. These discs are corrugated type and their diameter is 470 mm



Figure 7. Stubble cutter disc

DKC5783 FAO 500 was used as seed and the machines were adjusted so that the distance between the rows was 20 cm, the distance between the rows was 70 cm, the forward speed was 5 km h⁻¹ and the theoretical sowing depth was 5 cm. As fertilizer, 20x20 compound fertilizer was used with sowing.

The experiments were set up on a field with approximately 3.5 decare of wheat stubble, according to the randomized parcel trial design, with 3 replications (each parcel 3x100m). After sowing, the plots were irrigated with sprinkler irrigation at 4-day intervals during the germination period of the seeds.

A New Holland TD110D brand tractor was used in the trials. Trials were carried out on both foot types at a working speed of 5.4 km.h⁻¹. In order to clearly measure the instantaneous fuel consumption of the tractor during operation, one of the fuel meter connection hoses is connected to the fuel line from the fuel tank to the engine, and the other is connected to the return line from the engine to the fuel tank. The fuel meter measures the amount of fuel going to the engine fuel line and the return line to the fuel tank separately, and the net amount of fuel consumed by the tractor is determined as l h⁻¹ and calculated as l ha⁻¹.



Figure 8. Fuel consumption meter and display

A rod profile meter was used to determine surface unevenness after stubble sowing. The profile meter has rods placed at 2.5 cm intervals on a 1 m long profile material (Çarman 1997).

By measuring the surface profile with a profilometer placed perpendicular to the working direction, field surface unevenness was calculated with the help of the following equation (Kuipers 1957).

$$R=100 \log_{10} S$$

Here;

R: Soil surface roughness (%) after trials and

S: Standard deviation of measured values.



Figure 9. Profilometer

Soil moisture was measured with a TDR device calibrated by the gravimetric method.



Figure 10. Humidity measuring device (TDR)

A penetrometer device with digital display was used to measure the density of the soil. Soil resistance was measured as MPa at every 1 cm up to a depth of 80 cm in vertical condition with a penetrometer. A cone tip with an apex angle of 30° and a cone base area of 1cm² was used in the measurements. Among the values obtained in the measurements, values taken from a depth of 0–20 cm were taken into account.



Figure 11. Penetrometer

In order to determine the field sprout emergence rate, the sprouts emerging on the soil surface were counted by observing during the germination period on 3 randomly selected strips of 10 m length from 2 different rows in each plot, and the TFS was calculated with the following relation using the obtained data (Işık et al., 1986).

$$FSE = \frac{\text{Total number of seeds germinated in one meter}}{\text{Total number of seeds planted in one meter}} \times 100 \quad (1)$$

FSE: Field sprout emergence degree (%)

Since the depth measurement was done under field conditions, the grass length measurement method was used. Plants that had completed the germination period were removed from the soil and the distance between the seed residue and the point where the germinated plant turned from green to white was measured. Measurements were made on 25 plants randomly selected among the rows in each replication, and the data obtained from these measurements were entered into the MS Excel program and the average planting depth and vertical seed distribution coefficient of variation (VK, %) values were calculated in this program (Özmerzi and Keskin 1983).

3. Results and Discussion

The pneumatic precision stubble planter used in the trial was tested to determine the uniformity of distribution in the row under both laboratory and field conditions. The results regarding the adhesive tape test regarding the planter arrangement and the evenness of distribution of the corn seeds obtained in the field according to the planter foot type are given in Table 2. Under laboratory and field conditions, acceptable seed spacing values of 0.5-1.5 Z were found to be over 80%, and 0.5<Z and 1.5>Z were found to be below 10%.

In the adhesive tape experiments carried out in the laboratory, the expression 0.5<Z, which expresses the twinning rate of the seeds on the row, was found to be 0% at a row distance of 20 cm, and the expression 1.5>Z, which expresses the spacing ratio between the seeds on the row, was found to be 2.5%. These values were

found to be below the 10% limit value. It was determined that the ratio of seeds expressing the ideal row distance 0.5-1.5. Z was 97.50%, and this expression was above the 80% limit value (Table 2).

When evaluating single-grain precision sowing systems, the relative proportions of (0.5-1.5) Z plant spacings within the total plant spacing are taken into account. In the distribution of plants on the row (0.5-1.5). Z ratio is considered as acceptable plant spacing on the row. In modern single-grain sowing machines, it is required that the ratio of plants in the Z range (0.5-1.5) be greater than 80%, and the ratio of plants in the <0.5. Z and >1.5.Z range should not be more than 10% (Önal 1987).

In order to determine the field sprout emergence values, the distances between the sprouts emerging on the soil surface on different strips from the plots during the germination period were measured and post-sowing twinning and gap ratios were determined. The twinning rate ($0.5 < Z$) of ax foot and disc foot P.P.S.S. was determined as 6.10% and 8%, respectively. It was observed that these values were below the 10% limit value. The clearance ratio ($0.5 < Z$) for ax foot and disc foot P.P.S.S. was determined as 6.60% and 6.50%, respectively. It was observed that these values were below the 10% limit value (Table 2).

Table 2. Regularity of distribution over the row of the machine

Application	Sowing distance (Z) (cm)	Working speed (km h ⁻¹)	0.5<Z (%)	0.5-1.5Z (%)	1.5>Z (%)
Laboratuvar			0.00	97.50	2.50
Field					
Ax-foot	20	5	6.10	87.30	6.60
Disc- foot			8.00	85.50	6.50

Parish and Bracy (2003) reported that increasing the seed drop height in vacuum pneumatic single-grain sowing machines negatively affects seed distribution.

Karayel and Özmerzi., (2005) stated that due to the structural features of the single-disc and double-disc burying feet used in their research, the seed falling height increases compared to the axe-type and hoe-type burying feet. mm, and 265 mm in the double-disc seeding foot type, they reported that this caused the double-disc burying foot to provide worse horizontal plane seed distribution than expected.

When the applications were compared in terms of field sprout emergence rate, the highest field sprout output value was obtained as 87.30% from the P.P.S.S. with ax type burying feet and 85.50% from the P.P.S.S. with disc type burying feet. The FSE rate of the P.P.S.S. with ax-type burying feet is 2% higher than that of the P.P.S.S. with disc-type burying feet.

According to the results of field trials, the coefficient of variation of the depth distribution of corn and the average sowing depth at 20 cm in-row sowing distance; Values for different applications and foot types are given in Table 3. The ax type burying foot gave better results than the disc foot in terms of both coefficient of variation and sowing depth in both applications (Table 3).

Table 3. % CV and sowing depth of treatments

Applications	Burying foot type	Coefficient of variations (% CV)	Sowing depth (cm)
	Ax- foot	13.5	4.1
	Disc- foot	15.4	3.7

As a result of the depth measurement performed after the germination period, it was reported that the coefficient of variation value of the sowing depth distribution should be at most 25% (Anony-mous 1999).

According to Slattery (1997), in order to obtain a high-yield, sowing should be done at a depth as close to the optimum sowing depth as possible.

Heege (1993) stated the negativities that would occur as a result of insufficient sowing depth and stated that the increase in the coefficient of variation in sowing depth negatively affects the field sprout emergence rate.

The amount of stubble in the cultivation plots of the trial area was determined as 800 pieces.m². Sin-ce the amount of weeds was almost non-existent, no herbicide application was made before sowing.

The ratio of 0.5-1.5.Z, which indicates the appropriate sowing distance relative ratio, was determi-ned to be 87.30% in the Ax foot P.P.S.S. and 85.50% in the disc foot P.P.S.S. It has been observed that the ideal sowing distance ratio in the P.P.S.S. with both foot types is over 80%. Accordingly, it was determined that the distribution uniformity in the row in the P.P.S.S. with ax feet was better than in the P.P.S.S. with disc feet.

Table 5. Evaluation of field germination emergence degree (Anonymous 1999)

FSE (%)	Assessment
>80	Very good
>70-80	Good
>60-70	Middle
>50-60	Sufficient
=50	Insufficient

Karayel and Özmerzi., (2005) examined the effect of different seeding feet on seed distribution in precision sowing machines, and when they evaluated the coefficients of variation related to row distance, they generally found that the coefficients of variation of hoe, single and double-disc sowing feet were higher than the axe-type burying feet and improved soil penetration. They repor-ted that the increase in resistance decreased the coefficient of variation of the embedded piers.

It was observed that the FSE values in the P.P.S.S. of both foot types were good at > 80% within the evaluation criteria given in Table 5.

In the study, the fuel consumption of the tractor was measured as 10.90 l ha⁻¹ from the pneumatic precision seeder with ax-type burying foot and 9.81 l ha⁻¹ from the pneumatic precision seeder with disc-type burying foot. The fuel consumption of the P.P.S.S. with an ax-type burying foot consumed approximately 11% more fuel than the P.P.S.S. with a disc type foot. It can be said that this is due to the contact of the burying foot with the soil during operation. Aykas et al. (2007) and Korucu and Kirişçi (2003) reached similar results.

The soil surface roughness values of the soil for the applications varied between 14.28% and 16.35%. In the ax foot type, there was a 12.66% decrease in soil surface roughness compared to the disc foot type. This decrease had a positive effect on field sprout emergence, depth distribution coefficient of variation and sowing depth. In the cultivation of hoe crops, it is desired that the field surface be as smooth and smooth as possible after the stubble sowing process. This is especially important in terms of sowing depth and field output being of the desired quality. When examined from this perspective, we can state that the surface unevenness values obtained from the axe-type burying foot are appropriate.

Based on the results obtained from the experiments, the following evaluations can be made.

-The on-row distribution uniformity values obtained from the adhesive tape test conducted in the laboratory of the ax and disc-footed P.P.S.S. were within acceptable limits.

-When the results of field sprout emergence were evaluated, the best result was obtained from the machine with an ax-type burying foot.

-According to the results of field trials, the coefficient of variation of depth distribution and average sowing depth were obtained better with ax-type burying feet.

-Among the applications, the highest fuel consumption was obtained on the machine with an ax-type burying foot.

-In terms of soil surface roughness, the ax type burying foot gave better results than the double disc burying foot.

-The ax-type burying foot, which has a high field sprout output, can be recommended for direct sowing conditions.

Author Contributions: The authors have an equal contribution. All authors have read and agreed to the published version of the manuscript.

Conflicts of Interest: The authors declare no conflict of interest.

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Determination of Energy Utilization Efficiency and Greenhouse Gas Emission in Apple Production: Case of Isparta

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HIGHLIGHTS

- Energy use and greenhouse gas emissions in apple production were determined.
- Energy use efficiency indicators in apple production were determined.
- Greenhouse gas emission calculations were made in apple production.
- Energy utilization indicators were determined.

Abstract

The purpose of this study is to determine the energy utilization and greenhouse gas emission in apple production. Within the scope of the study, energy utilization efficiency indicators and greenhouse gas emission calculations were made in apple production. Total energy input in apple production has been calculated as 35338.97 MJ/ha, total energy output as 60038.50 MJ/ha, energy utilization efficiency as 1.70, specific energy 1.39 MJ/kg, energy productivity as 0.72 kg/MJ and net energy value as 24699.53 MJ/ha. Energy inputs in apple production consisted of direct energy with 11958.05 MJ/ha (33.84%), indirect energy with 23380.92 MJ/ha (66.16%), renewable energy with 3486.55 MJ/ha (9.87%) and non-renewable with 31852.42 MJ/ha (90.13%). In apple production greenhouse gas emission arising from inputs has been calculated as 1718.90 kgCO_{2eq}/ha while greenhouse gas emission rate has been calculated as 0.07 kgCO_{2eq}/kg. In respect to energy utilization efficiency, it is possible to claim that apple production was efficient for the 2021 production season.

Keywords: Apple; Isparta; greenhouse gas emission; specific energy; energy utilization efficiency.

1. Introduction

Türkiye is one of the leading manufacturers of fruit. Türkiye accounted for 2.67% of the world fruit production, which was 865590060 tons in 2017. With this production share, Türkiye ranks fifth after China, India, Brazil and the USA (Anonymous 2020; Bayav and Karlı 2020). According to the 2021 data of the Turkish Statistical Institute in Türkiye, apple cultivation is carried out on an area of 1688105 decares and

Citation: Gökdoğan O, Uysal Ö (2024). Determination of energy utilization efficiency and greenhouse gas emission in apple production: case of Isparta. *Selcuk Journal of Agriculture and Food Sciences*, 38(2), 216-225. <https://doi.org/10.15316/SJAFS.2024.020>

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Received date: 05/03/2024

Accepted date: 04/06/2024

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4493264 tons of apples (Golden, Starking, Amasya, Granny Smith, Other apples) have been produced. In Isparta province, 1130424 tons of apples were produced in an area of 222852 decares and Isparta province provides 25.16% of Türkiye's apple production. In Gönen district, where the study was conducted, apple production is carried out on an area of 9000 decares and 32345 tons of apples were produced (TÜİK 2024).

Apple is a fruit that is widely grown in the world due to its high yield per decare area, its variety, its resistance to cold climate conditions and its ability to be used in many different ways in the industry. In this context, it is the third most produced fruit after bananas and grapes in terms of production. The homeland of the apple, which is a very beneficial fruit containing many vitamins and minerals, is the South Caucasus, which includes Anatolia. While the number of apple varieties grown in the world exceeds 6500, this number is around 460 in Türkiye (Şenyurt et al. 2015; Karakaya and Kızıloğlu 2021).

Apple is an important nutritional element due to the mineral nutrients and vitamins it contains. 84% of fresh apple fruit consists of water. Dry matter includes carbohydrates, proteins, vitamins, pectins and mineral substances. Vitamins A and C found in apples and elements such as potassium, calcium, magnesium and sodium combine to form a number of salts. When the organic parts of these salts, that is, organic acids, are oxidized in the blood to provide energy, base components remain behind. Thus, apple has a positive effect on the acid-base balance in the blood. As a matter of fact, a study conducted in England found that eating an apple a day significantly reduces the risk of cancer (Anonymous 2008; Oğuz and Karaçayır 2009).

In terms of agricultural production diversity, Türkiye is among the few countries in the world. The products grown in each production area and the techniques used in their cultivation vary within certain limits. When assessing the carbon footprint resulting from crop production, it is important to obtain basin and product-based calculations with real field data and monitor them in line with the targets. The impact of the same product produced in different basins on environmental pollution is another topic that needs to be evaluated. In addition, determining the change in the carbon footprint of a single product produced in the same basin as a result of using different production techniques is also important in long-term monitoring and planning (Pan 2023).

In order to indicate how effectively energy is used, the total energy input consumed for agricultural production in a hectare area, including the main product and by-products taken as output, must be compared with its energy equivalent. The decrease in the total energy input for any agricultural production branch compared to the total product energy evaluated proportionally as output means that the level of mechanization increases. Most of the problems, from increasing production costs to disruption of natural balance and global warming, are related to ineffective energy utilization. For this reason, in the coming years, it is highly likely that it will be effectively used as an indicator of agricultural mechanization in all areas of agricultural production, without being limited to field and garden agriculture (Güceyü 2020). Related to the subject, energy balance and greenhouse gas emission studies have been conducted by Yılmaz et al. (2010), Rafiee et al. (2010), Çelen et al. (2017), Aydın et al. (2019), Ekinçi et al. (2020) on apple, by Aydın et al. (2017) on pear, by Ozkan et al. (2004a) on citrus, by Saltuk et al. (2022) on orange, by Ozkan et al. (2005) on grape, Mardani and Taghavifar (2016) on grape, by Gökdoğan et al. (2022) on avocado, Baran (2022) on persimmon, by Şimşek et al. (2022) on grape, by Demir (2023) on watermelon etc.

2. Materials and Methods

Gönen is 24 km north of Isparta Province (Fig 1). It neighbours Atabey in the east, Keçiborlu in the west, and Uluborlu in the north. There is also the province of Burdur in the southwest. The district's surface area is 372 km² and its altitude is 1020 m. Mediterranean climate prevails in the district (Anonymous, 2024a). This current study has been conducted in Gönen district of Isparta of Türkiye during the 2021 production period. The studied area spans over a 0.75 ha area where apple is cultivated. Granny Smith (0.435 ha), Breaburn (0.1 ha) and Pink Lady (0.215 ha) apple varieties grafted on M9 rootstock were selected as material in this area. Randomized complete-block design with three replications has been employed in the study. The amount of fuel consumption has been calculated and full-tank method has been used to achieve this. The amount of fuel used per unit area has been determined to measure the trial area and the amount of fuel that has been

placed inside the tank (Göktürk 1999; El Saleh 2000; Sonmete and Demir 2007). The work productivity for the area has been calculated and the productivity level has been deemed to be effective. Work productivity in (ha/h) has been achieved by calculating the effective working time (tef) (Güzel 1986; Özcan 1986; Sonmete 2006). Time durations have been measured in the study with the help of a chronometer (Sonmete 2006).

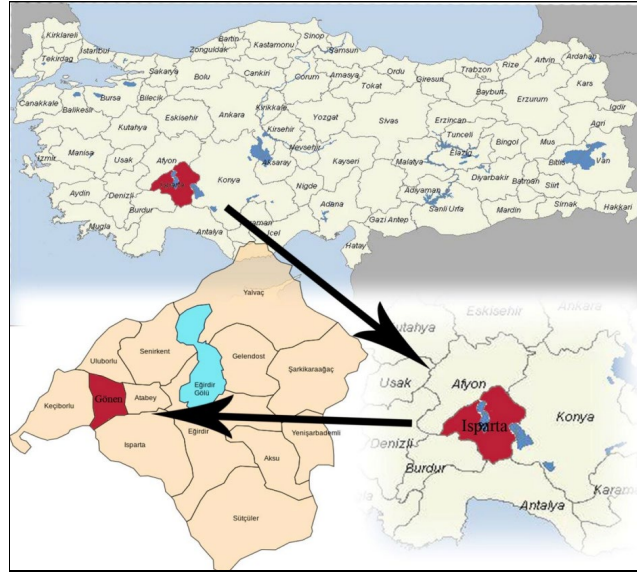


Figure 1. Gonen's location on the map (Anonymous, 2024b).

Human labour, tractor/machinery, chemical fertilisers, chemicals, diesel fuel, electricity and irrigation water have been used as energy inputs in apple production. The total of energy inputs has been calculated by multiplying the use of these inputs per hectare and their energy equivalents. Apple fruit has been deemed to be the energy output. Along with the energy balance sheet, energy utilization efficiency, specific energy, energy efficiency and net energy calculations have been made in apple production. Energy utilization efficiency, specific energy, energy efficiency and net energy calculations in apple production have been calculated by using the following formulas (Mandal et al. 2002; Mohammadi et al. 2008; Mohammadi et al. 2010).

$$\text{Energy utilization efficacy} = \frac{\text{Energy output} \left(\frac{\text{MJ}}{\text{ha}} \right)}{\text{Energy input} \left(\frac{\text{MJ}}{\text{ha}} \right)}, \quad (1)$$

$$\text{Specific energy} = \frac{\text{Energy input} \left(\frac{\text{MJ}}{\text{ha}} \right)}{\text{Product output} \left(\frac{\text{kg}}{\text{ha}} \right)}, \quad (2)$$

$$\text{Energy productivity} = \frac{\text{Product output} \left(\frac{\text{kg}}{\text{ha}} \right)}{\text{Energy input} \left(\frac{\text{MJ}}{\text{ha}} \right)} \quad (3)$$

$$\text{Net energy} = \text{Energy output (MJ/ha)} - \text{Energy input (MJ/ha)}, \quad (4)$$

The energy input types in apple cultivation have been calculated in terms of direct, indirect, renewable and non-renewable as per Yilmaz et al. (2010). Energy balance and GHG in apple production were created using the calculations given in Table 1-2. In calculating greenhouse gas emissions resulting from inputs in

apple production, the following formula adapted from Hughes et al. (2011) by Karaağaç et al. (2019) was used.

$$GHG_{ha} = \sum_{i=1}^n R(i) \times EF(i), \quad (5)$$

GHG_{ha} : Greenhouse gas emission (kgCO_{2eq}/ha)

$R(i)$: Application amount of i input (unit_{input}/ha)

$EF(i)$: GHG emission equivalent of i input (kgCO_{2eq}/unit_{input})

GHG rate is an index defined as the amount of GHG emissions per kg of yield. In calculating the GHG rate, the following formula, adapted by Karaağaç et al. (2019) from Houshyar et al. (2015) and Khoshnevisan et al. (2014) has been used.

$$I_{GHG} = \frac{GHG_{ha}}{Y} \quad (6)$$

I_{GHG} : GHG rate (kgCO_{2eq}/kg)

Y : Yield (kg/ha)

Table 1. Energy equivalents in apple production

Inputs	Unit	Energy Equivalent (MJ/unit)	References
Human labour	h	1.96	Mani et al. 2007; Karaağaç et al. 2011
Tractor	h	25.40	Singh, 2002; Akbolat et al., 2014
Rotary tiller	h	23.60	Singh, 2002; Akbolat et al., 2014
Sprayer	h	21.40	Singh, 2002; Akbolat et al., 2014
N	kg	60.60	Singh, 2002; Demircan et al., 2006
S	kg	1.12	Nagy, 1999; Mohammadi et al., 2010
Chemicals			
Fungicide	kg	99	Fluck, 1992; Ekinci et al., 2020
Insecticide	kg	363.60	Pimentel 1980; Mrini et al., 2002
Diesel fuel	L	56.31	Singh, 2002; Demircan et al., 2006
Irrigation water	m ³	0.63	Yaldız et al., 1993; Ozalp et al., 2018
Electricity	kWh	3.60	Ozkan et al., 2004b
Apple fruit (Output)	kg	2.37	Ekinci et al., 2020

Table 2. GHG emissions coefficients in apple production

Inputs	Unit	GHG Equivalent (kgCO _{2eq} /unit)	References
Machinery	MJ	0.071	Dyer J.A. and Desjardins 2006; Ekinci et al. 2020
N	kg	1.300	Lal 2004; Ozalp et al. 2018
S	kg	0.370	Maraseni et al. 2010; Eren et al. 2019
Fungicide	kg	3.900	Graefe et al. 2013; Ozalp et al. 2018
Insecticide	kg	5.100	Lal 2004; Ozalp et al. 2018
Diesel fuel	L	2.760	Clark et al. 2016; Eren et al. 2019
Electricity	kWh	0.608	Khoshnevisan et al. 2013; Ozalp et al. 2018

3. Results

The energy balance of apple production is given in Table 3. A total of 25332.70 kg of apples has been produced. Inputs include human labour, tractor/machinery power, chemical fertilisers, chemicals, diesel fuel, electricity and irrigation water. Apple fruit was obtained as output. Pruning in apple production was not done in this production season.

Table 3. Energy balance of apple production

Inputs	Unit	Energy Equivalent (MJ/unit)	Input per unit area (ha)	Energy Equivalent (MJ/ha)	Rate (%)
Human labour	h	1.96	252.07	494.05	1.40
Tractor and machinery				2353.72	6.66
Tractor	h	25.40	49.40	1254.76	3.55
Rotary tiller	h	23.60	19	448.40	1.27
Sprayer	h	21.40	30.40	650.56	1.84
Chemical fertilisers				17376.40	49.02
N	kg	60.60	280	16968	48.01
S	kg	1.12	320	358.40	1.01
Chemicals				3700.80	10.47
Fungicide	kg	99	8	792	2.24
Insecticide	kg	363.60	8	2908.80	8.23
Diesel fuel	L	56.31	64.20	3615.10	10.23
Irrigation water	m ³	0.63	4750	2992.50	8.47
Electricity	kWh	3.60	1349	4856.40	13.74
Total				35338.97	100
Output					
Apple	kg	2.37	25332.70	60038.50	100

In apple production, the total energy input was calculated as 35338.97 MJ/ha and the energy output was calculated as 60038.50 MJ/ha. Energy inputs were, respectively, chemical fertilisers energy with 17376.40 MJ/ha (49.02%), electricity energy with 4856.40 MJ/ha (13.74%), chemicals energy with 3700.80 MJ/ha (10.47%), diesel fuel energy with 3615.10 MJ/ha (10.23%), irrigation water energy with 2992.50 MJ/ha (8.47%), tractor/machinery energy with 2353.72 MJ/ha (6.66%) and human labour energy with 494.05 MJ/ha (1.40%).

In other similar studies conducted on energy inputs, Yılmaz et al. (2010) reported the highest energy input in apple cultivation to be chemical fertiliser input with 17974.79 MJ/ha (44.97%), Çelen et al. (2017) reported the highest energy input in apple cultivation to be chemical fertiliser input with 17 078 MJ/ha (29.02%), Ozkan et al. (2004a) reported the highest energy input in lemon cultivation to be chemical fertiliser input with 31290.97 MJ/ha (49.68%), Baran (2022) reported the highest energy input in persimmon cultivation to be chemical fertiliser input with 20950.42 MJ/ha (44.04%) while Mohammadshirazi et al. (2012) reported the highest energy input in tangerine cultivation to be chemical fertiliser input with 32630.30 MJ/ha (2.40%). Energy input, energy output, energy utilization efficiency, specific energy, energy efficiency and net energy calculations in apple production are presented in Table 4.

Based on the energy utilization efficiency calculations in apple production, a total of 25332.70 kg of apple have been produced, total energy input has been calculated as 35338.97 MJ/ha, total energy output has been calculated as 60038.50 MJ/ha, energy utilization efficiency has been calculated as 1.70, specific energy has been calculated as 1.39 MJ/kg, energy productivity has been calculated as 0.72 kg/MJ and net energy value has been calculated as 24699.53 MJ/ha. In other studies related to the energy utilization efficiency in apple cultivation, Yılmaz et al. (2010) calculated the energy utilization efficiency in apple cultivation as 2.26, Rafiee et al. (2010) calculated the energy utilization efficiency in apple cultivation as 1.16, Çelen et al. (2017)

calculated the energy utilization efficiency in apple cultivation as 1.56, Aydın et al. (2019) calculated the energy utilization efficiency in apple cultivation as 1.36, and Ekinci et al. (2020) calculated the energy utilization efficiency in traditional apple cultivation as 3.31.

Table 4. Energy utilization efficiency indicators in apple production

Indicators	Unit	Values
Product (Apple)	kg	25332.70
Energy input	MJ/ha	35338.97
Energy output	MJ/ha	60038.50
Energy utilization efficiency	-	1.70
Specific energy	MJ/kg	1.39
Energy productivity	kg/MJ	0.72
Net energy	MJ/ha	24699.53

Energy inputs in apple production are grouped as direct, indirect, renewable and non-renewable energies (Table 5). In apple production, direct energy inputs have been calculated as 11958.05 MJ/ha (33.84%), indirect energy inputs have been calculated as 23380.92 MJ/ha (66.16%), renewable energy inputs have been calculated as 3486.55 MJ/ha (9.87%), and non-renewable energy inputs have been calculated as 31852.42 MJ/ha (90.13%).

Table 5. Energy input types in apple production

Energy types	Energy input (MJ/ha)	Rate (%)
Direct energy	11958.05	33.84
Indirect energy	23380.92	66.16
Total	35338.97	100
Renewable energy	3486.55	9.87
Non-renewable energy	31852.42	90.13
Total	35338.97	100

In other similar studies conducted, Yılmaz et al. (2010) calculated the renewable energy input in apple production as 8.38%, Rafiee et al. (2010) calculated the renewable energy input in apple production as 34.07%, Aydın et al. (2019) calculated the renewable energy input in non-good practise apple production as 8.53%, Ekinci et al. (2020) calculated the renewable energy input in traditional apple cultivation as 13.58%, and Baran (2022) calculated the renewable energy input in persimmon production as 21.79%. They all reported the non-renewable energy inputs to be higher than the renewable energy inputs.

In apple production, greenhouse gas emission consisted of electricity with 820.19 kgCO_{2eq}/ha (47.72%), of N with 364 kgCO_{2eq}/ha (21.18%), of diesel fuel with 177.19 kgCO_{2eq}/ha (10.31%), of tractor/machinery with 167.11 kgCO_{2eq}/ha (9.72%), of S with 118.40 kgCO_{2eq}/ha (6.89%), of insecticide with 40.80 kgCO_{2eq}/ha (2.37%) and of fungicide with 31.20 kgCO_{2eq}/ha (1.82%). Total greenhouse gas emission has been calculated as 1718,90 kgCO_{2eq}/ha and emission rate has been calculated as 0.07 kgCO_{2eq}/kg (Table 6). In other studies conducted, Ekinci et al. (2020) calculated the greenhouse gas emission rate in traditional apple cultivation as 0.04 kgCO_{2eq}/kg, Baran (2022) calculated the greenhouse gas emission rate in persimmon cultivation as 0.18 kgCO_{2eq}/kg and Saltuk et al. (2022) calculated the greenhouse gas emission rate in orange cultivation as 0.08 kgCO_{2eq}/kg.

Table 6. Greenhouse gas emission equivalents in apple production

Inputs	Unit	Greenhouse gas equivalent (kgCO _{2-eq} /unit)	Input per unit area (br)	Greenhouse gas emission (kgCO _{2-eq} /ha)	Rate (%)
Tractor, machinery	MJ	0.071	2353.72	167.11	9.72
N	kg	1.300	280	364	21.18
S	kg	0.370	320	118.40	6.89
Fungicide	kg	3.900	8	31.20	1.82
Insecticide	kg	5.100	8	40.80	2.37
Diesel fuel	L	2.760	64.20	177.19	10.31
Electricity	kWh	0.608	1349	820.19	47.72
Total				1718.90	100
GHG rate (per kg)				0.07	

4. Conclusions

In this study, conducted in Isparta province, the local energy utilization efficiency, specific energy, energy efficiency, net energy values, greenhouse gas emissions and greenhouse gas emission rate have been calculated in apple production.

The summarised conclusions of the study are presented below.

Total energy input in apple production was calculated as 35338.97 MJ/ha and energy output was calculated as 60038.50 MJ/ha.

In apple production, an average of 25332.70 kg of apples was produced per hectare. According to energy utilization efficiency calculations, energy utilization efficiency was calculated as 1.70, specific energy as 1.39 MJ/kg, energy productivity as 0.72 kg/MJ and net energy value as 24699.53 MJ/ha.

Chemical fertilisers rank first among energy inputs in apple production with 17376.40 MJ/ha (49.02%).

In apple production, energy inputs consisted of direct energy with 11958.05 MJ/ha (33.84%), indirect energy with 23380.92 MJ/ha (66.16%), renewable energy with 3486.55 MJ/ha (9.87%) and non-renewable energy with 31852.42 MJ/ha (90.13%). Non-renewable energy inputs in apple production have been found to be higher than renewable energy inputs.

Total greenhouse gas emission has been calculated as 1718.90 kgCO_{2eq}/ha and greenhouse gas emission rate has been calculated as 0.07 kgCO_{2eq}/kg.

In order to increase energy utilization efficiency and reduce greenhouse gas emissions, it is necessary to increase the use of renewable energy sources and increase the use of organic and farm fertilisers in production inputs instead of chemical fertilisers.

With respect to energy utilization efficiency (1.70), apple cultivation has been a profitable one based on the 2021 production season data.

In conclusion, despite the fact that several energy indicators have been taken into account in this study, in addition to others that are commonly being used in energy analysis research, a fundamental next step ought to be taken to achieve a correct cropping system design, to help at the policy level (Alluvione et al. 2011).

Author Contributions: The authors contributed equally to the study.

Funding: This research received no external funding.

Conflicts of Interest: The authors declare no conflict of interest.

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Drawing the Phylogenetic Tree of *Tenothrips* species Using MOLE-BLAST: A Phylogenetic Analysis Approach

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HIGHLIGHTS

- This research employs MOLE-BLAST to build a tree for *Tenothrips* species, focusing on COI / 18S genes.
- MOLE-BLAST aids in identifying closely related sequences for phylogenetic analysis.
- This study improves our grasp of *Tenothrips* species' phylogeny, offering future research insights.

Abstract

The search for the neighboring sequence of the query for the construction of the phylogenetic tree above led to the use of MOLE-BLAST. The construction of the phylogenetic tree was demonstrated using *Tenothrips* species from Muğla, Konya and Antalya in 2023. The *Tenothrips* species were found to show some degree of relationships based on the tree diagram in all regions. The study aimed to draw the phylogenetic tree of *Tenothrips* species using MOLE-BLAST, a computational tool widely used for phylogenetic analysis by COI and 18S Ribosomal RNA gene regions.

Keywords: Neighbor-Joining tree; MOLE-BLAST; *Tenothrips*; COI; 18S Ribosomal RNA

1. Introduction

MOLE-BLAST, serving as a proximity-based exploration tool, facilitates taxonomists in locating the most closely related database neighbour for submitted query sequences. It can be described as a mechanism that imparts taxonomic context by computing multiple query sequence alignments (Altschul et al. 1990) along-side their top BLAST database hits (NCBI 2016). This process uncovers relationships among these sequences and subsequently illustrates the outcome of the neighbourhood search in a phylogenetic tree (Adebule 2018).

MOLE-BLAST is a specific subset of the broader BLAST (Basic Local Alignment Search Tool), designed to identify similarities between biological sequences, whether nucleotide or protein. The tool conducts a comprehensive comparison with sequences in the database, subsequently evaluating the statistical significance of these matches. BLAST's utility extends to the identification of gene family members, leveraging functional and evolutionary associations recorded within the GenBank database (Wolfe et al. 2014).

Distinguishing itself from other BLAST tools, such as smartBLAST (targeting highly similar proteins), primer-BLAST (for PCR primer design), Global Align (comparing sequences across their entirety), and MOLE-

Citation: Şahin Negiş İ. (2024). Drawing the phylogenetic tree of tenothrips species using MOLE-BLAST: A phylogenetic analysis approach. *Selcuk Journal of Agriculture and Food Sciences*, 38(2), 226-230. <https://doi.org/10.15316/SJAFS.2024.021>

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Received date: 05/09/2023

Accepted date: 10/06/2024

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BLAST stands out. It serves as a valuable resource for taxonomists, ecologists, and other researchers by allowing the submission of sequences to the National Centre for Biotechnology Information (NCBI) to verify the precise taxonomic annotation of these sequences (Adebule 2018). However, it is important to note that one disadvantage of using NCBI databases for sequence data is the possibility of inaccuracies, which could lead to incorrect analysis results. Therefore, researchers should carefully scrutinize and validate the data before conducting analyses.

Furthermore, MOLE-BLAST empowers users to evaluate sequence membership within taxonomy groups, discover neighbouring sequences, visualize the relatedness of reference specimens to sequences, and segment a substantial set of sequences into distinct genes or loci (Adebule 2018). Understanding the evolutionary relationships among species is crucial for unravelling the patterns of biodiversity and comprehending the processes that have shaped life on Earth.

In the context of Thysanoptera, a diverse order of insects commonly known as thrips, investigating their phylogenetic relationships provides insights into their evolutionary history, adaptive strategies, and ecological roles. The genus *Tenothrips* Bhatti, comprising 19 species globally, originates from the Mediterranean region. However, its distribution has expanded to encompass numerous regions worldwide (ThripsWiki 2023). The species within this genus demonstrate associations with a variety of Asteraceae species (Zhang et al 2018). Within the territory of Türkiye, three species of *Tenothrips* have been documented: *Tenothrips discolor* (Karny), *Tenothrips frici* (Uzel) and *Tenothrips anatolicus* (Priesner) (Tunç and Hastenpflug-Vesmanis 2016). In this study was aimed to draw the phylogenetic tree of *Tenothrips* species using MOLE-BLAST, a computational tool widely used for phylogenetic analysis by COI and 18S Ribosomal RNA gene regions. The study conducted by Şahin Negiş and others (2022) focuses on the molecular characterization of species belonging to the Thripidae family, whereas the present study introduces a method for constructing phylogenetic trees of *Tenothrips* species using a tool called MOLE-BLAST. This study specifically highlights the use of MOLE-BLAST for thrips phylogenetic analysis.

2. Materials and Methods

The phylogenetic analysis was conducted using a MUSCLE multiple alignment was computed for MOLE-BLAST. The dataset consisted of one sequence retrieved from NCBI, MW579077 *T. frici*. All *Tenothrips* species were collected from Muğla (Ula/Akyaka¹), Konya (Karatay/Yarma²), and Antalya (Serik³) in 2023, following the methodology described by Şahin Negiş et al. (2022). The samples were collected by shaking method on a white plate and transferred into tubes containing 70% ethanol. Sampling was done on many weeds, ornamental plants, and grains. Table 1 shows that sample information including date, coordinates and GenBank accession numbers. In the initial step, a pre-diagnosis was performed under a stereo microscope, and a sample was allocated to represent each region, some for morphological purposes and some for-DNA isolation. The DNA isolation was carried out individually for all specimens using the 'CTAB' protocol developed by Doyle and Doyle (1987). For the mitochondrial Cytochrome Oxidase Subunit, I (COI) gene region (~350 bp), as described by Timm et al (2008), were employed. Additionally, the 18S Ribosomal RNA (~650 bp) primers used were the same as those detailed in Şahin Negiş et al study (2022). Sequencing was performed using the Sanger sequencing method after the PCR stage in both gene regions (COI and ITS) by BM Labosis.

Table 1. The samples information collected during the 2023 in many weeds, ornamental plants, and grains.

Location	Coordinate		Species name	n		Date	GenBank Accession Number	
	N	E		♀	♂		COI	ITS
1	37°03'19.4"	28°20'52.2"	<i>Tenothrips discolor</i> (Karny)	47	19	03.09.23	PP537394	PP554182
2	36° 55' 18.56"	31° 6' 4.58"	<i>Tenothrips frici</i> (Uzel)	13	2	15.05.23	PP537391 PP537392	PP554181 -
3	37°48'27.33"	32°53'14.67"	<i>Tenothrips anatolicus</i> (Priesner)	42	0	01.08.23	PP537393	PP554183

3. Results and Discussion

In this study, the MOLE-BLAST analysis encompassed all sequences (GenBank accession numbers are given in Table 1) from different gene regions as a unified dataset. One of MOLE-BLAST's notable features is its capability to manage input sequences originating from distinct genes or loci. Faced with such diverse inputs, MOLE-BLAST efficiently clusters them and conducts separate Multiple Sequence Alignments (MSAs) and phylogenetic tree constructions for each locus (NCBI 2023). Additionally, this feature can enable the generation of Neighbor-Joining (NJ) trees for different loci by employing MOLE-BLAST immediately after multiple gene sequencing processes, such as Next-Generation Sequencing (NGS).

The initial phylogenetic analysis encompassed all sequences, including both COI and 18S Ribosomal RNA, and was conducted using MOLE-BLAST in NCBI website. It is an experimental tool meticulously crafted to aid taxonomists in the quest to pinpoint the closest database neighbors for their submitted query sequences (NCBI 2016). And the results were generated separately under distinct locus headings (Figure 1 and 2).

After multiple alignment under locus 1, the NJ tree based on the COI region was constructed using MOLE-BLAST. One GenBank sequence and two *T. frici* samples clustered together on the tree, while the other *Tenothrips* species recorded in Türkiye were in different clades. One of the most well-known barcoding gene regions is Cytochrome Oxidase Subunit I (COI), which serves as the cornerstone of a global bio identification system for insects (Hebert et al 2003). In the MOLE-BLAST tree (Figure 1) made in the study, *T. discolor* species was located close to the *T. frici* species, and this result was parallel to the COI result by Şahin Negiş et al. (2022).

For two of the *T. frici* specimens, there was a strong concordance with the reference specimens for *T. frici*. Similarly, the data for *T. anatolicus* formed another cluster, consistent with the findings of Şahin Negiş et al (2022). Figure 1 demonstrates that *T. discolor* occupies a position nearby but in a distinct clade compared to the other *Tenothrips* species.

Additionally, when examining locus 2 in the multiple alignment, the NJ tree based on the 18S r-RNA gene region revealed that *T. discolor* and *T. anatolicus* shared common branches, while *T. frici* appeared in a separate branch (Figure 2). The same similar branches were observed when analysing the COI gene region.

18S Ribosomal RNA gene sequences have proven invaluable for exploring phylogenetic relationships, finding extensive use in tracing evolutionary history across a broad spectrum (Hillis & Dixon 1991). These sequences are particularly useful for examining relationships among species within the same genus and among closely related genera (Hao et al 2013). Furthermore, the NJ tree based on the 18S Ribosomal RNA gene region, specifically locus 2 in the multiple alignment, revealed that *T. discolor* and *T. anatolicus* share common branches, while *T. frici* forms a separate branch (Figure 2). Besides *T. discolor*, which is in the same branch as *T. frici* taken from the gene bank (KC513013), *T. anatolicus* is located near this branch, which consists of the genus *Tenothrips* (75% bootstrap value) (Şahin Negiş et al 2022). The MOLE-Blast *T. frici* branch formed in the current study is located at an equal distance from both species. Such phylogenetic trees built with the MOLE-BLAST tool should be reconstructed with more samples and broader sequence data, as well as other sequences of the same species and the same gene region in the GenBank.

This analysis showcases the versatility of MOLE-BLAST in handling diverse genetic datasets and highlights its utility in elucidating the phylogenetic relationships among *Tenothrips* species. Understanding these relationships contributes significantly to our comprehension of biodiversity patterns and the processes that have shaped the evolutionary history of this diverse group of insects. Additionally, that kind of a tool can facilitate easier comparison of the obtained data, enhancing the efficiency and precision of genetic analyses.

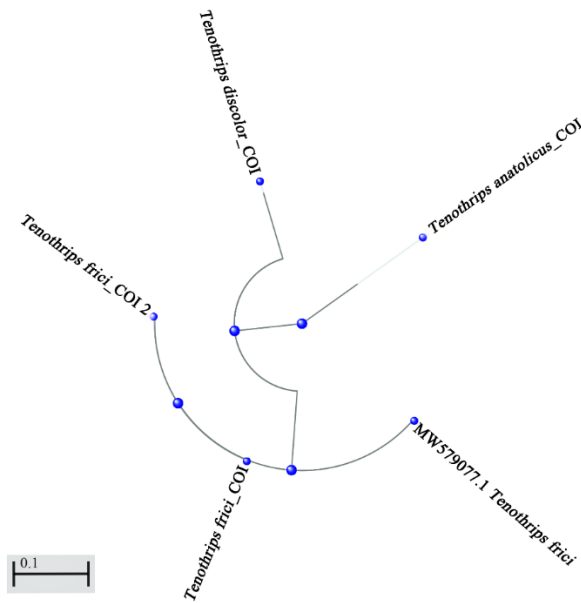


Figure 1. The Neighbor-Joining (NJ) circular phylogenetic tree of *Tenothrips* species, based on the COI gene region, was constructed using MOLE-BLAST.

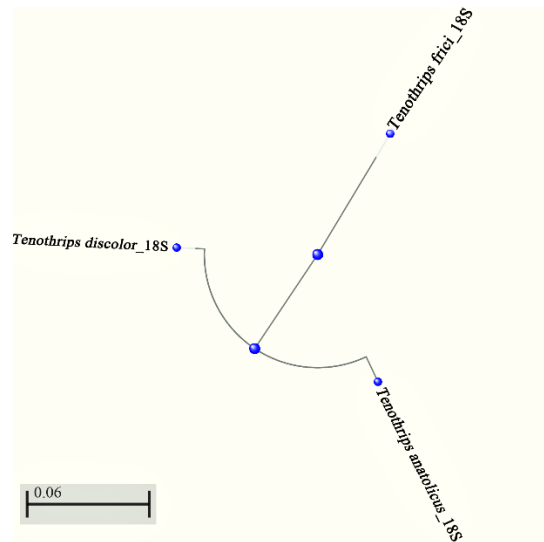


Figure 2. The Neighbor-Joining (NJ) circular phylogenetic tree of *Tenothrips* species, based on the 18S Ribosomal RNA gene region, was constructed using MOLE-BLAST.

Author Contributions: The author has read and agreed to the published version of the manuscript.

Funding: This research received no external funding.

Conflicts of Interest: The authors declare no conflict of interest.

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Determination of CAST/*MspI* Polymorphism in Cattle by PCR-RFLP Method

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HIGHLIGHTS

- The calpastatin (CAST) gene is one of the genes associated with meat quality.
- In this study, the CAST/*MspI* polymorphism was examined among cattle breeds.
- CAST/ *MspI* polymorphism can be used as a molecular marker for meat quality

Abstract

The calpastatin (CAST) gene is one of the genes associated with meat quality. Studies have shown that this gene is associated with quality traits such as body weight gain, carcass yield, meat tenderness and fat content in meat. In this study, the polymorphism of the CAST gene was determined using the PCR-RFLP method in Holstein, Simmental and Brown Swiss cattle. As a result of genotyping, three genotypes (MM, MN and NN) and two alleles (M and N) were determined. It was found that the MM genotype and the M allele had the highest frequency in all breeds examined, while the NN genotype had the lowest frequency. The NN genotype was only found in the Brown Swiss breed. All Simmental animals used in the study belonged to the MM genotype.

Keywords: Calpastatin; cattle; PCR-RFLP; holstein; simmental; brown swiss

1. Introduction

Until a few years ago, most studies focused on meat yield. Today, meat quality has become an important criterion alongside meat yield. In the studies on meat quality, the taste, texture, nutritional value and shelf life of the meat are emphasized (Munekata et al. 2021). These characteristics can be improved through care and feeding, but the genotype of the animal can limit this improvement. Better meat quality can be obtained from animals with genotypically good meat quality. As a result of studies on meat quality, the calpastatin (CAST) gene has been identified as one of the genes associated with meat quality.

Calpastatin, first described in the 1960s, is a protease inhibitor and specifically inhibits calpain enzymes, calcium-dependent proteases that occur in cells (Guroff 1964; Kotova et al. 2023). Calpain enzymes are involved in many functions in cells, but their uncontrolled activity can cause cell stress and even apoptosis

Citation: İlhan F, (2024). Determination of CAST/*MspI* polymorphism in cattle by PCR-RFLP method. *Selcuk Journal of Agriculture and Food Sciences*, 38(2), 231-236. <https://doi.org/10.15316/SJAFS.2024.022>

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Received date: 26/03/2024

Accepted date: 24/06/2024

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(programmed cell death). Therefore, calpastatin maintains the cellular balance by regulating calpain activity. In particular, calpastatin maintains the integrity of muscle fibers in muscle tissue. Increased calpain activity in muscle fibers can lead to the deterioration of muscle fibers and muscle breakdown (Bai et al. 2023; Huff-Lonergan et al. 1996). Therefore, calpastatin is important for the healthy function of muscle tissue. In particular recent studies have shown that the calpain system plays an important role in the initiation of proteolysis in the myofibril protein cycle; therefore, the calpain system acts as a regulator of muscle protein accumulation in domestic animals (Muroya et al. 2012). The CAST gene, which encodes calpastatin, is located on chromosome 7 in cattle and consists of 36 exons (Casas and Kehrlı Jr 2016).

The CAST gene has been studied in more detail in sheep. Studies using RFLP (Bozhilova-Sakova and Dimitrova 2021; Kolosov et al. 2021; Ramadevi et al. 2020), SSCP (Esteves et al. 2020) and DNA sequencing (Machado et al. 2020; Muhana et al. 2021) techniques have shown that the CAST gene is associated with meat quality and growth. There are few studies on this gene region in cattle (Curi et al. 2010; Curi et al. 2008; Juszczuk-Kubiak et al. 2008; Wicińska and Szreder 2004). In studies conducted at the DNA level in cattle, the polymorphism of the CAST gene was found to be associated with meat tenderness (Curi et al. 2009), fat content (Schenkel et al. 2006), shear force (Chung and Davis 2012; Natrass et al. 2014) in meat.

In this study, the polymorphisms of the CAST gene were determined in the Brown Swiss, Holstein and Simmental cattle breeds.

2. Materials and Methods

Blood samples from Brown Swiss (n=54), Holstein (n=50) and Simmental (n=52) cattle were used for this study. The blood samples were stored at -20 °C in the biotechnology laboratory of the Selcuk University Department of Animal Science. DNA isolation was performed according to the salting out procedure developed by Miller et al. (1988).

Genotyping of the obtained DNA samples was performed using the PCR-RFLP method. Primers with the sequences 5'-TGGGGGCCCAATGACGCCATCGATG-3' and 5'-GGGTGGAGCAGCAGCACTTCTGATCACC-3' were used for PCR (Juszczuk-Kubiak et al. 2008). For 10 µL PCR solution, 2 µL DNA (50-100 ng/µL), 0.25 µL (10 pmol/µL) of each primer, 5 µL Taq green PCR Master Mix and 2.5 µL ddH₂O were used. The PCR protocol was as follows: initial denaturation at 94°C for 5 minutes, followed by 35 cycles of 94 °C for 45 seconds for denaturation, annealing at 60 °C for 45 seconds and an extension step at 72 °C for 1 minute. Finally, the PCR process was completed by holding at 72 °C for 10 minutes.

The obtained PCR products were then digested overnight at 37°C by adding *MspI* enzymes, and the bands were detected on a 2% agarose gel. Allele and genotype frequencies were determined using the POPGEN 3.1 program.

3. Results and Discussion

In the study, exon 12 and 13 regions including intron 12 of the CAST gene (Juszczuk-Kubiak et al., 2008) were genotyped by PCR-RFLP method, using DNA obtained from Holstein, Simmental and Brown Swiss cattle breeds. The 622 bp region of the CAST gene was amplified and the *MspI* enzyme was used to recognize the C[^]CGG sequence. As a result of the *MspI* enzyme digestion, MM: 336, 286; MN: 622, 336, 286; NN: 622 bp, 3 genotypes were obtained (Figure 1).

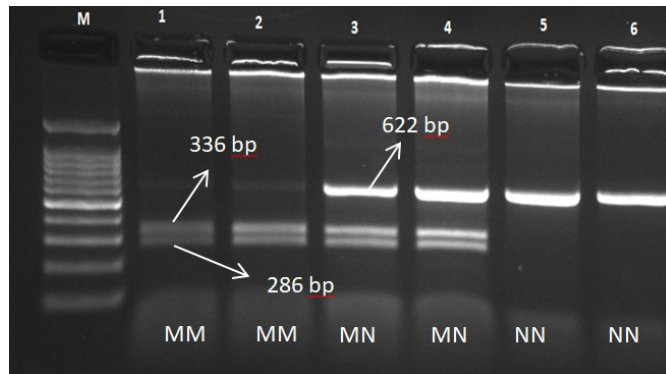


Figure 1. PCR-RFLP pattern of for CAST gene with *MspI* enzyme Lane M: 100 bp DNA ladder marker.

Table 1. The genotype and allele frequencies obtained as a result of PCR-RFLP analysis, the expected and observed heterozygosity values, and the χ^2 values

Breeds		Genotype			He	Ho	Allele frequency		χ^2
		MM	MN	NN			M	N	
Brown Swiss	Frequency	0.80	0.13	0.07					12.3
	Obs.	43	7	4	0.24	0.13	0.86	0.14	p:0.00*
	Exp.	39.98	13.04	0.98					
Holstein	Frequency	0.76	0.24	0					0.84
	Obs.	38	12	0	0.21	0.24	0.88	0.12	p:0.36
	Exp.	38.67	10.67	0.67					
Simmental	Frequency	1	0	0					-
	Obs.	52	0	0	0	0	1	0	
	Exp.	-	-	-					

χ^2 =chi-square value; Obs.: observed frequencies; Exp.: expected frequencies He= expected heterozygosity; Ho= observed heterozygosity

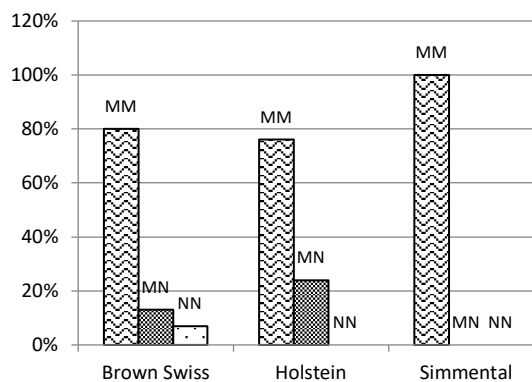


Figure 2. The genotype frequencies obtained as a result of PCR-RFLP analysis.

