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The Effect of Water Retaining Polymer Applications on Agricultural Characteristics of Hemp (*Cannabis sativa* L.)

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HIGHLIGHTS

- Hemp is a plant that can make significant contributions to the development of our country. Today, various parts of hemp such as leaves, flowers, seeds, stems, fibers and roots are used in different fields of industry.
- Water-retaining polymers contribute to the growth and development of plants in water-scarce regions due to their high water retention capacity.
- The study found that water-retaining polymer doses did not have a statistically significant effect on the examined characteristics of hemp.

Abstract

Hemp (*Cannabis sativa* L.) is a plant that usage areas and importance have increased rapidly in recent years in the world and in Turkey. This study investigated the effects of water-retaining polymers on yield and quality characteristics of hemp cultivation in Yozgat ecological conditions. Narlısaray population and Vezir variety, which are local dioecious genotypes of Turkey, were used. The experiment was established with Split Plots Experimental Design in Coincidence Blocks with three replications. Varieties were applied in the main plots, and water-retaining polymer doses (0, 2.5, 5, 7.5, 10, 12.5 kg da^{-1}) were used in the sub-plots. According to the results obtained, the measurements were as follows: plant height 194.11-224.11 cm, stem diameter 9.70- 10.66 mm, dry stem yield 1720.55 - 2181.11 kg da^{-1}, fiber yield 338.88 - 480.00 kg da^{-1}, seed yield 200.55 - 272.77 kg da^{-1} and oil content 29.20 - 31.44% in Narlısaray population. In the Vezir variety, plant height ranged between 200.16- 214.03 cm, stem diameter between 8.76 - 9.89 mm, dry stem yield between 1688.88 - 1949.88 kg da^{-1}, fiber yield between 333.33 - 412.77 kg da^{-1}, seed yield between 228.88 - 271.66 kg da^{-1} and oil content between 29.59 - 32.07%. As a result, there was no statistically significant effect of water retaining polymer doses on the examined traits of hemp plants. Still, significant differences were observed among the varieties regarding these traits.

Keywords: Fiber, hemp, seed, yield, water-retaining polymer, quality

1. Introduction

Hemp (*Cannabis sativa* L.) is an annual plant belonging to the *Cannabinaceae* family, originating from Central Asia, with different species. Among these species, *C. sativa* L. is mostly used for its fiber and seeds with thin leaves and low medicinal use. *C. indica* L. is a species with broader leaves mostly used for medicinal purposes.

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On the other hand, *C. ruderalis* L. is a wild species with thinner leaves and shorter stature compared to other species (Yılmaz and Yazıcı 2022).

The hemp plant has been cultivated throughout history for its fiber, medicinal applications, and seeds. Archaeological studies have revealed that fiber remains of hemp dating back to 10,000 BC. The wild species were used first, and cultivation has been practiced for at least 5,000-6,000 years (Schultes and Hofmann 1980; Fleming and Clarke 1998; Yazıcı and Yılmaz 2021).

Today, various parts of hemp, such as leaves, flowers, seeds, stems, fibers, and roots, are used in different fields of industry. Hemp leaves and flowers are used in cosmetic products such as medicine, skin, and hair care; fibers are used in textile products, paper production, home textiles, construction, plastics, and packaging materials. The seeds are used especially in food products such as pasta, cakes, biscuits and bread, and the remaining meal after the oil is extracted from the seed can be used in animal feeding and some rations (Gül 2008; Townshend and Boleyn 2010; Baldini et al. 2018; Yazıcı and Yılmaz 2021).

Hemp seeds contain essential fatty acids, linoleic acid (omega 6) and alpha-linolenic acid (omega 3). The importance of 3-6% alpha-linolenic acid in hemp oil is increasing in nutrition, cosmetics, and biomedical fields. Since the human body cannot synthesize these essential fatty acids, they must be taken through food. It is reported to provide health benefits such as cardiovascular diseases, diabetes, resistance to inflammation, night vision, and embryo brain and eye development during pregnancy (Smith 2000; Ozgul et al. 2005).

Worldwide hemp cultivation increased to 1 million hectares in the 1950s and then steadily declined. Between 2015 and 2020, the average hemp cultivation area for fiber purposes was reported to be 40-45 thousand hectares and 30-35 thousand hectares for seed purposes (Anonymous 2021). In Turkey, according to TUIK data, 15.66 tons of industrial hemp fiber production was made on an average area of 175 decares between 2017 and 2022, and the yield was calculated as 111.5 kg/decare. In the same period, 79.33 tons of industrial hemp seed production was realized on an average area of 1191 decares, and the yield was 56.5 kg/decare. In 2022, the highest fiber hemp production in Turkey was performed in Samsun (188 decares), Sakarya (100 decares), Sivas (47 decares), Sinop (15 decares), Burdur (9 decares), Tokat (4 decares) and Uşak (2 decares) respectively. The highest cultivation area and the production for seed purposes