The genotype and allele frequencies obtained as a result of PCR-RFLP analysis, the expected and observed heterozygosity values, and the χ^2 values calculated to test whether the population is in Hardy-Weinberg equilibrium are shown in Table 1. The analysis of Figure 2 shows that the MM genotype and the M allele have the highest frequency in all breeds; the genotype with the lowest frequency is the NN genotype. Among the available breeds, the Simmental breed was found to be monomorphic for the CAST gene and all animals of this breed had the MM genotype. The NN genotype was only found in the Brown breed among these three breeds in this study. The observed heterozygosity value was higher in the Holstein breed (0.24) than in the Brown Swiss breed (0.13). When the chi-square results were analyzed, it was found that only the Brown Swiss breed was not in balance ($P < 0.05$) and the other breeds were in balance. These results are consistent with the literature. The CAST gene has been examined in more detail in sheep and as a result of the *MspI* enzyme digest of the CAST gene, the NN genotype was not found or was found in very low frequency in the majority of the breeds examined (Asadi et al. 2014; Avanus 2015; Balcıoğlu et al. 2014; Bayraktar and Shoshin 2022; Gorlov et al. 2016; Ibrahim and Kali 2017; Saeed-ul-Hassan et al. 2012; Suleman et al. 2012). Similar results were obtained in several studies in cattle. In a study on Holstein cattle, the CAST gene was cut with the *MspI* enzyme and 44% MM and 56% MN genotypes were determined and no NN genotype was found (Yousefi and Mojtaba 2012). In another study on Sistani cattle, the proportion of CAST/*MspI* MM, MN and NN genotypes was 62%, 29% and 9%, respectively (Tahmoorespur et al. 2007). For this gene, genotyping was performed in cattle using RFLP with various enzymes, in addition to the *MspI* polymorphism, to determine the relationship of these genotypes with productivity. Studies conducted to determine the relationship between CAST/*AluI* polymorphism and yield in cattle, it was found that the CAST gene was associated with traits such as live weight, daily live weight gain and fattening period (Ardicli et al. 2017; Ardıçlı et al. 2017).

4. Conclusions

In this investigation, the CAST/*MspI* polymorphism was examined among Brown Swiss, Holstein, and Simmental cattle breeds. While both the Brown and Holstein breeds exhibited polymorphism in this gene, the Simmental breed was observed to be monomorphic. The CAST gene, known for its association with meat quality, emerges as a potential marker gene for marker-assisted selection studies targeting traits such as meat tenderness, fat content, shear force, as well as growth parameters including live weight and daily weight gain.

Author Contributions: The author confirms sole responsibility for the following: conceptualization, methodology, formal analysis, investigation, interpretation of results, writing—original draft preparation, writing—review and editing and visualization. The author has read and agreed to the published version of the manuscript.

Funding: This research received no external funding

Acknowledgments: Author gratefully thanks Assoc. Prof. Dr. İbrahim Aytekin for providing the experimental material.

Conflicts of Interest: The authors declare no conflict of interest.

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Comparative Analysis of Phenolic Profile and Mineral Constituents of *Datura stramonium* L. and *Datura innoxia* Mill.

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HIGHLIGHTS

- Two medicinal plant species (cultivated same plot) were compared belongings genus *Datura*.
- Total Phenolic and Flavonoid content, Total antioxidant activity of *D. stramonium* and *D. innoxia* was examined.
- Macro and Micro element content, as well as heavy metal content of *D. stramonium* and *D. innoxia* was clarified.

Abstract

Datura sp. is a genus of 14 species, poisonous plants belonging to the family *Solanaceae*. Distributed throughout the world and it is widely cultivated in Asia, America, Europe, South Africa and other tropical and subtropical regions and primarily used as an intoxicant and hallucinogen. *Datura stramonium* and *Datura innoxia* two medicinal plant known as Jimson weed and Thorn apple cultivated all over the world for many properties. This study revealed that total phenolic content (196.52 and 183.08 mg GAE g⁻¹), total flavonoid content (16.72 and 17.12 mg QE 100 g⁻¹), total antioxidant activity (91.74 and 88.97 µmol TE g⁻¹) and nutritional element content (Mg, K, Ca, Fe, Mn, Zn, Cu, Ni, As, Cd, Co, Cr and Pb) of two different medicinal plant species *Datura stramonium* and *Datura innoxia*.

Keywords: Alkaloids; dual value; heavy metal content; mineral content; nutritional properties

1. Introduction

Datura sp. is a genus of 14 species, poisonous flowering plants belonging to the family *Solanaceae* distributed throughout the world (Bhardwaj et al. 2016). Primarily used as an intoxicant and hallucinogen (Alabri et al. 2014). It is widely cultivated in Asia, America, Europe, South Africa and other tropical and subtropical regions (Gaire and Subedi 2013).

Datura stramonium is annual, leafy herbaceous, poisonous, medicinal plant commonly known as Jimson weed or *Datura*. *D. stramonium* is a plant that is a powerful hallucinogen that causes health problem. Its combination of atropine, scopolamine, and hyoscyamine also results in anticholinergic toxicity. Tachycardia, hyperthermia, dry skin and mucous membranes, skin reddening, visual impairment, speech impediment,

Citation: Nohutçu L, Şelem E, Tunçtürk R, Tunçtürk M (2024). Comparative analysis of phenolic profile and mineral constituents of *Datura stramonium* L. and *Datura innoxia* Mill. *Selcuk Journal of Agriculture and Food Sciences*, 38(2), 237-242. <https://doi.org/10.15316/SJAfS.2024.023>

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Received date: 01/04/2024

Accepted date: 26/06/2024

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reduction in intestinal sounds, urine retention, agitation, disorientation, and hallucinations are the classic signs of poisoning. Depending on gastric depletion, the symptoms typically start 1-4 hours after consumption and last for 24-48 hours (Vanderhoff and Mosser, 1992).

Datura innoxia Mill. also belonging to the family Solanaceae, commonly named as Thorn-Apple and native of Asia, United States, Mexico and China (Fatima et al. 2015). Many illnesses, including tumors, earaches, headaches, and asthma, have been treated with plant parts (Vermillion et al. 2011). The plant's antibacterial, anticancer, antihyperglycemic, and antioxidant qualities have enormous potential. (Arulvasu et al. 2014; Banu et al. 2014; Tandon et al. 2016).

The objective of the current study was to investigate total antioxidant, total phenolic and total flavonoids content, nitrogen balance index (NBI), chlorophyll, flavonol, anthocyanin, nutrient elements and heavy metal content of *D. stramonium* and *D. innoxia*.

2. Materials and Methods

2.1. Plant material

The study material consists of *Datura stramonium* and *Datura innoxia* grown in the Medicinal and Aromatic Plants Garden of Van Yuzuncu Yil University, Faculty of Agriculture, Department of Field Crops. The samples were taken first week of July (10.00 am) then shade dried. The dried samples were then cut into smaller pieces and grounded into moderately coarse powder.

2.2. Determination of heavy metal, and nutrient contents

The minerals (including macro elements: K, Ca and Mg; micro elements: Fe, Zn, Cu and Mn) and some heavy metals (As, Cd, Co, Ni, Cr and Pb) were determined. The mineral constituents of the plant samples were investigated as follows: Dried samples were ashed in a furnace with hydrochloric acid and nitric acid (AR) (AOAC 2000). Then, distilled water (50 ml) were added to samples in a volumetric flask. All assays were performed triplicate and the standard materials were being utilized for chemical analyses. Atomic Absorption Spectrometry (AAS) was used to estimate the K, Ca, Mg, Fe. ICP-OES (Inductively coupled plazma-Optical emission spectrometer) was used to determinate for other micro element and heavy metals constituents (Mn, Zn, Cu, Ni, As, Cd, Co, Cr and Pb). The results and standard deviations (Sd) of chemical analyses have been shown in Table 2.

2.3. Total antioxidant, total phenolic and total flavonoids content

Total phenolic compounds content was measured according to Obanda, Owuor (1997) method. The antioxidant activity was also performed based on the Antioxidant Power (FRAP) (Iron (III) antioxidant power reduction) method (Benzie, Strain 1996) followed by readings the absorbance at 593 nm and antioxidant activity values were recorded as Trolox equivalent (TE)/mg. The total flavonoids content was determined with some modifications according to the method developed by Quettier-Deleu et al. (2000). The total amount of flavonoid was measured at 415 nm and calculated in mg quercetin equivalent (QE) 100 g-1 DM by using the calibration curve prepared using standard quercetin.

2.4. Determination of Nitrogen balance index, chlorophyll, flavonol and anthocyanin Evaluation of Data

The Nitrogen balance index (NBI), chlorophyll, flavonol and anthocyanin content were measured on the leaf non-destructively using and in real time the Dualex scientific+ (FORCE-A, France) device before harvesting.

2.5. Evaluation of Data

All data were subjected to Analysis of Variance and the significance of mean values was tested by Duncan Multiple Range Test using COSTAS (version 6.3) software.

3. Results and Discussion

Phytochemicals are the bioactive substances that are produced by plants as secondary metabolites. Both edible and medicinal plants frequently contain phenolic compounds, which have been shown to have a variety of biological effects, including anti-inflammatory and antioxidant properties as well as anti-aggregatory and vasodilating properties (Kahkonen et al. 1999). Total phenolic content obtained from the *D. stramonium* and *D. innoxia* are presented in Table 1. Total flavonoid content in *D. stramonium* and *D. innoxia* was found to be 16.72 ± 2.82 mg QE 100 g⁻¹ and 17.12 ± 2.37 mg QE 100 g⁻¹, respectively. Presence of total flavonoids reported in the previous studies for *D. innoxia* 19 ± 0.17 mg QE g⁻¹ (Bagewadi et al. 2019) and for *D. stramonium* 13.19 mg QE g⁻¹ (Alper and Cennet 2022). Our obtained result of flavonoid content for both *Datura* species are in accordance with literature. Total antioxidant content of *D. stramonium* 91.74 ± 6.59 μmol TE g⁻¹ and *D. innoxia* 88.97 ± 5.53 μmol TE g⁻¹. In a previous study, antioxidant activity of *D. innoxia* reported 221.25 ± 1.06 mg AAE g⁻¹ (Bhardwaj et al. 2016) and *D. stramonium* 63.19 ± 1.02 mg TEs g⁻¹ (Alper and Cennet 2022). Total phenolic content of *D. stramonium* and *D. innoxia* found 196.52 ± 9.69 mg GAE g⁻¹ and 183.083 ± 0.95 mg GAE g⁻¹, respectively. Alper and Cennet (2022) reported total phenolic content of *D. stramonium* as 25.77 mg GAE g⁻¹ and Bhardwaj et al. (2016) reported total phenolic content of *D. innoxia* as 70.26 ± 1.12 mg GAE g⁻¹. Phenolic profile of plants may vary depending on many factors. Some of our results are compatible with previous studies and some of them do not seem to be compatible. This may be due to environmental conditions, harvesting or extraction method. In general, our results seem to be in accordance with the literature.

Table 1. Phenolic profile of *D. stramonium* and *D. innoxia*

Properties	<i>Datura stramonium</i>		<i>Datura innoxia</i>		CV
	Mean	S.D.	Mean	S.D.	
Total Flavonoid Content (mg QE 100 g ⁻¹)	16.72	± 2.82	17.12	± 2.37	4.17ns
Total Antioxidant Activity (μmol TE g ⁻¹)	91.74 a	± 6.59	88.97 b	± 5.53	1.10*
Total Phenolic Content (mg GAE g ⁻¹)	196.52 a	± 9.69	183.083 b	± 0.95	1.58**
Nitrogen Balance Index (NBI)	23.57 a	± 1.15	21.86 b	± 2.23	0.06**
Chlorophyll	43.47 a	± 2.39	37.23 b	± 3.51	3.50**
Flavonoid	31.05 a	± 0.02	1.7 b	± 0.09	4.34**
Anthocyanin	0.04 b	± 0.01	0.123 a	± 0.038	19.63**

S.D. represents Standard Deviation for three replications (n = 3)

Dualex value of *D. stramonium* and *D. innoxia* were found as Nitrogen balance index 23.57 and 21.86, Chlorophyll 43.47 and 37.23, Flavonoid 31.05 and 1.7, Anthocyanin 0.04 and 0.123, respectively. In a study by Uçar et al. (2023), NBI 26.32 dx, chlorophyll 21.32 dx, flavonoids 0.71 dx, and anthocyanin 0.074 dx were detected in the control group of *Salvia officinalis* plants. It was determined that the obtained results were different from the relevant literature. This situation is thought to be due to genetic structure and environmental condition.

Macro nutrients Mg, K and Ca levels of *D. stramonium* and *D. innoxia* found to be 4.33 ± 0.16 g kg⁻¹ and 5.5 ± 1.35 g kg⁻¹, 31.01 ± 3.73 g kg⁻¹ and 23.67 ± 2.62 g kg⁻¹, 30.99 ± 2.53 g kg⁻¹ and 17.66 ± 1.03 g kg⁻¹, respectively. *D. stramonium* has higher value in case of macro nutrients except Mg. Micro nutrients Fe, Mn, Zn and Cu results respectively; 623.60 ± 18.42 mg kg⁻¹ and 819.64 ± 20.57 mg kg⁻¹, 74.96 ± 2.60 mg kg⁻¹ and 83.88 ± 21.21 mg kg⁻¹, 69.32 ± 1.04 mg kg⁻¹ and 51.53 ± 13.22 mg kg⁻¹, 21.29 ± 0.84 mg kg⁻¹ and 13.92 ± 1.27 mg kg⁻¹. In a previous study while mineral content of *D. stramonium* reported as K 49.6 mg g⁻¹, Ca 23.1 mg g⁻¹, Mn 0.132 mg g⁻¹, Cu 0.032, Fe 1.91 mg g⁻¹, Zn 0.89 mg g⁻¹ (Butnariu et al. 2012), *Datura metel* nutrient content reported K 37.9 g kg⁻¹, Ca 30.0 g kg⁻¹, Mg 11.5 g kg⁻¹, Zn 66.8 mg kg⁻¹, Mn 77.6 mg kg⁻¹, Cu 12.5 mg kg⁻¹, Fe 0.9 mg kg⁻¹ (Bhattacharjee et al. 2004).

Table 2. Mineral and Heavy metal content of *D. stramonium* and *D. innoxia*

Elements	<i>Datura Stramonium</i>		<i>Datura innoxia</i>		CV
	Mean	S.D.	Mean	S.D.	
Mg (g kg ⁻¹)	4,33 b ± 0,16		5,5 a ± 1,35		7.19*
K (g kg ⁻¹)	31,01 a ± 3,73		23,67 b ± 2,62		5.78**
Ca (g kg ⁻¹)	30,99 ± 2,53		17,66 ± 1,03		10.84ns
Fe (mg kg ⁻¹)	623,60 b ± 18,42		819,64 a ± 20,57		2.71**
Mn (mg kg ⁻¹)	74,96 b ± 2,60		83,88 a ± 21,21		4.47*
Zn (mg kg ⁻¹)	69,32 a ± 1,04		51,53 b ± 13,22		1.65**
Cu (mg kg ⁻¹)	21,29 a ± 0,84		13,92 b ± 1,27		4.01**
Ni (mg kg ⁻¹)	1,12 b ± 0,24		1,84 a ± 0,56		1.98**
As (mg kg ⁻¹)	0,57 b ± 0,28		0,97 a ± 0,15		2.59**
Cd (mg kg ⁻¹)	0,12 ± 0,16		0,156 ± 0,17		13.05ns
Co (mg kg ⁻¹)	0,19 b ± 0,26		0,44 a ± 0,06		9.18**
Cr (mg kg ⁻¹)	1,68 b ± 1,91		3,2 a ± 0,08		6.47**
Pb (mg kg ⁻¹)	0,78 b ± 0,78		2,53 a ± 0,57		4.35**

S.D. represents Standard Deviation for three replications (n = 3)

The presence of heavy metals differed for both species studied. In *D. stramonium*, the heavy metal content was Cr > Ni > Pb > As > Co > Cd (1.68 mg kg⁻¹, 1.12 mg kg⁻¹, 0.78 mg kg⁻¹, 0.78 mg kg⁻¹, 0.57 mg kg⁻¹, 0.57 mg kg⁻¹, 0.19 mg kg⁻¹, 0.12 mg kg⁻¹), while Cr > Pb > Ni > As > Co > Cd in *D. innoxia* (3.2 mg kg⁻¹, 2.53 mg kg⁻¹, 1.84 mg kg⁻¹, 0.97 mg kg⁻¹, 0.44 mg kg⁻¹, 0.15 mg kg⁻¹, respectively). Ibiyam et al. (2017) in research that different part of *D. stramonium* plant investigated, reported that Cd and Pb not detected on leaves of plant. Bhattacharjee et al. (2004) revealed that level of Co and Ni in *Datura metel*, 25.4 mg kg⁻¹ and 8.3 mg kg⁻¹, respectively. Content of heavy metal in plant vary, depend on their contamination of environment such as soil, water or air. In this case, our obtained results in accordance with previous study.

4. Conclusions

Genus *Datura* have widely used in phytomedicine and popular all over the world for their antispasmodic and hallucinogenic properties. This study revealed the phenolic content and the presence of some minerals and heavy metals in two species belonging to the genus *Datura*. The two studied medicinal plants did not show major differences in the observed parameters. The variation in results are may be attributed to the differences in genetic structure. Further studies on the concentrations of active ingredients, especially the presence of alkaloids and the level of toxicity are recommended.

Author Contributions: LN and MT planned the study, cultivated plant material and collected the samples. RT and EŞ performed laboratory studies. LN, MT, RT and EŞ wrote and edited the manuscript.

Funding: This research received no external funding

Conflicts of Interest: The authors declare no conflict of interest.

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Impact of Young Farmer Support Program for Livestock Enterprises

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HIGHLIGHTS

- This project is supported by the General Directorate of Agricultural Research and Policies named "Impact Analysis of the Grant Program for Supporting Young Farmer Projects within the Scope of Rural Development Supports" and numbered TAGEM/TEPAD/G/18/A8/P3/001.

Abstract

It is indispensable for young farmers to remain in agriculture and/or to sustain farm holding operations. In this study, the impact of the Young Farmer Support Program (YFSP) implemented in Turkey is tried to be define by the counterfactual impact assessment methodology in holdings with livestock such as cattle, sheep/goat. The study was conducted throughout Turkey, the questionnaire with the farmers was completed at the end of 2018. The data obtained intended for the YFSP, which was used in 2016, was taken as basis for the impact assessment. Considering the distribution and number of project subjects from each region, the provinces that would represent the region in terms of its differences were purposefully selected and sample selection was made. The focus is on two important outputs, machine equipment purchased value per livestock unit (LSU) and average income value of cattle and sheep/goat sold per LSU in the last three years which are important in the study. As explanatory variables, age, gender, marriage, educational status, number of family members, non-agricultural income, etc. are employed in the models. According to the two important indicators, Average Treatment Effect (ATE) and Average Treatment Effect on the Treated (ATET), it is revealed that YFSP has positive effects on the dependent variables examined in cattle and sheep / goat farms. This knowledge and accumulation of experience can be transferred to other main components of rural development programs.

Keywords: Treatment effects; impact evaluation; young farmer; sustainability; counterfactual framework

Citation: Çitak Birol D, Çobanoğlu F, Akdemir HA, Yılmaz Hİ (2024). Impact of young farmer support program for livestock enterprises. *Selcuk Journal of Agriculture and Food Sciences*, 38(2), 243-264. <https://doi.org/10.15316/SJAIFS.2024.024>

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Received date: 23/04/2024

Accepted date: 27/06/2024

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1. Introduction

The unique importance of the sustainability of agricultural production has been more pronounced during the covid 19 pandemic. If humanity exists, it is inevitable that there will be agricultural production. Ensuring food supply security can only be achieved if the farmers, who are the most important actors, can continue their activities. For the farmers to continue their activities with pleasure, they and their first-degree family members should continue their lives as happily as possible. Especially for the future, the continuation of the agricultural production activities of young farmers can only be guaranteed by the opportunities, advantages and rural development supports that can be offered to them. At this point, young farmers exit from agriculture is seen intensely in Turkey, as in the world. Lack of motivation tools for young people to farming, negative developments in agricultural input and output prices, attractiveness of city life for households, etc. Such factors appear as the most important constraints encountered by young people in farming. Accordingly, aging in the agricultural population manifests itself as an important problem.

The fact that approximately half of the population of Turkey is under the age of 30 requires the employment opportunities of young people to be further increased and to be sustainable. Young people are moving away from agriculture due to reasons such as insufficient income, limited social opportunities in villages, fragmented or very small lands, and lack of alternative job opportunities in rural areas. The divergence of young people from agriculture brings along problems such as the aging and decline of the rural population. In Turkey, the general employment level in agriculture has been decreasing in recent years, and it is seen that the separation of the young population from the agricultural sector has gained an important momentum as in many other countries.

In addition to the low yield of agriculture in rural areas, the inadequacy of education, health, transportation services and the lack of social opportunities, which are the problems experienced in the past, push the youth to leave the countryside and agriculture. Due to all these reasons, migration from rural areas to urban areas is increasing and the population in the villages is getting older. Apart from these known problems, the problems of the changing century also affect the villages. The inability of young people in the villages to make use of internet facilities, the problem of young male individuals of marriageable age not to find a spouse to live in the village environment is seen as a reason for young female individuals to think that living in the village and working in agriculture requires heavy physical strength, and to abandon village life and agriculture. This circumstance is reported extensively in many studies in developed countries such as the United States of America (USA), Europe and Asia (Mills-Novoa 2011; Chen et al. 2014; Mihi-Ramirez and Kumpikaite 2014; Zagata and Sutherland 2015; Bednariková et al. 2016; Duesberga et al. 2017; Leonard et al. 2017; Morais et al. 2017; Faysse et al. 2019).

This rapid aging problem in the villages is getting more and more attention and raises concerns for the sustainability of agriculture. In addition, due to the migration from the village to the city, the population pressure in the cities makes urban life difficult. This situation experienced in agricultural areas regarding sustainability, which is one of the most important issues on the world's agenda, is one of the priority issues that need to be resolved. On the other hand, young farmers who continue agriculture have a lack of knowledge about entrepreneurship, adaptation to new techniques and technologies, business planning, marketing, risk management, organization, and environmentalist agricultural approaches in terms of the sustainability of agriculture. Within the framework of all these problems, young people should be supported in terms of not leaving agriculture and increasing the awareness of those who remain in agriculture.

Zagata and Sutherland (2015) brought up the debates on the definition of the young farmer. Under the concept of young farmers, they have focused specifically on the concepts being applied and / or the content of which is being discussed in the European Union. These concepts; new entrant and retirement schemes, young sole holder, farm-decision maker, multifunctionality, succession. While they defined under 40 as both young sole holders and farming successors, they considered the age of 65 and over as 'older farmers'. As a result, the following conclusions are drawn. (i) The concept of young farmer should be conceptualized based on Eurostat data. (ii) The issue of old age is examined in a social and economic framework. (iii) It is stated that it would be

beneficial to fill the content of young sole holders' concept according to agricultural management practices according to farm structures. (iv) It is recommended to address the 'young farmer problem' by region, according to mountainous areas. (v) It is suggested that the terms new entrant and young people should be evaluated within the framework of sociology based on agricultural literature. (vi) It is emphasized that small-scale farming should be considered in a separate status.

Hamilton et al. (2015) stated that young farmers are better motivated and devised business plans than older ones. At the same time, it was emphasized that they are more open to innovations, take more risks, and use more loans to grow their business. It is also declared that young farmers are more sensitive to food security and global warming issues and can take on more important tasks (EC 2013; Davis et al. 2013). On the other hand, Katchova and Ahearn (2016) notified that due to factors such as high land prices, difficulties in accessing loans, support policy tools, young people do not continue their activities as farmers in the agricultural sector and / or cannot enter the agricultural sector.

In the EU, it is reported that since the 1980s, young farmer support is offered through various support policy tools. Some of these instruments are listed below. Measure 12 'Setting up of young farmers' (Kontogeorgos et al. 2017), Measure 6 'Business start-up aid for young farmers' (Zagata and Sutherland 2015), early retirement motivations to provide farm succession (Mazorra, 2000), Measure 113 'Early retirement scheme' (Zagata and Sutherland 2015) and finally Young Farmer Payment (Zagata and Sutherland 2015). However, some criticisms are brought in terms of the effectiveness of these policy tools. For example, when the applications of Measure 112 support for the period 2007-2012 are examined, it is stated that there is an imbalance in the number of young farmers supported in the EU and that new entrants are given lower priority to the sector. Again, it was emphasized that Measure 113 does not provide inter-generational transfer (Davis et al. 2013; Zagata and Sutherland 2015).

May et al. (2019) explored the effectiveness of young farmer payment to prevent young farmers from quitting farming using a behavioral approach. According to the study findings, a clue is obtained that the Young Farmers' Motivation structure corresponds to a mediating variable between the payment and the thoughts of young farmers to leave the farm, and it is emphasized that the effectiveness of this support could be potentially strengthened by adding motivational meanings to the payment. This is stated to indicate that the farmers who received the payment gained recognition and prestige compared to the control groups and therefore are less likely to abandon the farm. It is notified that the purpose of these strategies is to prevent pessimism, to support community and family integration, to facilitate the participation of young farmers in the decision-making process of the enterprise, and to link the payment to the idea that this is not only an economic incentive but also a tool, can facilitate the achievement of motivational goals. The main obstacles to address to provide a strategy package of this nature are identified as problems of farm succession and lack of access to credit to young farmers.

Faysse et al. (2019) analyzed policy tools for supporting young farmers in Thailand. They stated that a workshop was held on access to land, capital and markets, capacity building and what should be done to make farming a more attractive profession. Emphasis was placed on the need to prioritize subsidies for starting farming and land lease agreements. In Thailand, as in many other countries, it is stated that to prevent the rapid decline of young farmers, awareness should be raised not only for young farmers, but also for the whole public.

Ma (2014) notified that in South Korea, within the scope of the Farm Successor Fostering program, the policy of granting a 10-year loan to young farmers was implemented. However, the program was not successful, especially as there were many applications for young farmers who are new to farming (Kang 2010). In Japan, on the other hand, they used a policy tool for farmers between the ages of 60-65 to receive additional payments if they retire and give their farms to their successors (Uchiyama 2014). Approaches where the amount of this payment is further increased if the successor to which the farm is given is under 35 years of age (Uchiyama and Whitehead 2012). Another supportive approach is to establish connections between families

who are interested in farming, who do not do farming, and those who want to retire but do not have a successor in the family (Nagatani and Sakamoto 2017).

In France, it is reported that many policy tools are used since the 1960s to enable young farmers to continue their activities in a sustainable manner. It is notified that the first condition to benefit from these supports is that the farm must exceed the threshold values varying according to the regions. One of the main aims of these policy tools is to prevent excessive growth of farm sizes. It is known that there are two main programs for medium-sized farms to be accessible to young farmers. First, the Land Development and Rural Establishment Societies (French acronym SAFER) are private companies whose shareholders are public organizations and farmers' unions (Sencebe 2012). In summary, SAFER buys the lands to be sold, then resells these lands, following consolidation and land reclamation efforts, giving priority to young farmers with shrinking business scale (Hennessy 2014). It is declared that some non-governmental organizations (NGOs) have also taken this action since 2014 (Ravenscroft 2014).

In the US, government support programs use the term "beginning farmer" rather than a young farmer. There is no age limit for them, but the condition that they have been farming for less than 10 years. The Department of Agriculture's farm service agency offers long-term loans for beginning farmers. If the applicant meets the proposed project criteria, this agency provides 100% of the credit required to purchase the farm (Dodson and Koenig 2007; Kaufmann 2013).

Aggelopoulos and Arabatzis (2020) examined the case study of the EU Young Farmer Program implemented in Greece. Accordingly, he discovered that, after the implementation of the financing Program, the approach of farms, by taking advantage of suitable crops, led to a shift towards taking advantage of the comparative advantages of the various regions.

According to the literature reviews, it is discovered that in almost all countries where agricultural production is important, special attention is paid to the efforts and struggles of young farmers to stay in agriculture. Although it is explored that many different support programs and tools are used for this purpose, it is clarified that a very limited number of studies are conducted to determine the success of these. For example, Pavic et al. (2020) analyzed the impact of the Young Farmers Support Program for the development of the dairy farming sector in Slovenia within the framework of an econometric model. As a result, it is discovered that the supports have positive effects on the number of workers employed full time, the number of cattle, total income, and net added value.

The rest of the work is organized as follows. After the introduction section, Young Farmer Support Program (YFSP), which was implemented in Turkey, is briefly mentioned in the content, and some scientific studies are examined. Then, data collection approach used in the study is presented. In the following stage, the conceptual framework and model approach are represented. The research results and discussion section are then included, and finally the conclusions section and recommendations are highlighted.

Young Farmer Support Program implemented in Turkey

Average age of the farmers in Turkey is increasing. Turkish farmers are on average 46 years old, and as such, they are 5 years younger than farmers in the EU and 12 years younger than those in the USA. It is clarified that the average agricultural experience of the Turkish farmer, who usually takes over his business (farm) from the family, is 23 years (CRO, 2019). For this reason, approaches to support young farmers have come to the agenda and started to be implemented in Turkey, as in many countries. The first regulation on supporting young farmers in Turkey was published in 2016. The purpose of this regulation is to determine the principles and procedures for ensuring sustainability in agriculture, supporting the entrepreneurship of young farmers, increasing the income level, creating alternative sources of income, and supporting rural agricultural production projects that will contribute to the employment of young people in rural areas. It covers the procedures and principles regarding the payment of grants up to thirty thousand TRY to the projects for the production, processing, storage and packaging of plant, animal, local agricultural products, medicinal and aromatic plants to be implemented by young farmers living in rural areas (OGRT 2016). Considering that 1

dollar was approximately 5 TRY during the survey period (Wikipedia, 2021), it turns out that this support is equivalent to 6000 dollars. In this regulation, the young farmer; it is defined as a real person between the ages of 18-40 who lives in a rural area or wants to engage in agricultural activities. The project subjects of the program are also framed. For animal production, the following are: (i) Cattle breeding and small ruminant breeding projects, (ii) Bee and bee products breeding projects, (iii) Poultry and silkworm breeding projects. For plant production, i) Closed orchard facility projects, ii) Seedling, sapling, indoor and outdoor ornamental plant growing projects, iii) Controlled greenhouse cultivation projects, iv) Cultivated mushroom production projects. For the production, processing, storage and packaging of local products and medicinal and aromatic plants; i) Production, processing, storage, and packaging of medicinal and aromatic plants, ii) Projects on vegetable and animal production with organic or good agricultural practices, using geographically indicated, local gene resources, iii) Projects on the production of foods with geographical indications. The conditions sought for young people to apply are as follows: i) Being a citizen of the Republic of Turkey. ii) Being over the age of 18 and under the age of 41 as of the publication date of this regulation. iii) Being literate. iv) Not being a paid employee as of the application date. v) Not attending formal education as of the application date. vi) Not being a taxpayer for VAT (Value Added Tax), real and simple procedure as of the application date. vii) Not benefiting from other grant programs of the Ministry for the same project. As the application area of this program, settlements with a population of less than twenty thousand were identified (OGRT 2016). The second regulation on young farmer support was published in 2017 (OGRT 2017). In this regulation, the definition of young farmer has been developed as follows: It refers to real persons between the ages of 18-40, who reside / want to reside in rural areas and who do / want to carry out agricultural activities. In this regulation, the grant support to be given for animal production has been developed as follows: Not having more than 15 cattle or 50 sheep and/or goat for animal production project applications on cattle and small breeding, facility construction and animal purchase as of the date of publication of this regulation. The third regulation on this subject was published in 2018. As a matter of fact, Young Farmers' Support in Turkey has been carried out over a three-year period covering 2016, 2017 and 2018. In this regulation, it is enriched with additional practices such as silkworm breeding and facility construction, free system laying hen breeding and facility construction, perennial forage crop cultivation (OGRT 2018). In the three-year period, 47775 people benefited from the young farmer support, and it was stated that the total amount of grant support provided was 1.5 billion TRY (MAF 2018). Most of this support was provided to cattle breeding farms and sheep/goat breeding farms.

There are several scientific studies that try to reveal the positive and negative aspects of the said support program or that make various recommendations. The main findings are summarized below. Doğan et al. (2018) discovered that the main factors affecting the level of benefiting from Young Farmer Project (YFP) support in Turkey are they defined that there was gender, marital status, farming status, the family's occupation in farming, the population of the place of residence, social security status, agricultural education certificate and land ownership. Unakitan and Başaran (2018) explored that the YFP is not sufficient to keep young people in rural areas and to ensure reverse migration. Although 67.20% of the farmers stated that the YFP had an encouraging contribution to agricultural production, a high portion of them, such as 82.80%, stated that they could not reach the income level they expected because of the project. Kan et al. (2018) declared that the YFP support provided an important step in terms of encouraging young people in agriculture in rural areas. They emphasized that the integration of this support, especially with its entrepreneurship feature, will be very beneficial. Altıntaş et al. (2019), in a study examining the migration tendencies of the rural young population, it was determined that those who have a high education level, have income from non-agricultural areas, think that there are difficulties in village life, find the living standard in the village low and cannot make a living because they earn there are more likely to migrate than others. Gedik (2019) made the following important conclusions about YFP. The most important contribution of the project to young farmers; it was clarified that for the first time, it is the fact that women farmers must own enterprises on their own, and for male farmers they have grown their existing businesses. The main goals of young farmers with their projects are to continue their projects and expand their businesses. Women farmers, on the other hand, are more determined to want to expand their businesses. In livestock enterprises, it has been determined that there is

no increase in income levels, and they have difficulties in meeting their borrowing and operating input costs. Birol et al. (2020) used the preference test method to determine the parameters that can be a criterion for supporting young farmers in Turkey with a study conducted throughout Turkey. It is determined that the biggest needs of farmers are marketing support, later Social Security Institution (SSI) support. In case of grants + SSI support + Marketing support, it became clear that young farmers should be given 51000 TRY grant. Çağlayan et al. (2020), in the evaluation of the young farmer program, which was implemented for three years, in terms of animal breeders, they found this program successful despite its shortcomings, and the most important deficiency observed was that the amount of the grant was insufficient. According to the audit report prepared by the Turkish Court of Accounts (TCA, 2019), it is investigated that there are some inadequacies in young farmer supports in Turkey. These are: (i) Some animals purchased do not have the characteristics specified in the technical specifications of the work, (ii) In the YFP, the animals that were disposed of by young farmers during the follow-up period are shown as if they were purchased from close relatives.

In summary, according to the literature reviews, it is explored that YFP, which was implemented in Turkey in 2016, 2017 and 2018, had positive effects on many issues, but it also had parts that were found to be inadequate.

2. Materials and Methods

2.1. Data collection

In this study, the impact of YFP applied throughout Turkey on cattle breeding and small ruminant (sheep and goat) breeding farms is examined. In other words, in case of using the 30000 TRY provided to the farmers within the scope of this support in cattle and small ruminant breeding holdings, this effect is evaluated. Basically, the main population of the study is formed since the farms benefiting from the YFSP in 2016 and the control group farms with similar characteristics and without support. In 2016, it is defined that there were 14977 farms in Turkey benefiting from this support. 8514 of these beneficiaries are cattle farms (Figure 1) and 2680 are sheep/goat farms (Figure 2). As can be seen, it is revealed that the said support is mostly used for livestock production.



Figure 1. The farmers benefiting from YFSP for cattle breeding by regions in 2016

Considering the distribution and number of project subjects from each region, the provinces that would represent the region in terms of its differences were purposefully selected and sample selection was made. If the number of projects per region is small compared to the distribution of subjects in the provinces we selected by region, the full count method was used. On the other hand, 10% samples were taken from livestock projects with many projects. As a result of the sampling, a survey was conducted on farmers who did not benefit from the program as a control group, as well as the number of samples obtained in cattle breeding projects.

In the selection of the young farmers to be surveyed, the distribution of YFSP according to the regions is taken as a basis for use in cattle breeding and sheep/goat breeding. At this point, the selection of the producers according to the regions is acted upon according to the distribution of the producers by regions. In the selection of the producers in the comparison group, although the distribution by regions is considered, it is paid attention that the producer and/or farm characteristics are as similar as possible to the producers and/or farms benefiting from the support. Thus, the selection was made according to the counterfactual selection methodology. Although the status of benefiting from the YFSP in 2016 is considered, the survey studies were accomplished in 2019. In the light of all these explanations, it can be stated that the quasi-experimental method was used in the study in question.

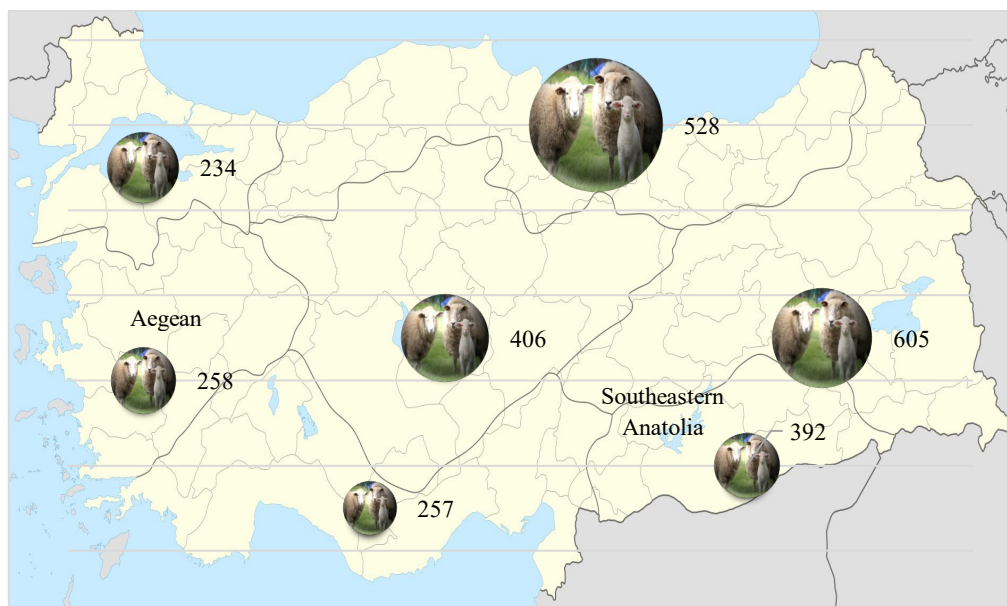


Figure 2. The beneficiary farmers from YFSP for sheep/goat breeding by regions in 2016

The number of surveys conducted on the number of young farmers who benefited from the support (treated) and who did not (control, comparison group) are described below (Table 1). In total, a questionnaire is conducted with 494 young farmers who benefited from the YFSP, 428 young farmers who were the control group as did not benefit from this support for the cattle breeding production. A survey study is performed with 210 young farmers who could benefit from YFSP, and 203 young farmers did not benefit from the support in the sheep/goat breeding.

The input and output variables examined under different models in the study are explained in the sections where they are analyzed. Covarrubias et al. (2012), and Sagbo (2019) notified that to determine the productive effects of supports at the farmer household level, it would be more appropriate to focus on components with high income generating capacity. In this case, they stated that the increase in productive farm capital assets would be an important indicator in determining the impact of the relevant support. In addition, the focus is on the impact of young farmer support used for cattle breeding and sheep/goat breeding on two outputs. These are: (i) The impact on the machine equipment value purchased (MEV) per livestock unit (LSU) obtained

in the last three years (EUROSTAT 2020), (ii) The effect on the average income value of cattle and sheep/goat (AVCSG) sold per LSU in the last three years (2016-2019). It is also quite natural to get this printout. Because, if the animal will not be used for milk production or if there is a defect that prevents it from living in a healthy way, it is sold naturally. This variable is used in similar studies. Agricultural and rural household income coefficients, which are among certain economic indicators, are considered as a differential in evaluating the impact of various development policies. Product income, livestock income and gross household income indicators can be used to assess the impact of different projects on household welfare. Livestock income is income from animals and animal products (Garbero et al. 2018). The income from this sales activity will also have an increasing effect on the income from farm activities. Livestock unit, abbreviated as LSU (or sometimes LU), is essentially a reference unit that facilitates the collection of livestock of various species and ages, using specific coefficients determined by nutritional or feed requirement. It is used for all types of animals. The reference unit (1 LSU) used for the calculation of livestock units is considered the grazing equivalent of an adult dairy cow producing 3000 kg of milk per year, without the use of additional concentrated foodstuffs. LSU is calculated only for cattle, goats, sheep, horses, pigs, poultry, and female breeding rabbits. In this study, it is calculated for cattle and sheep / goats (EUROSTAT 2020) (Table 2). As of the end of 2018, the exchange value of 1 dollar has a value equal to approximately 5 TRY (Wikipedia 2021). The machine-equipment value owned for both livestock production systems; purchased soil cultivation tools such as tractors, trailers, plows, fertilizer machine, harvester, irrigation equipment, etc.). Acquired in the period of 2016-2019 and priced considering the market values of the end of 2018. It is since the use of technology in agricultural production and thus the accumulation of capital is very important in outputting the value of machinery and equipment, and it is indispensable for the sustainability of all agricultural production systems.

Table 1. Distribution of young farmers surveyed by the regions

Regions	Cattle farms		Sheep/goat farms	
	Treated (n)	Control (n)	Treated (n)	Control (n)
Mediterranean	83	10	32	6
Eastern Anatolia	37	45	23	21
Aegean	52	53	18	23
Southeastern Anatolia	46	55	29	39
Central Anatolia	116	111	36	37
Black Sea	111	108	52	51
Marmara	49	46	20	26
Total	494	428	210	203

Table 2. Livestock unit coefficients

Category	Description	Coefficient
Bovine animals	Under 1 year old	0.400
	1 but less than 2 years old	0.700
	Male, 2 years old and over	1.000
	Heifers, 2 years old and over	0.800
	Dairy cows	1.000
	Other cows, 2 years old and over	0.800
Sheep and goats		0.100

Source: EUROSTAT, 2020.

All statistical and econometric analyzes were performed with the STATA/SE 14.2 package program.

For both livestock farms, descriptive statistics of some farmer and household characteristics are presented, and the impact assessment analyzes are explained in detail in the following stage. First, definitions and abbreviations of the variables used in the study are presented. These variables generally vary in the different

econometric models used in the study. The functional form of each model and the variables used are defined in the relevant section (Table 3).

Table 3. Variable abbreviations and explanations

Variables	Description	Abbreviation	Measurement
Dependent variables			
Machine equipment value	Machine equipment value per livestock unit (LSU) obtained in the last three years	MEV	TRY/LSU
Average value of cattle and sheep/goat	Average value of cattle and sheep/goat sold per LSU in the last three years	AVCSG	TRY/LSU
Treatment variable			
Benefiting from the YFSP	1: If the farmer is received support from YFSP 0: Otherwise	TREAT	
Independent variables			
Age	Year	AGE	Year
Gender	1: If the farmer is male 0: Otherwise	GENDER	-
Marital status	1: If the farmer is married 0: Otherwise	MS	-
Education	1: Primary school 2: Secondary school 3: High school 4: Associate degree 5: Undergraduate	EDU	-
Family members (including the farmers)	Number	FM	Number
Family farming	1: If the family is farming 0: Otherwise	FF	-
Family members engaged in agriculture	Number	FMEA	Number
Farming experience	Year	FE	Year
Family farming experience	Year	FFE	Year
Earning non-agricultural income	1: If the farmer earns non-agricultural income 0: Otherwise	ENAI	-

2.2. Conceptual framework

Different methods are available to estimate the increased effects of utilizing a support program on beneficiaries. These methods, called semi-parametric estimators, have become an established standard for estimating the causal effects in question. Compared to parametric regressions, semi-parametric estimators include covariates more flexibly, allowing heterogeneous effects, narrowing the "covariable information" to a single parametric function (Handouyahia et al. 2013). Huber et al. (2010) indicated that these methods are "semi-parametric". Because they explained that while the trend score was based on a parametric model, the relationship between the outcome variables and the trend score was not parametric. These authors stressed that it would be appropriate to examine popularly used estimators by dividing them into four classes: (i) Parametric estimators (such as OLS or Probit (Robins et al. (1992))), (ii) Inverse (selection) probability weighting estimators (Horvitz and Thompson, 1952), (iii) Direct matching estimators (Rubin 1974; Rosenbaum and Rubin

1983), (iv) Kernel matching estimators (Heckman et al. 1998). This study focuses on the estimators in the second and third groups.

In this study, the effect of YFSP on some outputs considered to be important in cattle and sheep/goat farms is analyzed. The treatment effect is evaluated using the concept of potential outcomes, also called the counterfactual framework (Salvioni and Sciulli, 2011). The variables such as inputs and outputs used in the econometric models are explained in detail in the data definition section and in the parts where the models are clarified. Outcomes expected to vary mainly for both animal production systems; (i) The impact of YFSP on the MEV per livestock unit (LSU) obtained in the last three years. (ii) The effect of YFSP on the AVCSG sold per LSU in the last three years (2016-2019).

The MEV per LSU and the AVCSG per LSU are outcome variables. In the survey study, young farmers are organized under two groups. Let the beneficiaries of YFSP be expressed as I , and those who do not benefit as J . If a young farmer has received support (i.e. benefited from the Young Farmer Support Program), an indicator variable T is defined, which is equal to one, otherwise zero. Also, let us define the Q result variables for each farmer, the use young farmer support in the study. In this case, by writing Q_i ($T_i = 1$). It will be emphasized that the farmer is defined as a farmer who has benefited from the support. Benefiting from YFSP has two possible consequences for each farmer: It is expressed as Q_1 if the farmer benefits from YFSP, and Q_0 if the farmer does not. For the farmers who benefited from the YFSP, the average treatment effect on the treated ($ATET$) obtained by the farmers who benefited from this support, according to the case of not benefiting from this support, is the difference between Q_1 and Q_0 for this group of farmers:

$$ATET = E(Q_1^1 - Q_0^0) = E(Q_1^1 - Q_0^0 | T_i = 1) = E(Q_1^1 | T_i = 1) - E(Q_0^0 | T_i = 1) \tag{1}$$

The average treatment effect (ATE) for benefiting from the support is for all farmers who benefit from YFSP and who do not; if all farmers benefit from this support, it reveals the expected change in outputs according to the status of not benefiting from it. This is called the average treatment effect and is formulated as follows (2):

$$ATE = E(Q_1^1 | T_i = 1) - E(Q_0^0 | T_i = 0) \tag{2}$$

In this case, the connection between ATE and $ATET$ is also explained below:

$$ATE = [E(Q_1^1 | T_i = 1) - E(Q_0^0 | T_i = 1)] + [E(Q_0^0 | T_i = 1) - E(Q_0^0 | T_i = 0)] \tag{3}$$

$$ATE = ATET + [E(Q_0^0 | T_i = 1) - E(Q_0^0 | T_i = 0)] \tag{4}$$

Unfortunately, since the data obtained from the survey study were obtained only once (in 2018), in other words, since the data were obtained after the YFSP was already applied, Q_1 was observed only for the farmers who benefited from the support and Q_0 for the control group who did not benefit from the support.

Therefore, to forecast $ATET$, if the farmers benefiting from the support had not benefited from the YFSP, it is necessary to estimate the status $E(Q_0^0 | T_i = 1)$. However, it would not be entirely correct to simply use the difference between groups of farmers to estimate $ATET$, for farmers who benefit from YFSP and those who do not. Since applying to the YFSP is optional, there are typically systematic differences between these two groups. Therefore, the average selection of the group that does not benefit from the support is a biased indicator of the choices the group benefiting from when they cannot participate in the YFSP. Response impact estimators try to obtain unbiased estimates of $ATET$.

For the predictors to objectively provide $ATET$ estimates, two conditions must be met. The first condition, also called the unconfounded assumption, is the Heckman et al. (1998). If the unconfoundedness is satisfied, conditionally on the set of observable common variables, the output obtained without treatment (YFSP), Q_0 is

estimated independently of treatment, i.e., $Q_0 \perp T_i | x_i$. In other words, considering the x covariates, this condition is met when the farmers will do in the absence of YFSP, regardless of whether the farmer is in the group that benefited from the support or not. Rosenbaum and Rubin (1983) showed that $P(T_i = 1 | x_i)$, propensity scores of individuals can be used as conditional statistics to calculate *ATE*. In this article, the propensity score is an estimate of the probability of a farmer participating in the YFSP as a function of the covariates factor x_i .

The second necessary condition is that the data set has sufficient overlap. This implies that the propensity score for both the beneficiary and the control group farmers who did not benefit from the support was neither zero nor one, $0 < P(T_i = 1 | x_i) < 1$. If satisfied, for each farmer who has benefited from the support, there is a possibility to find a farmer belonging to the control group who has essentially the same tendency to benefit from the support. In practice, the overlap condition is imposed, leaving observations that are not satisfied. In other words, if there are no farmers belonging to the control group who have a similar common variable set for some farmers, who did not benefit from the support, then these farmers are removed from the data set and thus *ATE* cannot be estimated for this group.

In Equation (1), for $E(Q_i^1 | T_i = 1)$ and $E(Q_i^0 | T_i = 1)$ the matching estimators are:

$$E(Q_i^1 | T_i = 1) = \frac{1}{I} \sum_i (Q_i^1) \tag{5}$$

$$E(Q_i^0 | T_i = 1) = \frac{1}{I} \sum_i (Q_i^0) = \frac{1}{I} \sum_i \sum_j w(i, j) (Q_j^0) \tag{6}$$

where Q_j^0 is the observed outcome for the farmers who did not benefit from treatment, $w(i, j)$ is the weights of the j th farmer who did not benefit from YFSP and used to estimate outputs, the MEV and AVCSG of the i th farmer who did not benefit from YFSP. In the Inverse Probability Weighting (IPW) estimators used in Stata Statistical Software 14.0 (StataCorp 2015), the weights $w(i, j)$ change inversely with the difference between the propensity score for the i th treated observation and the j th control observation. Therefore, Q_i^0 is predicted to place the more weighted control group observations which are the most like i th observation. The matching estimator used to calculate *ATE* in equation (1) using equations (5) and (6) is given below.

$$ATE = E(Q_i^1 | T_i = 1) - E(Q_i^0 | T_i = 1) = \frac{1}{I} \sum_i \{ (Q_i^1 - \sum_j w(i, j) (Q_j^0)) \} \tag{7}$$

There are many matching algorithms to calculate $w(i, j)$. In this study, inverse-probability weighted (ipw) and propensity score matching (psm) estimators are used, which are the most widely used in the international literature and to make comparisons at a certain level.

3. Results

3.1. Descriptive statistics for the farmer and household characteristics

Table 4 represent some precise characteristics of cattle and sheep / goat farmers by the control and treatment assignments. The statistical difference between the groups is defined using the t test, according to the p significance value. In both farm groups, the age of the farmers in the control group is higher, while those who benefited from the support are lower ($p < 0.01$). This circumstance is thought to stem from the characteristics of YFSP. In many studies conducted to evaluate the effects of various supports provided to rural areas, it is determined that the age of the farmers in the control group is higher, whereas the age of them is relatively lower in the farms using various support tools. This interesting finding suggests that younger farmers have higher entrepreneurial qualities and can take risks (Boone et al. 2013; Mwambi et al. 2016; Onyeneke et al. 2018; Ambler et al. 2020)

It is defined that the number of family members engaged in agriculture is higher in sheep / goat farms and this value is statistically significant ($p < 0.05$). This result can be explained by the increase in the size of the enterprise, albeit to a certain extent, as well as employing more wage workers from outside. On the other hand,

it can be stated that the family workforce is still used more in sheep and goat enterprises. Although there are many positive benefits of using family workforce in agricultural production, there are also opinions that it decreases productivity after a certain threshold point (Kostov et al., 2018). For this reason, the owner of the business will decide on the ideal number and combination of family members' use in agricultural activities. It reveals that in both farm groups, the farmer's own farming experience ($p < 0.01$) and the family's farming experience are higher on the farms that do not benefit from the YFSP ($p < 0.01$ and $p < 0.05$ in cattle farms and sheep/goat farms, respectively). This interesting inference can reveal that farmers and families who have less farming experience to YFSP have a higher tendency to benefit from this support.

Table 4. Descriptive statistics for the farmers

Variables	Cattle farms				Sheep/goat farms			
	Treatment	Control	t	p	Treatment	Control	t	p
AGE	32.23	36.35	59.581	0.000***	31.76	36.08	29.437	0.000***
FM	5.13	5.03	1.377	0.241	5.07	5.29	1.059	0.304
FMEA	2.62	2.54	0.545	0.461	2.54	2.90	5.320	0.022**
FE	11.39	15.92	70.399	0.000***	11.65	16.17	28.913	0.000***
FFE	26.47	31.12	15.667	0.000***	27.28	31.38	4.314	0.038**
GENDER	0.27	0.78	320.374	0.000***	0.32	0.87	180.388	0.000***
MS	0.93	0.86	14.404	0.000***	0.89	0.83	2.854	0.092*
FF	0.91	0.89	0.526	0.469	0.90	0.93	1.661	0.198
ENAI	0.27	0.31	2.144	0.143	0.16	0.27	7.455	0.007***

*, **, *** denotes statistical significance at $p < 0.10$, $p < 0.05$, and $p < 0.01$, respectively that can be drawn.

The difference between gender, marital status, number of family members and non-agricultural income circumstance is analyzed statistically. For both farms, the difference between groups is statistically significant for variables of gender ($p < 0.01$), marital status ($p < 0.01$ and $p < 0.10$ in cattle and sheep / goat farms, respectively). For both groups, it is determined that female farmers are more concentrated on the farms benefiting from the support. It is quite appropriate to make a positive choice especially for female farmers, who are quite difficult in nature and have a lower probability of sustainable earning than male farmers (Rahman, 2014). It is observed that the rate of marriage is higher among the farmers in the treatment group compared to the control group. Only in sheep / goat farms, the difference between the groups in terms of non-agricultural income is statistically significant ($p < 0.01$). It can be inferred from that non-agricultural income is more common in the control group farms.

The distribution of data showing dual character such as gender, marital status, family farming, non-agricultural income can be demonstrated more clearly graphically (Figure 3). When evaluated cumulatively; It is observed that non-agricultural income earning is less, the number of male farmers is higher, and married farmers are more concentrated.

The education level of farmers also varies to a certain extent according to farm groups totally (Figure 4).

When evaluated cumulatively, it is observed that the education levels of the farmers are concentrated at the primary, secondary, and high school levels. Schultz (1964) elaborated on the importance of education in agricultural development. Education increases farmers' ability and productivity capacity (Weir 1999). It contributes to the use of many chemicals and inputs at the most appropriate dose and time (Appleton and Bolihuta 1996; Huang and Luh 2009). Although there is no consensus in many studies, it would not be wrong to state that the level of education increases agricultural productivity and efficiency (Paltasingh and Goyari 2018). In this study, it is determined that the education levels of the farmers who benefit from and apply to the YFSP are concentrated at the primary, secondary, and high school levels. This is due to the general agricultural structure of Turkey. On the other hand, as of 2019, the "Expert Hands in Rural Development Project" instead of YFSP aims to provide incentives to farmers and/or farmer candidates with a relatively higher education level.

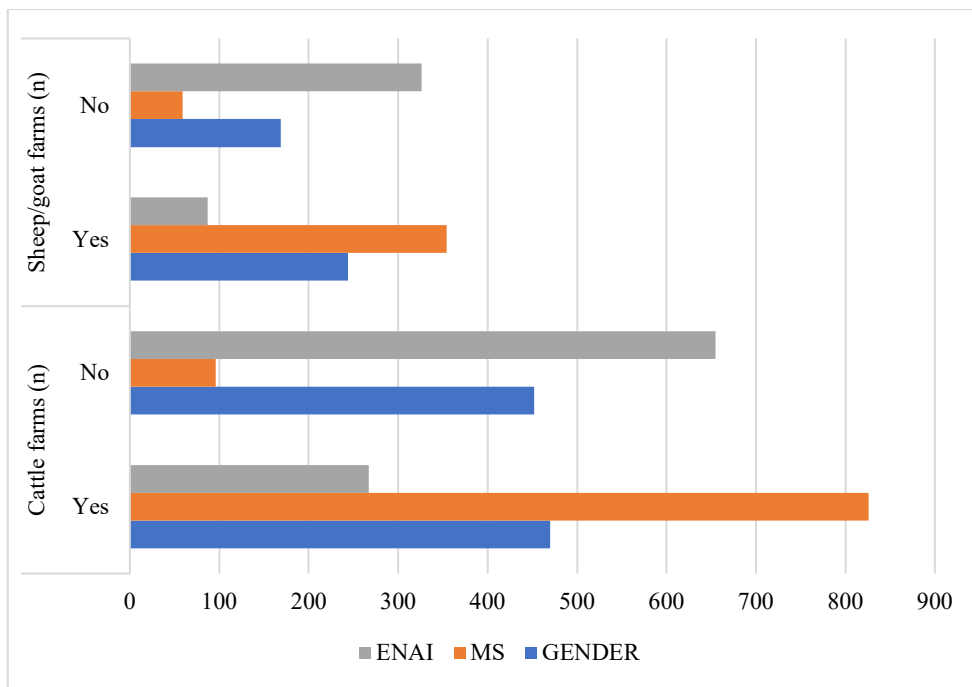


Figure 3. The farmer and household characteristics by the cattle and sheep/goat farms

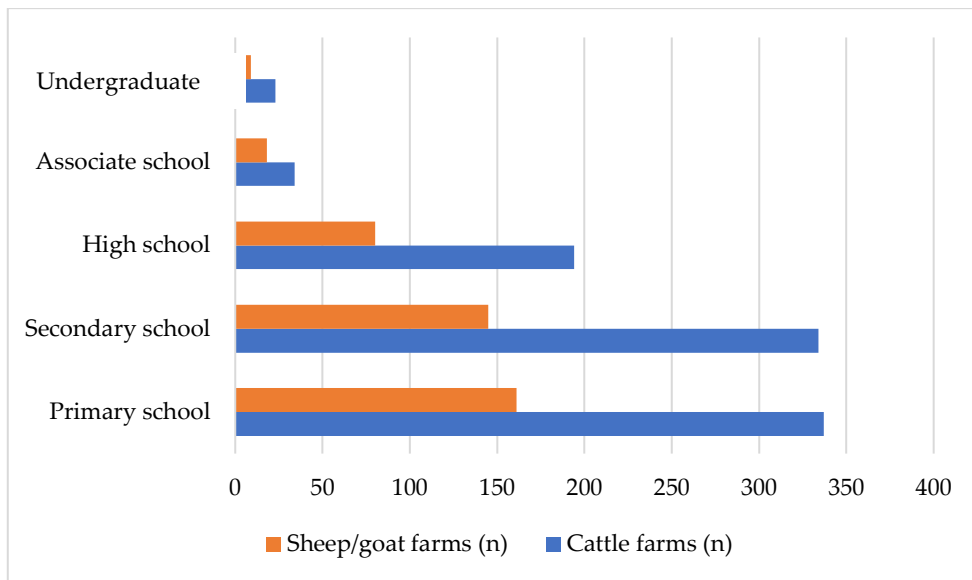


Figure 4. Farmer education characteristics by the farm groups

In the following section, the treatment effect of YFSP has been tried to be revealed.

3.2. Empirical estimations

Using a specific data set, it is very difficult to choose the most appropriate variables and their functional form in models for determining the treatment effect, as is the case for many purposes. In the context of treatment-effects, Cattaneo et al. (2013) could try to demonstrate that only choosing a model works by

minimizing an information criterion. The authors discuss a method and a user-typed command to facilitate the process. In this study, the most appropriate functional form and set of variables are used separately according to the analyzed livestock production system and the data obtained from the survey studies in the field.

3.3. Treatment effects of YFSP on cattle meat production

First, the effect of YFSP used for cattle meat production is analyzed (Table 6).

Table 6. Treatment effects of YFSP on the MEV and AVCSG in cattle farms

Outputs	ATE				ATET			
	ipw		psm		ipw		psm	
	coefficient	p	coefficient	p	coefficient	p	coefficient	p
MEV	2004.75	0.003***	2069.01	0.009***	1581.41	0.049**	1713.40	0.063*
AVCSG	701.50	0.088*	2129.53	0.050**	1023.00	0.008***	1438.47	0.075*

*, **, *** denotes statistical significance at $p < 0.10$, $p < 0.05$, and $p < 0.01$, respectively.

After the psm estimator, the box plots are drawn and examined to check the balance in the paired samples in MEV output. According to the paired box plot, it is understood that the covariates are in balance (Figure 5).

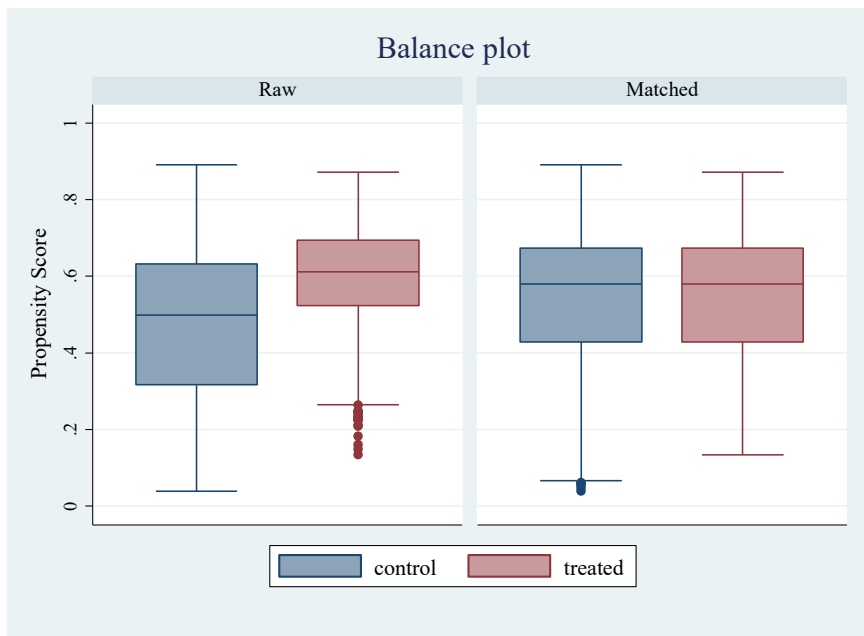


Figure 5. Box graph showing the balancing of all factors using psm estimator for MEV in cattle farms

After the psm estimator, the box plots are drawn and analyzed to detect the balance in the paired samples in AVCSG output. When considering the box chart, it is clarified that the covariates are in balance (Figure 6).

Regarding the MEV output, it is determined that the ATE and ATET coefficients obtained by ipw and psm estimators are quite close to each other. Considering the ATE coefficients, it is defined that if all farms would receive support from YFSP, they would have more machinery and equipment value of 2050 TRY/LSU [2004 TRY/LSU – 2069 TRY/LSU] (410 dollars/LSU) per farm, almost as if none of them would benefit from this support. In the case of ATET coefficients, it is estimated that the farmers who would benefit from the YFSP had an average of 1650 TRY/LSU [1581 TRY/LSU – 1713 TRY/LSU] (330 dollars/LSU) more machine equipment value per LSU, as they would benefit from this support. With an YFSP of 6000 dollars per farm, it is very

important to ensure an increase in value of machinery equipment of 410 \$/LSU for ATE and 330 \$/LSU for ATET per farm at the end of a period of approximately 2.5 - 3 years.

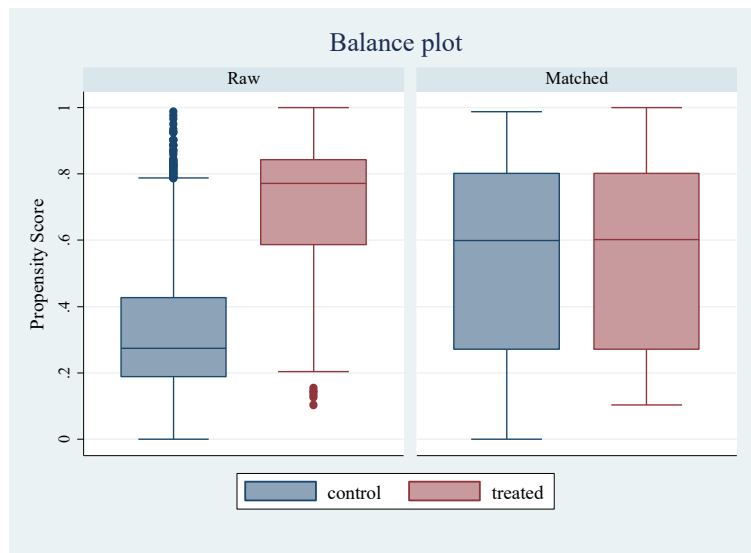


Figure 6. Box graph representing the balancing of all factors using psm estimator for AVCSG in cattle farms

Jiang et al. (2017) notified that in the land transfer policy process, farmers need financial resources, albeit at a certain level, in land acquisition, albeit with the support of the state, and this leads to the purchase of more machinery and equipment and encourages higher machinery and equipment investment costs. An important inference to be drawn from this is that the YFSP, which is used in our study, led to the need for more machinery for farms. Lopez et al. (2017), in their study to measure agricultural input supports and productivity, found that the treated group spent significantly more (34 percentage points) on machinery and equipment compared to the control group. Sagbo (2019) analyzed the impact of borrowing on the purchase of agricultural machinery and equipment. This study emphasized that the farmer could invest in agricultural machinery to fulfill many tasks and this decision can make agricultural activities more efficient. In the study, it is emphasized that loans taken especially for agricultural machinery can significantly reduce the expenditures made for the workforce employed on the farm, but with the new machinery purchased, more family members can be employed on the farm, while the use of family labor for other credit categories can be significantly reduced. In line with this inference, it can be inferred from that the increase in the value of machinery and equipment obtained by YFSP not only increases productivity in agricultural production, but also provides more intensive use of family workforce in the farm.

When the effect of YFSP on AVCSG output is evaluated, the following important conclusions have been reached. The ATE coefficient was determined to be on average 1500 TRY [701 TRY - 2129 TRY] (300 dollars) and the ATET coefficient to be 1250 TRY [1023 TRY - 1438 TRY] (250 dollars) on average. The ATE coefficient value reveals that if all their farm's benefit from YFSP, they will get an AVCSG output of 300 \$/LSU more than if none of them benefit. ATET coefficient value is also determined that the farms benefiting from YFSP obtained 250 \$/LSU more AVCSG since they benefited from this support. Ambler et al. (2016) evaluated the impact of cash grant on agricultural production among small-scale farmers for farm management practices in Senegal. Beneficiaries of the grant support have been found to have higher farm productivity and livestock asset accumulation. The analysis also revealed that the cash grant allows farmers to purchase agricultural inputs, that is, to invest in chemical fertilizers to increase crop yields. These findings are also consistent with the results obtained in our study. Garbero et al. (2018) conducted the impact assessment study of SPAM, a comprehensive support package that includes certified inputs (seeds, fertilizers, and pesticides), agricultural machinery, best production practices training, innovative practices, post-harvest management, in Senegal. The results

determined that the main effect occurred positively on the income of animals (animal and animal product sales) in particular. Arslan et al. (2020), given that the effects on wage employment are similar, they find that the impact of women's empowerment on animal production value is not very different in the comparison between female employment and male employment, as indicated by the analyzed women's empowerment indicators. In other words, they suggested that women in animal husbandry can be employed as wage workers within the framework of certain rules. Eroglu et al. (2020) examined the effects of livestock subsidies on the production and income of beef farms. The survey data were collected from 171 randomly selected cattle farms in Samsun, Turkey. It is discovered that the cattle breeding supports increase the meat production by 11760 kg and the gross profit by 8025.75 \$ on average. Among the supported (processed) farms, the beef production of a farm increases by 12620 kg compared to the unsupported case, and the production coefficient is determined to be statistically significant. In addition, although the gross profit increased by 7811.15 \$, it is determined that the gross profit coefficient is not statistically significant. As a result, it is demonstrated that cattle breeding supports significantly increase the average cattle meat production.

3.4. Treatment effects of YFSP on sheep/goat production

Treatment effect of YFSP used for sheep/goat production is analyzed (Table 7).

Table 7. Treatment effects of YFSP on the MEV and AVCSG in dairy/goat farms

Outputs	ATE				ATET			
	ipw		psm		ipw		psm	
	coefficient	p	coefficient	p	coefficient	p	coefficient	p
MEV	3344.71	0.007***	2858.63	0.035**	2754.55	0.048**	2942.88	0.039**
AVCSG	627.54	0.085*	↓		704.40	0.025**	↓	

*, **, *** denotes statistical significance at $p < 0.10$, $p < 0.05$, and $p < 0.01$, respectively.

↓ These coefficients could not be included, since the results are not statistically significant in the counterfactual impact assessment evaluation analysis performed using psm estimator.

The box plots are drawn and analyzed to control the balance in the paired samples in MEV output after using psm estimator. When considering the box chart, it is clarified that the covariates are in balance (Figure 7).

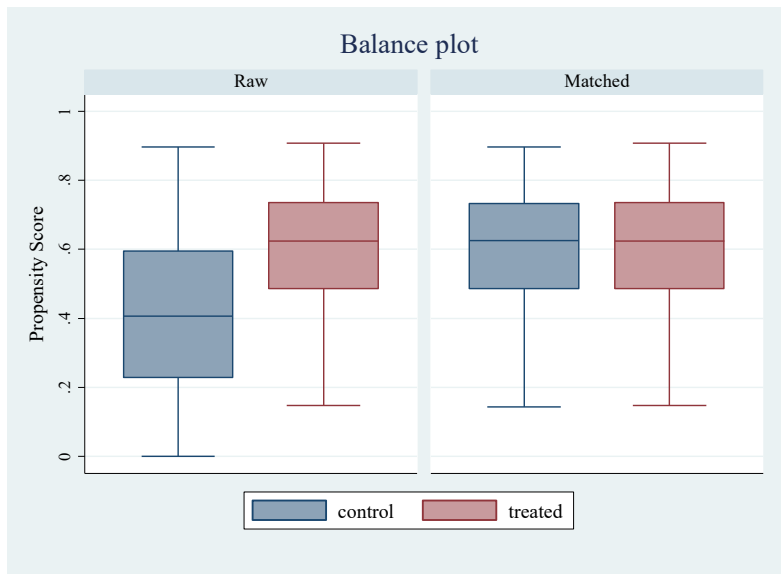


Figure 7. Box graph indicating all factors' balance using psm estimator for MEV in sheep/goat farms

When MEV output is considered, ATE and ATET coefficients estimated by ipw and psm estimators are relatively close values. For the ATE coefficients, if all farms would benefit support from YFSP, they would have more machinery and equipment value of 3000 TRY/LSU [2858 TRY/LSU – 3344 TRY/LSU] (600 dollars/LSU) per farm, as if none of them would benefit from this support. In the case of ATET coefficients, the farmers who would benefit from the YFSP had an average of 2800 TRY/LSU [2754 TRY/LSU – 2942 TRY/LSU] (560 dollars/LSU) more machine equipment value per LSU, as they would benefit from this support. With an as the YFSP support is 6000 dollars per farm, it can be stated that 600 \$/LSU for ATE and 560 \$/LSU for ATET per farm values are quite valuable to ensure sustainable farming during 2.5 - 3 years.

When the effect of YFSP on AVCSG output is evaluated, the following important conclusions are reached. The ATE coefficient is defined to be on average 627 TRY (125 dollars) and the ATET coefficient to be 704 TRY (141 dollars) on average. The ATE coefficient represents that if all their farms would receive YFSP, they will get an AVCSG output of 125 \$/LSU more than if none of them would benefit. ATET coefficient is also indicated that the farms would benefit from YFSP obtained 141 \$/LSU more AVCSG since they would benefit from this support.

Especially, after YFSP, it is observed that the value of machinery equipment purchased in the last three years is higher in sheep/goat farms than in cattle farms. It is thought that there are several reasons for this. First, in sheep/goat farms, the fixed capital components owned are less than that of cattle farms, therefore it is obtained from sheep and goat sales, which are purchased with the benefited YFSP and then sold more. Another reason is that animals such as sheep and goats have less life expectancy than cattle, and that these animals produce very little by-products such as milk and fleece and generate income. For this reason, it is not considered as an output. As a footnote, in the following period, it is estimated that farmers with cattle left in their hands will earn a main crop such as milk.

Paolantonio et al. (2018) assessed the impact of the project Plan VIDA-PEEP (PPV), an initiative financed jointly between IFAD and the Bolivian Government as part of the country's National Development Plan. To this end, this post-evaluation approach has applied a mixed method approach that combines non-experimental statistical methods and qualitative analysis to compare a sample of project beneficiaries with non-participants (control group). The results clearly demonstrated a positive impact on a range of economic mobility indicators related to asset ownership, as well as agricultural income, livestock sales which makes up the larger share of total income for households in the sample. INDECON (2019) analyzed the impact of various support programs on the physical capital of the farm, including Young Farmer's Capital Investment Scheme. It is determined that Indecon's completed and previous Rural Development Program (RDP) support components have led to results confirming the positive impact of capital grants on farm output and productivity, with counterfactual modeling involving capital investment. Overall, it is argued that through modeling and analysis, RDP support will contribute to enhancing the competitiveness of farms. It is emphasized that with increased competitiveness, it will most likely be mainly through capital investment measures.

Cavatassi and Mallia (2018) performed an impact assessment analysis for the Government of Tajikistan launched the Livestock and Pasture Development Project (LPDP) in August 2011, a project financed jointly by IFAD and the Government of the Republic of Tajikistan. Thus, this ex-post evaluation experimental design was used to design and define a valid counter-case. Thus, a non-experimental approach is utilized, combining quantitative methods and qualitative analysis used to enrich the project. As a result of the treatment effects, the effects of the project on the beneficiary group are positive and the effects on productive assets are important as well as the increase in income. This is particularly true when referring to livestock income or assets related to livestock, although of a smaller size, the positive effects also apply to crop income. The positive results showed that it is clearly reflected in the weight of the animals as well as the number of cattle on the farm. These positive results were highlighted in the form of better access to water and lower costs as well as tractor services. However, it was noted that these positive effects were not only due to the project, but also to the adoption of improved or controlled breeding and mating techniques.

4. Conclusion

An important motivation tool for young people to continue their agricultural activities and / or to attract young people to agriculture throughout Turkey, the Young Farmers Support Program (YFSP) was implemented in 2016, 2017 and 2018. However, the data obtained in this project were obtained from the 2016 YFSP. On the other hand, as of 2019, the "Expert Hands in Rural Development Project" instead of YFSP aims to provide incentives to farmers and/or farmer candidates with a relatively higher education level. In the YFSP, a fixed amount of cash of 30000 TRY (approximately 6000 dollars) was provided for each farmer in a three-year period, although this cash money cannot be used freely, but it is obligatory to be used in agricultural production branches directed by the Ministry of Agriculture and Forestry. In Turkey, especially in animal production, as there is a certain level of inadequacy in both meat and milk production, the usage preference and/or direction of the supports have mostly focused on cattle and sheep/goat breeding. For this reason, the research in question is conducted on the mentioned livestock farms. The project work is carried out throughout Turkey and it is thought that important implications are reached with this study. First, it is observed that in the farms benefiting from this support, relatively significant increases have been achieved in both cash assets and physical capital items such as animal assets, machinery, and equipment assets. Although it is observed that the sheep and goats are largely disposed of at the end of the project implementation period, it is clarified that this decrease in the number of cattle was less. The decrease in the number of sheep and goats is realized through the sale of them, and a certain accumulation is achieved in the equity of the farm because of this activity. On the other hand, from the unsold cattle and sheep/goat assets, in the following period, meat, milk, etc. it can be declared that there are significant increases in productive capital assets that will provide an increase in income for animal products.

As a result, it would not be a false statement to state that with the YFSP, an important awareness is achieved to attract young people's interest towards agricultural production.

Author Contributions: The authors have an equal contribution. All authors have read and agreed to the published version of the manuscript.

Funding: This research was funded by the General Directorate of Agricultural Research and Policies, grant number TAGEM/TEPAD/G/18/A8/P3/001.

Conflicts of Interest: The authors declare no conflict of interest

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Effect of Land Consolidation of Land Value

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HIGHLIGHTS

- Land consolidation is an important tool for increasing land value.

Abstract

In this study, the effects of land consolidation projects on land value were examined and these effects were evaluated with their positive and negative aspects. Land consolidation is an important rural planning strategy that includes objectives such as increasing agricultural productivity, regulating water management and improving infrastructure in rural areas. However, the effects of this strategy on land value are complex and diverse. In addition to the positive effects of the projects, the study also discussed their negative effects such as social inequality, ecosystem changes, water resource imbalances and loss of cultural heritage. The research evaluated the effect of land consolidation on land value in a selected area and determined that land values in the region increased after consolidation. Recommendations were made to mitigate the the negative effects of land consolidation, such as participatory planning processes, environmental impact assessments and the implementation of long-term sustainability strategies.

Keywords: Land consolidation; land value; rural planning

1. Introduction

Worldwide, population growth, urban expansion and an increase in agricultural demand have brought with them a complex set of issues that put pressure on land use and management. In this sense, land consolidation has emerged as an effective land management strategy, particularly in agricultural areas. Land consolidation aims to transform fragmented and dispersed land ownership arrangements into a more organized, efficient and sustainable land structure. However, there are gaps in the understanding of the effects of these consolidation processes, especially on land value.

"Land Consolidation" is the process of combining, shaping and reorganizing the parcels of land that are fragmented, scattered and deformed in a way that does not allow for economic agricultural activities for various reasons, according to the principles of modern agricultural management and in a suitable way for the development of irrigation services (ATOM 2015).

Citation: Saygılı BC, Çakmak B (2024). Effect of land consolidation of land value. *Selcuk Journal of Agriculture and Food Sciences*, 38(2), 265-273. <https://doi.org/10.15316/SJAFS.2024.025>

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Received date: 24/04/2024

Accepted date: 09/07/2024

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According to the Land Consolidation Implementation Regulation No. 30265 (2017), land consolidation refers to preventing the degradation and fragmentation of lands by natural and artificial influences, and in fragmented lands, combining more than one piece of land and creating new parcels that are more functional in economic, ecological and social terms by considering their natural characteristics, integrity of use and property rights, and determining the usage patterns of these parcels by evaluating their land characteristics and area and providing land development services.

Land consolidation refers to an arrangement and planning process that is generally applied to agricultural lands in rural areas (Boyras and Üstündağ 2008). This process aims to transform fragmented and scattered land ownership arrangements into a more organized and efficient structure (Akşit 2013).

A project carried out by the Tennessee Valley Authority in the US focused on water resource management, electricity generation, and the regulation of agricultural land in the early 20th century. This project involved a comprehensive planning and consolidation process to support economic development in the region (Erikli and Bayat 2016).

The European Union implements the Common Agricultural Policy (CAP) to manage agricultural land in a sustainable manner. Within the CAP, agricultural consolidation projects include objectives such as developing rural areas, providing support to farmers and increasing agricultural productivity. These cases illustrate land consolidation projects across the world, tailored to different geographies and needs. Each has been planned and implemented taking into account local conditions and objectives (ABDT 2023).

Land value is a concept that refers to the financial worth of a land or property determined by economic, social and environmental factors. This value often varies depending on market conditions, physical characteristics of the land, environmental factors, regional demands and a range of other factors. According to the American Society of Appraisers, "Land value is a value estimated as of a specific date for a specific intended use, usually realized as part of a financial transaction." This value is based on market conditions and land characteristics.

The value of agricultural land can be affected by a number of factors. Understanding the effects of consolidation on land value is important not only in increasing agricultural productivity, but also in a broad context such as property regulations, regional economic development and environmental sustainability. Land consolidation aims to reduce soil erosion and increase the sustainability of agricultural areas. This protects soil quality by reducing the risk of erosion and supports the long-term use of agricultural land. Afforestation projects that prevent wind and water erosion or agricultural practices that reduce erosion can increase land value by preventing soil erosion (Karakayacı et al. 2016).

Land consolidation is the process of combining and organizing small and fragmented agricultural lands in a region. This process generally aims to increase agricultural productivity, use water resources more effectively, prevent soil erosion, improve infrastructure and support rural development (Takka 1993). Land consolidation is a planning strategy implemented in many countries to increase the sustainability and efficiency of the agricultural sector (Khamrabaeva 2020). However, the effects of this process on land value may vary depending on a number of factors.

The main purpose of this study is to reveal in detail the effects of land consolidation on the value of agricultural lands and to explain how these effects may vary depending on various factors. Land valuation is carried out to provide necessary information for the agricultural sector, local governments, policy makers and land use decision makers. Understanding the impacts of land value should be considered a fundamental step in developing sustainable land management strategies.

This study evaluates the effect of land consolidation on land value within a general framework. A review of previous studies is conducted to better understand the effects of land consolidation on land value. This review will help understand findings from studies in similar contexts and identify the unique contributions of this study.

2. Land Valuation Methods

2.1. Traditional land evaluation methods

Traditional land valuation methods are techniques that are frequently used by real estate professionals and assessors and are generally intended to determine market value. Some of the traditional land valuation methods are as follows (Khamrabaeva 2020):

- *Comparative Market Analysis (CMA)*: This method makes an assessment by comparing the sale prices of other similar properties with similar characteristics. With this method, market analysis is performed, sales prices of similar properties are analyzed and the value of the property is determined according to similar characteristics. For example, to determine the value of a house, the sale prices of other houses with similar characteristics in the same area are analyzed. A comparison is made with the prices of similar properties based on factors such as the location, size, age and general condition of a property.
- *Cost Approach*: This method is based on determining the cost required to build a property. To determine the value of a property, the materials, labor and other costs required to build a similar property are calculated. To these costs, factors such as wear, age and functional deterioration are added to determine the value of the property. The value of the property is calculated by taking into account the cost of construction, land value and factors such as wear and aging.
- *Income Approach*: This method assesses the income generating potential of a property and determines the value of the property using this income. The value of the property is calculated using rental incomes, return on investment and financial performance analysis. To determine the value of a rental commercial property, rental income and investment returns are analyzed. The value of the property is determined using income capitalization rates and net present value calculations.
- *Decision Tree Analysis*: This method evaluates different valuation scenarios based on combinations of various factors. A decision tree is created that includes variables such as property characteristics, environmental factors and market conditions.
- *Narrative and Research Analysis*: This method makes an assessment based on expert opinions and market research. The value of the property is determined by taking into account local market conditions, environmental impacts of the property and expert assessments. The value of a property is analyzed by considering the general state of the real estate market in a region, future plans and environmental factors. This method usually involves expert opinions and detailed research.
- *Financial Analysis and Square Meter Valuation*: This method determines the property value by analyzing the costs per square meter. The square meter size of the property, building costs and similar factors are examined. Each valuation method is more appropriate for different types of properties and different market conditions. Often a property appraisal may involve a combination of these methods, as each one assesses from different angles and can complement each other.
- *Price per Square Meter*: This method makes an assessment by analyzing the costs per square meter of a property. When determining the value of a plot of land or building, the value of the property is calculated by taking into account the costs per square meter (e.g. construction cost) and the selling prices of similar properties. These traditional land valuation methods are often used in combination. For example, both the comparison method and the cost method can be considered when determining the value of a property. The combination of these methods can provide a more comprehensive assessment of a property.

2.2. Modern Land Valuation Methods

Modern land assessment methods have become more precise and sophisticated with technological advances and the use of analytical approaches. Descriptions and examples of some of the modern land assessment methods are as follows:

- **Geographic Information Systems (GIS):** GIS is a technological tool used for the collection, analysis and visualization of geographical data. In land assessment, it is used for the analysis and integration of factors such as land characteristics, location, environmental impacts on the map (Karakayacı 2011). GIS can be used in the assessment of a property by mapping factors such as infrastructure, transportation, green areas around it. For example, when determining the value of a house, GIS can be used to analyze factors such as infrastructure quality and transportation access in that region (Dölek and Avcı 2016).
- **Precision Assessment Models:** By using advanced statistical and mathematical models, it allows for a more precise assessment of various factors. Methods such as regression analysis and artificial intelligence algorithms can fall into this category. When determining the value of a property, a regression model can be used that includes the physical characteristics of the property, environmental factors and economic indicators. This model provides a more accurate estimate by analyzing the factors that affect the value of the property.
- **Artificial Intelligence and Machine Learning:** It has the capacity to make future value predictions by using the ability of computers to learn by analyzing data (Gündoğdu 1993, Küsek 1995). In determining the value of a property, machine learning algorithms can be applied on large data sets such as property characteristics, environmental factors and past sales data. These methods can predict the value of similar properties in the future by learning from data sets.
- **Crowdsourcing:** It is a method of making assessments using feedback and data from a wide range of users over the internet. Real estate evaluation platforms can create a large data set by collecting users' individual property evaluations and feedback, and make value estimates based on this data.
- **Sensitivity Analysis:** It is a method of understanding the impact of various factors on value and making value estimates according to the changes in these factors. In property valuation, sensitivity analysis can be used to understand the effects of factors such as interest rates and economic indicators on value and to make value estimates according to changes in these factors. Modern land valuation methods provide more accurate and detailed assessments based on more data and analytical power than traditional methods.

3. Materials and Methods

In this study, the land consolidation project, which was completed by registering to the title deed on 30.10.2023 in Mescit District of Kadı Hanı District of Konya province, was taken as material (Figure 1). In the current project, it was observed that the land consolidation was completed by registering to the land registry on 30.10.2023. In the first stage, it was determined that the parceling plans of the consolidation process were made and put on 1st display.

The peer comparison method was used to determine the effect of land consolidation on land value. In the research conducted in the region according to the peer comparison method, it was observed that the lands, which were in the range of 30-50 TL/m² depending on their location and surface area during consolidation, were marketed and sold in the range of 70-80 TL/m² after the consolidation was completed. According to the precedent comparison method, random lands were selected in the research area and their value was investigated according to precedents before and after 30.10.2023, and the value change of the same parcel before and after consolidation was examined.

4. Results and Discussion

Considering factors such as the zoning status of the real estate subject to appraisal, its location, transportation conditions, the formation of its surroundings, and land structure, it has been learnt that sales are not made through advertisement sites, but were made among the local people themselves, since the region is a region with dense agricultural lands. For this reason, in the comparable research, the sales prices of the

neighborhood where the real estate is located, and the headmen of the nearby neighborhood were learnt and the comparables were examined.



Figure 1. Locations of precedent real estates

In the light of this information, the handover price of 10,000 m² of agricultural land located on the island parcel taken as an example before 30.10.2023 (Consolidation Date) and its equivalents in the region were examined, and the unit prices of 10,000 m² of real estate taken as a sample before consolidation were examined and adapted. According to the precedent comparison method, the value of the same agricultural land before and after consolidation is given in Table 1.

While taking the precedents, the precedents of 2023, the year in which the land consolidation took place, were adapted by taking the precedents before and after the land consolidation. Sales values before and after land consolidation were shown in Table 2 and Table 3. Calculations according to the Peer Comparison method were stated above, and it was determined that the value of the parcel increased significantly after land consolidation. In interviews with local people and real estate agents, it was stated that land values in the region increased after land consolidation, adjusted for inflation, and this situation was confirmed according to peer research. As a result, it seems that there is a significant increase in the value of the regional infrastructure and land size and the lands with irrigation facilities after land consolidation, free from inflation, as a result of the completion of the land consolidation.

Land consolidation is an important rural planning strategy that includes various purposes such as increasing agricultural productivity, using water resources more effectively, and preventing soil erosion. However, debates about the effects of this strategy on land value are inevitable. The positive and negative effects of the land consolidation projects discussed in this study on land value were evaluated.

Table 1. Value of the same agricultural land before and after consolidation according to the peer comparison method in the research area

Precedent	Area (m ²)	Sale price (TL)	Sale date	Unit price (TL/m ²)	Zoning Legend	Description
Subject Real Estate	10.000	-	-	-	Agriculture	A comparable study was carried out for the real estate located on the same island parcel before and after consolidation.
1	10.000	750.000	After Consolidation 27.12.2023	75	Agriculture	After consolidation in the region where the subject property is located, the m ² unit prices of fields without boreholes vary between 50.00 TL and 75.00 TL, and the m ² unit prices of fields with irrigation facilities vary between 75.00 TL and 100.00 TL, depending on the surface area, soil structure, irrigation facilities and location. He declared that it could be sold between. It has been reported that the value of the real estate before consolidation was around 30-40 TL/m ² , but its value increased rapidly after consolidation. The precedent was taken from the Neighborhood Headman and contact information is not included within the scope of KVKK.
2	10.000	350.000	Before Consolidation (01.01.2023-30.10.2023)	35	Agriculture	In the region where the real estate subject to appraisal is located, the m ² unit prices of the fields that can be irrigated from the neighboring parcel vary between 40.00 TL and 50.00 TL, and the m ² unit prices of the fields that can be irrigated vary between 60.00 TL and 80.00 TL, depending on the surface area, soil structure, irrigation possibility and location. It has been declared that. It has been reported that the value of the real estate before consolidation was around 30-50 TL/m ² , but its value increased rapidly after consolidation. The precedent was taken from the Neighborhood Headman and contact information is not included within the scope of KVKK.
3	10.000	400.000	Before Consolidation (01.01.2023-30.10.2023)	40	Agriculture	In the region where the real estate subject to appraisal is located, the m ² unit prices of the fields that can be irrigated from the neighboring parcel vary between 40.00 TL and 50.00 TL, and the m ² unit prices of the fields that can be irrigated vary between 60.00 TL and 80.00 TL, depending on the surface area, soil structure, irrigation possibility and location. It has been declared that. It has been reported that the value of the real estate before consolidation was around 30-50 TL/m ² , but its value increased rapidly after consolidation. The precedent was taken from the Neighborhood Headman and contact information is not included within the scope of KVKK.
4	10.000	800.000	Meeting	80	Agriculture	It has been declared that the m ² unit prices of fields that can be irrigated from the neighboring parcel can be sold between 50.00/60.00 TL, and the m ² unit prices of fields with wells can be sold between 70.00 TL and 90.00 TL, although it varies depending on the surface area, soil structure, irrigation possibility and location in the region where the real estate subject to appraisal is located. It has been reported that the value of the real estate before consolidation was around 30-40 TL/m ² , but its value increased rapidly after consolidation. The precedent was taken from the Neighborhood Headman and contact information is not included within the scope of KVKK.

Table 2. Sales value before land consolidation

Summary of analysis results before land consolidation (30.10.2023 before)	
Appraised Average Unit Value (TL/m ²)	35
Area (m ²)	10.000
Achieved Value (TL)	350.000
Final Value Rounded (TL)	350.000

Table 3. Sales value after land consolidation

Summary of analysis results after land consolidation (Value on 27.12.2023 after 30.10.2023)	
Appraised Average Unit Value (TL/m ²)	80
Area (m ²)	10.000
Achieved Value (TL)	800.000
Final Value Rounded (TL)	800.000

Accordingly, factors such as land consolidation projects, increase in agricultural productivity, water management improvements, infrastructure regulations and reduction of soil erosion can positively affect land value. These projects can increase economic value by ensuring more efficient use of agricultural land in a region. Additionally, infrastructure improvements can increase land value, improving the overall quality of life in the area.

On the other hand, the negative effects of land consolidation projects, such as social inequality, ecosystem changes, water resource imbalances and loss of cultural heritage, cannot be ignored. Following the implementation of these projects, situations such as loss of land by small farmers, damage to local ecosystems and unbalanced use of water resources may reduce the land value in the region.

This study addressed the effects of land consolidation projects on land value in a multifaceted manner. Although some aspects of the projects have an increasing effect on land value, a complex picture emerges when other aspects are evaluated together with the negative effects. Therefore, regional differences, social needs and environmental impacts should be taken into account in the planning and implementation process of land consolidation projects. Recommended measures include a participatory planning process, identifying strategies that take into account the views of local communities, environmental impact assessments and implementation of long-term sustainability strategies. In this way, the positive effects of land consolidation projects can be increased and the negative effects can be minimized. This analysis presented in the study will contribute to future research, planning and policy making on land consolidation.

Land consolidation aims to increase the productivity of agricultural lands, use water resources more effectively, increase irrigation efficiency, optimize irrigation channels, save water, strengthen rural infrastructure, reduce soil erosion, ensure the sustainability of agricultural lands and irrigation, increase agricultural productivity, improve infrastructure and environmental sustainability. It has positive effects. In addition, there are negative effects such as increasing social inequalities, ecosystem changes, decreasing natural habitats, deterioration in soil health, and decreasing soil fertility, which may threaten the sustainability of agricultural lands in the long term.

Water resources are an important factor to be considered in land consolidation projects. Although land consolidation aims to use water resources more effectively, this process may negatively affect the groundwater balance and water basins. Falling groundwater levels or overuse of water resources can lead to environmental imbalances. These impacts emphasize that land consolidation projects must be carefully planned and implemented. A sustainable land consolidation process should aim to both maximize positive impacts and minimize negative impacts. This requires a comprehensive planning and implementation process that takes into account the needs of local communities and environmental sustainability.

5. Conclusion and Recommendations

Land consolidation aims to use agricultural lands more effectively. This strategy can combine small and fragmented agricultural lands to create large, organized and unified agricultural lands. This can lead to an increase in agricultural productivity, positively affecting the value of land. Larger plots of land allow modern agricultural machinery to be used and managed more effectively.

Land consolidation generally involves streamlining irrigation systems and using water resources more effectively. This contributes to a more sustainable management of water resources. By optimizing irrigation systems and using water-saving technologies, it can contribute to the protection of water resources by affecting land value.

Land consolidation aims to improve infrastructure in rural areas. Arrangement of roads, irrigation canals, water storage systems and other infrastructure elements can increase the overall land value in the area. These improvements can market agricultural products more effectively, increase trade, and improve the living standards of people living in rural areas.

Land consolidation aims to improve the quality of life in rural areas and support rural development. This strategy can increase overall economic activity in the region, strengthen the agricultural sector, and positively impact land value by providing employment and income to rural communities. However, the effects of land consolidation may vary regionally. The consolidation process may vary depending on local conditions, agricultural practices, government policies and community needs. Therefore, land consolidation projects do not always produce positive results and must be planned carefully.

The uncertainties and debates that may arise on the mentioned issues emphasize that consolidation projects should be planned carefully, adopt a participatory process and take into account the opinions of various stakeholders. Additionally, transparent communication and information sharing during the implementation of projects can also help reduce uncertainties and debates. Therefore, it is important to consider a broad perspective and the principle of sustainability when creating land consolidation strategies.

Author Contributions: Conceptualization, C.B.S. and B.C.; methodology, C.B.S.; resources, C.B.S.; data curation, C.B.S.; writing—original draft preparation, C.B.S.; writing—review and editing, C.B.S. and B.C.

Funding: This research received no external funding.

Conflicts of Interest: The authors declare no conflict of interest.

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Effects of Bacteria, Molybdenum and Sulphur Fertilization on Agronomic and Quality Characteristics of Mung Bean

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HIGHLIGHTS

- Bacterial and molybdenum application increases the yield and yield elements, nodule formation and quality characteristics of mung bean.
- The application amount of sulphur fertilization increased, agronomic parameters such as yield, hundred grain weight, protein content, ash content and nodulation activities increased.
- To increase yield and quality in mung bean, 30 kg S ha⁻¹ bacteria and molybdenum should be used.

Abstract

Mung bean is one of the most important edible legumes in the world and although its consumption is increasing day by day, its production is still low. Important cultural practices in mung bean cultivation are inoculation of seeds with symbiotic bacteria and optimum fertilization with nutrients. Therefore, the effects of sulphur (0, 10, 20 and 30 kg ha⁻¹) molybdenum (0 and 5 g kg⁻¹) and bacteria application on agronomic and quality characteristics of mung bean and nodule formation were determined in this study. The study was conducted in Isparta University of Applied Sciences, Faculty of Agriculture, Department of Field Crops in 2021. In the study, the differences between plant height, number of grains and pods per plant, plant grain yield, grain yield, harvest index, hundred grain weight, ash content, protein content, number and weight of nodules according to bacteria treatment; all traits except harvest index and hundred grain weight according to molybdenum treatment and all traits according to sulphur doses were found statistically significant. According to the results obtained, it was determined that molybdenum and sulphur treatments together with bacterial inoculation increased the yield, yield components and nodule formation of mung bean. It was determined that the increase in sulphur doses increased agronomic parameters such as yield, hundred grain weight, ash content, protein ratio and nodulation activities. In the study, it was concluded that 30 kg S ha⁻¹ should be used together with bacteria and molybdenum to obtain higher yield and quality product in mung bean cultivation.

Keywords: Mung bean; bacteria; molybdenum; sulphur; nodule properties

1. Introduction

Edible legumes are one of the most important crop groups in sustainable agricultural systems. The mung bean (*Vigna radiata* L.), commonly known as green gram in the world and mung bean or mung in Turkey, is one of the traditional legume crops. It is an important legume of Asian origin and is widely cultivated in

Citation: Karaman R, Turkey C (2024). Effects of bacteria, molybdenum and sulphur fertilization on agronomic and quality characteristics of mung bean. *Selcuk Journal of Agriculture and Food Sciences*, 38(2), 274-287. <https://doi.org/10.15316/SJAFS.2024.026>

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Received date: 17/03/2024

Accepted date: 18/07/2024

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countries in Asia, Australia and Africa (Yang et al. 2008). In Turkey, mung bean production is generally carried out using local varieties for family needs or for local markets. It is also grown locally in the Mediterranean and Southeastern regions of Turkey (Karaman and Türkay 2022). In the world and in our country, mung bean seeds (fresh or dried) can be used whole or processed to make bread, noodles, porridge, soup, snacks and even ice cream (Mogotsi 2006). Mung beans have high nutritional value as they have high and easily digestible proteins (Baraki et al. 2020). It also contains about 61% carbohydrates, 23.8% protein, 1.2% fat, 3.5% ash and 4.5% fiber on a dry weight basis (Dahiya et al. 2015) and is complementary to cereal grains of essential amino acids (lysine, cysteine, methionine, threonine and tryptophan) (Asaduzzaman et al. 2008). In addition, its biological N fixation ability similar to other legumes, short growth period, tasty and highly digestible roughage source, and its inclusion in crop rotations in different regions make mung bean a very advantageous crop (Bell et al. 2015; Taylor et al. 2016; Langworthy et al. 2018; Rawnsley et al. 2019; Karaman et al. 2022). Due to these features, the consumption of mung bean in our country and in the world is increasing day by day. Its popularity is also increasing day by day compared to other edible grain legume species (Karaman and Türkay 2022). However, the grain yield of local mung bean varieties in the world is 400 kg ha⁻¹ and the grain yield of new varieties developed can be 2 tons per hectare (Schafleitner et al. 2015; Nair and Schreinemachers 2020). In this direction, in order to meet the expected increase in demand in the coming years, increasing the production and yield of mung bean with appropriate fertilization methods is very important in meeting the global food need.

Mung beans are able to assimilate atmospheric nitrogen through *Rhizobium* bacteria. This improves soil fertility and reduces the need for increasingly expensive synthetic nitrogen fertilizers. *Rhizobium* bacteria can vary in number and activity, nodulation and nitrogen fixation. Tahir et al. (2011) suggested that *Rhizobial* populations living in the soil are not sufficient and therefore their efficiency in biological nitrogen fixation is low. To establish an optimum *Rhizobial* population in the root zone, legume seeds should be inoculated with an effective *Rhizobial* strain. Indeed, *Rhizobium* inoculation increased 57% effective nodules, 77% dry matter production, 64% grain yield and 40% hay yield in mung bean compared to control (Hossain et al. 2011).

Nitrogen fixation process is influenced by many factors, sulphur and molybdenum being one of them. Although mung beans require all essential plant nutrients, sulphur plays a very important role in the production and quality of mung beans. Sulphur is also essential for protein and enzyme synthesis and is a component of the amino acids methionine and cysteine (Scherer 2001). Insufficient sulphur supply can negatively affect the yield and quality of mung bean (Scherer et al. 2006). It also increases the formation of nodules in legumes, which results in greater sulphur utilization throughout the vegetative and generative growth phase (Mum et al. 2004). In addition, sulphur application to mung bean can increase plant height, number of main branches and leaves, dry matter, plant seed yield, nodule number and nodule weight (Singh and Yadav 1997). Molybdenum application plays a vital role in enhancing the N fixation process of *Rhizobium* bacteria. Molybdenum is required for nitrate reductase and nitrogenase enzyme activity (Westermann 2005) and it is also part of the nitrogenase enzyme and *Rhizobium* spp., which fixes nitrogen, requires molybdenum during the fixation process (Vieira et al. 1998). Molybdenum is a highly effective micronutrient especially in nodule formation, increasing N fixation and flowering number, improving pod tying and early flowering (Prasad et al. 1998; Singh et al. 2017). Therefore, it is very important for sustainable agriculture to determine the changes in yield and quality characteristics of *Rhizobium* inoculation with molybdenum and sulphur application to mung bean. Accordingly, this study was aimed to investigate the effect of sulphur, molybdenum and bacteria application on agronomic and some quality traits of mung bean.

2. Materials and Methods

2.1. Materials

This study was conducted in Isparta University of Applied Sciences, Faculty of Agriculture, Department of Field Crops 37°45'N and 30°33' E, 997 m) in 2021. Partow mung bean variety was used as seed in the experiment. *Rhizobium leguminosarum* bacteria used in the study were obtained from Ankara Soil and Fertilizer Research Institute Directorate in the form of peat culture, Ammonium heptamolybdate tetrahydrate

$(\text{NH}_4)_6\text{Mo}_7\text{O}_{24}\cdot 4\text{H}_2\text{O}$ (Sigma) chemical as molybdenum source and elemental sulphur (98%) as sulphur source from a private company.

2.2. Methods

2.2.1. Experimental site

When the climatic characteristics of the experimental area are examined, it is seen that there is no significant difference between the average temperatures during the vegetation period in many years. According to the long-term average, the lowest monthly average temperature during the mung bean planting season (May-October) was 10.7°C in May and the highest monthly average temperature was 23.4°C in July. Total rainfall during the vegetation period (291.7 mm) was higher than the long-term average (231.3 mm). Soil samples were taken from different parts of the experimental area at 0-30 cm depths and analyzed. Soil analysis were carried out in 5 different areas of the study area at 0-30 cm depths according to Rowell (1996). The texture of the soils of the test area is clayey-loamy and belongs to the slightly saline ($322 \mu\text{S cm}^{-1}$) group with a slightly alkaline reaction (pH value 7.7) and a salt content (EC) of $322 \mu\text{S cm}^{-1}$. It was found to be rich in lime (28.7%) but poor in organic matter (1.54%).

2.2.2. Experimental design and treatments

The experiment was conducted in 3 replicates according to the split plots experiment design. In the experiment, bacterial inoculation was allotted to main plots (B_0 = non inoculated, B_1 = inoculated), molybdenum treatment was assigned to sub-plots (M_{00} = osmopriming without molybdenum, M_{01} = molybdenum at 5 g kg^{-1} seed (Aslam et al. 2009) and sulphur doses were distributed in sub-sub-plots (S_0 = 0 kg ha^{-1} (no sulphur treatment), S_1 = 10 kg ha^{-1} , S_2 = 20 kg ha^{-1} , S_3 = 30 kg ha^{-1}).

In order to prevent contamination of bacteria, molybdenum and sulphur applications from plot to plot, a distance of 2 meters was left between the plots. Sulphur was applied uniformly to the rows opened with sowing. For osmopriming, seeds were kept in molybdenum solution for 6 hours and sowing was done without losing time. The inoculated seeds were moistened with sugar water (10%) just before sowing and inoculated with bacterial culture at the rate of 1 kg bacteria per 100 kg seeds. Inoculation and sowing were carried out in the evening hours when the direct rays of the sun lost their effect. Control plots were sown first to prevent bacterial contamination. The sown seeds were immediately covered and pressed.

The experiment was established in the first week of May. Mung bean seeds were sown in $40 \times 10 \text{ cm}$ sowing norm with 3 m length and 6 rows in each plot. The area of a plot was 7.2 m^2 ($3 \text{ m} \times 2.4 \text{ m}$). Di ammonium phosphate (18:46:0) and urea (46%) fertilizers were applied equally to all plots with the calculation of 40 kg ha^{-1} pure nitrogen and $60 \text{ kg ha}^{-1} \text{ P}_2\text{O}_5$. In the experiment, irrigation was carried out with drip irrigation considering the water requirement of the plants, soil moisture status and climatic conditions. Weed control was done with hand hoe and no chemical control was applied in the experiment. Harvesting was carried out in the first week of October after leaving 50 cm from the outermost rows in the sub-sub-plots and 50 cm from both sides of the plant rows to prevent edge effect.

2.2.3. Determination of agronomic and quality characteristics

In the study, plant height (cm), first pod height (cm), number of pods per plant (number of pods), number of grains per plant (number of grains), plant grain yield (g), harvest index (%) and grain yield (kg ha^{-1}), hundred grain weight (g), number of nodules (g plant^{-1}) and nodule weight (g plant^{-1}) were examined in 10 randomly selected plants. Yield and yield components were determined according to the methods described by Karaman (2019). During the flowering period, when nodule formation was the highest, the plants from each plot were removed from the soil 30-40 cm deep with the help of a belt and washed carefully to determine the number of nodules and nodule weight. Nitrogen content in mung bean grains was determined according to the Kjeldahl method (Kacar and İnal 2010) and protein content (%) was calculated by multiplying the obtained values with a coefficient of 6.25 (Bremner 1965). The ground mung bean grains were subjected to incineration in a muffle furnace at 550°C for 5 hours and the ash content was obtained by multiplying the obtained value by % (Yılmaz 2005).

2.2.4. Data evaluation

The data obtained were analyzed in the SAS statistical package program according to the split plots experimental design in randomized blocks. The differences of the averages calculated for each treatment were grouped according to the LSD test, some at 1% and some at 5%.

3. Results

In this study, the effects of sulphur, molybdenum and bacteria application on agronomic and some quality traits of mung bean were investigated and the results of analysis of variance are given in Table 1.

Table 1. Analysis of variance results for the effects of bacteria, molybdenum and sulphur treatments on yield and yield components in mung bean

SD	DF	Plant Height	First Pod Height	Pods per Plant	Grains per Plant ¹
Replication	2	0.19	6.25	1.93	170.04
Bacteria (B)	1	195.62*	72.40 ns	1429.76**	99748.19**
Error-1	2	5.77	7.72	1.04	92.17
Molybdenum (Mo)	1	166.18**	26.78*	124.90**	6441.96**
B x Mo Interaction	1	1.80 ns	0.17 ns	1.93 ns	1000.37 ns
Error-2	4	4.62	2.93	1.91	206.73
Sulphur (S)	3	47.00**	19.03**	64.31**	4185.73**
B x S Interaction	3	1.83 ns	0.76 ns	17.74**	577.05**
Mo x S Interaction	3	9.17 ns	2.33 ns	6.44**	854.47**
B x Mo x S Interaction	3	1.34 ns	2.81 ns	5.83**	268.41*
Error-3	24	5.58	2.61	1.14	67.43
SD	DF	Plant Grain Yield	Grain Yield	Harvest Index	Protein Content
Replication	2	1.29	12962.0	5.24	0,15
Bacteria (B)	1	239.77**	7925954.3**	315.70**	119,39**
Error-1	2	0.03	36967.4	2.98	0,13
Molybdenum (Mo)	1	13.06**	988060.7**	10.03 ns	8,02**
B x Mo Interaction	1	0.99 ns	168189.6 ns	1.15 ns	0,08 ns
Error-2	4	0.21	41949.2	1.91	0,16
Sulphur (S)	3	12.50**	399170.7**	22.98**	9,36**
B x S Interaction	3	1.69*	148434.4*	29.10**	0,94 ns
Mo x S Interaction	3	0.31 ns	208011.8**	1.86 ns	0,55 ns
B x Mo x S Interaction	3	0.30 ns	78062.3 ns	1.64 ns	0,41 ns
Error-3	24	0.52	38194.7	3.94	0,55
SD	DF	Hundred Grain Weight	Ash Content	Number of Nodules	Nodule weight
Replication	2	0.12	0.01	0.83	0.00
Bacteria (B)	1	4.24*	0.89 **	418.07**	20.66**
Error-1	2	0.13	0.01	0.96	0.01
Molybdenum (Mo)	1	1.08 ns	0.22**	54.70**	1.18**
B x Mo Interaction	1	0.01 ns	0.01 ns	12.77**	0.00 ns
Error-2	4	0.16	0.01	0.25	0.02
Sulphur (S)	3	1.42**	0.23**	29.03**	1.93**
B x S Interaction	3	0.11 ns	0.01**	7.94**	0.41**
Mo x S Interaction	3	0.01 ns	0.01**	0.58 ns	0.03 ns
B x Mo x S Interaction	3	0.11 ns	0.02**	0.05 ns	0.06 ns
Error-3	24	0.21	0.01	0.23	0.02

*: P<0.05; **: P<0.01; ¹: Mean squared values of traits; SD: Sources of variation; DF: Degrees of freedom

According to the variance analysis results of Rhizobium bacteria, molybdenum and sulphur (S) doses applied to mung bean on yield and yield components; the differences between plant height, number of grains and pods per plant, plant grain yield, grain yield, harvest index, hundred grain weight, protein content, ash content, number and weight of nodules were found statistically significant different level (P<0.01 and P<0.05). Differences between all traits except harvest index and hundred grain weight according to osmopriming treatment with molybdenum and differences between all traits according to sulphur doses were statistically significant. On the other hand, bacteria (B) x molybdenum (Mo) interaction for biological yield and nodule number; B x S interaction for number of pods per plant, number of grains per plant, plant grain yield, grain

yield, harvest index, ash content, number of nodules and nodule weight; Mo × S interaction for number of pods per plant, number of grains per plant, ash content and grain yield; B × Mo × S interaction for number of pods per plant, ash content and number of grains per plant were statistically significant (Table 1).

3.1. Plant Height

The mean plant height of mung bean plants varied between 62.03-66.06 cm and 62.18-65.91 cm according to bacteria and molybdenum treatments, respectively, and the plant height of the plants in the plots treated with bacteria and molybdenum was higher than the plots not treated with bacteria and molybdenum. Plant height averages varied between 61.75 cm and 66.42 cm according to sulphur doses. The lowest plant height was determined at 0 kg ha⁻¹ S dose and the highest plant height was determined at 30 kg ha⁻¹ S dose, and there was no statistical difference between them and 20 kg ha⁻¹ S application. However, plant height values of mung bean increased with increasing sulphur doses (Table 2).

Table 2. Mean values of plant height and first pod height traits of mung bean according to bacteria, molybdenum and sulphur treatments

Boron Appl.	Mo Appl.	Plant Height (cm)					First Pod Height (cm)				
		S (kg ha ⁻¹)					S (kg ha ⁻¹)				
		0	10	20	30	Mean	0	10	20	30	Mean
B ₀	Mo ₀	59.07	59.20	60.00	61.60	59.97	29.67	29.60	30.40	32.80	30.62
	Mo ₁	60.67	63.13	64.40	68.13	64.08	30.00	32.67	32.40	32.90	31.99
Mean		59.87	61.17	62.20	64.87	62.03^{B*}	29.83	31.13	31.40	32.85	31.30
B ₁	Mo ₀	62.53	64.73	64.90	65.40	64.39	32.05	32.70	33.20	33.87	32.95
	Mo ₁	64.73	66.33	69.33	70.53	67.73	32.40	33.53	35.93	36.40	34.57
Mean		63.63	65.53	67.12	67.96	66.06^A	32.23	33.12	34.57	35.13	33.76
Mo × S	Mo ₀	60.80	61.97	62.45	63.50	62.18^b	30.86	31.15	31.80	33.33	31.79^b
	Mo ₁	62.70	64.73	66.87	69.33	65.91^a	31.20	33.10	34.17	34.65	33.28^a
Mean		61.75^C	63.35^{BC}	64.66^{AB}	66.42^A		31.03^C	32.13^{BC}	32.98^{AB}	33.99^A	

*: There is no statistical difference between averages with the same letters.

3.2. First Pod Height

The height of the first pod is an important trait affecting harvest losses in machine harvesting and it is one of the most important agronomic traits for mung bean. In the study, the first pod height of mung bean increased with the increase in S doses. However, there was no statistical difference between 20 and 30 kg ha⁻¹ S treatments. According to molybdenum treatments, the first pod height was higher in the plots where Mo was applied compared to the plots where Mo was not applied. The first pod height increased with bacteria application, but this increase was not statistically different (Table 2).

3.3. Number of Pods per Plant

Bacteria, Mo and S fertilizer applications had a significant effect on the number of pods in mung bean plants. It was determined that the number of pods in the plants in the plots where bacteria and molybdenum were applied (30.75 and 26.90 pcs, respectively) was higher. However, the number of pods in the plant increased with the increase in S doses, the highest number of pods was determined in 30 kg ha⁻¹ S application and the lowest number of pods was determined in 10 kg ha⁻¹ S application and control (0 kg ha⁻¹ S) application. The number of pods per plant varied between 18.27-34.90 (S₀R₀-S₃R₁) according to S doses and bacteria application, and it was determined that the number of pods of the plants in the plots inoculated with bacteria was higher with increasing S doses. When the Mo × S interaction was examined, the highest number of pods was determined in 30 kg S fertilization per hectare with Mo application (29.69), followed by 20 kg S fertilization per hectare with Mo application (28.05). The lowest number of pods was determined in plots without Mo and S application (22.93). In the B × Mo × S combination, the highest number of pods was obtained in plots with 3 kg S application and molybdenum and bacteria inoculation (35.80) and the lowest number of pods was obtained in plots without molybdenum and bacteria inoculation with 0 and 10 kg S application (17.27 and 17.33 pcs, respectively) (Table 3).

Table 3. Mean values of number of pods per plant and number of grains per plant according to bacteria, molybdenum and sulphur treatments to mung bean

Boron Appl.	Mo Appl.	Number of Pods per Plant (pcs)					Number of Grains per Plant (pcs)				
		S (kg ha ⁻¹)					S (kg ha ⁻¹)				
		0	10	20	30	Mean	0	10	20	30	Mean
B ₀	Mo ₀	17.27 ^g	17.33 ^g	19.27 ^{fg}	18.20 ^{fg}	18.02	165.60 ^j	171.33 ^{ij}	172.53 ^{h-j}	217.33 ^{ef}	181.70
	Mo ₁	19.27 ^{fg}	20.73 ^{ef}	23.00 ^e	23.58 ^e	21.64	183.87 ^{g-j}	195.47 ^{f-i}	201.00 ^{f-h}	202.62 ^{e-g}	195.74
Mean		18.27^f	19.03^{ef}	21.13^d	20.89^{de}	19.83^B	174.73^d	183.40^d	186.77^d	209.98^c	188.72^{B*}
B ₁	Mo ₀	28.60 ^d	23.87 ^e	30.87 ^{b-d}	34.00 ^{ab}	29.33	252.80 ^{cd}	230.53 ^{de}	276.33 ^{bc}	295.30 ^{ab}	263.74
	Mo ₁	29.40 ^d	30.33 ^{cd}	33.10 ^{a-c}	35.80 ^a	32.16	267.47 ^{bc}	287.67 ^{ab}	313.33 ^a	315.70 ^a	296.04
Mean		29.00^c	27.10^c	31.98^b	34.90^a	30.75^A	260.13^b	259.10^b	294.83^a	305.50^a	279.89^A
Mo x S	Mo ₀	22.93 ^d	20.60 ^e	25.07 ^c	26.10 ^{bc}	23.68^b	209.20 ^{de}	200.93 ^e	224.43 ^{cd}	256.32 ^{ab}	222.72^b
	Mo ₁	24.33 ^{cd}	25.53 ^c	28.05 ^{ab}	29.69 ^a	26.90^a	225.67 ^{cd}	241.57 ^{bc}	257.17 ^{ab}	259.16 ^a	245.89^a
Mean		23.63^C	23.07^C	26.56^B	27.89^A	217.43^C	221.25^C	240.80^B	257.74^A		

*: There is no statistical difference between averages with the same letters.

3.4. Number of Grains per Plant

Number of grains per plant, which is effective on seed yield in mung bean, varied significantly according to sulphur, molybdenum and bacteria treatments. It was determined that the average number of grains per plant in the plots treated with bacteria and molybdenum (279.89 and 245.89 plants, respectively) was higher than the plots without treatment (188.72 and 222.72 plants, respectively). According to S doses, the number of grains per plant varied between 217.43-257.74 and the highest number of grains per plant was determined at 30 kg ha⁻¹ S dose, while the lowest number of grains per plant was determined in control (0 kg ha⁻¹) and 10 kg ha⁻¹ S treatments. However, the number of grains per plant increased with increasing S doses and 30 kg ha⁻¹ S dose increased the number of grains per plant by 19% compared to the control treatment. On the other hand, when the S x B interaction was examined, the number of grains per plant varied between 174.73- 305.50, the highest number of grains per plant was determined in the plots where 30 kg ha⁻¹ S application and bacteria were applied and there was no statistical difference between the number of grains per plant in the plots where 20 kg ha⁻¹ S application and bacteria were applied together. Again, the lowest number of grains per plant was determined in the plots without S and bacteria application, while they were in the same statistical group with 10 and 20 kg ha⁻¹ S and non-bacteria inoculated treatments. According to the Mo x S treatment, the highest number of grains per plant was determined in the Mo treatment with 30 kg ha⁻¹ S and the lowest number of grains per plant was determined in the plants without Mo treatment with 10 kg ha⁻¹ S application. When the S x Mo x B interaction was examined, the highest number of grains per plant was determined in plots with 3 kg S application and Mo and bacteria inoculation (315.70 pieces), while the lowest number was obtained from plants without molybdenum and bacteria inoculation with 0 kg ha⁻¹ S application (165.6 pieces). It was determined that S, bacteria and Mo applications and their co-applications significantly increased the number of grains per plant of mung bean (Table 3).

3.5. Plant Grain Yield

Plant grain yield is one of the important agronomic traits that directly affect grain yield. In the study, average plant grain yield varied between 10.65-15.12 g and 12.36-13.41 g according to bacteria and Mo treatments, respectively. Especially the grain yields of the plants treated with bacteria and Mo were found to be higher. According to S doses, average plant grain yield varied between 11.56 (S₀)-14.02 (S₃) g. It was determined that plant grain yield of mung bean increased with the increase in S doses. While the S x B interaction significantly changed the plant grain yield of mung bean, the highest plant grain yield was determined in plants inoculated with bacteria with 30 kg ha⁻¹ S application, followed by plants inoculated with bacteria with 10 and 20 kg ha⁻¹ S application. However, the lowest plant grain yield was detected in plants that were not inoculated (B₀) with 0 kg ha⁻¹ (control) S dose (Table 4).

Table 4. Mean values of plant grain yield and grain yield traits of mung bean according to bacteria, molybdenum and sulphur treatments

Boron Appl.	Mo Appl.	Plant Grain Yield (g)					Grain Yield (kg ha ⁻¹)				
		S (kg ha ⁻¹)					S (kg ha ⁻¹)				
		0	10	20	30	Mean	0	10	20	30	Mean
B ₀	Mo ₀	8.94	9.85	10.27	12.02	10.27	1867.0	1947.7	2031.7	2136.4	1995.7
	Mo ₁	14.05	16.44	16.22	16.43	11.03	2202.6	2554.7	2200.6	2646.1	2401.0
Mean		9.56^d	10.27^d	10.58^d	12.19^c	10.65^B	2034.8^d	2251.3^d	2116.1^d	2391.2^{cd}	2198.4^{B*}
B ₁	Mo ₀	13.05	14.35	15.12	15.29	14.45	2868.8	2665.5	3165.4	3007.5	2926.8
	Mo ₁	10.18	10.68	10.89	12.35	15.78	2494.5	3199.8	3334.5	3352.7	3095.4
Mean		13.55^b	15.40^a	15.67^a	15.86^a	15.12^A	2681.6^{bc}	2932.7^{ab}	3249.9^a	3180.1^a	3011.1^A
Mo x S	Mo ₀	11.00	12.10	12.70	13.66	12.36^b	2367.9 ^c	2306.6 ^c	2598.5 ^{bc}	2571.9 ^{bc}	2461.2^b
S	Mo ₁	12.12	13.56	13.55	14.39	13.41^a	2348.5 ^c	2877.4 ^{ab}	2767.5 ^{ab}	2999.4 ^a	2748.2^a
Mean		11.56^C	12.83^B	13.12^B	14.02^A		2358.2^C	2592.0^B	2683.0^{AB}	2785.7^A	

*: There is no statistical difference between averages with the same letters.

3.6. Grain Yield

Increasing grain yield in mung bean is one of the most important breeding objectives. In this direction, it was determined that S, Mo and bacteria applied to mung bean were highly effective on grain yield. Grain yield varied between 2358.2- 2785.7 kg ha⁻¹ according to S doses. The highest average grain yield was determined at 30 kg ha⁻¹ S dose and this dose was statistically in the same group with 20 kg ha⁻¹ S dose. The lowest grain yield was determined at 0 kg ha⁻¹ (control) S treatment. However, it was determined that 30 kg ha⁻¹ S application increased grain yield by 18% compared to the control application. On the other hand, average grain yield varied between 2198.4 (B₀)-3011.1 (B₁) kg ha⁻¹ and 2461.2 (Mo₀)-2748.2 (Mo₁) kg ha⁻¹ according to bacteria and Mo treatments, respectively. It was determined that the grain yields of the plants treated with bacteria and molybdenum were higher than the untreated plots. When S x Mo was analysed, the highest grain yield was determined in plots inoculated with bacteria with 3 kg ha⁻¹ S application (3180.1 kg ha⁻¹), followed by plots inoculated with bacteria with 20 and 10 kg ha⁻¹ S application (3249.9 kg ha⁻¹ and 2932.7 kg ha⁻¹, respectively). The lowest grain yield was recorded at 0, 10 and 20 kg ha⁻¹ (control) S doses and in plots without bacteria inoculation (2034.8 kg ha⁻¹, 2251.3 kg ha⁻¹ and 2116.1 kg ha⁻¹, respectively) (Table 4).

3.7. Harvest index

Harvest index is calculated by the ratio of grain yield to biological yield and increasing the harvest index value of mung bean is among the important breeding objectives. In this study, harvest index values varied between 24.08-27.45% according to S doses. While the highest harvest index value was found at 30 kg ha⁻¹ S dose, it was in the same statistical group with 10 and 20 kg ha⁻¹ S doses. However, the lowest harvest index value was found at 0 kg ha⁻¹ S dose. On the other hand, according to the bacterial treatments, the highest harvest index value was found in the plots with bacterial treatment (B₁). Although molybdenum application was not statistically effective on harvest index, it was determined that the harvest index value (26.35%) of the plants in molybdenum treated plots (Mo₁) was higher. When the S x B interaction was analysed, harvest index values varied between 20.99% (S₀B₀) and 30.44% (S₁B₁). There was no statistical difference between 10 kg ha⁻¹ S dose and bacteria treatment, 20 and 30 kg ha⁻¹ S dose and bacteria treatments (Table 5).

3.8. Hundred Grain Weight

Hundred grain weight, which is important for marketing, is an important quality criterion for determining large grains in mung bean. Bacteria treatments significantly changed the hundred grain weight of mung bean and the highest hundred grain weight was determined in bacterial inoculation (6.39 g). In molybdenum treatment, the highest hundred grain weight was determined in seeds osmoprimed with molybdenum, while there was no statistical difference between molybdenum treatments. On the other hand, according to the sulphur treatments, facial grain weight varied between 5.64-6.43 g. The highest facial grain weight was determined at 30 kg ha⁻¹ S dose, followed by 20 (6.27 g) and 30 (6.04 g) kg ha⁻¹ S dose, respectively. In the study, it was determined that especially bacteria and sulphur treatments significantly increased the hundred grain

weight. However, facial grain weight increased according to molybdenum treatment and interactions (Mo x S, B x Mo, B x S, B x Mo x S, B x Mo x S), but there was no statistical difference (Table 5).

Table 5. Mean values for harvest index and hundred grain weight traits of mung bean according to bacteria, molybdenum and sulphur treatments

Boron Appl.	Mo Appl.	Harvest Index (%)					Hundred Grain Weight (g)				
		S (kg ha ⁻¹)					S (kg ha ⁻¹)				
		0	10	20	30	Mean	0	10	20	30	Mean
B ₀	Mo ₀	19.09	21.59	23.94	26.24	22.72	5.28	5.35	5.81	6.12	5.64
	Mo ₁	22.88	21.96	23.54	27.39	23.94	5.33	5.90	6.20	6.36	5.95
Mean		20.99^d	21.77^d	23.74^{cd}	26.82^{bc}	23.33^B	5.31	5.62	6.01	6.24	5.79^{B*}
B ₁	Mo ₀	27.08	30.51	27.09	27.94	28.16	5.68	6.34	6.44	6.51	6.24
	Mo ₁	27.28	30.37	29.17	28.22	28.76	6.25	6.56	6.63	6.71	6.54
Mean		27.18^{a-c}	30.44^a	28.13^{ab}	28.08^{ab}	28.46^A	5.97	6.45	6.53	6.61	6.39^A
Mo x S	Mo ₀	23.09	26.05	25.52	27.09	25.44	5.48	5.85	6.12	6.31	5.94
S	Mo ₁	25.08	26.16	26.36	27.80	26.35	5.79	6.23	6.42	6.53	6.24
Mean		24.08^B	26.11^{AB}	25.94^{AB}	27.45^A		5.64^B	6.04^{AB}	6.27^A	6.43^A	

*: There is no statistical difference between averages with the same letters.

3.9. Protein Content

In the study, bacteria, molybdenum and S fertilizer applications had significant effect on the protein content of mung bean. It was determined that the protein ratio was higher in the plots where bacteria and molybdenum were applied (20.70% and 17.54%, respectively). In addition, the protein content also increased with the increase in S doses, the highest protein content was determined in 20 and 30 kg ha⁻¹ S treatments and the lowest protein content was determined in the control (0 kg ha⁻¹ S) treatment. On the other hand, it was determined that the mung bean grains obtained from the plots where S doses increased and bacteria and molybdenum treatments were applied had higher protein content (Table 6).

Table 6. Mean values of protein and ash content traits of mung bean according to bacteria, molybdenum and sulphur treatments

Boron Appl.	Mo Appl.	Protein Content (%)					Ash Content (%)				
		S (kg ha ⁻¹)					S (kg ha ⁻¹)				
		0	10	20	30	Mean	0	10	20	30	Mean
B ₀	Mo ₀	16.45	17.05	17.50	17.72	17.18	4.47 ^h	4.54 ^g	4.64 ^f	4.86 ^d	4.63
	Mo ₁	16.98	17.93	18.22	18.53	17.92	4.53 ^{gh}	4.77 ^e	4.86 ^d	4.89 ^{cd}	4.76
Mean		16.71	17.49	17.85	18.13	17.54^{B*}	4.50^g	4.65^f	4.75^e	4.87^{cd}	4.69^{B*}
B ₁	Mo ₀	18.84	19.29	21.23	21.65	20.25	4.75 ^e	4.85 ^d	4.95 ^c	5.03 ^b	5.03
	Mo ₁	19.67	21.32	21.61	22.00	21.15	4.94 ^c	4.95 ^c	5.04 ^b	5.20 ^a	4.89
Mean		19.26	20.31	21.42	21.82	20.70^A	4.84^d	4.90^c	5.00^b	5.12^a	4.96^A
Mo x S	Mo ₀	17.64	18.17	19.37	19.69	18.71^b	4.61 ^g	4.69 ^f	4.79 ^d	4.95 ^b	4.76^b
S	Mo ₁	18.33	19.63	19.92	20.27	19.53^a	4.74 ^e	4.86 ^c	4.95 ^b	5.04 ^a	4.90^a
Mean		17.99^C	18.90^B	19.64^{AB}	19.98^A		4.67^D	4.78^C	4.87^B	4.99^A	

*: There is no statistical difference between averages with the same letters.

3.10. Ash Content

Crude ash content, which is an indicator of inorganic matter content of seeds, varied between 4.69% (B₀)-4.96% (B₁) and 4.76% (Mo₀)-4.90% (Mo₁) according to bacteria and molybdenum treatments, while bacteria and molybdenum treatments increased ash content. As the application amount of S doses increased, ash content also increased. The highest ash content was determined at 30 kg ha⁻¹ S dose and the lowest at 0 kg ha⁻¹ (control) S dose. On the other hand, when the S x B interaction was analysed, the highest ash content was determined in S₂B₁(5.12%) and the lowest in S₀B₀ (4.50%). According to Mo x S interaction, the highest ash content was determined in Mo₁S₃(5.04%) and the lowest in Mo₀S₀ (4.61%). In the B x Mo x S combination, the highest ash rate was obtained in molybdenum and bacterial inoculation (B₁Mo₁S₃, 5.20%) with 30 kg S application per hectare, and the lowest ash rate was obtained in molybdenum and bacterial inoculation (B₀Mo₀S₀, 4.47%) with

0 S application per hectare. Rhizobium bacteria, molybdenum and sulphur applications to mung bean increased the ash content (Table 6).

3.11. Number of Nodules

In the study, it was determined that *Rhizobium* bacteria inoculated to mung bean seeds before sowing (9.12 pieces plant⁻¹) increased the number of nodules compared to the plants in the plots without inoculation (3.21 pieces plant⁻¹). Molybdenum application also encouraged nodule formation and the number of nodules was higher in the treated plots (7.24 pieces plant⁻¹). According to S doses, the number of nodules in plants varied between 4.23 (S₀)-7.74 (S₃) pieces plant⁻¹ and the number of nodules in plants increased in parallel with the increase in S doses. When the S × B interaction was analysed, the number of nodules varied between 2.32 (S₀B₀)-11.08 (S₃B₁) pieces plant⁻¹. However, S₀B₀ and S₁B₀ treatments with the lowest nodule number and S₃B₁ and S₂B₁ treatments with the highest nodule number were in the same statistical group. It was found that increasing S doses and inoculation with bacteria increased the number of nodules (Table 7).

3.12. Nodule Weight

In the study, average nodule weight values varied between 0.79 (R₀)- 2.10 (R₁) g plant⁻¹ and 1.28 (Mo₀)-1.60 (Mo₁) g plant⁻¹ according to bacteria and molybdenum treatments, respectively, while bacteria and molybdenum treatments increased nodule weight values. According to S doses, the average nodule weight varied between 0.90 (S₀) g plant⁻¹ and 1.82 (S₃) g plant⁻¹. However, it was determined that the nodule weight of mung bean increased with increasing S doses. When the S × B interaction was analysed, nodule weight varied between 0.50 (S₀B₀)- 2.51 (S₃B₁) g plant⁻¹. S₂B₁ and S₃B₁ treatments, which had the highest nodule weight, were in the same statistical group. Similar to the nodule number, especially the increase in S doses and bacterial inoculation were effective in increasing the nodule weight of the plants (Table 7).

Table 7. Mean values for nodule number and nodule weight traits of mung bean according to bacteria, molybdenum and sulphur treatments

Boron Appl.	Mo Appl.	Number of Nodules (pcs plant ⁻¹)					Nodule Weight (g plant ⁻¹)				
		S (kg ha ⁻¹)					S (kg ha ⁻¹)				
		0	10	20	30	Mean	0	10	20	30	Mean
B ₀	Mo ₀	2.14	2.36	2.60	3.57	2.67 ^d	0.33	0.58	0.64	0.94	0.62
	Mo ₁	2.50	3.45	3.90	5.23	3.77 ^c	0.66	0.82	0.97	1.33	0.95
Mean		2.32^e	2.90^e	3.25^e	4.40^d	3.21^B	0.50^e	0.70^{de}	0.81^d	1.14^c	0.79^{B*}
B ₁	Mo ₀	4.80	6.78	9.20	9.37	7.54 ^b	1.07	1.80	2.48	2.43	1.94
	Mo ₁	7.47	10.08	12.47	12.80	10.70 ^a	1.55	2.33	2.55	2.57	2.25
Mean		6.13^c	8.43^b	10.83^a	11.08^a	9.12^A	1.31^c	2.06^b	2.51^a	2.50^a	2.10^A
Mo × S	Mo ₀	3.47	4.57	5.90	6.47	5.10 ^b	0.70	1.19	1.56	1.69	1.28 ^b
	Mo ₁	4.98	6.77	8.18	9.02	7.24 ^a	1.11	1.58	1.76	1.95	1.60 ^a
Mean		4.23^D	5.67^C	7.04^B	7.74^A		0.90^D	1.38^C	1.66^B	1.82^A	

*: There is no statistical difference between averages with the same letters.

4. Discussion

In this study, the changes in some agronomic and quality characteristics of mung bean as a result of sulphur, molybdenum and bacteria application were determined. It was observed that sulphur, molybdenum and bacteria treatments were highly effective on plant height and first pod height of mung bean and the treatments increased plant height and first pod height. However, plant height and first pod height increased with the increase in sulphur doses, and the highest plant height was determined in 20 and 30 kg ha⁻¹ sulphur application (Table 2). Rhizobium bacteria are well known to affect plant growth and development through a wide variety of mechanisms such as N₂ fixation, production of plant growth regulators, mineral uptake and suppression of plant diseases (Kennedy et al. 2004; Ahmad et al. 2013). Sulphur fertilization improves nitrogen uptake efficiency of plants and prevents nitrogen loss from the soil (Brown et al. 2000). Therefore, the development of plants increases. In this study, it was determined that sulphur, bacteria and molybdenum applications increased plant height and first pod height of mung bean (Table 2). As a matter of fact, other

studies have also reported significant increases in plant growth with sulphur, *Rhizobium* inoculation and molybdenum application (Iqbal et al. 2012; Togay et al. 2008; Bahadur and Tiwari 2014).

In the study, changes in yield and yield components according to sulphur, molybdenum and bacteria applications applied to mung beans were examined. Especially the application of sulphur, molybdenum and bacterial inoculation alone and in combination had a significant effect on the number of pods and grains in the plant, plant grain yield, grain yield and harvest index, especially of mung bean. *Rhizobium* bacteria and molybdenum application applied to mung bean increased the number of pods and grains in the plant, plant grain yield, grain yield, harvest index and hundred grain weight compared to the control application. However, with the increase in S doses, it was determined that the number of pods and grains per plant, plant grain yield, grain yield, harvest index and hundred grain weight of mung bean increased, and 30 kg S application per hectare gave the highest values (Tables 3; 4; 5). The beneficial effect of this increase in S, Mo and bacterial application on yield and parameters contributing to yield can be attributed to the vital role of enzymes in the function of biological processes that lead to an increase in yield components in plants. Molybdate, the dominant form present in plants, is included in enzymes by participating directly or indirectly in nitrogen metabolism, especially in plants, as a part of the protein complex molybdenum cofactor (Kaiser et al. 2005). Along with these, the findings in the study are similar to Rabbani et al. (2005) who found that the number of pods and grain yield of the plant increased with *Rhizobium* inoculation in combination with micronutrients is consistent with the findings.

In the study, protein and ash content of mung bean showed significant changes according to the treatments. Especially protein and ash content of mung bean grains increased with increasing S doses. However, protein and ash content increased according to bacteria and molybdenum treatments (Table 5). In this context, the importance of sulphur in pulse nutrition has been well understood in recent years. It is involved in various metabolic and enzymatic processes, including protein formation, photosynthetic activity, and symbiotic nitrogen fixation between legume and *Rhizobium* (Becana et al. 2018). In addition, sulphur plays an important role in the synthesis of coenzyme A (CoA), biotin, thiamine (vitamin B₁) and glutathione (Kahraman et al. 2016) found a significant negative relationship between protein content and nitrogen-free substances and a significant positive relationship between sulphur content. In this respect, they stated that nitrogen-free substances and sulphur content should be emphasized in order to increase the protein content in legumes. They stated that environmental conditions and genotypes are effective on the protein and ash content of mung bean grains (Baraki et al. 2020; Karaman 2019; Yadav et al. 2023); and that the protein content may vary according to various nutrients such as sulphur, bacteria and molybdenum (Ahmad et al. 2021; Ahmad et al. 2022; Junaid et al. 2022). In this context, the findings obtained in this study are in agreement with the data obtained by the researchers.

In the absence of *Rhizobium* bacteria, mung beans are not able to assimilate atmospheric nitrogen, which increases soil fertility and minimizes the need for nitrogen fertilizers. Weisany et al. (2013) observed that biological nitrogen fixation provides an economic advantage by improving the quality and quantity of the product by reducing nitrogen input as fertilizer. In this context, inoculation of *Rhizobium* bacteria increased the nodule number and weight of mung bean. In addition, S and Mo treatments were also found to be effective in increasing the nodule number and weight of mung bean (Table 6). The increase in nodule formation and weight is due to bacterial inoculation, sulphur and molybdenum, which are required by *Rhizobium* bacteria to fix atmospheric nitrogen in legumes. Indeed, *Rhizobium* inoculation was found to increase the number of effective nodules in mung bean by 57% compared to the control (Hossain et al. 2011). In particular, molybdenum is an important element that helps protein synthesis and the fixation of atmospheric nitrogen by nodule bacteria in the roots of legumes. In addition, since Mo has low mobility, it is present at higher levels in the root zone of the plant than in the stem and leaves. The Mo element nitrogenase enzyme acts as a key enzyme in the functions of nitrogen fixing microorganisms, while nitrate reductase enzyme acts as a key enzyme in the reduction of nitrate to nitrite in plants. Nicoloso and Dos Santos (1990) determined the effects of different nitrogen doses, *Rhizobium* inoculation and molybdenum applications on bean plants and found that molybdenum did not increase nodule weight in nitrogen-free applications, while inoculation and molybdenum increased dry matter content, total nitrogen accumulation and grain yield. It was also found that

S and Mo application can increase nodule formation in legume plants (Vieira et al. 1998; Yadav, 2004; Singh et al. 2017).

5. Conclusions

In this study, some agronomic and quality traits and changes in nodule development were determined as a result of sulphur, molybdenum and bacteria treatments applied to mung bean. In the study, significant differences were found in terms of the traits examined according to sulphur, molybdenum and bacteria treatments. It was determined that bacteria and molybdenum application increased the yield, yield components and nodule formation of mung bean. It was determined that agronomic parameters such as yield, hundred grain weight, protein content, ash content and nodulation activities increased as the application amount of sulphur fertilization increased. As a result, it can be recommended to use 30 kg S ha⁻¹ with bacteria and molybdenum for higher yield and quality in mung bean cultivation. Encouraging the use of S and Mo-containing fertilizers and bacterial inoculation and providing the necessary support in this regard will make significant contributions to the development of mung bean cultivation.

Author Contributions: The authors have an equal contribution. All authors have read and agreed to the published version of the manuscript.

Funding: This research received no external funding.

Conflicts of Interest: The authors declare no conflict of interest.

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Utilizing of Plant-Smoke Solution to Alleviate Drought Sensitivity on Forage Peas

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HIGHLIGHTS

- Drought stress is a meteorological event that most threatens agricultural production and food security in the future.
- Therefore, urgent measures need to be taken to preserve the quality and yield of existing genotypes.
- In this context, smoke solutions in which both natural and agricultural wastes are recycled can be used.
- Because, the results of this study determined that priming with smoke solution prevented the effects of drought on forage pea.

Abstract

The study was conducted to determine the sensitivity of forage pea to drought stress intensities and the contribution of smoke solutions in preventing this sensitivity. For the smoke solution, dried poppy harvest residues were prepared with a special mechanism. Within this scope, two different concentrations (1 and 10%) of poppy smoke solution were utilized in priming of two distinct forage pea varieties (Gap pembesi and Özkaynak). After priming, the seeds were sowing in pots and it was exposed to moderate and severe drought conditions after 21 days. Effects of smoke solutions and drought stress in varieties were assessed by physically (shoot length) and chemical parameters (crude protein, ADF, NDF, Ca, Mg, P and K). Both forage pea varieties have been observed that crude protein and mineral substance (except Ca) contents decrease significantly under drought stress, especially in severe drought. This loss was increased again by eliminating both doses (especially 10%) of smoke solutions. In fact, under normal growing conditions (control), 10% smoke solution application had the highest crude protein in both varieties. However, the effect of drought and the defense mechanism of smoke solution created differences in forage pea varieties based on parameters. In general, the Özkaynak was more resistant to drought and the response to smoke solution was stronger. In conclusion, by incorporating the poppy-smoke solution into the drought stress mitigation strategy, this approach not only curbed environmental losses but also mitigated the impacts of drought stress.

Keywords: Smoke solution; drought; forage quality; priming.

Citation: Çopur Doğrusöz M, Mut H (2024). Utilizing of plant-smoke solution to alleviate drought sensitivity on forage peas. *Selcuk Journal of Agriculture and Food Sciences*, 38(2), 288-297. <https://doi.org/10.15316/SJAIFS.2024.027>

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Received date: 01/02/2024

Accepted date: 25/07/2024

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1. Introduction

The formation of growth and quality in plants is primarily an extremely complex process determined by both genetics and the environment. While there may not be genetic variability, the environment, especially due to climate change, is constantly undergoing changes. In recent years, with global climate change, the consequences of commonly observed extreme meteorological events have been gaining increasing attention worldwide. Drought, along with other abiotic stresses, is a meteorological event that has shown a growing trend in terms of its duration, frequency, and spatial scope and significantly affects agricultural production.

By the year 2050, it is estimated that the global population will exceed 9.7 billion, with more than 65% of individuals relying solely on agriculture for their livelihoods. In developing countries like Turkey, this ratio is predicted to reach as high as 90% (Castañeda et al. 2016). In contrast, 22.5% of Turkey is experiencing high desertification, while 50.9% is showing a moderate tendency towards desertification. This situation reflects the severity of the threats that will emerge in the future. Therefore, making a paradigm shift towards sustainable agriculture and finding solutions to water scarcity and its impact on food security are of vital importance (Khaleghi et al. 2019). The need to develop methods that can increase a plant's resistance to drought stress, improve crop growth, and contribute to environmentally friendly and sustainable agriculture has become essential.

Drought severely limits the morpho-physiological functions of plants, having a negative impact on plant growth, productivity, reproduction, and survival. It is known that the increase in reactive oxygen radicals within plant cells, induced by abiotic stresses, is at the core of these negative impacts (Zandalinas et al. 2020). Plants adapt or respond to changing environmental conditions through various morphological, physiological, anatomical, and biochemical changes. Mechanisms and responses specific to drought stress can vary widely among different plant species and even varieties. Therefore, in our study, two different varieties of forage pea were examined in drought conditions.

The protein content of forage peas, which can reach up to 40%, indicates that it is a quality forage crop. It plays a significant role in improving animal nutrition, and in the future, there will be an increased need for high-quality and high-yielding varieties due to drought stress. Yield and quality are directly influenced by environmental factors and their interactions. Understanding the expression of these effects at the phenotypic and biochemical levels and predicting the segregation of field-assessed products are of great importance. In this context, it is necessary to first evaluate the responses of forage peas, which are sensitive to drought, to drought severity and to apply techniques that will mitigate or eliminate these responses.

Various approaches have been employed to mitigate drought stress, as documented in studies by Saha et al. (2020) and Zhang et al. (2020). Nevertheless, an emerging and relatively underappreciated method within the realm of agricultural mitigation strategy involves the use of plant-derived smoke solutions. This innovative approach contributes to environmentally sustainable solutions aimed at ensuring global food security. These smoke solutions, derived from burning agricultural waste and preserving the resulting smoke in water, can be applied directly to seeds through priming or to the soil. Several researchers have demonstrated the significant positive impact of plant-derived smoke solutions on germination and seedling growth, as evidenced in studies by Jefferson et al. (2008), Dixon et al. (2009), and Doğrusöz (2022). Furthermore, it has been established that smoke solutions enhance plant tolerance to abiotic stresses, as highlighted in studies by Li et al. (2017), Shah et al. (2021), Shah et al. (2020), and Khan et al. (2017). However, it should be noted that the efficacy of these smoke solutions varies depending on factors such as the type and quantity of plant material burned, the solution concentration, and the specific plant genotype employed (Doğrusöz et al. 2022).

This study focused on assessing the sensitivity of two distinct forage pea varieties to moderate and severe drought stress using chemical parameters. Additionally, two different concentrations of poppy smoke solution were utilized in priming to alleviate drought sensitivity. By incorporating the poppy-smoke solution into the drought stress mitigation strategy, this approach not only curbed environmental losses but also mitigated the impacts of drought stress.

2. Materials and Methods

The varieties 'Gap Pembesi and Özkaynak' of forage pea (*Pisum sativum* spp. *arvense* L.) were used by plant material. For the smoke solution, dried poppy harvest residues were used. The separate trials for both varieties were performed under fully controlled climate room (light, temperature and humidity) at the Agriculture Faculty of Yozgat Bozok University.

Priming Application with Smoke Solutions

Smoke solutions were generated from the poppy residues. The 1 kg of poppy straw was subjected to controlled combustion using a specialized system, following the methodology detailed in studies by Ghebrehiwot et al. (2009) and Basaran et al. (2019). The resulting smoke was then entrapped by passing it through 4 liters of distilled water. The initial smoke solution was subsequently diluted to achieve concentrations of 1% and 10% using additional distilled water. Forage pea seeds were soaked for 18 hours at a temperature of 22°C in these smoke solutions, while distilled water served as the control group.

Forage pea Growth Conditions, Drought Stress Treatments

The study encompassed three distinct treatment groups: severe and moderate drought, and normal conditions (irrigation; control). Certainly, the drought stress conditions were created by subjecting the plants to reduced water availability. Specifically, severe drought stress was simulated by withholding water for the final eight days of the experiment, while moderate drought stress was imposed by withholding water for the last four days. This manipulation aimed to replicate varying degrees of water scarcity to study the plants' responses under different levels of drought stress. That is, drought stress was achieved by completely cutting off water from the seedlings. The primed seeds of these plants were sown in 8-liter pots filled with a consistent mixture of peat and soil. Employing randomized plots trial design, the experiment featured three replications, each comprising 10 plants. The trials were conducted separately. Both varieties of forage pea were meticulously cultivated under controlled conditions, maintaining a temperature of $25 \pm 2^\circ\text{C}$. Subsequently, all plants were harvested 28 days after the experiment's initiation.

The Parameters Examined in Forage Pea Seedlings

Twenty-eight days later, all plants in the pots were removed and shoot length (cm) was determined. The samples were dried in a drying cabinet at 65 °C for 48 hours and ground to a diameter of 1 mm. Dried samples were ground and passed through a 1 mm sieve. The ground samples were processed in NIRS (Foss 6500) device (near infrared reflectance spectroscopy; Silver Spring, MD, USA) with the IC0904FE program to determine crude protein (PRT;%), ADF (acid detergent fiber,%), NDF (neutral detergent fiber;%). and mineral matter (Ca, P, K and Mg;%) were analyzed.

Statistical analyses

Statistical analyses of data were conducted with SPSS software, version 20.0 Duncan's test was performed to assess differences between averages at a significance level of $p \leq 0.05$. All parameters' data were further subjected to correlation analysis and a principal component analysis (PCA), with separate for each variety. The heat map, PCA and ballon plot graphics were created with SRplot (Tang et. al. 2023), separately for each variety

3. Results

Moderate and severe stresses of drought significantly influenced forage pea plant growth and chemical contents. However, the smoke solution applied alleviated the negatives of these effects. In both varieties, the longest shoot length was determined in 0 dose of the control treatment. The expected decrease in shoot length was observed with drought stress. However, smoke solutions significantly reduced shoot length in both varieties, both in normal irrigation and drought stress treatments. Therefore, smoke has a negative effect on the solution.

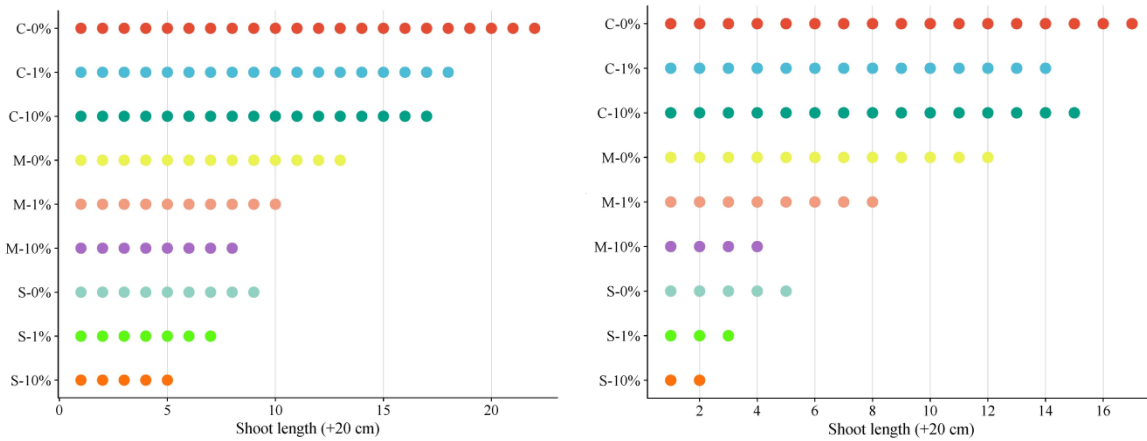


Figure 1. Changes in shoot length of smoke solution applied in forage pea under drought stress (*Gap pembesi*: left graphic, *Özkaynak*: right graphic)

In both varieties, chemical contents were significantly ($p < 0.01$) affected by priming with smoke solutions under applied drought stress (Table 1 and 2). The highest protein content in both varieties was obtained under normal irrigation conditions and 10% solution application. Additionally, Crude protein content increased in moderate drought as a defense mechanism against stress but decreased in severe drought. However, it was observed that the protein content increased in parallel with the increase in smoke solution in drought applications as with normal irrigation.

Table 1. Chemical composition (%) of smoke solution applied in *Gap Pembesi* under drought stress

Treatments	PRT	ADF	NDF	Ca	Mg	P	K
C-0%	31.98 bc	14.30 bc	27.69 ab	1.72 cd	0.54 c	0.39 cd	2.38 c
C-1%	32.16 bc	14.76 b	27.33 ab	1.97 bc	0.55 c	0.39 cd	2.44 c
C-10%	35.29 a	13.86 c	30.13 a	2.00 b	0.58 b	0.45 b	2.90 b
M-0%	32.17 bc	14.65 b	27.61 ab	1.63 d	0.48 d	0.38 cd	1.91 d
M-1%	32.85 b	10.20 d	20.70 c	1.81 c	0.50 cd	0.43 bc	2.93 b
M-10%	32.46 b	14.12 bc	25.72 b	2.27 ab	0.63 ab	0.48 a	3.37 a
S-0%	31.00 c	15.34 a	24.13 b	1.99 bc	0.52 cd	0.37 d	1.87 d
S-1%	31.97 bc	13.08 cd	21.67 c	2.16 b	0.59 b	0.40 c	2.33 cd
S-10%	32.70 b	9.56 d	17.57 d	2.41 a	0.67 a	0.41 c	2.69 bc

Different letters significant differences at $p \leq 0.05$ within one parameter. PRT; crude protein, C; control, M; moderate drought, S; severe drought.

An increase in the ADF of both varieties was determined under severe drought. However, in *Gap pembesi*, smoke solution application caused a decrease in ADF in all three cultivate conditions. In *Özkaynak*, while ADF increased in 10% dose under normal conditions, it increased in 1% dose under moderate drought. In *Gap pembesi*, NDF content was higher under normal irrigation condition than in other treatments. Drought stress and smoke solution applications generally reduced NDF content. In *Özkaynak*, NDF decreased in moderate drought and increased in severe drought. These values generally decreased in smoke solution applications. In smoke solution applications, these values decreased with 1% solution but increased again with 10% solution, in drought conditions (Table 1 and 2).

Table 2. Chemical composition (%) of smoke solution applied in Özkaynak under drought stress

Treatments	PRT	ADF	NDF	Ca	Mg	P	K
C-0%	29.29 c	21.58 b	37.56 ab	1.01 e	0.37 bc	0.46 c	3.48 d
C-1%	30.02 bc	19.60 cd	34.89 c	1.29 cd	0.37 bc	0.46 c	3.86 c
C-10%	35.94 a	23.59 ab	35.54 bc	1.49 a	0.40 a	0.56 a	4.27 b
M-0%	29.91 bc	21.53 b	33.79 cd	1.26 cd	0.31 e	0.48 bc	4.30 b
M-1%	29.81 bc	24.02 a	31.27 d	1.30 c	0.32 d	0.49 bc	4.40 ab
M-10%	31.61 b	19.53 cd	34.62 c	1.35 b	0.35 cd	0.51 b	4.49 a
S-0%	27.45 d	23.84 a	38.08 a	1.30 c	0.35 cd	0.46 c	3.88 c
S-1%	29.52 c	18.48 d	32.71 cd	1.35 b	0.35 cd	0.46 c	3.94 bc
S-10%	29.77 bc	20.32 c	34.89 c	1.47 a	0.38 b	0.47 bc	3.98 bc

Different letters significant differences at $p \leq 0.05$ within one parameter. PRT; crude protein, C; control, M; moderate drought, S; severe drought.

In *Gap pembesi*, mineral substance contents (Ca, Mg, P and K) decreased in parallel with the stress severity under drought conditions. As a result of priming with smoke solutions, it was determined that a 10% dose increased the mineral substance content in both moderate and severe drought. Moreover, M-10% application had the maximum potassium and phosphorus content, while M-10% and S-10% applications had the maximum calcium and magnesium content (Table 1). In *Özkaynak*, mineral matter contents decreased under drought stress. However, these decreases were prevented by smoke solution doses. Additionally, calcium, magnesium and phosphorus reached their maximum in C-10% application. Potassium content had the highest value in the M-5% and M-10% treatments (Table 2).

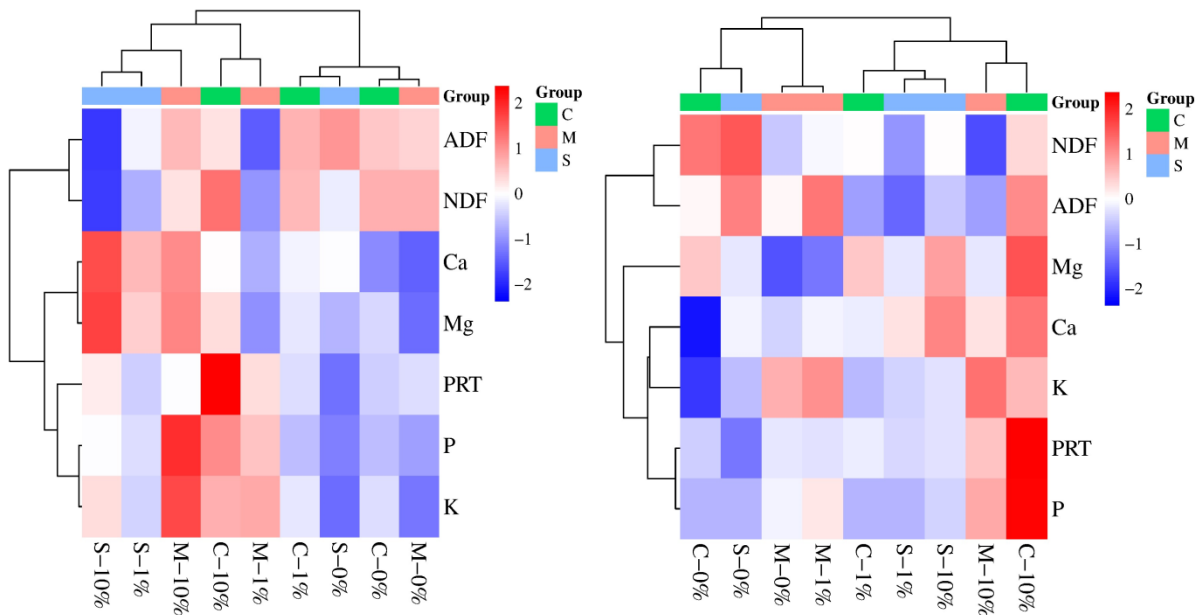


Figure 2. Representation with a heat map of the relationship between responses of smoke solution applied in forage pea under drought stress (*Gap pembesi*: left graphic, *Özkaynak*: right graphic), (The heat map was created with SRplot)

A heat map serves as a means to visually represent and analyze intricate data sets, commonly applied in various fields of data analysis and statistics. In this study, separate heat maps were generated for each genotype to illustrate the relationships between features and applications, as well as to depict the distribution of applications and features within the context of these relationships (Figure 2). The color patterns on the map distinctly highlight the influence of the growing environment and the impact of drought concerning the examined characteristics in pea genotypes. The influence of smoke solutions is evident in both varieties; however, this graph clearly demonstrates that the *Özkaynak* variety exhibits a notably positive response to

the 10% smoke solution. Balloon plot graphics, which classify the effects of features on treatments according to circle size and color tones, are also designed for each type (Figure 3). It is clearly seen that the PRT in both varieties increases in parallel with the smoke solution ratios and is high in the C-10% application. Circle sizes vary depending on the values of the features, and the highest effect was on the protein content in Gap pembersi, while it was on NDF in Özkaynak.

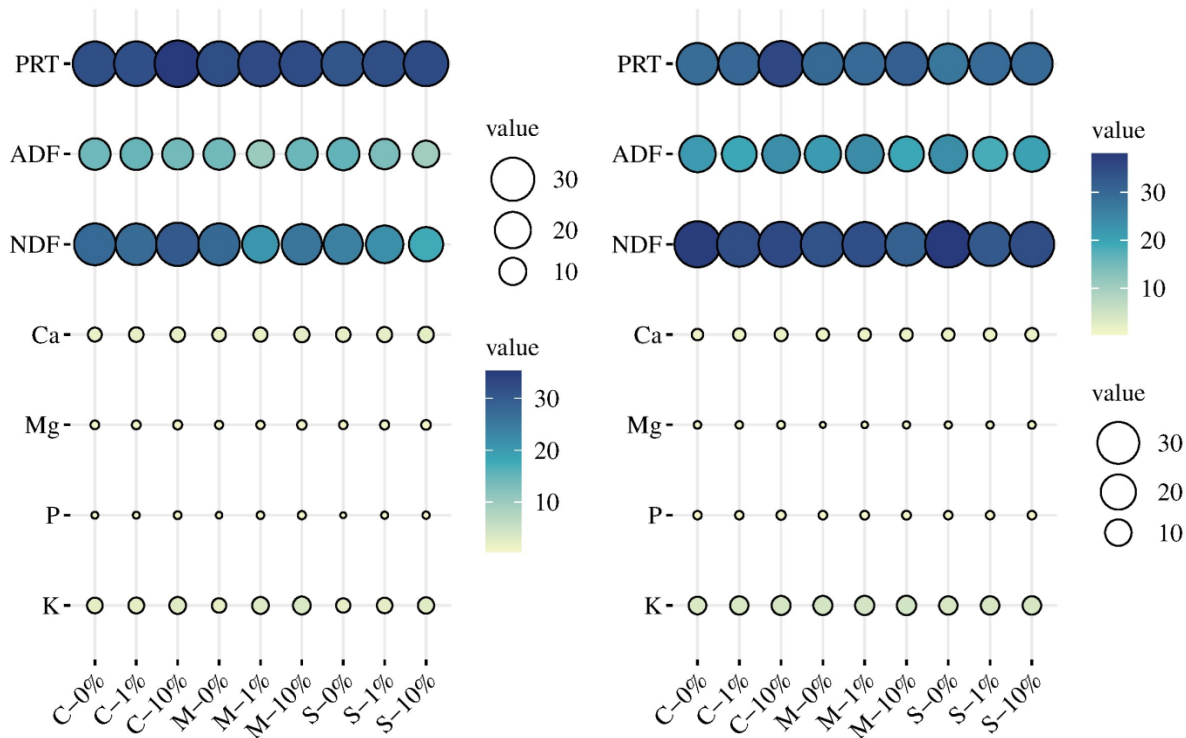


Figure 3. Representation with the balloon plot of the responses of smoke solution applied to forage pea under water deficiency conditions. The mean values of the replicates are shown (*Gap pembersi*: left graphic, *Özkaynak*: right graphic), (The balloon plot was created with SRplot).

In this research, principal component analysis (PCA) was utilized to delve into the intricacies of eight variables and unveil their fundamental characteristics. Additionally, PCA was employed to observe the collective impact of all transactions on graphs for each genotype, using two components. The PCA biplots provided a comprehensive insight into the biochemical responses of forage pea to water deficiency and smoke solutions. Separate PCA biplots were generated for both the population and variety of forage pea. For *Gap pembersi*, the biplot graphs elucidated 69.63% of the cumulative variance, with PC1 and PC2 contributing 68.6% and 14.7% respectively. Conversely, *Özkaynak* exhibited principal components PC1 and PC2, representing 44.9% and 28.3% of the cumulative variance, respectively (Figure 4). Vector contributions in the biplot indicated that the first component was heavily loaded by almost all traits, with a similar trend for the second component, except for SL. Analysis revealed four main groups in both biplots, as evidenced by the heat map. In both varieties, grouping was influenced by drought stress and smoke solutions, although the specifics varied primarily based on the dose of smoke solutions. Priming, particularly with smoke solution, led to a reduction in these fundamental distinctions, causing different treatments to converge into the same groups. Notably, in *Özkaynak*, treatments C-10 and MD-10% were placed in the same group. In *Gap pembersi*, the M-10% treatment occupied positive quadrants of both F1 and F2, whereas in *Özkaynak*, C-10% treatments did. Additionally, it was apparent that the response mechanism to drought stress in *Gap pembersi* was linked with an increase in PRT, K, and P, while in *Özkaynak*, it was associated with an increase in PRT, SL, and ADF.

These findings underscored the significant efficacy of poppy smoke solution in enhancing drought stress tolerance (Figure 2, 3 and 4).

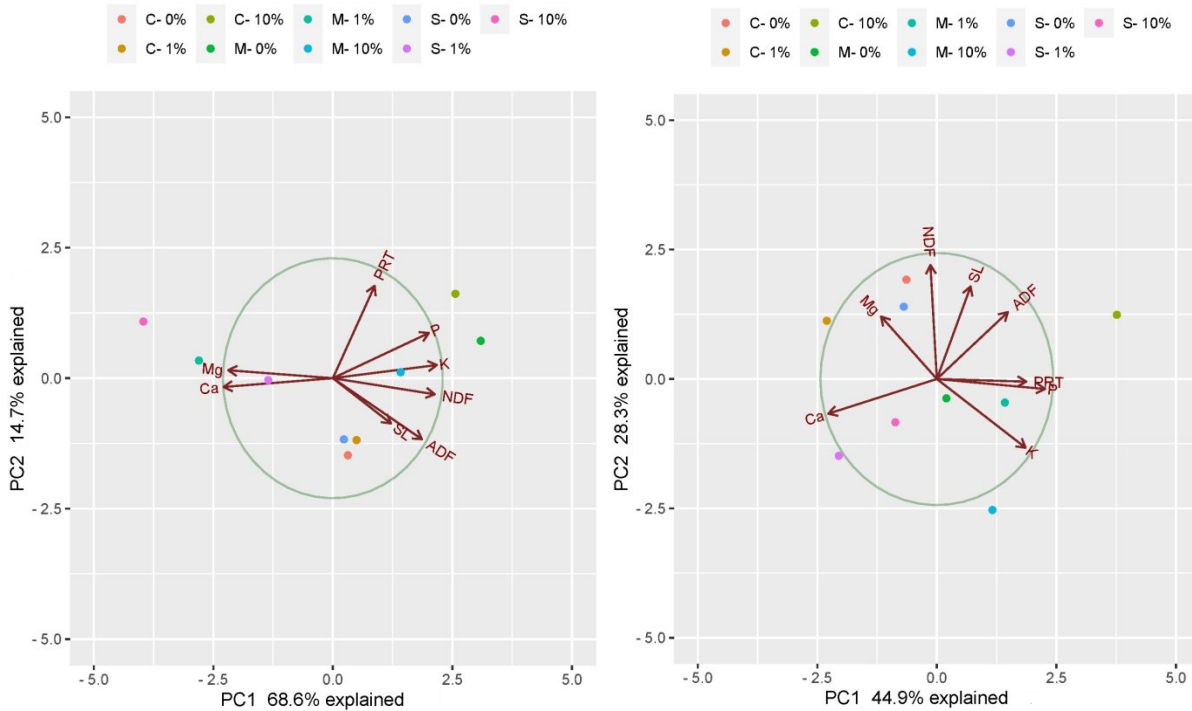


Figure 4. Principal component analysis of the responses of smoke solution applied to forage pea under water deficiency conditions. The mean values of the replicates are shown (*Gap pembesi*: left graphic, *Özkaynak*: right graphic).

4. Discussion

The study examined the physical and chemical reactions to water deficiency in various forage pea varieties, aligning with prior research (Gökmen and Ceyhan 2015; Ceyhan et al. 2012; Reinhardt et al. 2015; Yousofi et al. 2016; Tekin and Ceyhan 2022). The findings suggest that forage pea varieties exhibit sensitivity to unfavorable ion ratios, and osmotic stress, such as drought, impacts cell division. The parameters examined in the study were evaluated as defense mechanisms against drought stress and are very important in terms of animal nutrition. The balance between protein with minerals and fiber components with ADF and NDF values, is crucial for assessing the nutritional quality of forage crops. Livestock performance, including growth rates, milk production, and overall health, can be significantly influenced by the crude protein, minerals and fiber content in their diet. Therefore, understanding these parameters helps farmers and nutritionists make informed decisions about the suitability of forage crops for specific livestock needs. It may involve adjusting feeding practices, supplementing with additional nutrients, or seeking alternative forage sources during periods of drought to maintain the health and productivity of livestock. Instead, it is much more logical and profitable to preserve the existing potential of quality forages, such as forage peas, with practices that will prevent quality loss under drought conditions.

In both drought stress levels were generally negative effects on the nutritional composition of plants, including changes in crude protein, mineral content, and fiber fractions such as ADF and NDF. In moderate drought, there was a partial increase in crude protein concentration as the plant tries to concentrate nutrients in response to water scarcity. However, in other instance, drought stress increase was reducing protein synthesis and result in lower crude protein content. Drought stress in both forage pea was typically leads to

an increase in fiber content, including both ADF and NDF. As the plant experiences water scarcity, it may allocate resources to the production of structural components like cellulose and lignin, resulting in higher fiber concentrations. However, since the results obtained were within the range required for quality hay (NRC, 2001; 25-35% ADF and 40-50%), this increase was considered positive in the study. Water scarcity can limit the uptake of essential minerals (Ca, Mg, from the soil, affecting the overall nutrient profile of the plant. Mineral contents had variable effects on drought stress levels, decrease in especially severe drought, but Ca content increased in both varieties. Additionally, all of the mineral substance contents obtained in the study were found to be above the values (Kidambi et al. 1993; min. 0.8% K, 0.21% P, 0.3% Ca and 0.1% Mg) that should be in quality forage. These similar effects have been determined by many researchers in plants exposed to drought stress (Pei et al. 2010; Chen et al. 2011). The specific responses were vary among plant species and depend on the severity and duration of drought stress (Hu and Schmidhalter 2005). Additionally, some plants may exhibit adaptive responses to drought, such as altering their physiological processes to better withstand water scarcity (Seleiman et al. 20219). However, it is clearly seen in this study that decreased the PRT, Mg, K and P contents with drought stress in forage pea was increased again with poppy-derived smoke solutions. However, the effect of drought and the defense mechanism of smoke solution created differences in forage pea genotypes. In general, the Özkaynak was more resistant to drought and the response to smoke solution was stronger.

Plant-derived smoke solutions, known for their significant impact on germination, root growth and seedling development (Dogrusoz 2022), effectively mitigated the forage pea's response to drought stress (Li et al. 2017). Recent research has identified that the protective effect against drought is due to in strigolactones and karrikins butenolide molecules, present in plant-derived smoke solutions. These components have been shown to positively influence various processes related to plant drought responses, including the regulation of chemical composition (Li et al. 2017; Yang et al. 2020; Zheng et al. 2020). In the context of drought, the application of smoke solutions might help plants overcome stress and improve their resilience. However, the specific effects can vary depending on the plant species, doses and environmental conditions.

5. Conclusions

These results showed that in forage pea, poppy smoke solutions helped them cope with the adverse effects of drought by activating specific physiological and chemical responses. However, the defense mechanism against drought stress of the Özkaynak variety was stronger. It's important to note that the use of smoke solutions in agriculture and ecosystem restoration is an evolving field, and more research is needed to fully understand the mechanisms involved and the potential benefits across different plant species and environments. Therefore, plant-based applications of smoke solutions in drought stress need to be increased. Also, we think that smoke solutions should be evaluated under other biotic and abiotic stress conditions as well as drought stress. Additionally, the practical application of these solutions in agriculture may vary, and considerations such as concentration, timing, and compatibility with other agricultural practices need to be addressed.

Author Contributions: Conceptualization, H.M. and M.CD.; methodology, H.M.; software, H.M.; and M.CD.; formal analysis, H.M.; investigation, H.M.; resources, M.CD.; data curation, H.M.; writing—original draft preparation, H.M. and M.CD.; writing—review and editing, M.CD.; visualization, H.M.; supervision, M.CD. and H.M. funding acquisition, H.M. and M.CD. All authors have read and agreed to the published version of the manuscript.

Funding: This research received no external funding.

Conflicts of Interest: The authors declare no conflict of interest.

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Determination of the Factors Affecting the Department Preferences of Selcuk University Faculty of Agriculture Students

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HIGHLIGHTS

- Increasing the contribution of agriculture to the economic and social development of a country is only possible with agricultural education.
- With the high number of departments and the necessity of the department that individuals choose for their future lives to be suitable for them, it is an important issue that needs to be emphasized.
- Most of student families are farming. This situation can be interpreted as families directing their children to their own profession or wanting them to transfer their own business to their children and keep them going.

Abstract

Agriculture is one of the priorities and very important fields in Türkiye's economic development, and it should be carried out with scientific methods. In today's faculties of agriculture, education is given in different departments and agricultural engineers who can work in various fields of agriculture are trained. For this reason, it is an important issue that the department chosen by the students should be suitable for them and what the department selection is made according to. This study was conducted to find answers to the questions of whether the students' department preferences have changed according to factors such as age, gender, family work, monthly income of the family, number of siblings, family residence, and the order in which they prefer the Faculty of Agriculture. The data in the study were tested with Chi-Square analysis. As a result of the study, the effect of all factors except the number of siblings and the order of preference on the department preference was found to be statistically significant, and it was concluded that the students consider their personality traits, their interest in agricultural activities, the place of their preferred profession in the society and the employment rate of the profession.

Keywords: Faculty of agriculture; department preference; questionnaire; chi-square.

1. Introduction

Agricultural production is a process developed with education, training, and experience in order to meet the needs of humans and animals. Agriculture is an important sector that provides employment to millions of people, produces nutrients necessary for human life, contributes to national income and foreign trade, and

Citation: Taniş AN, İlhan F, Öndeş M, Özbay A, Keskin İ (2024). Determination of the factors affecting the department preferences of Selcuk University faculty of agriculture students. *Selcuk Journal of Agriculture and Food Sciences*, 38(2), 298-308. <https://doi.org/10.15316/SJA.FS.2024.028>

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Received date: 13/02/2024

Accepted date: 24/06/2024

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supports industry in terms of raw material supply and input demand (Cinemre and Kılıç 2002; Boz 2004; Kılıç *et al.* 2020). Increasing the contribution of agriculture to the economic and social development of a country is only possible with agricultural education. The success of the training can be measured by the fact that the training activities first lead to information change and then to behavioral change in farmers (Eryılmaz and Kılıç 2019).

In the context of agriculture and technology, agricultural education plays a crucial role in fostering interest in the agricultural profession, facilitating rural development, and guiding young individuals, particularly those in rural areas, towards agriculture (Soydan 2012; Baydar and Esmet 2022). Every activity carried out in the field of agriculture is of immense importance. One of these activities is the development of scientific methods for agriculture. Many agricultural schools have been opened in Turkey for this purpose (Kadioğlu 2005).

Modern agricultural practices in Türkiye started with the opening of the opening of the Agricultural School on January 10, 1846, at Ayamama Farm in Yeşilköy, Istanbul. The development of agriculture and agricultural schools continued with the opening of Bursa Agricultural School in 1891, which can be considered higher education today, and Istanbul Halkalı Agricultural Schools in 1893. These schools continued their activities until the first years of the Republic of Türkiye. Many of the young people who graduated from Halkalı Agricultural School since 1923 were sent abroad to study and learn different modern research methods. In 1927, a Scientific Committee composed of young people who went to Germany for education and training and educators from Germany analyzed the agricultural situation in our country. The committee, which made many suggestions, also suggested the opening of a modern agricultural school. Thereupon, with a law enacted in 1927, the foundations of agriculture and agricultural schools in higher education were laid (Yılmaz *et al.* 2023).

The Higher Agricultural Institute in Ankara established agriculture and agricultural laboratories shortly after its foundation. This led to the establishment of Türkiye's first modern higher education board. This board started to work on October 30, 1933, and was named Ankara University Faculty of Agriculture with the University Law dated 1946 and the University Supplementary Law enacted in 1948. After that, Ege University in 1955, Atatürk University in 1957, and Çukurova University Faculty of Agriculture in 1967 were opened. In 1975, Selcuk University Faculty of Agriculture opened its doors. Today, the number of faculty of agriculture is 40 (Öcal *et al.* 2022).

In today's agricultural faculties, education is given in different departments, and agricultural engineers who can work in various fields of agriculture are trained. Agricultural education is given in departments such as Agricultural Economics (AE), Agricultural Structures and Irrigation (ASI), Horticulture (H), Crop Protection (CP), Field Crops (FC), Soil Science and Plant Nutrition (SSPN), Agricultural Machinery (AM), Food Engineering (FE), Aquaculture, and Animal Science (AS), which are both different from each other and have common working areas. Given the abundance of departments and the importance of selecting a department that aligns with their future aspirations, it's crucial to underscore this significant issue. Researchers have conducted numerous surveys to determine the factors that influence students' preference for the departments they study in. Several ideas have emerged as a result of these studies. It is stated that the main factors affecting the choice of profession are the influence of social theories or communities such as culture, family structure, school, and friendship relations (Kuzgun 2004). In addition to these, in a study on the factors that general high school students pay attention to when choosing a profession, the factors affecting the choice of profession were determined as ability, interest, values, personality traits, the benefits of the profession (money, prestige, fame, etc.), and the wishes of the family (Kiyak 2006). In addition to these factors, the importance of political, economic, legal, and system-related features (state of the country, economic structure, laws, etc.) and chance factors (health conditions, natural events, etc.) has been emphasized (Korkut Owen 2008). The most important reasons for students to choose a department were the expectation of finding a job after graduation (Öztürk and İlman 2015), the sufficient score for this department (Gezgin 2015), as well as the interest in the field and the suitability of the field with personality traits, which are other factors affecting the choice of the department. In addition, it was observed that the chance factor affects department preference (Owen *et al.* 2012).

In studies on gender, which is another factor affecting the choice of department, it has been determined that female students take into consideration the suitability of the department for their gender and interests and that they are more affected by the ideas of their families when choosing the department than male students. It has been observed that male students take into account the good earnings that the department will bring (Owen et al. 2012). It has been determined that the gender factor does not have a direct effect on the choice of profession and that professional flexibility is important for men and women to give them the opportunity to develop themselves and show their talents (Edwards and Quinter 2011). Clutter (2010) stated that the most powerful factor affecting the choice of profession is the family of the individual. It is seen that the economic status of the family has a decisive effect on the factors affecting department preference. Families from different economic levels may have different expectations according to their economic level. Social classes of families influence the vocational and educational aspirations and interests of their children (Schoon and Parsons 2002). Studies conducted in Turkey show that the occupational choice made according to the socioeconomic level of the family changes. For example, it has been found that those who tend to education faculties have a middle socioeconomic level (Erden 1995; Akbayır 2003). Bahar (2002) reached similar results in her research with the students of the faculties of education and medicine and found that the students of the faculties of economics and administrative sciences come from a higher socioeconomic level.

The aim of this study is to seek answers to questions such as whether it changes according to factors such as age, gender, family job, family monthly income, number of siblings, family's place of residence, and in which order they prefer the Faculty of Agriculture.

2. Materials and Methods

This study was carried out at Selçuk University, Faculty of Agriculture, in the spring semester of the 2021-2022 academic year, with a total of 440 students from 9 departments (Horticulture: 63, Plant Protection: 97, Food Engineering: 26, Field Crops: 39, Agricultural Economics: 25, Agricultural Machinery: 47, Agricultural Structures and Irrigation: 12, Soil Science and Plant Nutrition: 32, Animal Science: 100) participating in the study.

The applied questionnaire consists of two parts. In the first part of the questionnaire, there are total of 7 multiple-choice questions about the information concerning the students, including students' age (<22, 23-25, >26), gender (male, female), family job (farmer, civil servant, worker, businessman), family monthly income (6001-10000, 10001-14000, 14001- 20000, >20001), number of siblings (1, 2-3, 4-5, >6), their family's place of residence (those who reside in Konya Center are abbreviated as KCenter, those who reside in Konya's district are abbreviated as KDistrict, those who reside in a center outside Konya are abbreviated as KOCenter, and those who reside in a district outside Konya are abbreviated as KODistrict) and their department preference order (1, 2-5, 6-10, >10).

In the second part, a total of 6 yes/no questions were asked, such as whether they had prior knowledge about the university and the department, which is thought to have an effect on their choice of the department, their interest in the department they studied and agricultural activities, and the employment rate of the department. The chi-square (χ^2) test was used to assess whether there was a statistically significant difference between the answers given to the survey questions. The control hypothesis was rejected for the χ^2 values calculated for the first part questions, which are less than the 5% significance level and the table value determined according to the degrees of freedom, and the opposite hypothesis was accepted (Keskin et al., 2023). We conclude that a difference exists between the sections concerning the relevant question. We conducted the analysis again, removing the department with the highest Chi-Square value, to determine which department or departments are responsible for the difference. The analysis was continued until the control hypothesis was accepted, i.e., there was no difference between the sections.

3. Results and Discussions

The number of factors affecting the department preferences of the students (age, gender, family job, family monthly income, number of siblings, family's place of residence, and department preference order) according to departments is given in Table 1.

Table 1. Factors affecting students' department preferences and Chi-Square analysis results.

Factors	Departments									General	
	AE	ASI	FC	SSPN	AS	CP	AM	H	FE		
Age	<22	17	10	27	22	57	72	32	53	4	294
	23-25	2	0	10	7	10	22	14	8	0	73
	>26	5	2	2	2	32	3	1	2	7	56
	General	24	12	39	31	99	97	47	63	15	423
	Chi-Square=73.030 (Yates Correction Chi-Square=62.80), DF=16, P<0.01										
Gender	Male	18	5	30	20	67	62	44	36	3	285
	Female	7	7	9	12	33	33	3	27	23	154
	General	25	12	39	32	100	95	47	63	26	439
	Chi-Square=57.345 (Yates Correction Chi-Square=51.63), DF=8, P<0.05										
Family Job	Farmer	11	5	12	14	38	51	12	10	3	156
	Civil Servant	5	1	9	7	29	13	7	20	9	100
	Worker	4	3	8	5	14	12	14	11	7	78
	Tradesman	4	3	10	6	18	21	13	20	7	102
	General	24	12	39	32	99	97	46	61	26	436
Chi-Square=50.593 (Yates Correction Chi-Square=40.97), DF=24, P<0.05											
Family Monthly Income	6001-10000	12	4	24	19	54	67	35	40	19	274
	10001-14000	4	1	4	3	16	8	6	13	5	60
	14001-20000	5	2	6	3	15	10	2	5	1	49
	>20000	4	4	5	7	15	12	3	4	1	55
	General	25	11	39	32	100	97	46	62	26	411
Chi-Square=33.620 (Yates Correction Chi-Square=22.53), DF=24, P>0.05											
Number of Siblings	1	3	4	6	4	17	18	9	18	2	81
	2-3	13	4	22	18	46	54	24	29	17	227
	4-5	2	4	9	6	25	16	10	12	6	90
	>6	4	0	2	3	9	6	3	3	1	31
	General	22	12	39	31	97	94	46	62	26	429
Chi-Square=22.099 (Yates Correction Chi-Square=13.52), DF=24, P>0.05											
Family's Place of Residence	Konya Center	9	8	16	9	31	18	12	14	8	125
	Konya District	8	3	12	10	30	15	5	4	3	90
	KO Center	4	1	7	8	17	15	20	29	11	112
	KO District	4	0	4	5	21	49	8	16	4	111
	General	25	12	39	32	99	97	45	63	26	438
Chi-Square=98.578 (Yates Correction Chi-Square=84.26), DF=24, P<0.05											
Department Preference Order	1	12	5	22	16	39	27	22	31	2	176
	2-5	8	5	11	9	38	35	16	18	13	153
	6-10	4	1	4	3	12	13	5	7	6	55
	>10	1	1	2	4	9	22	4	6	4	53
	General	25	12	39	32	98	97	47	62	25	437
Chi-Square=37.622 (Yates Correction Chi-Square=29.40), DF=24, P<0.05											

When Table 1 is examined, the effects of age ($P>0.01$), gender, family occupation, family residence and department preference order on department preference were found to be statistically significant ($P<0.05$). The monthly income of the family and the number of siblings were not effective in determining the department preference of the students.

The number of factors affecting the department preferences of the students (age, gender, family job, monthly income of the family, number of siblings, place of residence of the family, and department preference order) is given in Figure 1-7 as percentages. The majority of the students are in the <22 age group, followed by the 23-25 age group and the >30 age group, respectively (Figure 1-a). The age factor had an effect on department preference in the Animal Science and Plant Protection departments ($P<0.05$). Many associate degree students come to the Department of Animal Science to complete their undergraduate studies, which explains why the number of students in the >30 age group is higher than in other departments. Öztürk and İlman (2015) found that 24% of the students ($n: 36$) were between the ages of 17-19, 66.7% ($n: 100$) were between the ages of 20-22, and 9.3% ($n: 14$) were between the ages of 23-25. stated that. The age range of the students identified in this study is similar to the study conducted by Öztürk and İlman (2015).

Figure 1-b displays the gender distribution of the students by department. Figure 1-b reveals that the proportion of female students in ASI and FE departments surpasses that of male students in other departments. The reason for the high number of female students in the ASI and FE departments may be that these departments require less physical strength compared to other departments. As a result of the Chi-Square analysis, the effect of gender on department preference is mostly seen in the FE and AM departments ($P<0.05$). As a result of this study, it has been seen that male students give more importance to the personality traits of the department and their interest in agricultural activities than female students. According to Owen et al. (2012), as a result of their study, it is seen that male students consider their interest in the profession more than female students when choosing the department. Conversely, female students place a higher priority on the field's suitability for their gender.

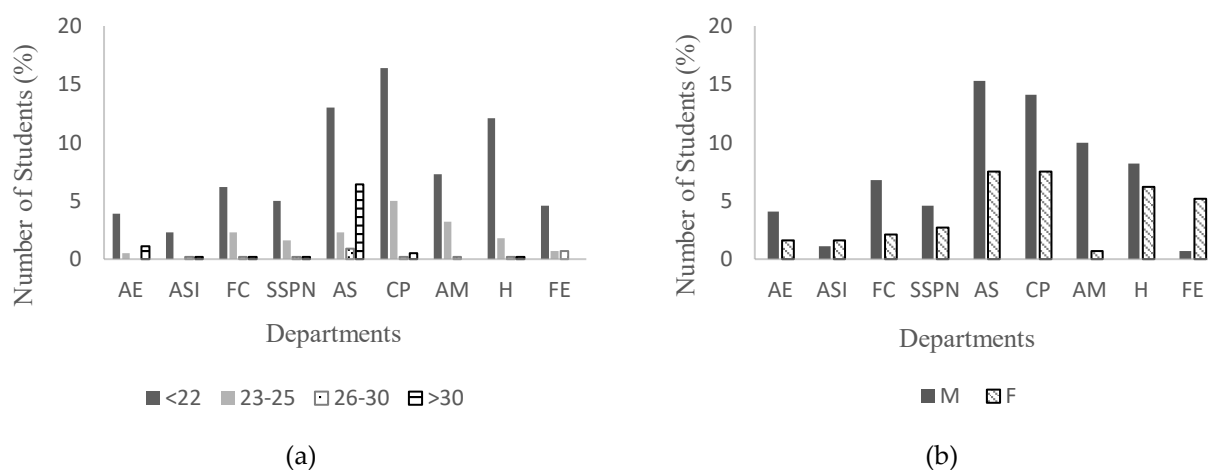


Figure 1. (a) Number of students by age (%); (b) Number of students in departments according to gender (%).

Figure 2-a shows the distribution of student families' work by departments. According to Figure 2-a, it is seen that the majority of student families are farming. Families may interpret this situation as guiding their children towards their own profession or encouraging them to carry on with their own business. While most of the students whose families are workers in the AM department, it was determined that the families of the students in the FE department were mostly civil servants. According to the Chi-Square analysis, the work done by the students' families was most effective in the Department of Plant Protection and Horticulture ($P<0.05$). In their study, Kıyak and Ölçer (2015) stated that some families can direct their children to do professions that they want but cannot do by influencing their children. In this study, a result was reached, as stated by Kıyak and Ölçer (2015).

Figure 2-b displays the distribution of student families' monthly income by department. According to Figure 2-b, it is seen that the monthly income of the most is in the range of 6001–10000. As a result of the Chi-Square Test, the monthly income of the students' families was most effective in the department of ASI ($P<0.05$).

As a result of their study, Kıyak and Ölçer (2015) stated that families with lower monthly income direct their children to jobs that will provide income in a short time. In this study, the fact that the majority has the lowest income level among the options shows the contrast between families directing their children to their own work despite not being able to earn a high income.

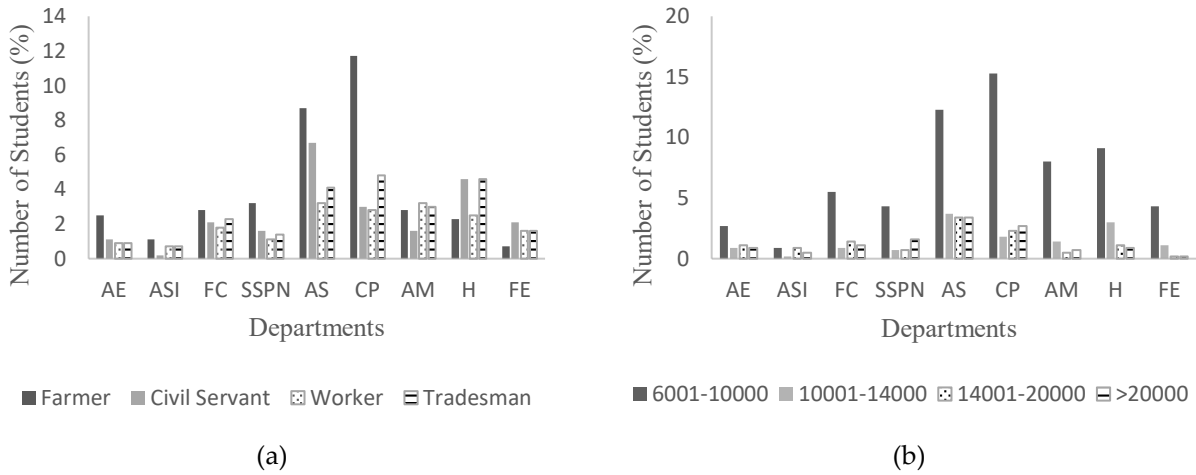


Figure 2. (a) Number of students in departments according to family's job (%); (b) Number of students in departments according to monthly income (%).

Figure 3-a displays the percentage distribution of students' siblings by department. According to Figure 3-a, it is seen that most of the students studying in other departments, except for the ASI department, are in the 2-3 group of siblings. Table 1 shows that the Chi-Square test revealed a statistically insignificant effect of the number of siblings on the students' department preference. This suggests that families prioritize their children's education, and the number of children has no bearing on this.

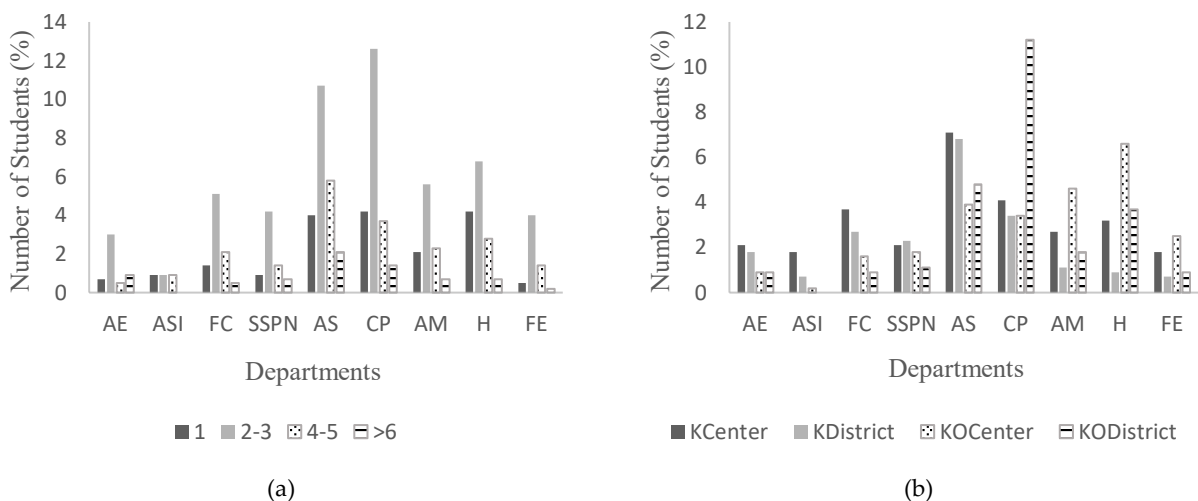


Figure 3. (a) Number of students in departments according to number of siblings (%); (b) Number of students by place of residence (%).

Figure 3-b shows the distribution of the student families according to their places of residence. According to Figure 3-b, it is seen that the residences of the families of the students studying in the AE, ASI, FC, and AS departments are mostly in Konya Center. Students who favor these departments believe

they don't need to travel to other cities to study them. It is seen that the residences of the families of the students studying in the AM, H, and FE departments are in the center outside of Konya. While most of the students in the SSPN department reside in the districts of Konya, it is seen that the students in the CP department mostly reside in the districts outside of Konya. As a result of the Chi-Square test, it was seen that the students' families' places of residence were mostly effective in the departments of H, AM, CP, AS, and ASI ($P < 0.05$).

Figure 4 displays the percentage distribution of the students' preference for the Faculty of Agriculture by departments. According to Figure 4, it is seen that the majority of the students (AE, FC, SSPN, AS, AM, and H) preferred the Faculty of Agriculture in the first place. The Chi-Square test did not reveal a statistically significant order in which the students preferred the Faculty of Agriculture (Table 1).

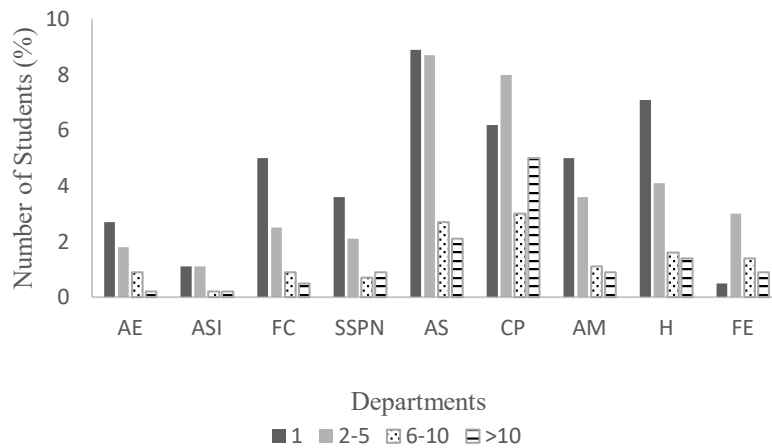


Figure 4. Number of students in departments according to order of preference (%)

The questions given are whether the students have prior knowledge about their university, whether they have any prior knowledge about the department, whether the chosen department is suitable for their personality traits, whether they have any interest in agricultural activities, whether the department you choose takes into account its place in society, and whether the department you choose takes into account the employment rate. Table 2 presents the distribution of the answers based on the departments and the Chi-Square analysis results.

As can be seen in Table 2, according to the Chi-Square analysis made, it was observed that most of the students in all departments had prior knowledge about their universities, the department they chose was suitable for their personality traits, they were interested in agricultural activities, and they took into account their place in society and the employment rate when choosing a department ($P < 0.05$). It has been determined that although the students have prior knowledge about the above subjects, they do not have enough prior knowledge about the department they are studying. Figure 5-a displays the percentage of students who possess prior knowledge about the university.

The distribution of students according to departments and whether they have prior knowledge about their departments is shown in Figure 5-b as a percentage. It is seen that most of the students in all departments have prior knowledge about the department they are studying. These rates are 76.0% in AE, 67.7% in ASI, 84.6% in FC, 75.0% in SSPN, 75.0% in AS, 72.2% in CP, 68.1% in AM, 76.2% in H, and 84.6% in FE.

Figure 6-a presents a percentage distribution of the students' department suitability based on their personal characteristics. As can be seen in Figure 6-a, most of the students in all departments are in line with their personal characteristics.

Table 2. Distribution of students’ answer to questions about university and department according to departments.

Factors	Answers	Departments									General
		AE	ASI	FC	SSPN	AS	CP	AM	H	FE	
Did you have any prior knowledge about Selçuk University?	Yes	20	8	29	25	76	55	31	44	24	312
	No	5	4	10	7	24	42	16	19	2	129
	General	25	12	39	32	100	97	47	63	26	441
	Chi-Square=19.185 (Yates Correction Chi-Square=16.09), DF=8, P<0.05										
Did you have any prior knowledge about your department?	Yes	19	8	33	24	75	70	32	48	22	331
	No	6	4	6	8	25	27	15	15	4	110
	General	25	12	39	32	100	97	47	63	26	441
	Chi-Square=29.769 (Yates Correction Chi-Square=26.31), DF=8										
Is the department you have chosen suitable for your personality traits?	Yes	17	11	32	29	89	73	35	56	24	366
	No	8	1	7	3	11	24	12	7	2	75
	General	25	12	39	32	100	97	47	63	26	441
	Chi-Square=18.205 (Yates Correction Chi-Square=13.75), DF=8, P<0.05										
Did you have any interest in agricultural activities?	Yes	20	10	31	27	85	73	27	53	13	339
	No	5	2	8	4	15	24	20	10	13	101
	General	25	12	39	31	100	97	47	63	26	440
	Chi-Square=28.795 (Yates Correction Chi-Square=24.37), DF=8, P<0.05										
Have you considered the place of your chosen department in society?	Yes	13	11	21	21	60	54	29	38	23	270
	No	12	1	18	11	39	43	18	25	3	170
	General	25	12	39	32	99	97	47	63	26	440
	Chi-Square=16.181 (Yates Correction Chi-Square=12.83), DF=8, P<0.05										
Have you taken into account the employment rate of department you have chosen?	Yes	13	10	24	24	55	45	28	40	20	259
	No	11	2	15	8	45	51	19	23	6	180
	General	24	12	39	32	100	96	47	63	26	439
	Chi-Square=17.140 (Yates Correction Chi-Square=13.84), DF=8, P<0.05										

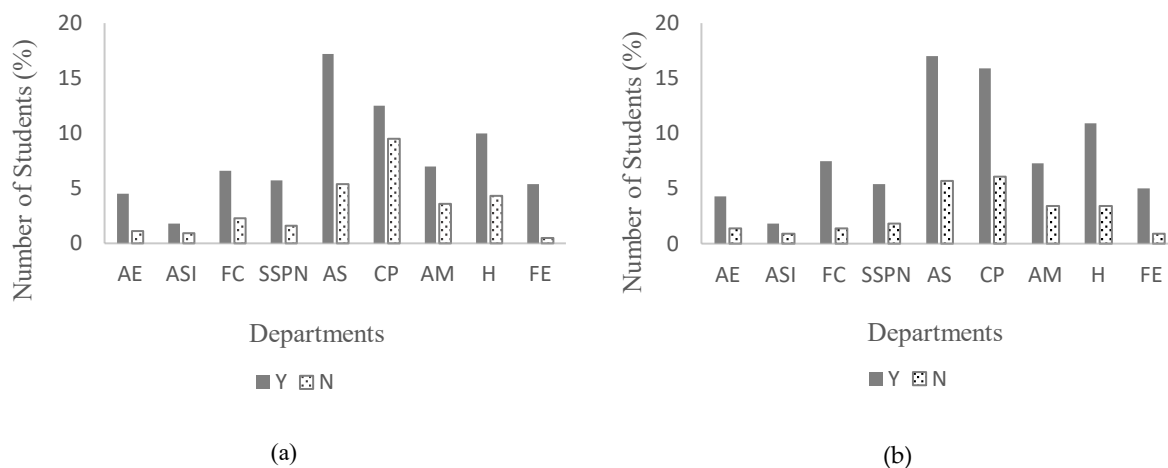


Figure 5. (a) Number of students in departments according to prior knowledge about the university (%); (b) Number of students in departments according to whether they have prior knowledge about the department (%).

The distribution of students according to departments and whether they have an interest in agricultural activities or not is given in Figure 6-b as a percentage. Figure 6-b reveals an interest in agricultural activities among students in other departments, with the exception of ASI. The ASI's field of study, which is less relevant to agricultural activities than other departments in the Faculty of Agriculture, could potentially explain this.

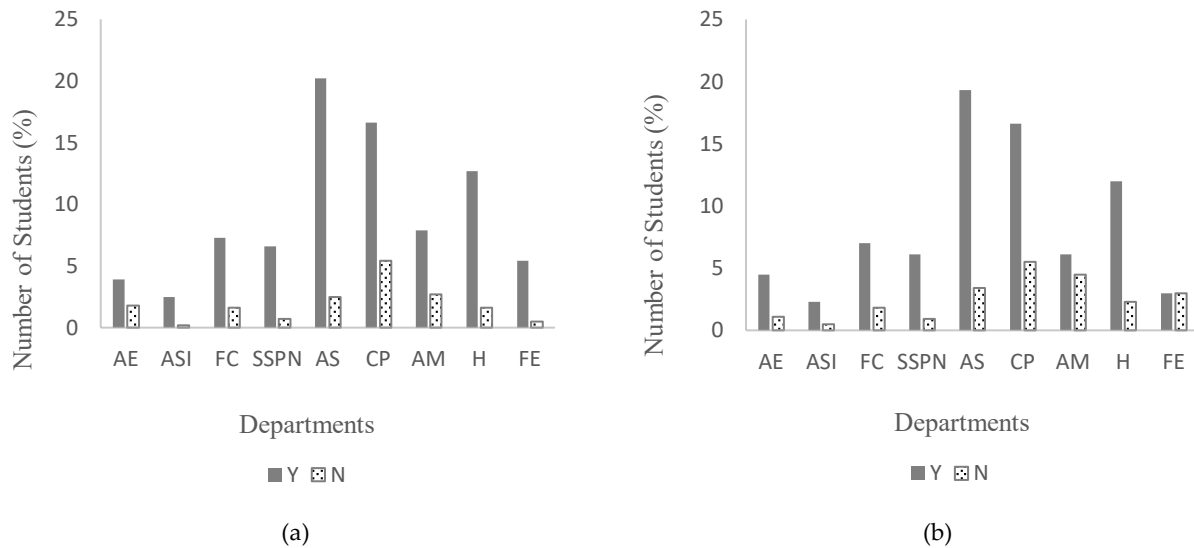


Figure 6. (a) Number of students in departments according to the suitability of the department to their personal characteristics (%); (b) Number of students in departments according to whether they have an interest in agricultural activities (%).

The distribution of the importance of the profession preferred by the students in society according to the calculated percentage values is shown in Figure 7-a. According to Figure 7-a, it is seen that the place of the profession they prefer in society is important for the students in all departments. Since it is well known that having a prestigious career is important for most people, it is expected that students should consider the place of the profession in society when choosing a department.

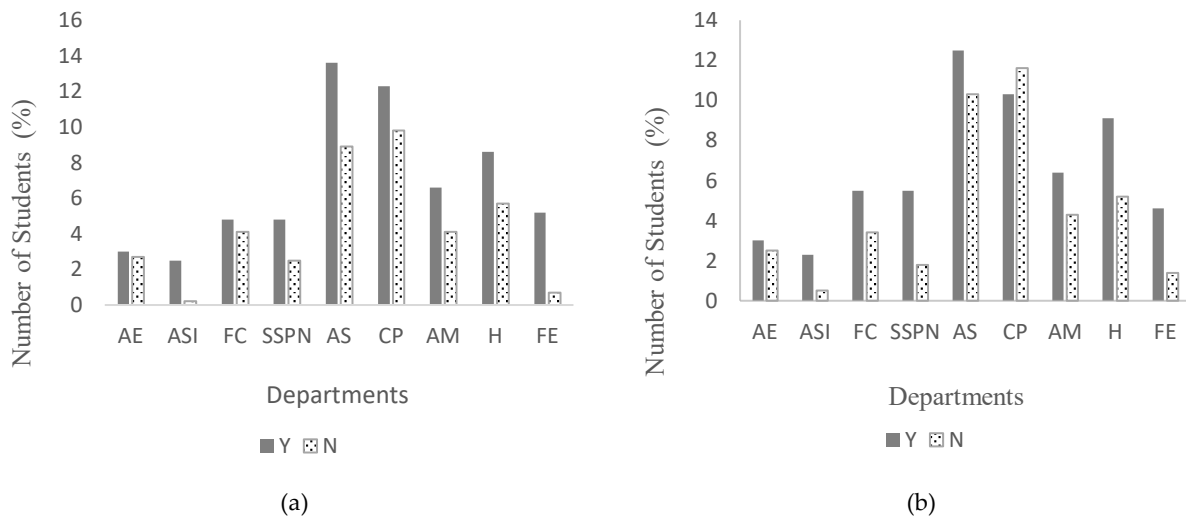


Figure 7. (a) Number of students in departments according to whether they consider the place of the profession in society (%); (b) Number of students in departments according to whether the employment rate is taken into account or not (%).

Figure 7-b shows the distribution by departments based on the calculated percentage values, regardless of whether the students consider the employment rate of their preferred profession. Figure 7-b reveals that students from other departments, apart from the CP department, consider the employment rate of their preferred profession. Most students who prefer the CP department may not consider the employment rate of the profession because they plan to continue their family business or because they prefer a department with an academic career plan.

4. Conclusions

At the end of the study, which was conducted to determine whether the department preferences of Selçuk University Faculty of Agriculture students change according to factors such as age, gender, family job, monthly income of the family, number of siblings, family residence place, and in what order they prefer the Faculty of Agriculture, the number of siblings and the order of preference, We found that all the factors, except for the department preference, had a statistically significant effect. It is seen that most of the students participating in the survey are in the <22 age group. Since the age to start higher education is close to this age group, it is expected that the majority will be in the <22 age group. Unlike other departments, the Department of Animal Science has more than 30 students. We interpret this situation as a result of many associate degree students coming to the Department of Animal Science to complete their undergraduate degrees. In terms of gender, it was determined that the number of male students was higher, except for the ASI and FE departments. It is thought that the ASI and FE departments may have been preferred by female students since they require less physical strength than other departments. As a result of the study, it was seen that male students gave more importance to the personality traits of the department and their interest in agricultural activities than female students. It was determined that the majority of the students in the FE department were civil servants, while the majority of the students in the AM department were workers, and the majority of the students in the FE department were civil servants. Families of students studying in the AE, ASI, FC, and AS departments are located in the Konya Center; families of students studying in the AM, H, and FE departments are located outside of Konya. While most of the students in the SSPN department reside in the districts of Konya, it has been determined that the students in the CP department mostly reside in the county outside of Konya. It was observed that most of the students preferred the Faculty of Agriculture in the first place. Most students across all departments possess prior knowledge about their respective universities and departments. Students across all departments take their personality traits into account when selecting a department. Students in other departments, with the exception of FE, exhibit an interest in agricultural activities. It is a well-known fact that having a prestigious career is important to most people. It has been determined that the students participating in this study attach importance to the place of their preferred profession in society. Students in other departments, with the exception of the CP department, show consideration for the employment rate of their preferred profession. This may be because they are already pursuing their family business.

This study aims to uncover the factors that influence the department and, consequently, the career choices of students at Selçuk University's Faculty of Agriculture. It is thought that the results obtained will be a guide for future studies.

Author Contributions: Conceptualization, İ.K.; methodology, F.İ.; validation, İ.K. and F.İ.; formal analysis, F.İ. and A.N.T.; investigation, A.Ö., M.Ö. and A.N.T.; data curation, A.Ö. and M.Ö.; writing—original draft preparation, A.N.T.; writing—review and editing, A.N.T. and İ.K.; visualization, A.N.T.; supervision, İ.K. and F.İ.; project administration, İ.K. All authors have read and agreed to the published version of the manuscript.

Acknowledgments: This research received no external funding.

Conflicts of Interest: The authors declare no conflict of interest.

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Harmful Insect and Mite Species and Natural Enemies in Strawberry Fields of Halkapınar (Konya) District

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HIGHLIGHTS

- The study was carried out to determine harmful mites strawberry areas in Halkapınar District of Konya Province, Türkiye in 2011.
- One mite species, 19 pest species from 5 orders belonging to 16 families and 8 beneficial insect species from 4 orders belonging to 6 families were determined.
- The most important harmful mite and insects were *Tetranychus urticae* Koch, *Thrips tabaci* Lindeman, *Brachycaudus helichrysi* Kaltenbach and *Chaetosiphon fragaefolii* Cockerell
- The most important natural enemies were *Coccinella septempunctata* L *Scymnus pallipediformis* Günther, *Scymnus rubromaculatus* Goeze and *Scolothrips longicornis*

Abstract

The aim of the study is to observe harmful mites and insect species and their natural enemies being in strawberry areas in Halkapınar District of Konya Province of Türkiye in 2011. Different methods were used to detect pests and useful insect species; picking up insects by using aerial net, examine of leaves, flowers, fruits and offshoots by inspection and lupe, in counting leaf determining of amount of insects and mites for per leaflet and pitfall trap methods. As a result, one mite species, 19 pest species from 5 orders belonging to 16 families and 8 beneficial insect species from 4 orders belonging to 6 families were determined. According to intensity and extensity, the important, harmful insects were *Tetranychus urticae* Koch, *Thrips tabaci* Lindeman, *Brachycaudus helichrysi* Kaltenbach and *Chaetosiphon fragaefolii* Cockerell; and beneficial insects were *Coccinella septempunctata* L *Scymnus pallipediformis* Günther, *Scymnus rubromaculatus* Goeze and *Scolothrips longicornis* Prisner.

Keywords: Halkapınar; harmful mites; harmful insects; natural enemies; strawberry

Citation: Kilinç A, Uysal M, Şahbaz A (2024). Harmful insect and mite species and natural enemies in strawberry fields of Halkapınar (Konya) district. *Selcuk Journal of Agriculture and Food Sciences*, 38(2), 309-325. <https://doi.org/10.15316/SJAfS.2024.029>

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Received date: 12/01/2024

Accepted date: 25/07/2024

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1. Introduction

Strawberry, a berry fruit, is consumed both domestically and exported to other countries. Strawberry plant, which is perennial, herbaceous and evergreen has entered the diet of millions of people around the world with its flavor, vitamin (A, B, C) and mineral substance (calcium, iron, phosphorus). Strawberry production was restricted to only a few countries in the past. However, this situation has changed with breeding studies and breeding new varieties that provide sufficient yield under various environmental conditions (Çakaryıldırım 2004)

The fact that many other fruit types can be found in the market in the months when they are not yet released to the market, and the attractiveness and high vitamin C content have caused this fruit well-known in some country markets and to be sold at high prices. In recent years, it has appealed to a wide range of consumers due to processing or frozen besides and its fresh consumption. It can also be grown in lands on the hillside and mountain villages where agricultural production is limited, such as intermediate agriculture cultivation (Çakaryıldırım 2004).

Strawberry, consumed commonly as a table fruit in the Halkapınar district, is also an alternative product to cherry cultivation. Strawberry cultivation has become widespread in this region in recent years and producers establish new strawberry plantations. Based on interviews with Konya Halkapınar farmers, Halkapınar District Directorate of Food, Agriculture and Forestry technical staff and field observations regarding the current research subject, it was concluded that pesticides were applied unconsciously against mite and insect species in the production (Kılınç 2013). Several studies reported pest and beneficial species in strawberry fields in different regions of Türkiye (Erkılıç et al. 1996; Gençer et al. 2004; Kaplan 2007). However, no study has been carried out on the pests found in strawberry fields in Konya Halkapınar district yet. In accordance with European Union legislation, a minimum amount of pesticide residue is required for directly consumed products. In recent years, some products exported abroad returned to Türkiye for this reason. With the current study, it is aimed to determine the mite and insect species that cause significant economic damage in the strawberry fields in the region and to collect information that will form the basis for the strawberry integrated pest management studies to be carried out in the coming years. In addition, natural enemies of common pests for biological control are also tried to be determined.

2. Materials and Methods

The main material of this study consisted of strawberry fields in Halkapınar district of Konya province, and the mite and pest insect species found in these areas and their natural enemies. In the study, 35 cm diameter standard aerial net, Eppendorf tubes, 70% alcohol, transparent polyethylene bags, loop, ethyl acetate, formaldehyde 37% and 500 ml plastic containers were used.

Sampling was carried out in six different locations in total- one location in Ivriz village, one in Yayıklı village, and four in Çakıllar village, Konya Province Halkapınar District, in 2011.

Observations were carried out between May and October in 2011 to determine the mite and insect species seen in different phenological periods of strawberry and their natural enemies. Specimens were collected four times a month during May-August and twice in September and October.

The aerial net method was used to collect mobile insects during the survey. The leaves, flowers, buds, fruits and shoots of the plant were examined visually and a 1 magnifying glass, and the existing mite and insect species, if any, were brought to the laboratory by taking them into killing bottles or in 70% alcohol.

Strawberry fields in the region are generally in parcels smaller than 1 decare. So, the samplings were done at 10 different points of an area of approximately 1 decare. The aerial net method was used to collect mobile insects. In this method, insects were collected by aerial nets a hundred times at 10 different points in the fields in a diagonal direction to scrape the upper part of the plants. Insects collected aerial nets were killed with ethyl acetate and brought to the laboratory, classified according to their order and families, and fixed for identification after needling and labeling.

In this study, buds, leaves, flowers, fruits and shoots of 25 plants were examined visually and with a loop by walking diagonally in the fields. Three leaves were taken from each of the plants and mixed and followed by taking 20 leaves from this mixture and counted with a magnifying glass, the species belonging to the existing mites, *Aphididae* and *Thripidae* families. In the leaf count, infestation rates and distribution areas of 6 locations in 3 different regions, per leaf of species belonging to mites, *Aphididae* and *Thripidae* families, were determined (Erkiliç et al. 1996). Insect species existed in the region and their population status was determined as a result of this survey.

Pitfall traps were used to detect soil pests (Önder 1979). This method has been used to detect pests dwelling in the soil such as wireworms, Carabids, and Tenebrionids. In the application of this method, 37% formaldehyde and a small amount of detergent were placed in 500 ml wide-mouthed plastic jars, and the insects were caught by burying jars in line with the soil surface. These traps were changed every 2 weeks and the adult insects in them were taken with the help of a gauze and prepared for identification.

3. Results

3.1. Mites and Insect Species Found in Strawberry Fields

In this study, one mite species and 19 harmful insect species from 16 families belonging to 5 orders were determined (Table 1).

Tetranychus urticae Koch, 1836 (Acarina: *Tetranychidae*)

Common name: Two-spotted spider mite

Description: The color of the adults is greenish-yellow, light yellow, or reddish. There is a pair of black spots on the upper part of the body and near the middle. Dorsal setae are spiny-like. Their nymphs are smaller, and their colors are greenish yellow, light yellow (Demirözer 2008).

Distribution: It is found in almost every part of our country (Anonymous 2024a).

Damage and Hosts: Cotton, melon, watermelon, cucumber, pumpkin, okra, pepper, eggplant, bean, peanut, strawberry, blackberry (Anonymous 2024a). This species has been found in all study areas. Two-spotted spider mites cause significant damage by sucking the leaf sap and feeding on the buds that will form the next year's fruit flower. They first produce white, then yellow-brown spots on the leaves they suck on. Later, these spots coalesce and cause the leaf to dry and fall, thus causing a significant loss of product. It was reported that Two-spotted spider mite constitute primary pest group in strawberries, and they damage mesophyll cells in the leaves, causing plants to be stunted, sometimes to death, and yield reduction of about 50% in cases where the population density is high (Kaplan 2007). Also, Kaplan (2007) was noted that in cases where *T. urticae* damage is severe, there would be a decrease in flower and fruit set and a general regression in fruit development in the coming years. Erkiliç et al. (1996), in addition found that *T. urticae* was the most detrimental species in their study in the strawberry fields of Mersin province.

In this study, a total of 249 *T. urticae* individuals were found, 33 in İvriz village, 41 in Yayıklı village and 175 in Çakıllar village strawberry fields.

Thrips tabaci Lindeman, 1889 (Thysanoptera: *Thripidae*)

Common name: Tobacco thrips

Description: The Body of adult females is brown-black, and antennae are 8 segmented. The most distinctive morphological characteristic is the light color of the third antennal segment.

Distribution: Kaplan (2007) reported that this pest has a cosmopolitan distribution and is more or less common in almost every part of our country.

Damage and Hosts: Kaplan (2007) reported that this species is polyphagous, and its adults and larvae break the epidermis of leaves, stems, and fruits of plants, tearing or rasping, absorbing coming juice, and feeding sites on plants acquire a whitish or silvery color.

Table 1. Harmful mite and insect species detected in strawberry fields in Halkapınar district of Konya province and their locations.

Order	Family	Species	Locations		
			Konya-Halkapınar		
			İvriz Village	Yayıklı Village	Çakıllar Village
Acarina	Tetranychidae	<i>Tetranychus urticae</i> Koch,1836	x	x	x
Thysanoptera	Thripidae	<i>Thrips tabaci</i> Lindeman, 1889	x	x	x
Hemiptera	Aphididae	<i>Chaetosiphon (Pentatrachopus) fragaefolii</i> Cockerell,1901	x	x	x
		<i>Brachycaudus helichrysi</i> Kaltenbach, 1843			
		<i>Empoasca decipiens</i> Paoli, 1930	x	x	x
	Cicadellidae	<i>Psammotettix</i> sp.	x	x	x
		<i>Lygaeus equestris</i> Linnaeus, 1758	x	x	x
	Pyrrhocoridae	<i>Pyrrhocoris apterus</i> Linnaeus, 1758	x	x	x
	Pentatomidae	<i>Nezara viridula</i> Linnaeus, 1758		x	x
	Coreidae	<i>Centrocoris variegatus</i> Kolenati, 1845	x	x	x
Orthoptera	Acrididae	<i>Truxalis robusta</i> Uvarov, 1916			x
		<i>Aiolopus strepens</i> Latreille, 1804	x	x	x
	Gryllidae	<i>Gryllus bimaculatus</i> De Geer, 1773	x	x	x
	Gryllotalpidae	<i>Gryllotalpa gryllotalpa</i> Linnaeus, 1758	x		
Coleoptera	Curculionidae	<i>Polydrusus</i> sp.	x	x	x
	Scarabaeidae	<i>Epicometis (Tropinota) hirta</i> Poda, 1761	x	x	x
	Chrysomelidae	<i>Chrysolina orientalis</i> Olivier, 1807	x		x
	Buprestidae	<i>Acmaeodera pilosellae</i> Bonelli, 1842		x	x
	Carabidae	<i>Harpalus (Pseudoophonus) rufipes</i> De Geer, 1774	x	x	x
Dermaptera	Forficulidae	<i>Forficula auricularia</i> Linnaeus, 1758	x	x	x

As a result of the study, a total of 196 individuals of the species were identified, 33 in İvriz village, 32 in Yayıklı village, and 131 in Çakıllar village strawberry fields.

Chaetosiphon (Pentatrachopus) fragaefolii Cockerell, 1901 (Hemiptera: *Aphididae*)

Common name: Strawberry aphid

Description: Wingless individuals are small, oval-shaped elongated, transparent yellowish white to light greenish yellow. Body length is 0.9-1.8 mm (Kocadal 2006).

In winged individuals, the head and thorax are black, the abdomen is very pale greenish-white, and there is a brownish-black pattern on the dorsal of the abdomen. Body length is 1.3-1.8 mm (Kocadal 2006).

Distribution: This species is native to North America and has spread throughout world (Kocadal 2006).

Hosts: *Fragaria* spp, *F. vesca*, *Potentilla* spp. and *P. anserina* (Kocadal 2006).

Kovanci et al. (2003) determined 3 aphid species in the strawberry fields of Bursa province. One of these species is *Chaetosiphon (Pentatrachopus) fragaefolii*. The other two species were identified as *Aphis forbesi* Weed and *Aulocorthum solani* Kaltenbach.

In this study, this species was detected on strawberry plants.

The aphid species *Brachycaudus helichrysi* and *Chaetosiphon (Pentatrachopus) fragaefolii* were detected and, a total of 230 individuals were identified, 34 in İvriz village, 37 in Yayıklı village and 159 in Çakıllar village strawberry fields.

Brachycaudus helichrysi Kaltenbach, 1843 (Hemiptera: *Aphididae*)

Common name: Leaf-curling plum aphid

Description: Wingless viviparous females are oval-shaped, yellowish green or yellowish brown in color and covered with a white waxy-powdered substance on the abdomen. Antennae are short and half the length

of the body. The -head, legs, and cornicles are black. Sometimes a black spot can be found at the end of the abdomen. body length is between 1.5 and 2.0 mm (Kocadal 2006).

The winged forms are long and oval in shape, the body is covered with a white waxy substance, the head and thorax are black, the abdomen and the cauda are green or greenish yellow, and the cornicles are brown. Body length varies between 1.3-1.8 mm (Kocadal 2006).

Distribution: It is distributed in Central and West Asian countries, Italy, France, the southern region of Switzerland, Spain, Israel, Portugal, Egypt, Lebanon and is also found in Türkiye (Kocadal 2006).

Hosts: *Agave* spp, *Achillea santolina*, *Aciphylla squarrosa*, *Albizzia lophantha*, *Anthemis nobilis*, *Apium oraveolens*, *Artemisia* sp, *Aster* sp, *Borago officinalis*, *Capsella bursa-pastoris*, *Carthali* spp. *cyaneus*, *Chrysanthemum* spp, *Gerbera* spp, *Gaillardia pulchella*, *Gnaphalium* spp, *Helianthus annuus*, *Matricaria parthenoides*, *Myosotis* spp, *Pallenis spinosa*, *Pyrus malus*, *Russelia juncea*, *Solanum tuberosum*, *Venidium venidium*. reported (Kocadal 2006).

Kovanci et al. (2003) reported 3 aphid species, except *Brachycaudus helichrysi*.

The aphid species *Brachycaudus helichrysi* and *Chaetosiphon (Pentatrachopus) fragaefolii* were found and, a total of 230 individuals were identified, 34 in İvriz village, 37 in Yayıklı village and 159 in Çakıllar village strawberry fields.

Empoasca decipiens Paoli, 1930 (Hemiptera: Cicadellidae)

Common name: Green leafhopper

Description: The aedeagus is simple, the ventral end of the pygopheric lobe has no protrusion, it tapers at the end, the ventral extension of the anal tube is directed posteriorly, there is an angular protuberance in the basal part, the pregenital sternite is rectangular in the female, the posterior edge is slightly wavy (Güçlü and Özbek 1994).

Its body color is completely green and can turn yellow or orange after death. The body length is 3.20 (3.00-3.35) mm for males and 3.55 (3.30-3.70) mm for females (Güçlü and Özbek 1994).

Distribution: The species have been reported from Afghanistan, Germany, Austria, Bulgaria, Czech Republic, Morocco, France, Netherlands, Iraq, England, Iran, Spain, Israel, Switzerland, Italy, Cyprus, Libya, Lebanon, Egypt, Pakistan, Poland, Romania, Russia, Tunisia, Türkiye, Jordan and Greece (Kaplan 2007).

It is found in all regions of Türkiye except the Black Sea Region and reported from Adana, Antalya, Aydın, Balıkesir, Çanakkale, Denizli, Erzurum, Eskişehir, Hatay, İzmir, Kahramanmaraş, Manisa, Mersin and Muğla provinces (Kaplan 2007).

Damage and Hosts: *Cicadellidae* species cause damage by sucking plant sap with their stinging-sucking mouth parts. They transmit plant pathogens to the plant with the effect of the toxic substances they give to the plant body during sucking. The damage caused by *Cicadellidae* species is firstly yellow or white spots seen around the sucking area due to the loss of chlorophyll, and the leaves are covered with these small white dots. Later, the white spots turn brown. Since the nutrition is mostly along the veins, the leaves gradually shrink and dry along the midrib (Kaplan 2007).

Allium spp, *Amaranthus* spp, *Avena* spp, *Capsicum* spp, *Castanea* spp, *Chenopodium* sp, *Citrus* spp, *Cyperus* spp, *Datura* spp, *Daucus* spp, *Granium* spp, *Juglans* spp, *Malva* spp, *Mercurialis* spp, *Sonchus* spp, *Xanthium* spp, *Vicia* spp, *Beta vulgaris* L, *B. vulgaris* var *rapa*, *Cannabis sativa* L, *Citrullus lanatus* L, *Crataegus oxyacantha* L, *Cucumis sativus* L, *Moschata lam*, *C. pepo* L, *Cynodon dactylon* L, *Ficus carica* L, *Glycine max* L, *Glycyrrhiza glabra* L, *Gossypium hirsutum* L, *Helianthus annuus* L, *Hibiscus esculentus* L, *Lactuca aculeata* L, *Malus domestica* L, *Medicago sativa* L, *Mentha piperita* L, *Nicotiana tabacum* L, *Olea europaea* L, *Oryza sativa* L, *Petroselinum sativum* Hoff., *Phaseolus vulgaris* L, *Pimpinella anisum* L, *Pisum sativum* L., *Polygonum aviculare* L, *Portulaca oleracea* L, *Prosopis stephaniana* Bieb, *Prunus amygdalus Dulcis*, *P. armeniaca* L, *P. avium* L, *P. domestica* L, *P. persica* L, *Raphanus raphanistrum* F, *R. sativus* L, *Ricinus communis* L, *Sesamum indicum* L, *Setaria glauca* L, *Solanum lycopersicum* L, *S. nigrum* L, *S. melongena* L, *S. tuberosum* L, *Sorghum halepense* L, *Spinacia oleracea* L, *Trifolium repens* L, *Vicia faba*, *V. sativa* L, *Vigna unguiculata* L, *Vitis vinifera* L. and *Zea mays* L. plants (Kaplan 2007).

In the current study, a total of 68 adults were collected from 6 in İvriz village, 10 in Yayıklı village and 52 in Çakıllar village strawberry fields.

Psammotettix sp. (Hemiptera: Cicadellidae)

A total of 20 adults were collected from 5 in İvriz village, 3 in Yayıklı village and 12 in Çakıllar village strawberry fields.

Lygaeus equestris Linnaeus, 1758 (Hemiptera: Lygaeidae)

Common name: Black-and-red-bug

Description: The bodies of adults are bright red or reddish orange. The antennae are four-segmented and black. There is a bright white spot in the form of a dot on the tip of the forewings. The legs are completely black (Demirözer 2008).

Distribution: Species is widespread in the Palearctic regions of the world. It is common almost everywhere in Türkiye (Kaplan 2007).

Damage and Hosts: the species feeds on plants and plant seeds, usually on the soil surface, under stones and debris, and on low plants. Although it has a wide host range, it has not economic importance (Kaplan 2007).

It is reported that feeding of *L. equestris* on sunflowers in Hungary in 2001 caused damage by increasing of 2.5% in the linoleic content of the grain (Kaplan 2007).

In Türkiye, *L. equestris* was recorded among the harmful species in pistachios in Şanlıurfa (Yanık and Yücel, 2001).

Gencer et al. (2004) determined 8 *L. equestris* individuals in their study conducted in the strawberry fields of Bursa province. Detecting this species on *Rubus* and *Rosa*, Gençer et al. (2004) reported that this species was *Fragaria* spp. It has been stated that it can also be found on the species, but it will not cause any economic damage in the study.

In this study, a total of 22 adults were collected from 4 in the village of İvriz, 2 in the village of Yayıklı and 12 in the strawberry fields of Çakıllar Village.

Pyrrhocoris apterus Linnaeus, 1758 (Hemiptera: Pyrrhocoridae)

Common name: Fire bug

Description: The body is oval, the dorsal has red and black markings, and the antennae and legs are black. The lower part of the abdomen is black and light-colored towards the edge. Its wings are short. There are no spines in the middle femur. Its body length is 7-11 mm (Lodos 1978).

Distribution: It can be found in almost every part of the Palearctic region including the Oriental sub-region and in North America (Lodos 1978).

It is a common species in almost every region of Türkiye, (Lodos 1978).

Damage and Hosts: The main host of the species is hibiscus. However, it can be sometimes seen in okra and some other *Malvaceae* plants. Adults and nymphs especially feed on the seeds of hibiscus. The damage is not significant.

Gencer et al. (2004) detected 2 *Pyrrhocoris apterus* L. in a strawberry fields study of Bursa province and stated that this species does not feed on strawberries.

Current study reported a total of 20 adults from 2 in İvriz village, 2 in Yayıklı village and 16 in Çakıllar village strawberry fields.

Nezara viridula Linnaeus, 1758 (Hemiptera: Pentatomidae)

Common name: Southern green stink bug

Description: Its general color is green, and in some individuals, the edges of the head and pronotum and the connexivum may be yellowish. Adults that emerge in autumn to spend winter are brownish in color. The body is flat and broad; It is covered with small, dense spots. There are 3 or 5 whitish calli at the base of the scutellum. The body length is 12-15 mm (Öncül 2006).

Distribution: It is a cosmopolitan species that is more or less common in almost all parts of Türkiye (Öncül 2006).

Damage and Hosts: It has economically importance species. It is common in agricultural areas, scrubs and meadows. It is highly abundant species. It feeds on a wide variety of vegetables, especially on tomatoes, peppers, beans, grasses (particularly corn, millet, rice), hemp, cotton, soybeans, sesame, clover, tobacco, hazel, citrus fruits, other fruit trees, and many wild and cultivated plants (Öncül 2006).

Kaplan (2007) detected this species in Elazığ strawberry fields, but did not mention any damage to strawberries.

In total of 20 adults were collected, 4 in the Yayıklı Village and 16 in the Çakıllar Village strawberry fields.

Centrocoris variegatus Kolenati, 1845 (Hemiptera: Coreidae)

Common name: Leaf-footed bug

Description: It is a species with a long oval body, a hard integument, dark brown color, yellowish spotted and patterned there are many spines on the sides of the pronotum. The proboscis reaches the middle coxa. It has a triangular projection on either side of the base of the pronotum, extending to the middle of the scutellum. The tip of the scutellum is convex, with a long carina in the middle. Femur and tibiae black. Its body length is 10-12 mm (Öncül 2006).

Distribution: It has been found in Ankara, Aydın, Balıkesir, Bursa, Denizli, İzmir, Manisa and Muğla provinces (Önder et al. 2006).

In the world, it has a presence in the Mediterranean, Caucasus and Cyprus (Önder et al. 2006).

Damage and Hosts: This species is not economically importance. It spreads in agricultural areas, scrubs and meadows. It is a rare species and feeds on plant (Önder et al. 2006). It occurs in plants of *Chenopodiaceae* family, particularly in cultivated and wild forms. There have also been reports of damage to sugar beets and spinach grown for seed (Lodos 1986).

In total of 13 adults were collected, 2 in the village of İvriz, 1 in the village of Yayıklı and 10 in the strawberry fields of Çakıllar Village.

Truxalis robusta Uvarov, 1916 (Orthoptera: Acrididae)

Description: The body length is 15-25 mm. The head is conical and elongated, and the muzzle is well developed with back wings (Sevgili et al. 2012).

Distribution: It is found in Iran, Russia and Azerbaijan.

It has been detected in Antalya, Kahramanmaraş, Gaziantep, Malatya, Amasya, Hatay, Elazığ, Van and Erzincan in Türkiye (Sevgili et al. 2012).

Damage and Hosts: It has been found in alfalfa, cotton, melon and cucurbits (Abıvardı 1965).

In a current study, one adult was identified in the strawberry fields of Çakıllar Village.

Aiolopus strepens Latreille, 1804 (Orthoptera: Acrididae)

Common name: Broad green-winged grasshopper

Description: The pronotum, 18-28 mm long, is slightly convex anteriorly, posteriorly triangular, central carina is prominent, the posterior femur is 3 times longer than the width, the inner surface is slightly reddish with two black spots, and the posterior tibia is red (Vidal 2000).

Distribution: It has been detected in Southern Europe, North Africa, North Asian Mountain ranges, plateaus and basins (Vidal 2000).

It has been found in the provinces of Adıyaman, İzmir, Diyarbakır, Samsun, Bursa and Tokat in Türkiye (Önder et al. 1999).

Hosts: It has been detected in bushes, false bromine, savanna grasslands, stony riverbanks, cultivated areas, riverine grasslands and grain boundaries (Vidal 2000).

Kovanci et al. (2003) found 12 individuals of *Aiolopus strepens* in the strawberry fields of Bursa province, but reported that there was no significant damage to the strawberry.

As a result, a total of 17 adults were found, 5 in İvriz village, 3 in Yayıklı village and 9 in Çakıllar village strawberry fields.

Gryllus bimaculatus De Geer, 1773 (Orthoptera: *Gryllidae*)

Common name: Two-spotted cricket

Description: The pronotum is almost equally wide anteriorly and posteriorly; body usually dark black, base of tegmina with yellow spots on both sides, these spots rarely take the form of a band, ventral of posterior femur red, tegmina black, sometimes brown; hind wings longer than tegmina; the body is 21-25 mm in males, 25-29 mm in females, and the ovipositor is 11-14 mm long (Özbek and Hayat 2003).

Distribution: It is a common species in the west and south part of Türkiye (Özbek and Hayat 2003).

Damage and Hosts: Causes damage to maize, rice, wheat, tobacco, cotton, cucurbits and some other plants. It gnaws on the cobs of the maize during the milk production period. They eat the stems and leaves of rice and wheat plants. It is also reported that they eat the newly planted seeds (Özbek and Hayat 2003).

As a result of the study, a total of 28 adults were collected, 2 in İvriz village, 6 in Yayıklı village and 20 in Çakıllar village strawberry fields.

Gryllotalpa gryllotalpa Linnaeus, 1758 (Orthoptera: *Gryllotalpidae*)

Common name: European mole cricket

Description: The general color of this species is gray brown, dark brown or reddish brown, the body is covered with short velvety hairs, the lower part of the body is yellowish, the pronotum is large and well developed, 4-5 spacing on the inner and upper part of the posterior tibiae. There are teeth or spines, the front legs are digging, the upper wings are short, the lower wings are well developed, the body length is between 35.0-50.0 mm, and sometimes larger individuals are encountered. The eggs are 2.5-3.0 mm in length and oval, first yellowish and later darker. Nymphs are very similar to adults except for the wings and size (Sönmezyıldız 2006).

Distribution: It has been reported spreading from the whole of Europe, Russia, West Asia, Java, China, Australia, North Africa, Somalia, Abyssinia, Israel, Iran, Iraq, Syria, Afghanistan, Pakistan to the Equator (Sönmezyıldız 2006).

This species is found in varying densities in almost every region of Türkiye

Damage and Hosts: *G. gryllotalpa*, an omnivorous species, eats some insects and worms living in the soil, but feeds mainly on cotton, tea, sugar beet, grass, potatoes, rice, carrots, tobacco, cucurbits, citrus fruits, olives, vines, various vegetables seedlings etc. Adults and nymphs cut the roots of the plants they come across while opening a gallery in the soil and hollowing out the tubers (Sönmezyıldız 2006).

Kovanci et al. (2003) detected only 3 *G. gryllotalpa* in their study in the strawberry fields of Bursa province.

One adult was found in İvriz village in this study.

Polydrusus sp. (Coleoptera: *Curculionidae*)

Damage and Hosts: Kovanci et al. (2004) reported 8 *Polydrusus* species in a study conducted in the strawberry fields of Bursa province. Species belonging to this genus are polyphagous, larvae feed on the roots of cultivated plants, and the adults feed on the leaves of various plants.

A total of 39 adults were collected from 6 in the village of İvriz, 9 in the village of Yayıklı and 24 in the strawberry fields of Çakıllar Village.

Epicometis (Tropinota) hirta Poda, 1761 (Coleoptera: *Scarabaeidae*)

Common name: Apple blossom beetle

Description: Adults are matte black. There are frequent and long yellow hairs on the body. There are white spots on the elytra (Demirözer 2008).

Distribution: This species spreads over a widespread from England to the Middle East and Central Asia, and can be found in almost every part of our country (Kaplan 2007).

Damage and Hosts: they generally cause damage by eating the flowers of various fruit trees, roses, peas, broad beans and other plants in the fields and gardens from the beginning of April (Kaplan 2007).

A total of 5 adults were found in current study, 1 in İvriz village, 2 in Yayıklı village and 2 in Çakıllar village strawberry fields.

Chrysolina orientalis Olivier, 1807 (Coleoptera: Chrysomelidae)

Description: Body length is 3-5 mm. Antennae are long and reddish-brown. The elytra is metallic green with numerous red-brown pits.

Distribution: It is reported from Hatay, Osmaniye, Amasya, Konya, Istanbul, Bilecik, Bursa and Afyon in our country (Özdikmen and Aslan 2009).

it can also be found in Syria, Israel, Cyprus, Iran, Iraq and Tunisia (Aslan et al. 2003).

Damage and Hosts: Field survey shave shown that it causes damage on strawberry leaves and Amaranthus spp. making holes in the leaves.

In the current study, a total of 6 adults were found, 2 in İvriz village and 4 in Çakıllar village strawberry fields.

Acmaeodera pilosellae Bonelli, 1842 (Coleoptera: Buprestidae)

Description: The body length of this species, belonging to the *Buprestidae* family, is 3-8 mm. This insect has a bright coppery bronze color and often has gray hairs on the sides of the body. The head is convex and the elytra is light yellow with black spots on the elytra (Thery 1942).

Distribution: It has been only found in İçel (Erdemli) in Türkiye (Tezcan 1995).

It has a global distribution and found in Greece, Macedonia, Bulgaria, Serbia, Montenegro, Austria, Iran, Albania, Czech Republic, Azerbaijan, France, Italy, Switzerland and Turkmenistan in the world (Anonymous 2024b).

Damage and Hosts: It is a polyphagous pest and generally prefers plants of the *Fagacea* family. Adults *Hieracium* sp, *Taraxacum* sp, *Convolvulus* sp and *Helianthemum* sp. It has been reported that it was found on (Thery 1942).

Prior to this study, this species had not reported in Türkiye, but 9 adults were found, and no damage was observed, 7 in the Yayıklı Village and 2 in the Çakıllar Village strawberry fields.

Harpalus (Pseudoophonus) rufipes De Geer, 1774 (Coleoptera: Carabidae)

Common name: Strawberry seed beetle

Description: The body length of the adult is 14-17 mm. Body color is matte black. The elytra is covered with yellowish hairs. Antennae and legs red-brown. The legs are covered with red-brown hairs (Anonymous2013b).

Distribution: In our country; Adana, Antalya, Ardahan, Artvin, Bartin, Bingol, Bursa, Cankiri, Diyarbakir, Erzincan, Erzurum, Giresun, Gumushane, Igdir, Isparta, Icel, Izmir, Kahramanmaras, Karaman, Kars, Konya, Kütahya, Malatya, Mugla, Tokat, Trabzon, Osmaniye, Yalova (Kesdek and Yıldırım 2003).

In the world; Kazakhstan, Latvia, Liechtenstein, Lithuania, Luxembourg, Malta, Macedonia, Moldova, Netherlands, Norway, Poland, Portugal, Romania, Slovenia, Spain, Russia, Sweden, Switzerland, Ukraine, Serbia, Montenegro, Algeria, Egypt, Morocco, Tunisia, it has been reported to be found in Afghanistan, Cyprus, Russia, Iran, Iraq, Kyrgyzstan, Tajikistan, Turkmenistan, Uzbekistan, Russia and China (Avgın 2006).

Damage and Hosts:

Cockscomb, legumes, cereals, raspberries and strawberries are the host of this species. Adults feed on strawberry seeds close to the soil and the flesh of the strawberry fruit (Anonymous 2013b).

In this study a total of 79 adults were found, 23 in İvriz village, 3 in Yayıklı village and 53 in Çakıllar village strawberry fields.

Forficula auricularia Linnaeus, 1758 (Dermaptera: Forficulidae)

Common name: Earwig

Description: They are bright brown-colored insects that are easily recognizable due to their short wings and pincer-shaped cerci at the end of the abdomen. The claws of females are straight, not curved like those of males. The forelimbs are very small, leathery and veinless. The hind wings are yellowish, with radial veins and fan-like. Their legs are reddish brown (Kansu 1999).

Distribution: It is seen in Türkiye, Bulgaria, USA, Greece and Europe (Sönmezyıldız 2006).

Damage and Hosts: Adults and nymphs are omnivores and feeds on the leaves, flowers and fruits of many plants. They cause damage, especially by eating the flowers of ornamental plants. They seek for food in the dark. They climb bushes and trees. They eat almost everything. (Sönmezyıldız 2006).

Forficula auricularia species cause extensive damage to forest trees and shrubs, ornamentals, vegetables and fruit trees. Damage is seen on almost all parts of the trees. It causes considerable damage to ornamental plants, especially dahlias, chrysanthemums, roses and carnations (Sönmezyıldız 2006). This insect eats small, soft-bodied insect other than plants. It especially feeds of aphids and small caterpillars. As a result of the study, a total of 32 adults were collected, 5 in İvriz village, 4 in Yayıklı village and 23 in Çakıllar village strawberry fields.

3.2. Description, habitat and distribution of natural enemies in strawberry fields

In the study conducted in 2011 in strawberry fields in Halkapınar district of Konya province, 8 beneficial insect species from 6 families belonging to 4 orders were identified.

Coccinella septempunctata Linnaeus, 1758 (Coleoptera: Coccinellidae)

Common name: Seven-spot ladybird

Description: It is a well-known species with an oval shape, 6-8 mm in length. The head and pronotum are black, with two small yellow spots on the head and two quadrangular yellow spots on the anterior corner of the pronotum. The elytra are red in color and has seven black spots on it. The right and left sides of the scutellum are lighter in color than the elytra (Uygun 1981).

Habitat: This species is an important enemy of aphids. It is very common on low and tall plants in gardens, fields, meadows, pastures and woods (Portakaldalı 2008).

In this study, a total of 5 individuals were identified, 1 in the Yayıklı Village and 4 in the Çakıllar Village strawberry fields.

Distribution: It is a widespread species in the Palearctic region. Uygun (1981) found this species almost every part of Türkiye.

Scymnus rubromaculatus Goeze, 1778 (Coleoptera: Coccinellidae)

Common name: Dusky ladybird

Description: It is a broadly oval species with a size of 2x1.8 mm. Elytra, pronotum and head completely black in females; In males, most of the pronotum and the middle back of the head are lighter in color (Uygun 1981).

Habitat: Yiğit and Uygun (1982), this species is a predator and a natural enemy of aphids (Yaşarakıncı and Hıncal 2000)

Bayram (2008) determined that this species feeds on *Eriosoma lanugierum*.

A total of 4 adults were identified, 1 in the Yayıklı Village and 3 in the Çakıllar Village strawberry fields in this study.

Distribution: Uygun (1981) reported as a common species in Türkiye, Adana, Afyon, Ankara, Antalya, Bursa, Hakkari, Hatay, İçel, Kahramanmaraş, Kastamonu, Kayseri and Marmara Regions

Table 2. Natural enemies detected in strawberry fields in Halkapınar district of Konya province and their locations.

Order	Family	Species	Locations		
			Konya-Halkapınar		
			İvriz Village	Yayıklı Köyü	Çakıllar Köyü
Coleoptera	Coccinellidae	<i>Coccinella septempunctata</i> Linnaeus, 1758		x	x
		<i>Scymnus rubromaculatus</i> Goeze, 1778		x	x
		<i>Scymnus pallipediformis</i> Günther, 1958		x	x
Thysanoptera	Thripidae	<i>Scolothrips longicornis</i> Prisner, 1926		x	x
	Nabidae	<i>Nabis pseudoferus</i> Remane, 1949	x		x
Hemiptera	Reduviidae	<i>Coranus griseus</i> Rossi, 1790	x	x	
Neuroptera	Chrysopidae	<i>Chrysoperla carnea</i> Stephens, 1836	x		x
	Raphidiidae	<i>Raphidia</i> sp.	x		

Scymnus pallipediformis Günther, 1958 (Coleoptera: Coccinellidae)

Common name: Bred minipiga

Description: It is an oval shaped species with a length of 1.85-2.8 mm. There are 2 or 4 round reddish spots on the elytra of this species. The number and size of the spots may vary. Specimens with two stains may be very similar to *S. apetzii*. Definitive species identifications can be made according to the condition of aedagus. The aedeagus and parameres of *S. pallipediformis* are equal or longer. The siphon tip is curved (Uygun1981).

Habitat: This species feeds on aphids, crustaceans and psyllids. It is a predator of *Lepidosaphes pistaciae* and *Agonoscaena pistaciae*. It has been reported that it is fed with the maize leaf aphid *Rhopalosiphum maidis* in the Çukurova region (Portakaldalı, 2008).

As a result of the study, a total of 7 adult individuals were identified, 1 in the Yayıklı Village and 6 in the Çakıllar Village strawberry fields.

Distribution: It has been found in Adana, Antalya, Bursa, Hakkari, Hatay, İçel, Istanbul, İzmir, Kırklareli, Malatya, Mardin, Muğla and Urfa (Uygun1981).

Scolothrips longicornis Prisner, 1926 (Thysanoptera: Thripidae)

Common name: Six-spotted thrips

Description: The first instar L1 larva is very small (approximately 0.2 mm), transparent white color and red eyes. After molting, the L2 larva looks very similar to the L1 larva and only the body grows significantly (Şengonca and Gerlach 1986). These 2 larval stages are followed by 2 dormant protonymph and nymph stages, due to typical neometobola metamorphosis of Thysanoptera. During this period, nymphs are white in color and do not take food, although they can move. Antennae are free in protonymph and attached to the head in nymphs (Şengonca and Gerlach 1986).

Adults are about 1 mm in length and light lemon yellow in color. There are 3 brownish spots evenly spaced on the front wings in the form of eyelashes. Although the adult male is similar to the female, the body is smaller and the wings are shorter (Şengonca and Gerlach 1986).

Habitat: *Scolothrips longicornis* has been found in areas of cotton, field vegetable and peanut production, also fruit growing and apple production areas on plateau, and in herbaceous plants in the coastal areas (Tunç 1990). It has been found together with *T. urticae* on fruit trees and herbaceous plants. It should be kept in mind that *S. longicornis* may also be related to *T. cinnabarinus* Boisduval (Şengonca and Gerlach 1986), which lives on herbaceous plants on the beach and is known to feed on it under controlled conditions. On plateau, *Bryobia rubrioculus* (Scheuten 1857))

Panonychus ulmi (Me Gregor), *T. urticae*, *T. viennensis* (Zacher) were mostly detected on fruit trees. However, it should be noted that there is no information feeding under controlled conditions with the exception of *T. urticae* (Şengonca and Gerlach 1986).

As a result of the study, a total of 5 adults were identified, 2 in the Yayıklı Village of Halkapınar district of Konya and 3 in the strawberry fields of Çakıllar Village.

Distribution: *Scolothrips longicornis* is a common species, and has been reported to attract attention in the Mediterranean and Southeastern Anatolia regions of Türkiye (Şengonca and Gerlach 1986).

In the world; it is found in Egypt, Romania, Hungary, Austria, France, Spain and North America (Tunç 1990).

Nabis pseudoferus Remane, 1949 (Hemiptera: *Nabidae*)

Common name: Almindelig nymfetaege

Description: It has an elongated body, grayish or dirty yellowish color. There is usually a longitudinal line on the back of the head and anterior to the pronotum. Hemelytra always fully developed and distinctly longer than abdomen, covered with dense and short hairs. Corium has less than 45 (usually between 25-35) hairs on the apex-lateral part. Aedeagus has one mating hook. Its body length is 7-8 mm (Lodos 1986).

Habitat: Öztemiz (2012) reported this predator insect feeds on the eggs and larvae of *Tuta absoluta*. It has also been reported that *Nabis pseudoferus* feeds on soft-bodied insects, aphids, *Cicadellids* and *Noctuidae* larvae (Lodos 1986).

As a result of the study, a total of 2 adult individuals were identified, 1 in the strawberry field of İvriz Village and 1 in the Çakıllar Village strawberry field.

Distribution: It has been reported to be found in the region from Europe to Iran. It is also common species in our country (Lodos 1986).

Coranus griseus Rossi, 1790 (Hemiptera: *Reduviidae*)

Description: Body length is 8.5-11.2 mm. Body color is gray, brown-gray and sometimes black. The underside of the elytra is red or orange. Antennae are gray and short (Straub and Günther 2006).

Habitat: They are found in grassy grasses, rocks and places with sparse vegetation. They feed on small arthropods (Anonymous, 2013 c).

A total of 3 adults were identified, 1 in the strawberry field of İvriz Village and 2 in the strawberry field of Yayıklı Village in the current study.

Distribution: In the world; Albania, Andorra, Bosnia and Herzegovina, Bulgaria, Germany, France, Greece, Italy, Croatia, Malta, Macedonia, Montenegro, Portugal, Romania, Russia, Serbia, Slovenia, Spain, Switzerland, Ukraine, Hungary,

It is reported in Algeria, Morocco, Tunisia, Afghanistan, Armenia, Azerbaijan, Georgia, Iran, Iraq, Israel, Kazakhstan, Syria, Tajikistan, Turkmenistan, Uzbekistan and Cyprus (Straub and Günther 2006).

It is found in Aydın, Bilecik, Çorum, Denizli, Diyarbakır, Eskişehir, İzmir, Konya, Malatya, Mardin, Muğla, Siirt, Tekirdağ and Şanlıurfa in our country (Önder 1980).

Chrysoperla carnea Stephens, 1836 (Neuroptera: *Chrysopidae*)

Common name: Green lacewings

Description: The head is green and there are very little dark spots on it. The chest and abdominal segments are green, and there are black, short hairs on it. The wing opening is 20-30 mm, 8-10 mm in height females and 6-8 mm in men (Bozdoğan et al. 2012).

Habitat: The wintering adults in the secluded places of the tree branches, in the walls of the houses and so on. They spend on the floor. Depending on the temperature of winter, the colors return from green to red. They fly in the light around the electrical lamps in the evening. During the day, they relax in the shade places of trees, shrubs or grasses close to aphids (Bahadıroğlu and Daymaz 2001). It is an important predator and was reported as a natural enemy feeding larvae and some types of mites (Lodos 1984).

As a result of the study, a total of 6 adult individuals were identified in İvriz Village of Halkapınar district of Konya and 5 in the strawberry areas of Çakıllar Village.

Distribution: Europe is located in North America with a large part of Asia. It is quite common in Türkiye (Koca 2009).

Raphidia sp. (Neuroptera: Raphidiidae)

Common name: Snakefly

Habitat: They usually prey on small insect such as weak aphids and small caterpillars. Larvae do not know how to eat well, but soft-bodied insects eat (Anonymous 2013 a).

As a result of the study, a total of 1 adult were identified as 1 in the strawberry area of İvriz Village.

4. Conclusions

One mite species, 19 harmful insect species from 16 families belonging to 5 orders and 8 beneficial insect species from 6 families of 4 orders were determined in this study carried out in İvriz in 2011 in Halkapınar district of Konya and in İvriz, Yayıklı and Çakıllar villages in 2011. Among insect species identified, *Tetranychus urticae* Koch, *Thrips tabaci* Lind *Brachycaudus helichrysi* Kaltenbach and *Chaetosiphon fragaefolii* Cockerell were important in terms of pest prevalence and density, and as predator *Coccinella septempunctata* L. and *Scymus pallipediula-niformis* longi were found important species in strawberry fields.

More species were identified in the strawberry fields of the village of Çakıllar than in the other two villages. It is thought that the reason for the detection of more species in Çakıllar village is due to the large number of sampled sites and the high altitude.

Kaplan (2007) determined 58 harmful species from 20 families belonging to 7 orders and 21 beneficial insect species from 11 families in a faunistic study conducted in strawberry fields in Elazığ province. Erkiç et al. (1996) detected 48 insect and mite species in their faunistic study in the strawberry fields of İçel province. In this study, fewer species were detected compared to previous studies, and this is thought to be due to late start of strawberry cultivation, low number of strawberry locations, and they are spraying surrounding orchards with broad-spectrum insecticides.

The most common pest is *T. urticae*. Yigit and Erkiç (1992), Schuster et al. (1979) and Lapre et al. (1982) stated in various studies that red spiders, which constitute the most important pest group in strawberries, damage the mesophyll cells in the leaves, causing the plants to be stunted, sometimes to death, and 50% yield loss, especially in cases where the population density is high. However, since it creates a low population, there is no obvious harm.

Aphid species *Brachycaudus helichrysi* and *C. fragaefolii* detected in all the sampling area are. Although these species were seen to a greater or lesser extent at all 1 sampled locations from the beginning of the season through July, August, September and October, they did not show a significant population densities. Kaplan (2007) reports that aphids primarily transmit viruses when they feed by stinging sucking sap but, they also cause deformities as a result of feeding when their populations are high.

T. tabaci is the only thrips species found to be harmful to strawberries in the current study. Although they are generally seen in gardens like aphids, their population level are low. Cultural measures should be given importance in the management of thrips species.

As a result, it was reported that the most common pests such as spider mites, aphids and thrips were also detected in this study in the strawberry fields of Halkapınar district of Konya province, have not yet reached the population density that requires management methods.

Strawberry cultivation in Halkapınar district has started to become widespread since 2010. As strawberry pest population are low, intensive spraying is not used. It is essential to take the necessary plant protection measures to maintain the equilibrium state, and to train and raise awareness of the practitioners in this context.

It was noted during the censuses that the presence of weeds had a great effect on the population increase of small arthropods in strawberry fields. Therefore, weed removal should be given special attention alongside other cultural measures.

With this research, detrimental insect species and their natural enemies have been revealed in the strawberry fields of Halkapınar district of Konya province, and it is thought that it will contribute to the studies to be carried out on this subject.

Author Contributions: The authors have an equal contribution. All authors have read and agreed to the published version of the manuscript.

Acknowledgments: This article is derived from master's thesis of Atalay KILINÇ. The author is grateful to his supervisors for their support during his study. The study has not applied to any funding organization.

Conflicts of Interest: The authors declare no conflict of interest.

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The Global Goose Meat Production Quantity Forecast for the 2023–2027 Years

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HIGHLIGHTS

- Goose meat
- Forecast
- ARIMA

Abstract

This study examines the growing acceptance of goose meat production, its nutritional value, and its varied cultural and gastronomic significance. The purpose of this study was to evaluate the global output of goose meat between 2023 and 2027. The investigation was carried out using statistical data websites, such as FAOSTAT. Forecasts for upcoming years were created by combining data on the production of goose meat from 1961 to 2022. The ARIMA model was used to create forecasts, and the most appropriate model was found using the SAS statistical program. Because it outperformed the other models on several metrics, including AIC, BIC, SSE, MSE, SBC, MAE, MAPE, DW, RMSE, HQC, and R², the ARIMA (3,1,1) model was determined to be the most suitable model. It is projected that the amount of goose meat produced worldwide will rise from 150 thousand tons in 1961 to 4 million 751 thousand tons in 2027. A change of -0.019% was computed based on the differences between the average of the 61 years that followed this period and the actual production figures for the 62 years between 1961 and 2022. The current study predicts that global goose meat production will increase by 246.32% in the five years between 2023 and 2027, compared to the average of the previous 62 years. The results of this study, which used advanced statistical methods and market analysis, suggest that goose meat production will increase over the next five years.

Keywords: Goose meat; production; forecast; ARIMA

1. Introduction

As the global population continues to grow, pressure on limited land and water resources will increase in the near future. Food supply, which was only an acute problem in isolated regions in the past, is now becoming a global threat. This increasing demographic trend requires the development of innovative and sustainable methods to increase food production and meet the nutritional needs of the growing world population in an efficient and responsible manner (Schneider et al. 2011). The link between future food security and human

Citation: Dumlu, B. (2024). The global goose meat production quantity forecast for the 2023–2027 years. *Selcuk Journal of Agriculture and Food Sciences*, 38(2), 326-341. <https://doi.org/10.15316/SJAFS.2024.030>

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Received date: 09/02/2024

Accepted date: 29/07/2024

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health will become increasingly important and the interaction between these two areas will become a key focus for sustainable development in the future. Meat is an important food source in the international economy, with the share of livestock farming in the national economy being more than 50 percent in leading agricultural countries (Ederer et al. 2023).

A balanced diet rich in essential nutrients not only serves as a way to prolong life span but more importantly, also plays a role in enhancing the health and vitality of an aging society (Wickramasinghe et al. 2020; Güner 2021). In recent years, changes in global food systems and dietary habits have increased the importance and value of various protein sources (Ambikapathi et al. 2022). For a healthy diet, not only the amount but also the source of protein consumed should be considered. Proteins are critical components of the human body and are critical for health and development. This term, derived from the Greek word "proteios", emphasizes the amino acid structure and digestibility of high-quality proteins, especially from animal sources, making them important for human growth, development, and overall health (Elmadfa and Meyer 2017; Wu 2019). Animal proteins generally have a higher biological value than plant proteins and are both more easily and efficiently utilized by the body (Reid-McCann et al. 2022). This biological value is particularly evident in the popularity of animal-derived proteins, such as goose meat, which is a rising product in the global food market. When it comes to poultry meat, chicken and turkey meat come to mind first. However, in the world of gastronomy, there are also fewer known alternatives, such as goose meat, which is rich in flavor and nutritional value. Goose meat, an important product in the global food sector, has gained popularity among consumers in various regions of the world in recent years (Kılıç 2021). The consumption of goose meat is increasing among consumers in various regions of the world and is a concrete example of the effective use of animal proteins in nutrition. This change emphasizes the vital role of various protein sources in meeting nutritional needs across different cultures and regions as well as evolving dietary preferences. Goose is of great importance not only for its meat, but also for its liver, fat, eggs, and feathers (Kozák 2021).

In this context, goose meat, which is a critical component of a healthy and balanced diet, attracts attention in terms of both nutritional value and sustainable production potential. Goose meat which is rich in protein, minerals, and vitamins, is becoming increasingly important for a healthy and balanced diet (Pereira and Vicente 2013; Goluch and Haraf 2023). Goose meat provides high-quality protein, contains significant amounts of B vitamins, especially vitamin B12, and has a relatively low collagen content ranging from 0.39 to 0.91%. It is also rich in minerals such as iron, zinc, and phosphorus. Moreover, the iron content of goose meat is particularly noteworthy for those with iron deficiency (Adamski and Wenczek 2012; Orkus et al. 2021; Goluch and Haraf 2023).

Goose meat which is known worldwide for its short production time and economic costs, has a rich and aromatic taste with intense flavour, nutritive value, and high fat content. The available fats are mostly rich in omega-3 unsaturated fatty acids and contribute to heart and muscle health, nervous system, and general immune system support (Buzala et al. 2014; Öz and Çelik 2015; Uhlířová et al. 2018). It should be noted that meat production statistics may fluctuate annually depending on a variety of factors, including consumer preferences, economic conditions, and changes in agricultural practices. According to FAO statistics, meat production worldwide was 360 million tons by the end of 2022. Of this figure, 39% is cattle production and 35% is ovine production. In 2023, world red meat production was 215.3 million tons in total. Of this production, 35.4% is cattle, 7.8% is ovine and 56.8% is pork (Yıldırım 2023).

While the world poultry meat production was 9 million tons in 1961, this production amount reached 139.2 million tons in 2022. These data on poultry meat production include goose, chicken, turkey, duck, pigeon, and other birds. According to FAO data, poultry meat production was 7.6 million tons of chicken, 898 thousand tons of turkey, 336 thousand tons of duck, 150 thousand tons of goose, 10 thousand tons of pigeon and other birds in 1961; 123.6 million tons of chicken, 6.1 million tons of duck, 5.1 million tons of turkey, 4.4 million tons of goose, 18.7 thousand tons of pigeon and other birds in 2022. According to the data for the years 1961-2022, goose meat ranks 4th in poultry production in the world. The leading countries in poultry meat production in the world are China 98.53%, Taiwan 0.33%, Egypt 0.29%, Madagascar 0.29%, Myanmar 0.17%, Ukraine 0.12%, Türkiye 0.09% and other countries 0.18%. China has the largest share in terms of goose meat consumption and production. According to FAO statistics, the goose meat production values for 2022 are as follows; China 4.3

million tons, Taiwan 14.5 thousand tons, Madagascar 12.8 thousand tons, Egypt 12.6 thousand tons, Myanmar 7.5 tons, and Turkiye 3.8 tons. Increasing goose meat production can contribute to improving the quality of nutrition, especially in developing and underdeveloped countries.

According to the 2023 data provided by the World Population Review on poultry meat consumption in the world food market, Israel is the country that consumes the most poultry meat in the world with 64.9 kg per capita per year. China ranks second in the world with an annual consumption of approximately 21.56 million tons of poultry meat, Brazil ranks third with 10.29 million tons and Mexico ranks fourth with 4.6 million tons. It is stated that Europe consumes approximately 11 million tons of poultry meat annually. These data explain how poultry meat consumption rates have caught a changing trend according to countries. Poultry meat consumption is particularly popular in China and Europe, but the amount of consumption in each country varies depending on factors such as population size and eating habits.

Consumption trends show that goose meat preference has been increasing among consumers worldwide due to its short production process and cost-effectiveness (Buzala et al. 2014). China stands out as the leading country in goose and duck meat consumption worldwide and accounts for 76% of total consumption. Approximately 5.5 million tons of goose and duck meat is consumed annually. France is the second largest consumer with 203 thousand tons per year. Myanmar ranks third and consumes 174 thousand tons. The countries with the highest per capita consumption of goose and duck meat are Taiwan (6.12 kg/person), China (3.77 kg/person) and Myanmar (3.23 kg/person). These data show that the global goose and duck meat market is expected to grow in the coming years (Anonymous 2024a).

In 2022, according to goose meat production and export shares, the countries are listed as Poland, Hungary, Belgium, Germany, France, Hong Kong, Austria, Netherlands, the United States of America, and South Africa. Poland has a large share (54.54%) of the global goose meat export market and exported 71.76 million USD. This represents an increase of 33.99 percent compared to the previous year and 63.54 percent over the three-year period. Hungary has a 41.11 percent share of the export market and exports 54.10 million USD. However, Hungary's export value decreased by 13.02 percent compared to the previous year. Belgium contributed 1.58 percent to the global export market, exporting 2.07 million USD.

Germany and Hong Kong are the largest importers of goose and duck meat products worldwide. The main suppliers of these imports were Hungary, China, Poland, and France, accounting for 66 percent of the total global export volume. Average import prices also varied among the leading importing countries. In 2022, France had the highest import price per tons, while the UK was among the lowest. The average import price in Europe increased by 24% year-on-year, reflecting increased demand and potentially higher production costs or supply constraints. These figures provide a comprehensive overview of the global goose meat market, showcasing the leading countries in production, consumption, exports, and imports, and market dynamics in terms of pricing and growth trends (Anonymous 2024a,b,c). Goose meat production estimates will be made by evaluating the available figures with time series analysis.

Time series analysis is a set of methods and techniques used to understand current data and predict future trends. One of the most widely used time series forecasting models is the ARIMA (Autocorrelated Integrated Moving Averages) model. This model is a powerful and flexible time series analysis method used to learn trends and seasonality in time series data and to predict future values (Cowpertwait and Metcalfe 2009; Wilson and Armitage 2012; Wilson and Armitage 2012; Box et al. 2016; Wilson 2016).

This study aims to analyze in detail the global goose meat production forecasts for the period 2023-2027 using Food and Agriculture Organization (FAO) data for the period 1961-2022, how it has changed worldwide and how it will change in the future, the causes and consequences of this change based on historical data and current market trends. The focus of the paper is to analyze in depth how the importance of goose meat production in terms of global food security and nutritional balance is shaped by both consumer health and economic, environmental, and social factors. It also identifies potential growth areas and challenges in the sector. This study has the potential to be a valuable resource for both academic researchers and industry experts and aims to shed light on the changing dynamics in global food production.

2. Materials and Methods

2.1. Material

This study's secondary data came from the Food and Agriculture Organization of the United Nations (FAOSTAT 2024). A variety of sources, including local and worldwide online resources, academic papers, journals, reports, essays, and theses, were also employed in the research in addition to the data on goose meat production, consumption, exports, and imports that was gathered from these foreign sources. Several approaches have been used in this study to analyze the data and make future projections.

2.2. Method

Based on statistical data from the FAOSTAT, data on the global production of goose meat between 1961 and 2022 were examined in this study. The study forecasted the output of goose meat from 2023 to 2027 using the ARIMA time series analysis methodology. Using SAS 9.4 software, the statistical data in this model were calculated. Microsoft Word was the primary tool for modifying the tables in the research, although Microsoft Excel was used for mathematical computations and graphing.

2.2.1. ARIMA models

The ARIMA (Auto-regressive Integrated Moving Average) model stands out among the effective linear statistical models that are frequently used in time series forecasting. This model was developed by Box and Jenkins in the early 1970s and is a common method used to analyse time series data (Reinsel 1994; Yüksel 2015; Brownlee 2020). The ARIMA model is used to forecast time series data. It is studied by utilising the past values of a series for future forecasts. This model is designed specifically for linear time series, characterised by three parameters and is usually expressed as p, d, q . Where "p" denotes the degree of the AR term, "d" the degree of integration and "q" the degree of the MA term. ARIMA essentially analyses the past values of a time series to make predictions about its future values. The ARIMA model utilises the past values of a variable to predict its future values (Alabdulrazzaq et al. 2021; Bai et al. 2023; Linardatos et al. 2023). Time series are generally divided into two categories: stationary and non-stationary. Stationary time series have statistical properties that do not change over time, while non-stationary time series have variable properties such as seasonal patterns. In non-stationary time series, the mean and variance may change over time. Non-stationary data can be converted to stationary by taking the differences between consecutive observations, a process called "differencing". By removing changes in the level of the time series, differencing removes trends and seasonalities and then uses these data to make forecasts (Nason 2006; ArunKumar et al. 2021; Uzundumlu et al. 2023). In the ARIMA model, an iterative process is followed and the data analysis and modelling stages are carried out carefully (Box et al. 2016; Fattah et al. 2018).

Time series models are generally categorised as ARMA (Stationary with Time), ARIMA (Non-Stationary with Time) and SARMA (Seasonal and Exponential Adjustment). When conducting statistical analyses based on time series data using the ARIMA model, several important hypotheses are considered. These include that the data are stationary over time, have a normal distribution, do not contain anomalies or outliers, and there is no missing data (Makridakis and Whellwright 1978; Palabıçak 2019).

ARIMA time series model has 4 basic steps. The first step starts with "Determination of the Model". First of all, the appropriate model type should be selected according to the characteristics of the data set and the structural behaviour of the time series. In determining the appropriate model for time series analysis, it is first necessary to evaluate whether the data are stationary or not. This is usually done through unit root tests. If the data are found to be non-stationary, differencing methods should be applied to achieve stationarity. The second step, "Estimation of Parameters", involves the calculation of the parameters of the selected model by statistical methods. Estimation of parameters, which is a very important step in model selection, usually involves the use of SCAN and ESCAF techniques to determine the most appropriate models. At this stage, the suitability of the model and the significance of the parameters are evaluated. The third stage, "Diagnosis and Audit", involves testing the assumptions of the model and examining its suitability for the data. Various information criteria and performance measures were used to determine the ARIMA time series model. AIC

(Akaike Information Criterion), BIC (Bayes Information Criterion), DW (Durbin-Watson), HQC (Hannan-Quinn Criterion), MAE (Mean Absolute Error), MAPE (Mean Absolute Error Percentage), SSE (Sum of Squares Error), The most appropriate models were determined according to the results of RMSE (Root Mean Square Error Squared), SBC (Schwarz's Bayesian Criterion), MSE (Mean Square Error Squared) and R^2 (Coefficient of Determination). Various tests, especially BIC and AIC, are used in the diagnosis and examination phase of model selection. The model that passes the highest number of these tests is generally accepted as the most appropriate model. These criteria and criteria helped to select the best model by focusing on factors such as predictive ability, fit and complexity of the model. However, normality tests and normality of residuals tests should be performed to accept the selected model. If these tests show non-normality, the model can be rejected in favour of another model that better meets the criteria. If the model does not fit the data set well, the process should go back to the first step to identify a new model. If the model is found to be appropriate, the final step, "Making the Prediction", should be taken and future predictions should be made using the model. This iterative process is designed to ensure that time series data are analysed and predicted with scientific rigour and rigour (Kohvakka 2017; Prabhakaran 2019; Silva et al. 2021; Andipara 2022; Hamel and Ismael 2022; Zhao et al. 2022; Uzundumlu and Dilli 2023; Wagner and Cleland 2023).

3. Research Findings

3.1. Goose Meat Production Estimates for 2018-2025 with ARIMA Model

3.1.1 Stagnation detection

The Dickey-Fuller test first analyses the stationarity of the data for the presence of a unit root in the application of the ARIMA model. Furthermore, the analysis reveals the absence of a unit root and white noise. Figure 1 illustrates the non-stationary nature of the goose meat production data between 1962 and 2022.

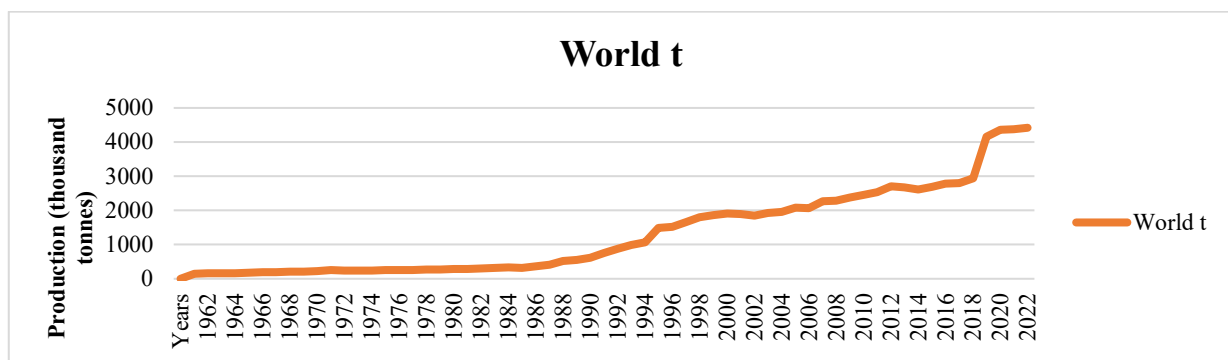


Figure 1. World goose meat production in the period 1962-2022 (thousand tons).

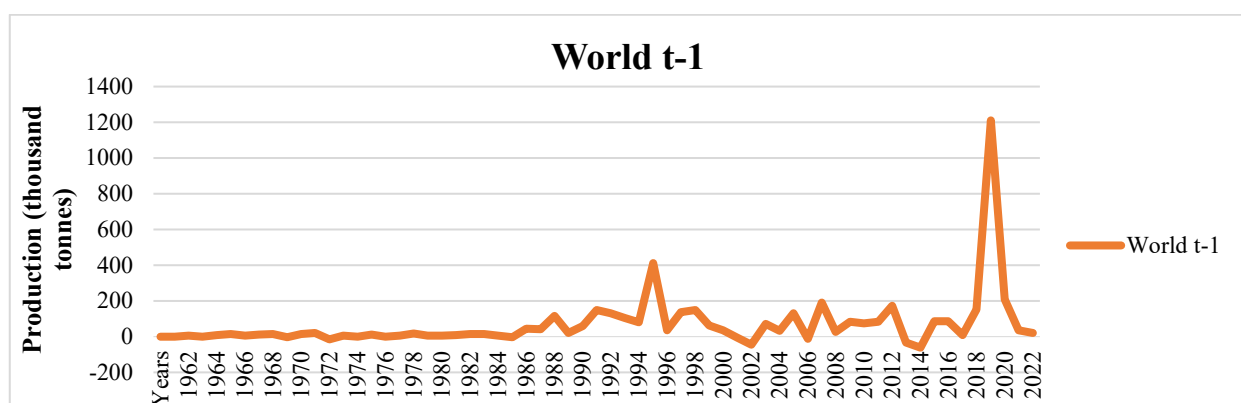


Figure 2. World goose meat production after one year of differential treatment in the period 1962-2022 (thousand tons)

It seems that the global goose meat production data is not stable and does not revolve around a specific point. We used a method that involves applying a one-year lag to the data in order to make it stationary. With a one-year lag, this process resulted in the achievement of stationarity.

As shown in Figure 2, the one-year lagged data analysis resulted in the use of a data set of 61 (62-1=61) years instead of 62 years, accounting for the differences in the previous year's data. This process has stabilized the goose meat production data, creating a zigzag pattern that resembles a heartbeat graph.

3.1.2. Parameter estimates for goose meat consumption

Within the scope of the research, the fluctuating tendency of goose meat data around the world necessitated the application of a one-year lag with the differencing process in order to reduce the effects of these fluctuations. This method enabled the data set to acquire a stationary form and then made it more suitable for time series analysis.

Table 1. Parameter Estimates.

Variable	DF	Forecast	Se	t Value	Prediction Pr > t	Variable Class
Stasis	1	-59380	54984	-1.08	0.2848	
t ₁	1	5955	3363	1.77	0.0820	t ₁
IWorldt ₁	1	-0.05136	0.05178	-0.99	0.3255	
IWorldt ₁₋₁	1	0.13614	0.14069	0.97	0.3374	

DF: The standard Dickey-Fuller, Se: Standard error.

ARIMA procedures are given in Table 2. As can be seen from the table, the number of observations analysed was determined as 61 by subtracting 1 from 62. This shows that one-year differences of the data are taken into account. The mean value of the annual goose meat production worldwide was determined as 69987.06 thousand tons and the standard deviation was calculated as 165296.4 thousand tons.

Table 2. ARIMA Procedure.

Variable name = Worldt	
Differentiation period (year)	1
Average of the study series (thousand tons)	69987.06
Standard deviation (thousand tons)	165296.4
Number of observations (year)	61
Observation reduced by differencing (years)	1

Table 3 shows the values of the ARMA(p+d.q) trial ranking criterion tests for world goose meat production.

Table 3. ARMA (p+d.q) Trial Ranking Criterion Tests

SCAN			ESACF		
p+d	q	BIC	p+d	q	BIC
0	0	24.01916	0	0	24.01916
			1	0	24.04953
			3	1	24.24378
			5	2	24.25891

SCAN: The smallest canonical, ESACF: Extended Sample Autocorrelation Function, BIC: Bayesian Information Criterion.

Minimum Table Value: BIC(0,0) = 24.01916

At 5% significance level

There are 1 p and q values in SCAN analysis and 4 p and q values in ESACF analysis. Considering the cases where both p and q are accepted as zero, ARIMA(0,1,0). ARIMA(1,1,0), ARIMA(3,1,1) and ARIMA(5,1,2) models were found to be the most appropriate models. SCAN and ESCAF determined the best models and selected them based on specific criteria. Table 4 provides these criteria.

Table 4 presents the results of ranking criterion tests based on p and q values for world goose meat production. As a result of the model comparisons based on the values. Choose the models with a DW of 2.00 or closer and the greatest R² value from these criteria, in addition to the other models having the smallest coefficient (Kurtoğlu et al. 2024). In addition, we conducted a comparison of the models, considering the discrepancies between the five-period forecast and the actual values over the past decade (Uzundumlu et al. 2023). The ARIMA (3,1,1) model, which exhibits superior performance in many criteria, is determined as the most appropriate model.

Table 4. Ranking Criterion Tests according to p and q Values.

p	q	BIC	SSE	MSE	SBC	MAE	MAPE	DW	RMSE	AIC	HQC	R ²	2013/2017	2018/2022
0	0	24.02	1.60	2.75	1.59	7.72	4.26	1.99	1.66	1.59	1.59	4.04	1.94	-6.30
1	0	24.05	1.60	2.75	1.59	7.71	9.98	2.00	1.66	1.59	1.59	4.98	1.78	-5.09
3	1	24.24	1.56	2.73	1.56	7.46	7.84	2.02	1.65	1.56	1.56	4.14	1.78	-5.11
5	2	24.26	1.57	2.70	1.59	8.27	14.2	1.98	1.64	1.59	1.59	4.07	1.38	-5.50

BIC: Bayesian Information Criterion, SSE: Sum of Squared Estimate of Errors, MSE: Mean Squared Error, SBC: Schwarz's Bayesian Criterion, MAE: Mean Absolute Error, MAPE: Mean Absolute Percentage Error, DW: Durbin Watson Test, RMSE: Root Mean Square Error, AIC: Akaike Information Criterion, HQC: Hannan-Quinn Criterion, and R²: Coefficient of Determination (Uzundumlu and Dilli 2023). Note: SSE 1012, MSE values 1010, SBC 103, MAE 104, MAPE 102, AIC and HQC 103, R² multiplied by 101 and RMSE values by 105.

Table 5 shows the autocorrelation check of the residuals. The fact that the P values in the table are not significant even at the 0.05% level indicates that there is white noise in the first difference of the data and thus the data are stationary.

Table 5. Autocorrelation Control of Residuals.

Delay	Chi-Sq	DF	Pr > ChiSq	Autocorrelation					
6	0.90	4	0.9251	0.002	0.007	0.002	0.062	-0.058	-0.076
12	3.81	10	0.9555	0.096	0.026	0.006	0.031	-0.068	0.150
18	5.34	16	0.9938	-0.078	0.067	-0.028	0.006	-0.044	-0.070
24	12.53	22	0.9454	-0.041	-0.003	0.051	0.026	-0.077	0.243

DF: The standard Dickey-Fuller.

Figure 3 displays the residual correlation indicators. White noise tests are used to determine whether residual series include information that can be assessed by a more sophisticated model (SAS 2014). Furthermore, residual correlation analyses confirm the suitability of the current model.

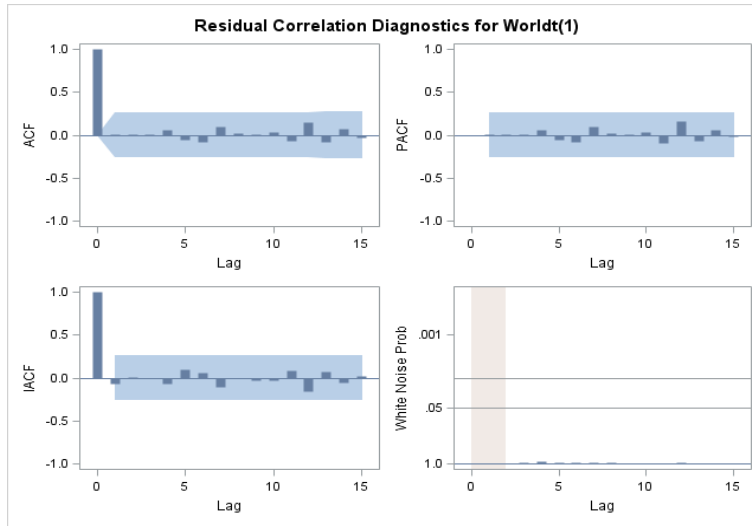


Figure 3. Residual correlation indicators.

Analysis of the residual normality indicators in Figure 4 reveals that, despite the model's diagnostic tests deviating from a completely normal distribution, many parameter estimates are significant and the residuals exhibit a white noise character. This situation prepared the ground for the transition to the estimation and diagnostic control phases.

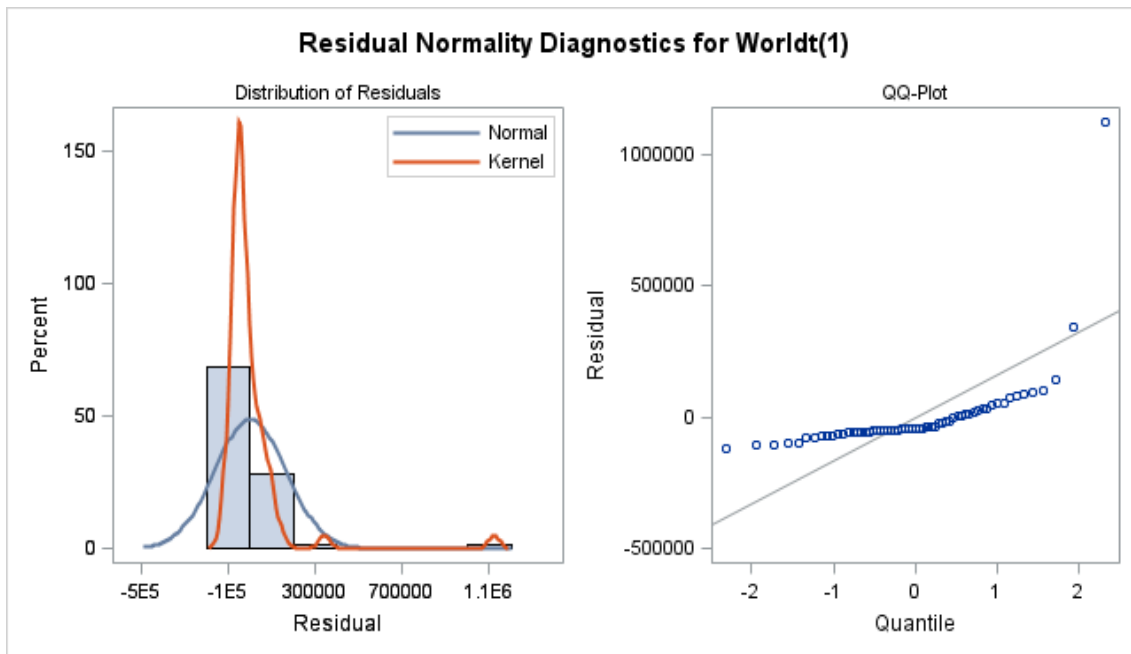


Figure 4. World residue normality indicators.

Table 6 shows the mean lag and AR. MA factor 1 value is given. Considering the determined estimation parameters. The mathematical expression of the ARIMA (3,1,1,1) model. It is presented in Equations 1 and 2 (SAS. 2014).

Table 6. Forecast Mean Average Lag and AR, MA Factor 1 Values.

Forecast Average (thousand tons)	69.438
Delay Value (years)	1
AR factor 1 value	1 - 0.02844 B**(3)
MA factor 1 Value	1 + 0.1979 B**(1)

$$W_t = \mu + \frac{\theta(B)}{\phi(B)} a_t \tag{1}$$

$$(1 - B)Y_t = \mu + \left(\frac{1 - \theta_1 B^1}{1 - \phi_1 B^1} \right) a_t \tag{2}$$

$$(1-B) \text{ World goose meat production } t = 69.438 * 10 + \frac{(1 + 0.1979 B)}{(1 - 0.02844 B)} * a_t$$

t: indexes time

Wt: is the response series Y, or a difference of the response series

Yt: are the original response variable observations

μ: is the mean term

B: is the backshift operator; that is, $BX_t = X_{t-1}$

φ (B): is the autoregressive operator, represented as a polynomial in the backshift operator:

$$\Phi (B) = 1 - \Phi_1 B - \dots - \Phi_p B^p$$

Θ (B): is the moving-average operator, represented as a polynomial in the backshift operator:

$$\Theta (B) = 1 - \Theta_1 B - \dots - \Theta_q B^q$$

a_t: is the independent disturbance also called the random error

Figure 5. The minimum, maximum and average goose meat production values estimated for 2023-2027 based on the world goose meat production data between 1961-2022.

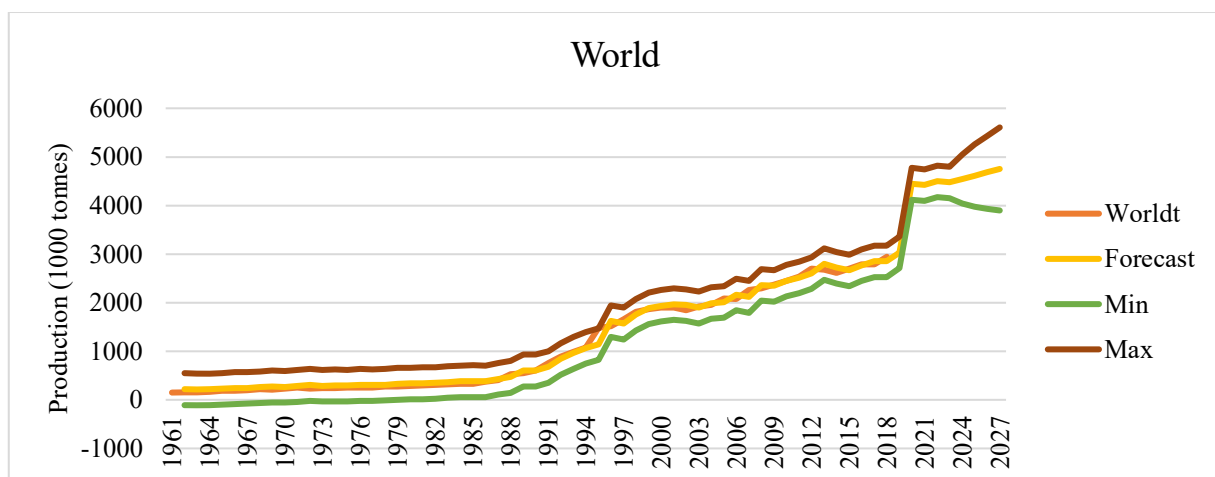


Figure 5. World goose meat production projections for 1962-2027 (thousand tons).

As can be seen, the amount of goose meat production realised on an annual basis between 1962 and 2022 worldwide was 1,351,672.61 and the estimated value calculated by ARIMA (3,1,1) for the same period was calculated as 1,351,418.50 tons. The deviation of the forecast values in the same periods was determined as -0.019%. With this prediction, it is expected that the deviation will be lower in future forecasts. While the world goose meat production was approximately 150 thousand tons in 1961, it is expected to increase to approximately 4 million 751 thousand tons in 2027. In this case, it is estimated that goose meat production will increase approximately 32 times in 66 years. In addition, world goose meat production is estimated to be at least 3 million 997 thousand tons and at most 5 million 231 thousand tons between 2023 and 2027 with 95% confidence interval. 4 million 477 thousand tons in 2023, 4 million 545 thousand tons in 2024, 4 million 613 thousand tons in 2025, 4 million 682 thousand tons in 2026 and 4 million 752 thousand tons in 2027. Formula 3 shows the change according to the two-period average, taking into account the average annual production forecasts for the 61 years between 1962 and 2022 and the 5-year period between 2023 and 2027.

$$\text{Increase Rate} = \frac{(4.613.955,214 - 1.332.286,94)}{(1.332.286,94)} * 100 \approx \%246.32 \tag{3}$$

The formula predicts a 246.32 percent increase in the five-year world goose meat production average between 2023 and 2027 compared to the 62-year average.

In Table 7, the value of goose meat production per capita in g was determined at 10-year intervals between 1961 and 2027. For this purpose, the amount of goose meat produced is proportioned to the world population.

Table 7. Goose meat production per capita by years (g).

Years	Goose Meat Produced Amount (million kg)	World Population (million people)	Per Person Production (g)
1961-1970	185.32	3.373.99	54.93
1971-1980	254.52	4.105.77	61.99
1981-1990	402.80	4.912.53	81.99
1991-2000	1.394.86	5.781.59	241.26
2001-2010	2.115.06	6.603.39	320.30
2011-2020	3.025.76	7.465.40	405.30
2021-2027*	4.555.49	8.120.13	561.01

*Forecast values for the years 2023-2027 are used.

When goose meat production per capita is evaluated by years, global goose meat production started from 185.32 million kg between 1961-1970 and increased to 254.52 million kg with an increase of 37.4% in the period 1971-1980. In the same period, the world population increased by 21.6% to 4.11 billion people and per capita goose meat production increased by 13.1% to 61.99 g. In 1981-1990, goose meat production increased to 402.80 million kg with an increase of 58.1%. The world population increased by 7.3% to 4.91 billion people and the per capita goose meat production increased by 25.3% to 81.99 g. In the period between 1991 and 2000, goose meat production increased significantly to 1.39 million kg, representing a large increase of 246.6%. The world population increased by 18.0% to 5.78 billion people and per capita goose meat production increased by 194.3% to 241.26 g.

In the early 2000s, global goose meat production increased by 51.9% to 2,115.06 million kg between 2001-2010. The world population increased by 7.9 per cent to 6,603.39 million people and the per capita goose meat production increased by 32.9 per cent to 320.30 grams. In the period between 2011-2020, goose meat production increased by 43.1% to 3,025.76 million kg. The world population increased by 13.0% to 7,465.40 million people, and goose meat production per capita increased by 26.6% to 405.30 grams. According to the estimated data

for 2021-2027 (2023 and beyond). global goose meat production is expected to reach 4,555.49 million kg with an increase of 50.5%. According to the data, the world population is expected to increase by 8.8% to 8,120.13 million people and goose meat production per capita is expected to increase by 38.9% to 561.01 grams. As a result, the amount of goose meat produced in 1961 was 185.32 million kg. According to 2027 year-end estimates, it will increase up to 4,555.49 million kg and will increase approximately 16 times in 66 years. The number of people in the world was 3,373.99 million in 1961. In 2027, it is estimated as a result of the analysis that it will reach 8,120.13 million people and will increase 2.5 times.

When the table is analysed, it is seen that global goose meat production has increased over time and this increase has accelerated especially in the 1990s and afterwards. The time period in which the amount of goose meat per capita increased the fastest was between 1981-1990 and 1991-2000. When the results are analysed, there is an increasing trend in the production values per capita in all of the values given in goose meat production per capita. When the import data of the goose meat produced are analysed, it is generally observed that the countries with high welfare level demand it. This situation can be explained by the high scarce demand and the increase in goose meat prices against this demand.

According to the agricultural products market report for 2022. In 2020, world goose and guinea fowl production increased by 2.4% compared to the previous year and reached 2.8 million tons. China alone accounted for 95.9% of this production, or 2.7 million tons. China was followed by Egypt with 1.0 per cent and Myanmar and Madagascar with 0.5 per cent. China, which is in an impressive position in this production, exported only 11 thousand tons of the 2.7 million tons of goose and guinea fowl meat, ranking first in production and second in exports. This situation. This situation reveals that China is an important player in goose and guinea fowl meat consumption. On the other hand, Poland and Hungary, which are in the top three in exports, export a large portion of the meat they produce (TEPGE 2024). In the same period, the world population has also increased, and in parallel, goose meat production per capita has increased. According to the estimated data, global goose meat production and per capita production are expected to continue to increase in the future. China has the biggest share in this increase. The large population and increasing demand in China has made this country the leader in goose meat production. According to FAO data, the increase in China's goose meat production, which was 63 thousand tons in 1961 and reached 4.3 million tons in 2022, is calculated in Equation 4;

$$\text{Increase Rate} = \frac{(4.314.516 - 63.800)}{(63.800)} * 100 = \approx 6.661,39 \quad (4)$$

This increase of $\approx 6.661,39\%$ has enabled China to increase its share in world production from 35% in 1961 to 98.5% in 2022. When the world goose meat production data between 1961-1970 and 2011-2020 are analysed, China's share increased from 56% to 93%. Global goose meat production. 4.4 million tons in 2022 and this amount is expected to continue its upward trend in the following years. When this information is compared with the information provided by "euromeatnews" and "global trade magazine" sources with FAO data; they reported that the market volume of goose and duck meat will reach 8 million tons by the end of 2025, as the compound annual growth rate (CAGR) for the next 7 years is expected to be +1.6% as of 2018 (Anonymous 2024a, d). When annual compound interest is applied to the goose meat production value of 2,945 thousand tons in 2018 according to FAO data in Equation 5;

$$2.944.656,98 * 1 + 0.016^7 = 3.290.718,003 \quad (5)$$

For 2025, the estimated goose meat production value is found as 3.290 thousand tons. In the present study, the estimated goose meat production value for 2025 was determined as 4.613 thousand tons. As a result, the market has tended to oligopolies over the years. There are differences between countries in the percentage changes between 1961-2022 and 2023-2027, which are the compared and estimated periods. These differences

are based on various factors such as agricultural policies, economic growth rates, climatic conditions, technological developments and changes in consumer demands. In this context, it can be said that fluctuations in goose meat production are complex and multifaceted, and this situation constitutes an important factor that will affect strategic planning for stakeholders in the sector. It is stated that the share of goose meat production of China, the leading country in the production period between 1961-2022, in total world production is 98.5 per cent. These data emphasise China's leading position in goose meat production and its significant share in global production.

The United Arab Emirates lacks production data for the years 1961–2022, and Poland, Hungary, and Germany, which are significant exporters, do not provide recent production data. However, it is noteworthy that the average annual imports and exports were 88 tons and 3502 tons in 2018 and 2022, respectively. In other words, the United Arab Emirates should have a place among the producing countries. While the 2013–2017 data of countries that did not have data in recent years was taken into account, the 2018–2022 averages were taken into account for China and England. Therefore, Hungary ranks first with 69.20% in terms of exporting the products it produces, followed by Poland with 67.93%, Germany with 36.81%, England with 14.25%, and China with 0.32%. In other words, Poland, Hungary, Germany, and even the United Kingdom do significant marketing based on their production.

Undoubtedly, climate change and illnesses have a significant impact on plant and animal production, leading to an elevated level of variability in the accuracy of forecasts. Furthermore, there may be unforeseen discrepancies in the projections for next year due to inadequacies or inaccuracies in secondary data sources.

This growth suggests that the goose meat industry has significant economic and commercial potential. This provides important information for stakeholders and policy makers in the sector in terms of strategic planning and resource management. The analysed data suggest that total goose meat production will continue to increase, but fluctuations may occur due to various reasons.

4. Conclusions and Recommendations

This article uses statistical data from FAOSTAT to forecast and assess the global output of goose meat for the years 2023–2027. The methods used to produce goose meat vary greatly between traditional and modern farming techniques. Thus, it is critical to examine the worldwide patterns in the production of goose meat over this time frame to shape consumer preferences, sustainable food supplies, and agricultural policy in the future. The average global goose meat production between 1961 and 2022 was 1.3 million tons. According to the projections, the average goose meat production is expected to be approximately 4.6 million tons in the 2023–2027 period. During the 1961-2022 production period, goose meat production per capita was 0.055 kg worldwide. According to estimates, this value is expected to increase to 0.561 kg between 2022 and 2027.

This increase indicates a significant growth in per capita goose meat consumption and shows changes in consumer habits and supply-demand dynamics at the global level. Countries with high affluence levels are generally known for their demand for more specific and specialized products. This can lead to an increase in imports of specialized and demanding products, such as goose meat. An increase in demand can contribute to higher prices and demand for imports from countries that produce these products. Germany and Hong Kong are the leading countries in imports. Although Poland, Hungary, and Belgium are the leading countries in goose meat exports, the leaders in production are China, Taiwan, and Egypt.

These countries generally focus on domestic markets and have a lower share of exports. This situation shows the need for various regulatory policies to protect producers and consumers. These policies can be effective in supporting local producers, setting quality standards, and regulating exports and imports. Policies to set and inspect food safety standards to protect consumer health and safety may also be important. Various measures can be taken to maintain a balance between imports and exports, ensure fair competition, and protect the interests of all stakeholders in the sector. The increasing trend observed in goose meat production worldwide has led producers to adopt various methods to obtain more products due to their high economic value. The use of correct methods in agricultural practice, especially the transfer of correct information to

producers on technology, irrigation, spraying, species, and variety selection, is important for taking environmental protection measures and preventing major environmental problems. In the future, measures such as storage, product diversification, and marketing infrastructure will increase competitiveness in the market to protect producers and increase their income. This will positively contribute to the welfare level of society through increased employment and gross domestic product.

Author Contributions: The authors have an equal contribution. All authors have read and agreed to the published version of the manuscript.

Funding: This research received no external funding.

Data Availability Statement: In this study, "FAOSTAT" data are used.

Conflicts of Interest: The author declare no conflict of interest.

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Determination of Fatty Acid Profiles and Antioxidant Activities of Some Edible Oils Consumed in Türkiye

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HIGHLIGHTS

- The purpose of this study was to determine the antioxidant activity of the samples and to identify the presence of β -sitosterol in their structure.
- The study identified the fatty acids present in the oils, as well as the ratio of saturated to unsaturated fatty acids.
- The oils were found to contain linoleic acid (C18:2) and oleic acid (C18:1) as the primary fatty acids.

Abstract

Vegetable-based edible oil in Turkey is produced primarily from olive oil, as well as oils obtained from various plants such as sunflower, corn, cotton, poppy, soybean, safflower and canola which are also among the main food sources. The quality of the oils used in our diet is primarily determined by the ratios of saturated and unsaturated fatty acids. Another important feature that determines oil quality is primarily antioxidant activity and its other biological activity capacities. This study was carried out to determine the fatty acid profiles and antioxidant capacities of 17 (seventeen) different oils of vegetable origin, which are widely consumed in Turkey and have economic importance. Fatty acid compositions were determined with GC-MS method. Antioxidant activities of different edible oils were determined by using 2,2'-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging activity method. According to the results of this research, the highest scavenging effect was detected at different concentrations was obtained from soybean oil with $94.30 \pm 1.57\%$, while the least scavenging effect was obtained from peanut oil with $32.34 \pm 1.00\%$. Major fatty acid components linoleic acid (C18:2) and linolenic acid (C18:3) were detected in 17 different oils. Also, the presence of beta-sitosterol was examined by Thin Layer Chromatography and it was determined that beta-sitosterol was present in almost all of them.

Keywords: Fatty acid; saturated fatty acid; unsaturated fatty acid; antioxidant activity; DPPH

1. Introduction

Essential nutrients are classified into two main groups: macronutrients and micronutrients. The feature distinguishing macronutrients from micronutrients is their having energy values, in another mean, they give us the energy that we need on a daily basis. On the other hand, micronutrients are auxiliary substances for

Citation: Çelik SA, Kan Y (2024). Determination of fatty acid profiles and antioxidant activities of some edible oils consumed in Türkiye. *Selcuk Journal of Agriculture and Food Sciences*, 38(2), 342-355. <https://doi.org/10.15316/SJA.FS.2024.031>

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Received date: 11/03/2024

Accepted date: 31/07/2024

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our body, such as minerals and vitamins. Macronutrients are carbohydrates, proteins and fats. Fat is one of the main nutrients required for many metabolic activities in our body and it is used as a building stone in the body therefore oils are especially important in hormones and cell membranes (Kolzeev 2022). In addition, vitamins A, D, E, and K are fat-soluble vitamins. A certain amount of oil consumption is needed daily by our body in order to use these vitamins. Since oils contain higher calories compared to other food groups, oil consumption was kept very low in diets (1 g ~ 9 calories) and in some diseases, or it could be zero. It could trigger other hormonal disorders in such case. According to performed studies in recent years, some essential oils should be used in diets and in diseases such as heart disease. The importance of oils with a high unsaturation rate in other diseases like losing weight, high cholesterol and heart diseases etc. has also been understood more clearly. Many studies and discussions show that a certain amount of oil included in an adequate and balanced diet should be taken every day. One of the important points is the type and amount of consumed oil. Whether the fatty acids are essential or not is determined according to the saturation and unsaturation ratio of consumed oil's structure. (Kaya 2018; Kayahan 2002).

The basic building stone of oils containing high amounts of carbon and hydrogen are structures called triglycerides. Triglycerides which are composed of fatty acids and glycerol are linked by ester bonds. These substances which are either solid or liquid at room temperature, are insoluble in water and are but they are not volatile. The oils are confidently classified as saturated, monounsaturated, and polyunsaturated based on the precise ratio of saturation and unsaturation in their fatty acid structure. Such as saturated oils are solid at room temperature, and coconut oil and butter are included this group while olive oil and sesame oil can be given as examples of monounsaturated oils which is known that monounsaturated oils have an effect on reducing total cholesterol. Corn oil, linseed, poppy oil, and grape seed are among the polyunsaturated oils that contain omega-3 and omega-6 in their structure. It is suggested that they be taken as supplements or applied topically, as these fatty acids are considered essential and cannot be synthesized by the body. Arachidonic acid, linoleic acid, and linolenic acid are considered essential fatty acids. Omega fatty acids have useful functions for brain development, strengthening of the immune system, prevention of coronary heart diseases (Kayahan 2003; Kalbini Dinlesen 2022). So fatty acids are important components for cell membranes of all tissues in our body. The main component of the brain after water is lipids, and about half of its dry weight is oil. For this reason, it is important to take the right oil in adequate amounts and to protect it with antioxidants. The harmful effects of reactive oxygen species and other radicals, which are formed as a result of certain mechanisms in our body, are controlled by antioxidants. When free radicals are formed at rates exceeding the capacity of the defense mechanisms, the balance between the oxidant and antioxidant system in our body becomes damaged. As a result of this case, it may cause many diseases such as hypertension, coronary heart disease, diabetes, premature aging, Alzheimer's. Therefore, antioxidants and essential oils with high antioxidant capacity, food supplements, etc. should be used as a protector against free radicals (Kurban and Mehmetoğlu2006) which is known that daily fat intake protects the antioxidant system of our body thanks to reducing the rate of oxidative metabolism by regulating the fatty acids composition of the cells (Leenen et al. 2002).

As it is known, most of the oils are obtained from oily seeds. For example, plant sterols/stanols are important bioactive components in the structure of plants with beneficial properties for human health. The three main plant-derived sterols are campesterol (24- α -methylcholesterol), β -sitosterol(24- α -ethylcholesterol), and stigmasterol (Δ 22, 24- α -ethylcholesterol).

Plant sterols are structurally similar to cholesterol. The foods having the highest plant sterol content are oilseeds, especially nuts. It contains sterols in its structure according to the oil seed from which it is obtained. All vegetable oils, especially safflower oil (Δ 7 stigmastanol), rapeseed oil (brassicasterol) and olive oil (β -sitosterol) have high plant sterols content. Although it has been known for years that especially the plant sterols have cholesterol-lowering effect (Çekici and Yıldırım 2019; Ateş and Veliöğlu2014; Köhler et al. 2017).

In this study, it was aimed to determine the fatty acid profile of 17 different oils that we consume directly in our daily life as flavoring in meals, salads, black cumin and safflower oil, as well as in diets applied in certain diseases, and also to determine the antioxidant activities and the presence of β -sitosterol in their structures. Besides, the fatty acids of the oils were detected, and the ratios of saturated and unsaturated fatty acids were determined.

2. Materials and Methods

2.1. Material

The samples used in this study were obtained from Zade oil factory in Konya, a brand that is widely recognized in our country. A total of 17 different oil types were used such as soybean, corn, safflower, canola, sunflower, cotton, linseed, black cumin, pumpkin seeds, walnut, olive oil, extra virgin olive oil, mixed pomace olive oil, grape seed, sesame, poppy and peanut oils.

2.2. Determination of Antioxidant Activity

The 2,2'-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging activity method was used to determine the antioxidant activity in oils which method was discovered by Blois (1958), and modified method was applied to the extracts by Hatano et al. (1989). The scavenging effects of oil samples against DPPH free radical were determined by measuring the color transformation spectrophotometrically at a wavelength of 515 nm in the UV/visible region. The DPPH stock solution was weighed in the required amount at a concentration of 6×10^{-5} mol/l and it was dissolved in ethanol (75%). 300 μ l of each oil sample was taken into the test tubes with the help of a micropipette and 2700 μ l of DPPH solution was added to them. Then, the tubes were incubated at room temperature in the dark for 20 minutes. At the end of the period, the absorbance of the samples was read in a blinded ethanol spectrophotometer (Agilent Technologies UV-Visible spectrophotometer, Germany) at 515 nm wavelength. The % inhibition of the samples against DPPH free radical was calculated according to the formula given below. Each sample was run at three different concentrations as 25 mg/ml, 50 mg/ml and 100 mg/ml. Each sample was run in 3 parallels and the results were given by calculating the standard deviation as the mean % scavenging effect of 3 parallels.

2.3. Determination of the Presence of Beta-Sitosterol by TLC (Thin Layer Chromatography)

The given statement is a definition of solid-liquid adsorption chromatography known as Thin Layer Chromatography. The mobile phase moves from the bottom to up over the stationary phase in this method. Chloroform:diethylether (1:1 v/v) solvents were used to examine the presence of beta-sitosterol in oils. The presence of beta-sitosterol in oils was determined by the retention factor, through reaching the highest level in the solvent system (European Pharmacopoeia 2021).

2.4. Determination of Fatty Acid Compositions

2.4.1. Methylation in Oils

The methylation process in oils is based on the extraction of fatty acids by separating the fatty acids and glycerol in their structure. The process of esterification was performed in accordance with the European Pharmacopoeia. First of all, 450 mg (0.45 g) oil sample was weighed into a 50 ml volumetric flask and 12 ml of 0.5 N methanolic NaOH was added on it and it was left in the heater for 10 minutes (approximately 80°C) until the oil and solution were mixed. When saponification took place, 20 ml of BF₃/MeOH was added to the mixture and it was kept in the heater for 2 minutes. At the end of the period, the flask was filled up to the 50 ml line of the volumetric flask with saturated NaCl solution. After adding 1 ml of hexane and performing the phase separation, the top part was taken and transferred to the vial and given to GC-MS for reading (Eryilmaz et al. 2015; Orhan Erdogan et al. 2013).

2.4.2. Chromatographic Conditions

The fatty acids were determined using a Gas Chromatography Mass Spectrophotometer (Agilent 6890N Network GC system combined with Agilent 5973 Network Mass Selective Detector). The device was

equipped with an Agilent 19091N-136 column (HP Innowax Capillary; 60.0 m x 0.25 mm x 0.25 mm), and Helium was used as the carrier gas with a flow rate of 1.2 ml/min. The injection volume was 1 µl with a split ratio of 50:1, and the injector temperature was set to 250°C. The scanning range was 35-450 atomic mass units (AMU) with ionization by electron bombardment (EI - 70 eV).

The fatty acid compositions were determined using data from the Famed 23, Wiley, and Nist Mass Spectral Library. Retention indices were calculated based on the peak emergence time relative to n-alkanes.

3. Results and Discussion

3.1. Antioxidant Activity

Antioxidant activities of oil samples were determined at 3 different concentrations, 25 mg/ml, 50 mg/ml and 100 mg/ml. As seen in Table 1, soybean, corn and safflower oil showed the highest scavenging effect at all 3 concentrations. It was determined that soybean, corn, safflower and canola oils among the run oil samples showed the best antioxidant activity at 100 mg/ml concentration due to their DPPH radical scavenging effect of approximately 90% and above. These oils are also followed by sunflower oil, cottonseed oil, linseed oil, black cumin oil, pumpkin seed oil and walnut oil, whose activities vary between 70-80%, respectively. Of the oils tested, it is worth noting that peanut oil exhibited a lower antioxidant activity of 47.30% compared to the others, which showed an intermediate antioxidant activity of less than 70%. Although it has a high amount of vitamin E in its structure, when its fatty acid profile is examined, the unsaturation/saturation ratio was found lower than other oils. When it is considered that fatty acids are associated with antioxidant activity, the result shows parallel. The soybean oil is consumed in certain diets in the world as a source of antioxidant activity due to the polyphenols and tocopherols (Vitamin E) in its structure. At the same time, safflower oil has high antioxidant activity because of its rich Vitamin E content. Also, the corn oil has high antioxidant activity because of its high Vitamin E (about 13%- Shoemaker 2019) and phyosterols content. The presence of high amounts of oleic and linoleic acids found in soy, corn and safflower oils is also effective in their high antioxidant activities because vegetable-based edible oils contain compounds having natural antioxidant activity. There is ascorbic acid, α -tocopherol, beta-sterol, β -carotene, flavonoids among these compounds Further investigation is necessary to determine the specific components in vegetable-based edible oils that contain antioxidant compounds and to better understand their benefits (Uluata 2010). Antioxidant activities of some oils are also high, especially due to the sterols, phenolic compounds and tocopherols found in their structures (Tuberosso et al. 2007; Stuchlik and Zak 2002).

3.2. Determination of Beta-Sterol Presence with TLC

The presence of beta-sitosterol was investigated in oils using thin layer chromatography and chloroform: diethylether (1:1 v/v) solvent system and it was determined that beta-sitosterol was present in all of them. Beta-sitosterol is one of the plant sterols and it is especially important in terms of lowering high cholesterol. Another subject about vegetable-based edible oils also has their own plant sterols they gain functionality to each oil. In this respect, the presence of beta-sitosterol in the oils used in this study shows that these oils can be used both in diets and in nutrition for heart disease and cholesterol.

3.3. Determination of Fatty Acid Compositions

Table 2 gives the fatty acid profiles of the oils used in this study. The fatty acids in the structure of oils are classified as saturated and unsaturated fatty acids in Table 3. Table 4 shows the relationship between saturated, mono and polyunsaturated fatty acid profiles and saturated and unsaturated fats in oils (Figure 1,2).

According to Table 2, the saturated fatty acids determined in oils are myristic (C14:0), palmitic (C16:0), stearic (C18:0), and arachidic (C20:0) acid. Myristic acid was found to be the most abundant, with a concentration of 0.634%, in peanut oil. The palmitic acid was determined at the highest rate with 20.857%, in cotton oil; it was found at the lowest rate with 5.33% in canola oil. Stearic acid was determined as 9.182 % in peanut oil as the highest and 1.851% in canola oil as the lowest rate. Arachidic acid was found at the highest

rate in mixed pomace oil with 0.654%, in canola oil with 0.637% and in pumpkin seed oil with 0.599%, respectively. No arachidic acid was determined in safflower oil.

Table 1. % Inhibition of oils to DPPH radical.

Oils	% Inhibition of Oils to DPPH Radical ^{a±SD^b}		
	Concentration		
	25 mg/ml	50 mg/ml	100 mg/ml
Soybean	52.16±1.04	72.95±0.52	94.30±1.57
Corn	52.06±1.99	72.49±0.33	94.27±0.78
Safflower	50.65±0.43	69.64±0.72	93.01±0.35
Canola	48.18±0.99	64.91±0.42	89.46±1.85
Sunflower	41.84±1.24	57.34±0.59	79.67±0.98
Cottonseed	42.18±1.15	56.59±0.49	77.88±0.46
Linseed	43.71±0.80	60.06±1.02	76.32±1.37
Black cumin	39.96±0.30	52.99±1.07	72.64±0.74
Pumpkinseed	40.35±1.22	52.73±0.65	72.44±1.21
Walnut	38.56±1.07	50.99±0.86	72.03±0.20
Mixed Pomace olive	38.74±0.65	48.41±1.28	66.11±0.93
Olive	37.38±0.38	47.98±1.38	65.03±0.48
Grapeseed	38.97±0.72	47.36±0.73	63.95±0.26
Sesame	37.71±0.69	47.97±0.82	63.62±0.37
Extravirgin olive	35.00±0.97	44.71±0.92	60.42±0.44
Poppy	36.23±1.73	43.62±0.89	57.33±0.89
Peanut	32.34±1.00	38.83±0.52	47.30±0.70

SD: Standard deviation

Monosaturated fatty acids were identified as palmitoleic acid (C16:1), oleic acid (C18:1) and eicosanoic acid (C20:1). Palmitoleic acid was highest in canola oil (1.851%) but it was not detected in safflower oil. As it is known, oleic acid is found the highest fatty acid in olive oil. The oleic acid was detected with 74.15% in olive oil, 70.952% in extra virgin olive oil and 67.72% in mixed pomace oil in this study. The lowest rate was detected in canola oil with 0.102% rate. While eicosanoic acid was detected highest in canola oil (1.233%), it could not be detected in safflower oil.

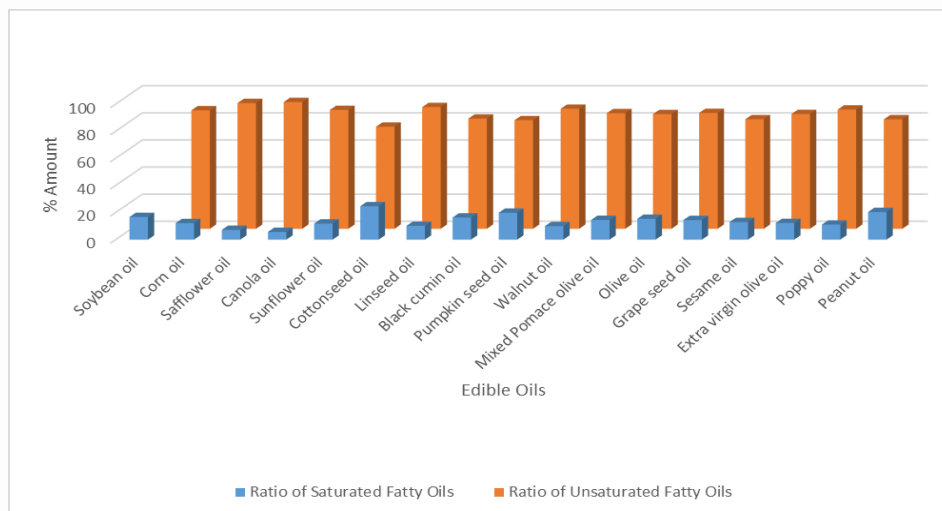


Figure 1. The Sum of Saturated and Unsaturated Fatty Acids in the Structure of Edible Oil

Table 2. Fatty acids methyl esters of oils (%).

Oils	Myristic Acid (C14:0)		Palmitic Acid (C16:0)		Palmitoleic Acid (C16:1)		Stearic Acid (C18:0)		Oleic Acid (C18:1)		Linoleic Acid (C18:2)		Linolenic Acid (C18:3)		Arachidic Acid (C20:0)		Eicosanoic Acid (C20:1)	
	RI*	%	RI	%	RI	%	RI	%	RI	%	RI	%	RI	%	RI	%	RI	%
	Soybean	-	0	1292	11,003	1296	0,076	1547	5,174	1560	25,991	1584	50,993	1754	5,967	1885	0,55	1921
Corn	-	0	1292	9,43	1296	0,054	1547	2,534	1560	28,965	1584	56,89	1754	1,32	1885	0,216	1921	0,231
Safflower	975	0,07	1292	5,485	-	0	1547	1,627	1560	28,727	1584	63,997	1754	0,093	-	0	-	0
Canola	975	0,05	1292	5,33	1296	1,851	1592	0,325	1618	0,102	1667	84,846	1755	5,556	1886	0,637	1921	1,233
Sunflower	975	0,049	1292	6,079	1296	0,056	1592	5,744	1618	28,23	1667	59,091	1755	0,186	1886	0,404	1921	0,16
Cotton seed	975	0,552	1292	20,857	1296	0,417	1592	2,906	1618	19,766	1667	54,628	1755	0,381	1886	0,332	1921	0,161
Linseed		0	1290	5,528	1296	0,072	1586	4,44	1610	24,965	1665	17,309	1752	47,315	1883	0,193	1921	0,178
Black cumin	975	0,149	1290	12,115	1296	0,18	1586	3,793	1610	24,646	1665	55,728	1752	0,442	1883	0,271	1921	0,34
Pumpkin seed	975	0,089	1290	11,067	1296	0,094	1586	8,098	1610	39,238	1665	40,531	1752	0,15	1883	0,599	1921	0,133
Walnut	975	0,022	1290	6,812	1296	0,129	1585	3,048	1609	18,044	1665	60,251	1752	9,939	1882	0,133	1921	0,243
Mixed Pomace olive	-	0	1290	12,03	1296	0,487	1584	1,869	1609	67,72	1665	15,993	1752	0,699	1882	0,654	1921	0,428
Olive	975	0,012	1290	11,511	1296	0,685	1584	3,301	1608	74,15	1664	8,849	1752	0,603	1880	0,542	1921	0,347
Grape seed	-	0	1290	9,041	1296	0,158	1583	5,217	1607	22,161	1662	62,634	1751	0,394	1884	0,233	1920	0,162
Sesame	-	0	1290	9,015	1296	0,142	1583	3,521	1607	41,683	1662	38,291	1751	0,537	1884	0,432	1920	0,174
Extra virgin olive	-	0	1290	9,203	1296	1,355	1583	2,771	1607	70,952	1662	11,33	1751	0,856	1884	0,327	1920	0,205
Poppy	975	0,05	1290	8,293	1296	0,135	1583	2,762	1607	14,84	1662	72,431	1751	0,558	1884	0,102	1920	0,061
Peanut	975	0,634	1290	9,171	1296	0,113	1583	9,182	1607	18,252	1662	58,110	1751	2,223	1884	0,22	1920	0,095

Table 3. Classification of saturated and unsaturated fatty acids (%).

Oils	Saturated Fatty Acids					Unsaturated Fatty Acids			
	Myristic Acid (C14:0)	Palmitic Acid (C16:0)	Stearic Acid (C18:0)	Arachidic Acid (C 20:0)	Palmitoleic Acid (C16:1)	Oleic Acid (C18:1)	Linoleic Acid (C18:2)	Linolenic Acid (C18:3)	Eicosanoic Acid (C20:1)
Soybean	0	11,003	5,174	0,550	0,076	25,991	50,993	5,967	0,246
Corn	0	9,430	2,534	0,216	0,054	28,965	56,890	1,320	0,231
Safflower	0,070	5,485	1,627	0	0	28,727	63,997	0,093	0
Canola	0,050	5,330	0,325	0,637	1,851	0,102	84,846	5,556	1,233
Sunflower	0,049	6,079	5,744	0,404	0,056	28,230	59,091	0,186	0,160
Cotton seed	0,552	20,857	2,906	0,332	0,417	19,766	54,628	0,381	0,161
Linseed	0	5,528	4,440	0,193	0,072	24,965	17,309	47,315	0,178
Black cumin	0,149	12,115	3,793	0,271	0,18	24,646	55,728	0,442	0,340
Pumpkin seed	0,089	11,067	8,098	0,599	0,094	39,238	40,531	0,150	0,133
Walnut	0,022	6,812	3,048	0,133	0,129	18,044	60,251	9,939	0,243
Mixed Pomace olive	0	12,030	1,869	0,654	0,487	67,720	15,993	0,699	0,428
Olive	0,012	11,511	3,301	0,542	0,685	74,150	8,849	0,603	0,347
Grape seed	0	9,041	5,217	0,233	0,158	22,161	62,634	0,394	0,162
Sesame	0	9,015	3,521	0,432	0,142	41,683	38,291	0,537	0,174
Extra virgin olive	0	9,203	2,771	0,327	1,355	70,952	11,330	0,856	0,205
Poppy	0,050	8,293	2,762	0,102	0,135	14,84	72,431	0,558	0,061
Peanut	0,634	9,171	9,182	0,220	0,113	18,252	58,110	2,223	0,095

Canola oil contains the highest percentage of linolenic acid at 84.846%, followed by the other oils which contain lower amounts of this polyunsaturated fatty acid. The analysis revealed that the oils contain two polyunsaturated fatty acids: linoleic acid (C18:2) and linolenic acid (C18:3). It was detected in poppy oil with 72.431% and safflower oil with 63.997%, respectively. The lowest linoleic acid content of 8.849% was found in olive oil. While flaxseed oil has the highest linolenic acid content with 47.315% rate. It was determined as 9.939% in walnut oil and 5.967% in soybean oil. It was determined at the lowest level as 0.093% in safflower oil.

The major fatty acid detected in soybean oil is linoleic acid with rate of 50.993%. While oleic acid was determined at a rate of 25.99% and palmitic acid was determined at a rate of 11.003%, myricitic acid was not detected. On the other hand, the main fatty acid of corn oil is linoleic acid and it was found at a rate of 56.89%. The oleic acid was determined as 28.965% and palmitic acid was determined as 9.43%. The linoleic acid was found as the primary fatty acid in safflower, canola, sunflower, cotton, black cumin, pumpkin seeds, walnut, grape seed, poppy and peanut oils. It was determined as 84.846%, the highest rate in canola oil what was determined lower in walnut oil at a rate of 40.531%. The main fatty acid in linseed oil is linolenic acid and it was determined at a rate of 47.315%. The main fatty acid is oleic acid in olive oil, extra virgin olive oil, mixed pomace oil and sesame oil. Oleic acid was determined as 41.683% in sesame oil and 74.15% in olive oil.

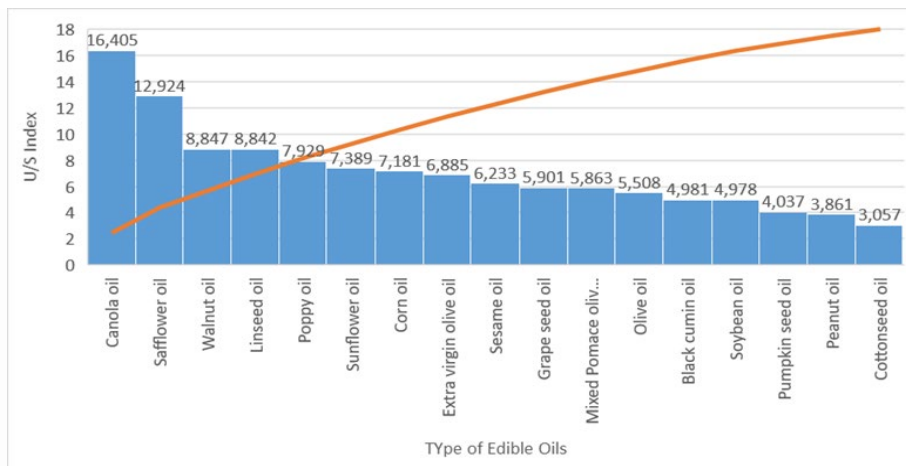


Figure 2. Ratio of unsaturated/saturated index of edible oils

The myristic (0.027-0.079%), palmitic (8.995-11.802%), stearic (4.035-6.800%), oleic (21.048-25.390%), linoleic (50.982-56.071), linolenic (5.162-6.494%) and arachidic (0.349-1.002%) acid were determined as main components in a study conducted with soybean oil (Jokic et al. 2013). In another study, it was determined that the palmitic and stearic acid contents of soybean oil were in the range of 10.63-11.43%, 3.76-4.61%, respectively (Esteves et al. 2010). Galao et al. (2014) determined the oleic, linoleic and linolenic acid ratios of soybean oil as 13.46-20.63%, 53.32-59.34%, 7.38-11.89%, respectively. Most of the fatty acids in corn oil are unsaturated same as in other oils. Corn oil contains palmitic acid (16:0), stearic acid (18:0), oleic acid (18:1), and linoleic acid (18:2) among its main fatty acids. The highest fatty acid in corn oil is linoleic acid (Moreau 2002). It was determined that palmitic, palmitoleic, stearic, oleic, linoleic, linolenic, arachidic and eicosanoid acid contents in corn oil are respectively; 11.39%, 0.10%, 1.83%, 29.90%, 54.57%, 0.97%, 0.40% and 0.25% (Kerr et al. 2016). Corn oil was detected that it contains high levels of linoleic acid (64.6%) and oleic acid (21.6%) in another study (Kar Eryilmaz 2009). Yılmaz (2018) determined oleic (8.805-34.266%) and linoleic (53.528-83.221%) acids as major fatty acids in the varieties of safflower oils in his thesis study. fatty acid compositions of safflower oil were determined as palmitic acid 3.90-6.80%, stearic acid 1.10-4.50%, oleic acid 6.20-81.90% and linoleic acid 11.00-83.10% (Johson et al. 1999), linoleic acid 70.30-78.80%, oleic acid 82.10-62.70% (Cazzato et al. 2001), palmitic acid 6.0-8.5%, stearic acid 2.0-3.1%, oleic acid 7.8-30.6% and linoleic

acid 6% 0.0-81.6% in other studies conducted on safflower (Uysal 2006), what was determined, myristic (<0.1%), palmitic (5.2-10.5%), stearic (4.4.-6.9%), oleic (23.2-59.5%), linoleic (18.8-15.2%), linolenic (11.9-44%) and arachidic (0.11-0.18%) acid in the study conducted on canola (Bauer et al. 2015). As a result of study canola can be classified as low linolenic acid, high oleic acid, high lauric acid content (Moreau 2002). The sunflower oil contains averagely 69-70% linoleic acid, 20% oleic acid, 10-11% palmitic and stearic acid (Sunflowersa 2022). Also, it is classified into 3 groups as low (10-29%), medium (30-59%) and high oleic acid (60-90%) sunflower oil (Pacurenau-Joita et al. 2005). It has been reported that 20-30% oleic, 60-70% linoleic and <1% linolenic acid in sunflower oil obtained from conventional seeds; 25-75% oleic, 15-35% linoleic and <1% linolenic acid in sunflower oil containing medium oleic acid; sunflower oil containing high oleic acid contains 80-90% oleic, 5-9% linoleic and <1% linolenic acid (Moreau 2002). Another detection that the cottonseed oil has 0.78-0.80% myristic, 24.85-25.63% palmitic, 0.54-0.57% palmitoleic, 3.01-3.13% stearic, 14.06-17% oleic, 52-55.82% linoleic, 0.12-0.14% linolenic, and 0.29%- 0.31 arachidic acid in a conducted study (Konuskan Bozdoğan et al. 2017). Flaxseed oil has low (about 7-10%) saturated fatty acids while it has high unsaturated fatty acids (80-93%). According to a study conducted in Flaxseed oil, what has been reported that it has averagely 15.8-62.5% oleic, 17.8-19.6% linoleic and 10-58.3% linolenic acid (Lewinska et al. 2015). In another study, fatty acid profiles of flaxseed varieties were examined, and it was determined that there were 17-24.8% oleic, 10.2-13.1% and 47.8-59.9% linolenic acids (Bertrand and Özcan 2017). Black seed oil is important as a functional oil due to its the thymoquinone content at the same time black seed oil also has a high unsaturation rate as an fatty acid profile. Lutterodt et al. (2010) found 2.56-2.80% stearic, 22.63-24.51% oleic, 58.83-61.20% linoleic and 0.21-0.28% linolenic acid in 6 groups of cold-pressed black cumin oil. The linoleic acid (18:2) was found 66.5% and oleic acid (18:1) was found 23.5% in another study (Çiftçi et al. 2011). The essential fatty acids of pumpkin seed oil were detested as oleic (17.0-39.5%), linoleic (18.1-62.8%), palmitic (12.6-18.4%) and stearic 5.1-8.5) acids. Additionally, the most striking feature of pumpkin seed oil in terms of fatty acid composition content is that the ratio of total unsaturated fatty acid (80.65%) is quite high compared to total saturated fatty acid (19.35%) (Ardabili et al. 2011). Akın (2016) determined 5.260-5.290% stearic, 27.520-27.590% oleic, 53.190-53.270% linoleic and 0.390-0.440% linolenic acid fatty acids in pumpkin seed oil in his thesis study. In a study on the fatty acids contained in walnut types, the linoleic acid content was determined as highest between 50.24-60.60%, respectively followed by 20.70-28.33% oleic acid and 10.93-15.04% linolenic acid. Besides, monounsaturated fatty acids between 22.17-29.73% and saturated fatty acids between 4.00-7.86% were obtained in the study (Şimşek 2016). Walnut is an important oil source for the brain because of its high amount omega-3 fatty acids content. According to Unver and Celik's (2005) research, the walnut types they examined exhibited different ratios of linoleic acid (ranging from 41.13% to 61.15%), oleic acid (ranging from 22.39% to 49.12%), palmitic acid (ranging from 6.01% to 10.21%), and stearic acid (ranging from 2.17% to 4.99%). About 90% of grape seed oil consists of mono and polyunsaturated fatty acids which contains especially high amount of linoleic acid (58-78%) and 3-15% oleic acid and contains about 10% saturated fatty acids (Bail et al. 2008, Rombaut et al. 2015; Konuşkan et al. 2019). It was reported that grape seed oil contains palmitic (6.6%), stearic (3.5%), oleic (14.3%), linoleic (74.7%) and linolenic acid (0.15%) in another study (Orsavova et al. 2015). The fatty acid components of sesame oil are approximately 7-12% palmitic, 0.35-6% stearic, 35-50% oleic, 35-50% linoleic and 0.30-0.80% linolenic acid (BÜF, 2022). Sesame is considered to be one of the oldest and most significant oil plants cultivated globally. Unlike other vegetable oils, sesame oil contains oleic and linoleic fatty acids, each ranging from 35 to 45% (Liu et al., 1992). The fatty acids in the sesame oil were determined as myristic, palmitic, palmitoleic, stearic, oleic, linoleic, linolenic and arachidic acids, and the main components were determined as oleic acid (39.67-41.05%) and linoleic acid (42.09-43.31%) in the study conducted in 2021 (Özpolat et al. 2021). Poppy seed fatty acids are linoleic (62-72%), oleic (10-30%), palmitic (9-10%), stearic (1.5-2.5%) and linolenic (0-5%) acids (Cameo 2022). In the 2014 thesis study examining the fatty acid distribution of oils from various poppy seeds, it was found that linoleic acid (C18:2) had the highest ratio. It was stated that it varied between 65.52-74.97%. The fatty acid with the highest ratio after linoleic acid was oleic acid (13.26-21.43%). The other fatty acid with the highest ratio was determined as palmitic acid (8.65-10.06%) (Abudak 2014). The main fatty acid component of peanut oil was oleic acid (45-53%), while linoleic acid (27-32%) and palmitic acid (11-14%) were other fatty acids (Ghazani and Marangoni 2016). Then the oleic acid was found as 37.7-82.2%, linoleic

acid 2.9-41.5%, palmitic acid 9.6-13.2%, stearic acid 1.6-3.7%, arachidic acid 1.2-1.7% in studies carried out in peanut oil (Dwivedi et al. 1996, Hassan and Ahmed 2012, Chowdhury et al. 2015). One of the differences of peanut oil from other oils is the ratio of oleic acid to linoleic acid (O/L value) and this ratio determines the nutritional value, storage time and shelf life of both the oil and the products in which peanut oil is used. If this value is high (>10:1), the shelf life is reduced, if it is low (1.5/1), the shelf life is extended (Chamberlin et al. 2014). Although the fatty acid ratios in the olive oil composition vary depending on various factors, it generally consists of 50-83% oleic acid, 7-20% palmitic acid and 3-20% linoleic acid (T.G.K. 2014). The fatty acid composition of 10 different samples of olive oils sold in Nizip and its surroundings was analysed in a 2012 study. The results showed that oleic acid had the highest rate (62.430-71.321%), followed by linoleic acid (7.216-11.825%) and palmitic acid (2.260-12.016%) (Türkoğlu et al. 2012). They determined that the oleic and linoleic acid values of a total of 103 olive oil samples produced with different systems (classical and modern) in Izmir during two different harvest periods varied between 67.68-74.16% and 8.72-13.89% (Diraman et al. 2009).

Table 4. Ratio of saturated, monounsaturated and polyunsaturated fatty acids (%).

Oils	Saturated Fatty Acids Ratio	Monounsaturated Fatty Acids Ratio	Polyunsaturated Fatty Acids Ratio	Unsaturated/Saturated Ratio	Total
Soybean	16,727	26,313	56,960	4,978	100
Corn	12,180	29,250	58,210	7,181	99,64
Safflower	7,182	28,727	64,090	12,924	99,99
Canola	5,705	3,186	90,402	16,405	99,293
Sunflower	11,872	28,446	59,277	7,389	99,595
Cotton seed	24,647	20,344	55,009	3,057	100
Linseed	10,161	25,215	64,624	8,842	100
Black cumin	16,328	25,166	56,170	4,981	97,664
Pumpkin seed	19,853	39,465	40,681	4,037	99,99
Walnut	10,015	18,416	70,190	8,847	98,621
Mixed Pomace olive	14,553	68,635	16,692	5,863	99,880
Olive	15,366	75,182	9,452	5,508	100
Grape seed	14,491	22,481	63,028	5,901	100
Sesame	12,968	41,999	38,828	6,233	93,795
Extra virgin olive	12,301	72,512	12,186	6,885	96,999
Poppy	11,102	15,036	72,989	7,929	99,127
Peanut	20,407	18,46	60,333	3,861	99,2

The literatures related to the oils used in this study are given respectively. Fatty acid components obtained from all edible oils except canola, flaxseed, black cumin and peanut oils show parallelism with studies by other researchers. Considering that the canola plant is classified according to its oleic acid content, it can be said that the canola used in this study is a low oleic acid canola variety. As it is known, there are 2 different types of flaxseeds (yellow and brown), and since it is not known which kind of flaxseed oil is used here, the main fatty acid is linolenic acid according to the results obtained. Palmitic, oleic and linoleic acids constitute the main fatty acid profile of peanut oil (Carrin and Carelli 2010). Therefore, the results of the study show parallelism with the literature. When the method used to obtain oil from black cumin seeds and the storage times of the seed are considered, it is seen that obtained data in other studies are close to each other.

Table 4 presents the relationship between unsaturation and saturation ratio of oil acids in the structure of oils. This parameter is crucial in determining the nutritional value of oils and whether they possess functional properties. When the unsaturated/saturated ratio is greater than 1, it means that the nutritional

value is high, and this means that this oil can be used in diets, cholesterol, high blood pressure, etc. states that it can be easily consumed in certain diseases (Lawton et al. 2000). According to result, the highest was determined in Canola (16.405%), Safflower (12.924%) and sunflower (7.389%), while the lowest was found in flaxseed (3.057%) and Peanut (3.861%) oil. It was determined 3.2-3.4% in canola, 10.55% in safflower, 6.76% in sunflower and 7.05% in flaxseed in the study conducted by Kostik et al. (2013), on the other hand, Zambiasi et al. (2007) determined it 4.10% in canola, 5.80-5.95% in sunflower, 6.56% in flaxseed and 1.70% in peanut. The obtained values according to these results show almost parallelism.

Changes in fatty acid compositions differ depending on the obtained oil seed type, climate and environmental conditions, soil structure and growing season. However, different maturation times of oilseeds should also be considered. In addition, it is expected that there will be a change in fatty acid profiles when the storage conditions of oilseeds are taken into account from the harvest to the period when oil is obtained. Finally, the extraction method of the oils can also affect the fatty acid composition.

As it is seen above mentioned, since the ratio of unsaturated fatty acids is high in all of the oils used in the study, it can be said that fatty acids can be easily used in certain diets, especially with cholesterol, diabetes and heart diseases.

Author Contributions: Methodology, software, formal analysis, investigation, resources, data curation, writing—original draft preparation, writing—review and editing, S.A.Ç. All authors have read and agreed to the published version of the manuscript.

Funding: This research received no external funding.

Conflicts of Interest: The authors declare no conflicts of interest.

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Use of An Animal-Derived Biostimulant for Alleviating the Effects of Drought Stress on Sugar Beet

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HIGHLIGHTS

- Recent studies have focused on the ability of biostimulants to alleviate plant stress.
- Sugar beet is extremely sensitive to the lack of water during the early growth stages.
- Foliar biostimulant applications may reduce the adverse effects of drought stress to the growth of sugar beet seedlings.
- The beneficial effects of animal-derived biostimulant were determined to mitigate drought stress.

Abstract

This study focused on mitigating effects of an animal-derived biostimulant on sugar beet plantlets subjected to drought stress. The experiment was performed at the Seed Science and Technology Laboratory of the Eskişehir Osmangazi University, Faculty of Agriculture, Department of Field Crops, in 2024. It was established by the randomized plot 2×5 factorial experimental design (ANOVA) with four replications. The sugar beet cultivar Mohican was sprayed by an animal-derived biostimulant (Andolamin®) containing 11% amino acids. Different levels of the biostimulant (control, 12.5, 25, 50, and 75 mL/L) were treated twice at 2-day intervals. Morphological and physiological measurements were made at 7 days after the first application on sugar beet plants grown under two irrigation regimes (water deficit (WD) 50% of field capacity and well-watered (WW) 80% of field capacity). The findings showed that drought had a hazardous impact on sugar beet's number of leaves (NL), fresh (LFW) and leaf dry weight (LDW), relative water content (RWC), and leaf area (LA). Leaf surface temperature (LST), chlorophyll content (Chl), and electrolyte leakage (EL) were higher in plants under water deficit. Foliar biostimulant application mitigated the effect of drought stress on seedlings through improving LFW, LDW, Chl, EL, and LA. On the other hand, biostimulant treatment had no significant effects on NL, and RWC in seedlings exposed to drought stress. It was concluded that animal-derived biostimulant application may be used for alleviating the harmful effects of drought stress and may stimulate the growth of sugar beet seedlings.

Keywords: *Beta vulgaris* L.; drought stress; tolerance; biostimulant

Citation: Kulan EG, Kaya MD (2024). Use of an animal-derived bio stimulant for alleviating the effects of drought stress on sugar beet. *Selcuk Journal of Agriculture and Food Sciences*, 38(2), 356-367. <https://doi.org/10.15316/SJAIFS.2024.032>

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Received date: 05/12/2023

Accepted date: 01/08/2024

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1. Introduction

The sugar beet (*Beta vulgaris* L.), a member of the *Chenopodiaceae* family, is a biennial plant adapted to arid and semi-arid climates (Khan et al. 2018). Its optimum development occurs at moderate or cool temperatures, typically between 10 and 20 °C (Lombardi et al. 2022). The crop has multiple uses in manufacturing industries, including sugar production (Mall et al. 2021).

Around the world, a wide range of environmental stresses such as heat, salt, flooding, and drought have an adverse impact on every stage of crop development during the life cycle (Ghadimezhad Shiade et al. 2023). Drought affects approximately fifty percent of the earth's arid and semi-arid areas (Nadeem et al. 2019) and is a prolonged period of drastically less rainfall in a certain region (Saeidnejad et al. 2016). A shortage of water may occur from many factors, including insufficient rainfall, unequal amounts of precipitation, the intensity and duration of droughts, and the rate of stress accumulation (Pessarakli and Marcum 2013). In response to drought conditions, plants reduce leaf pigments and close stomata reducing photosynthesis, growth, and other important physiological and biochemical processes (Yan et al. 2016; Saady et al. 2021; McDowell et al. 2022; Pepe et al. 2022) and have a considerable detrimental impact on plant growth, development, and production (Muhammad Aslam et al. 2022; Yahaya and Shimelis 2022).

Many field crops have a higher vulnerability to low temperatures and drought, particularly during germination and seedling growth. Drought stress hinders seed imbibition, thereby inhibiting germination (Islam et al. 2018). Also, it limits the beet yield (Monreal et al. 2007) and quality (Nause et al. 2020) of sugar beet production. Lack of water availability leads to reduced growth and yield (Salem et al. 2021). This occurs due to the generation of reactive oxygen substances (ROS), which begin lipid peroxidation of membranes and interaction with other macromolecules (Bistgani et al. 2017). Sugar beet is extremely affected by water limitations, mostly during its early growth stages, and there is a positive correlation between consumption of water and root yield. Also, pathogenic and natural factors can cause seedling damage, hence delaying seedling emergence prolongs the critical stage of growth and increases the risk. Genetic and environmental factors, as well as the effects of seed pre-treatment and seeding applications, strongly influence the stages of germination and seedling development in sugar beet (Helaly et al. 2009).

Different groups classify plant biostimulants, including seaweed extracts, humic and fulvic acids, beneficial chemical elements, chitin and chitosan, microorganisms, inorganic salts, peptides, and amino acids (Du Jardin 2015; Rouphael and Colla 2020). These biostimulants promote plant growth by stimulating natural processes like nutrient uptake performance and stress resistance (Abdel Latef et al. 2021; Attia et al. 2021). Also, they have practical applications in agriculture (Bulgari et al. 2019). Applying biostimulating substances in small quantities, either to the soil or to the leaves of plants, can have a beneficial impact on plant growth (Barone et al. 2019). These substances encourage the development of both roots and shoots, enhancing the rate of photosynthesis, and improving the quality of crops such as beans (Abrantes et al. 2011; Latif and Mohamed 2016), pepper (Palangana et al. 2012), wheat (Hammad and Ali 2014; Farooq et al. 2017), fenugreek (Abdel Latef et al. 2017), tomato (Colla et al. 2017), soybean (Kocira et al. 2018), and sorghum (Ahmad et al. 2016). Biostimulants improve the capacity of plants to tolerate drought stress (Ertani et al. 2013; Colla et al. 2015). Paul et al. (2019) documented enhanced tomato growth in response to biostimulant treatment during periods of drought induced. Hence, products containing biostimulant properties appear as an effective choice for combining seedling production. This leads to the growth of strong seedlings with optimal nutrient levels and well-established root systems. Additionally, these products promote an increase in leaf area and number, thereby enhancing photosynthetic capacity. Consequently, these seedlings demonstrate optimum growth, ultimately resulting in high-quality crop yields. (Bettoni et al. 2022).

2. Materials and Methods

An experiment was conducted at the Seed Science and Technology Laboratory of the Eskişehir Osmangazi University, Faculty of Agriculture, Department of Field Crops, in 2023. Seeds of cultivar Mohican were sown in vials filled with a mix of soil:peat:perlite:vermiculite (4:5:1:1). When the seedlings reached 2 true leaves, they

were transplanted to plastic cups (75×95 mm) and irrigated at field capacity. The plants were grown in a climate chamber at 24/18 °C with 16 hours of light / 8 hours of darkness. Andolamin® was used as an animal-derived biostimulant. The product is composed of 55% organic matter; 25% organic carbon; 8.5% organic nitrogen; and 11% free amino acids, with a pH of 5.8-7.8. The biostimulant application was carried out at intervals of 2 days, the first being at 27 days after sowing (DAS), via a foliar spray. At 34 DAS, all seedling samples of each replication were selected, and the following characters were evaluated as the number of leaves (NL), leaf fresh weight (LFW), leaf dry weight (LD), leaf surface temperature (LST), chlorophyll content (Chl), relative water content (RWC), electrolyte leakage (EL), and leaf area (LA). For LFW, samples were washed and weighed on an analytical balance. For the determination of LDW, samples were placed in an electric oven at 70°C ± 5°C for 48 h, then they were calculated. LST was determined by an infrared thermometer. Chl was read using a Konica Minolta SPAD-502 meter on the 3rd or 4th leaf of the seedlings. RWC was determined from two plants in each replicate, specifically from the mature leaves. Two leaves were taken from all replicates and promptly measured to determine the fresh weight (FW). Samples were soaked in distilled water in a falcon tube for 24 hours to regain turgidity and then the turgor weight (TW) was measured. The leaf samples were put in an air oven at 70°C for 48 h in order to determine the dry weight (DW). RWC of the leaves was detected in the formula by described Ghoulam et al. (2002), $RWC (\%) = (FW - DW)/(TW - DW) \times 100$ (Eq. 1). EL was analyzed by using the four discs of young leaf from each treatment. The leaf samples were washed with deionized water to get rid of any electrolytes remaining on the leaf surface. Four leaf disks with a 10 mm diameter were excised, weighed, and placed into glass tubes filled with 20 mL of deionized water. After the incubation period of 24 h at 25°C, the solution's EC (Lt) was directly read by the EC meter. Then, they were taken into the water bath for 45 minutes at 90°C, and the EC (Lo) was recorded again at 25°C after equilibration (Yadav et al. 2012). The electrolyte leakage was calculated by the formula (Ghoulam et al. 2002): $Electrolyte\ leakage (\%) = (Lt / Lo) \times 100$ (Eq. 2). After all leaves were cut and scanned in the scanner, leaf area (LA) measurements were calculated with the use of the Image J® software program.

The plots included two irrigation regimes of water deficit (WD) (50% of field capacity) and well-watered (WW) (80% of field capacity), and five doses of an animal-derived biostimulant (control, 12.5, 25, 50, and 75 mL/L). The Levene test was used to test for homogeneity. Data given in percentages were subjected to an arcsine transformation before statistical analysis. The data were analyzed by the randomized plot 2×5 factorial experimental design (ANOVA) with four replications. The MSTAT-C (Freed et al. 1991) statistical program was used for the analysis of variance, and a comparison of the means was performed using the Least Significant Difference (LSD) test ($p < 0.05$).

3. Results

For all characteristics except LST, there was a significant difference between the levels of drought, but biostimulant doses were significant for LFW, LDW, LST, Chl, EL, and LA (Table 1). Under drought stress, the NL, LFW, LDW, RWC, and LA were significantly suppressed; however, increasing biostimulant doses enhanced the LFW, LDW, RWC, EL, and LA. There was a significant interaction between drought stress and biostimulant doses for the morphological traits, except for NL. The highest (6.30 number per plant) and lowest (5.80 number per plant) NL were measured at a biostimulant dose of 25 mL/L, without and with drought stress, respectively. Sugar beet seedlings in well-watered conditions had a larger leaf size than in plants under drought (Fig. 1). LFW and LDW increased with gradually increasing biostimulant doses, and significant differences were determined (Fig. 2). Drought stress influenced all the physiological parameters of sugar beet seedlings. LST, Chl, and EL were higher in drought-stressed plants. The minimum Chl (53.2 SPAD) was recorded in seedlings applied with the lowest dose of biostimulant and it was enhanced by increasing doses, reaching the maximum value with 55.3 SPAD at a biostimulant level of 50 mL/L. Drought reduced RWC of leaves; however, increased biostimulant doses led to linearly induced RWC. For the EL, a significant interaction was observed (Fig. 3). The EL, with the minimum values (19.3% in drought and 13.9% in well-water) recorded at 25 mL/L of biostimulant, showed that higher ELs were observed under drought stress. Sugar beet seedlings produced a larger LA per plant as the biostimulant dose increased it considerably (Fig. 4). The LA per plant increased up to a dose of 50 mL/L, but it dropped at 75 mL/L, which was considered as overdose.

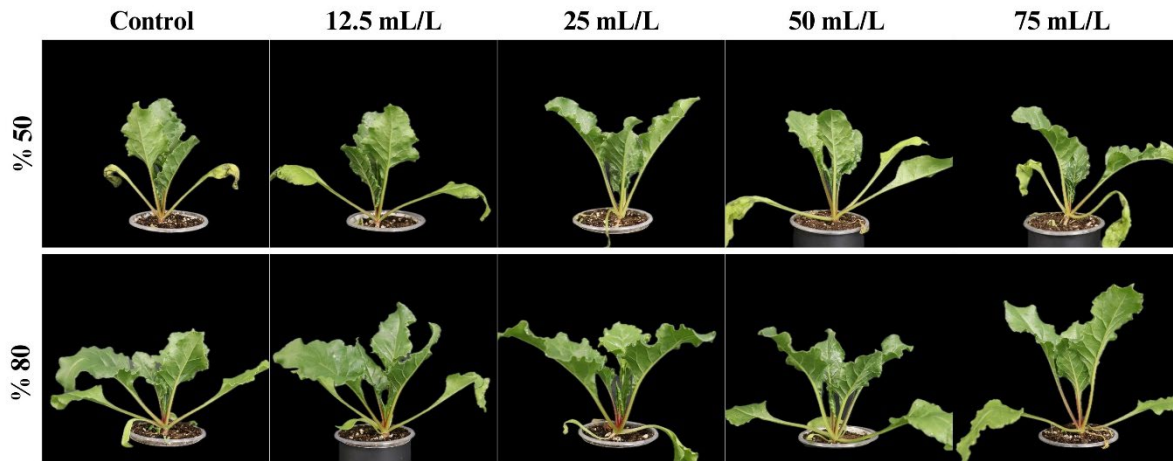


Figure 1. Front views of seedlings sprayed with different biostimulant doses under drought (50% of field capacity) and well-watered (80% of field capacity) conditions.

Table 1. Means and analysis of variance for the number of leaves, leaf fresh and dry weights, leaf surface temperature, chlorophyll content, relative water content, electrolyte leakage, and leaf area of sugar beet plants subjected to different biostimulant doses under well-watered and drought stress.

	Number of Leaves (number plant ⁻¹)	Leaf Fresh Weight (mg plant ⁻¹)	Leaf Dry Weight (mg plant ⁻¹)	Leaf Surface Temperature (°C)	Chlorophyll Content (SPAD)	Relative Water Content (%)	Electrolyte Leakage (%)	Leaf Area (cm ²)
Drought Stress (A)								
50%	5.40 ^b ± 0.13	4357 ^b ± 99.5	392 ^b ± 10.0	25.2 ± 0.15	60.9 ^a ± 0.55	63.4 ^b ± 0.55	22.9 ^a ± 0.63	93 ^b ± 2.30
80%	6.15 ^a ± 0.08	6658 ^a ± 64.1	518 ^a ± 10.0	25.0 ± 0.09	48.4 ^b ± 0.66	74.6 ^a ± 0.34	15.7 ^b ± 0.72	143 ^a ± 1.49
Biostimulant Dose (B)								
Control	5.88 ± 0.22	5375 ^c ± 509	457 ^a ± 36.4	25.1 ^{bc} ± 0.10	55.9 ^a ± 1.94	68.7 ± 2.27	18.6 ^b ± 2.18	115 ^b ± 8.71
12.5 mL/L	5.63 ± 0.26	5512 ^{bc} ± 537	464 ^{ab} ± 34.8	24.9 ^{cd} ± 0.15	53.2 ^c ± 2.58	68.8 ± 2.14	20.1 ^a ± 2.05	119 ^b ± 10.9
25 mL/L	6.00 ± 0.19	5650 ^b ± 377	489 ^a ± 15.3	25.3 ^b ± 0.15	54.8 ^{ab} ± 3.42	69.8 ± 2.29	16.6 ^c ± 1.04	120 ^b ± 10.4
50 mL/L	5.88 ± 0.23	5882 ^a ± 354	465 ^{ab} ± 18.4	24.5 ^d ± 0.10	55.3 ^{ab} ± 1.17	69.5 ± 1.57	21.1 ^a ± 0.46	126 ^a ± 6.55
75 mL/L	5.50 ± 0.19	5118 ^d ± 422	402 ^c ± 22.4	25.7 ^a ± 0.18	54.1 ^{bc} ± 3.03	68.4 ± 2.78	20.5 ^a ± 1.91	109 ^c ± 10.7
Analysis of Variance								
A	<0.01**	<0.01**	<0.01**	0.1ns	<0.01**	<0.01**	<0.01**	<0.01**
B	>0.23ns	<0.01**	<0.01**	<0.01**	<0.01**	>0.50ns	<0.01**	<0.01**
A × B	>0.26ns	<0.01**	<0.01**	0.15ns	<0.01**	<0.04*	<0.01**	<0.01**

Means ± SD of replicates connected with the same letter(s) in the same column are not significantly different by the LSD test at $p \leq 0.05$. **: significant at 1%; *: significant at 5%; ns: non-significant.

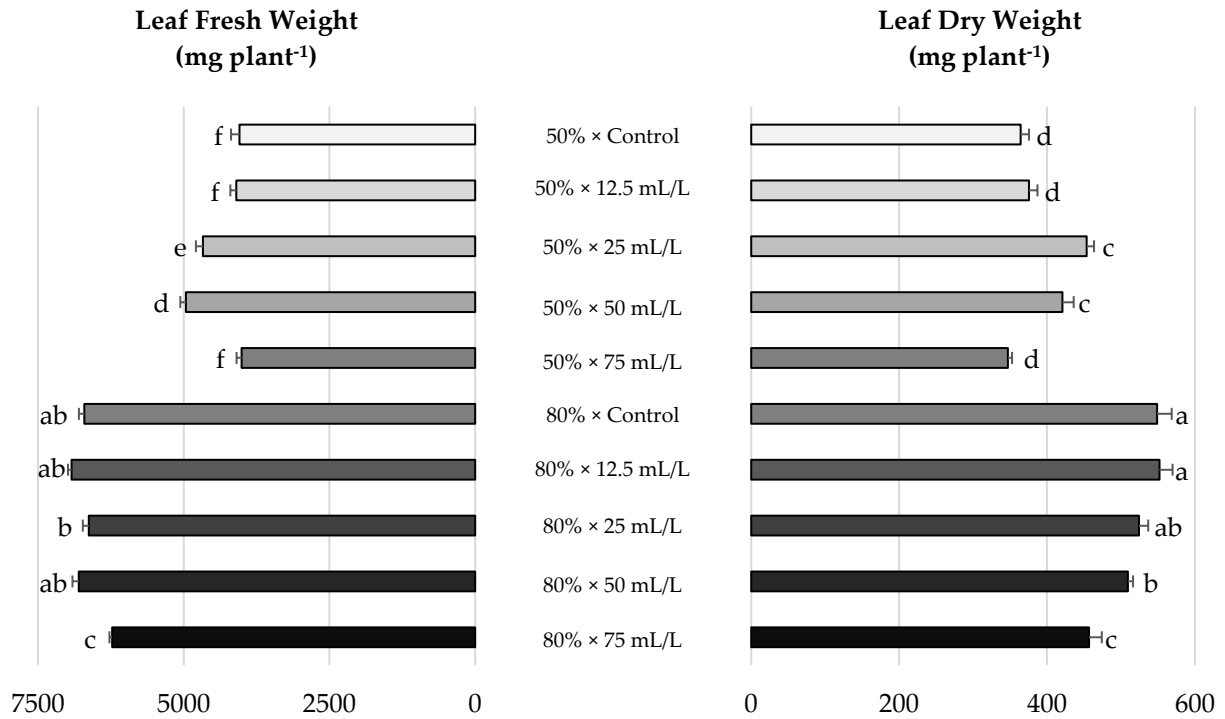


Figure 2. The interaction of drought stress and biostimulant doses on leaf fresh weight and leaf dry weight. Letters on each bar denote the significance level at $p < 0.05$.

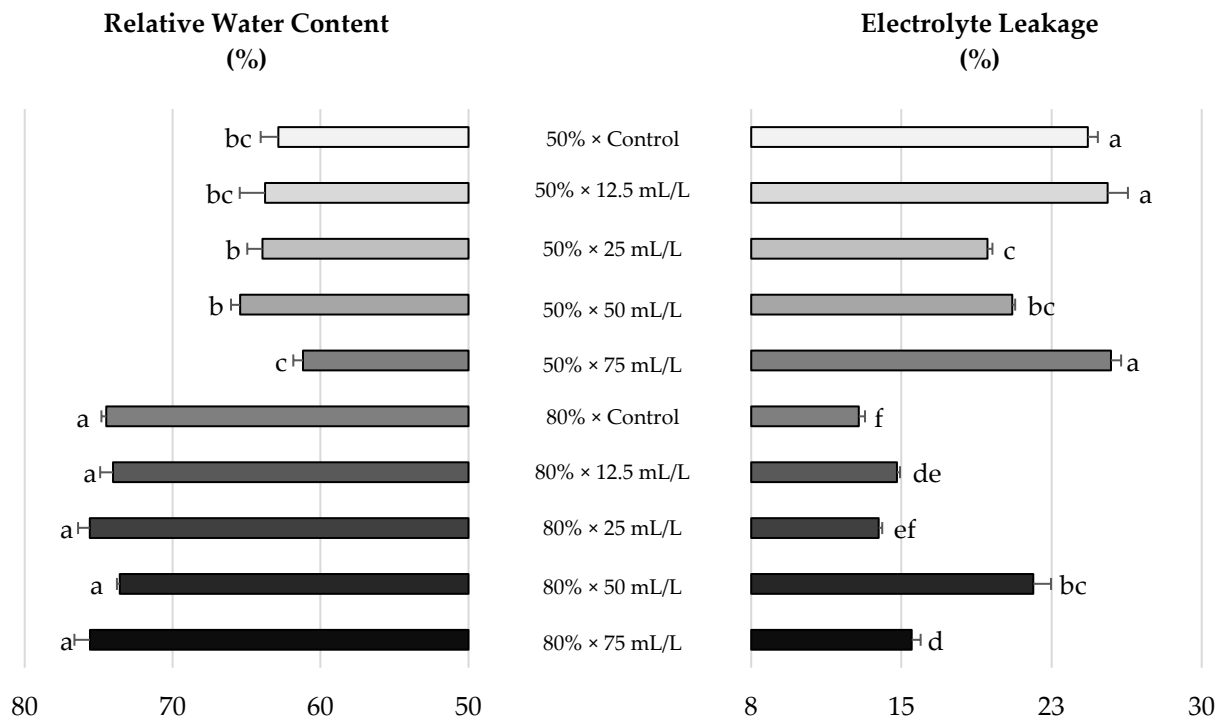


Figure 3. The interaction of drought stress and biostimulant doses on relative water content and electrolyte leakage. Letters on each bar denote the significance level at $p < 0.05$.

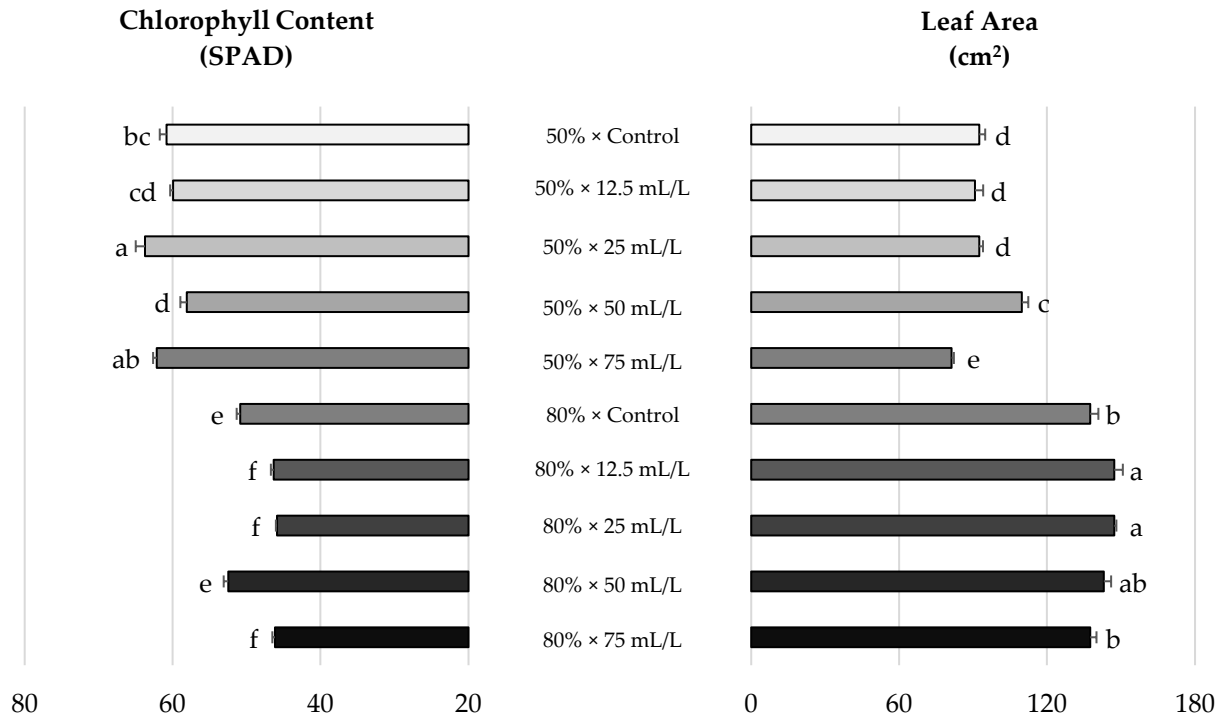


Figure 4. The interaction of drought stress and biostimulant doses on chlorophyll content and leaf area. Letters on each bar denote the significance level at $p < 0.05$.

4. Discussion

Drought stress harms plant growth and development at all stages, beginning at sowing and continuing until harvest. Sugar beet is vulnerable to drought stress during every stage of its life cycle. To produce high quality sugar and root yields, sugar beet requires much more water compared to other crops. In this study, sugar beet seedlings were treated with different levels of foliar animal-derived biostimulant to alleviate the hazardous effects of drought stress. In this study, there was a decline in NL, LFW, LDW, RWC, and LA under drought stress. According to Ahmad et al. (2019) and Paponov et al. (2020), the existence of water deficit conditions significantly decreased the NL in maize. Our results showed that LFW and LDW increased with increasing biostimulant doses, and the differences were significant. Similarly, Borcioni et al. (2016) observed that fulvic acid application increased LFW and LDW in lettuce. Santos (2014) observed a significant increase up to 85% in the LDW of plants, while investigating the effects of applying humic substances to the leaves of sweet pepper. Widuri et al. (2018) reported a significant decline in shoot dry weight in common bean under drought conditions. RWC plays an essential role in controlling many physiological processes in plants. The primary indication of the drought stress response is the decrease in RWC (Hussain et al., 2018). A decrease in RWC and LA in sugar beet was demonstrated by Khodadadi et al. (2020). Drought stress generally causes a decline in leaf RWC in barley, according to Istanbuli et al. (2020). The incidence of cell membrane damage and subsequent release of electrolytes from plant cells are significant indicators in the plants exposed to drought stress (ElBasyoni et al. 2017). Similarly, a considerable increase in EL was observed in sugar beet seedlings under drought. Drought stress reduced LA and whole plant biomass, as observed by Kumar et al. (2022). This decline was due to the limitations of leaf growth and the interruption of the photosynthetic process. For many crops, LA considerably reduced under drought stress, such as wheat (Naz and Perveen 2021) and maize (Cai et al. 2020). In this study, foliar animal-derived biostimulant positively induced RWC, EL, and LA. Amino acids may serve as osmoprotectant agents in plants suffering drought stress (Zulfiqar et al. 2020), hence contributing to the improvement of plant tissue's RWC. In our study, animal-derived biostimulant treatments increased the RWC of leaves. Similar results demonstrated an increase in RWC on soybean (Teixeira et al. 2020), tomato (Alfosea-Simon et al. 2020), and broccoli (Kaya 2023) leaves when a biostimulant containing amino acids was applied. In general, drought stress can reduce Chl by harming the membrane and structure

of chloroplasts (Shin et al. 2021). In our study, the beneficial effect of animal-derived biostimulant on Chl can be explained by the stimulatory effects of amino acids on chlorophyll biosynthesis (Noroozlo et al. 2019). Many studies showed a significant increase in Chl following treatment with biostimulants in bean (Sadak et al. 2015), soybean (El-Aal 2018), timothy (Radkowski and Radkowska 2018), and cotton (Ergin et al. 2024); however, insignificant differences in Chl were found in potato (Farhad et al. 2011), chamomile (Pirzad et al. 2011), and apple (Ping et al. 2015).

5. Conclusions

The use of biostimulants via foliar spray is a remarkable and new technology that is friendly to the environment and enhances agricultural sustainability. Furthermore, it has the potential for yield and quality improvement in crops under a wide range of conditions. Drought stress induced an important adverse effect on the evaluated morpho-physiological parameters in our experiment. Overall, the application of animal-derived biostimulant by spraying showed a promising effect on the development of sugar beet plantlets under drought stress. Especially at 25 and 50 mL/L doses of the biostimulant, improvements in RWC, EL, and LA characteristic were observed. In conclusion, foliar application of the animal-derived biostimulant may be beneficial to alleviate the negative impacts of drought and may stimulate the growth of sugar beet seedlings.

Author Contributions: Engin Gökhan KULAN planned, established, and conducted the study. Mehmet Demir KAYA analyzed the data obtained in the study, and Engin Gökhan KULAN and Mehmet Demir KAYA wrote the article. All authors have read and agreed to the published version of the manuscript.

Funding: This research received no external funding.

Data Availability Statement: The data that supports the findings of this study are available within the manuscript.

Acknowledgments: The authors are thankful to the staff of the Seed Science and Technology Laboratory, Department of Field Crops, Eskişehir Osmangazi University.

Conflicts of Interest: The authors declare no conflict of interest.

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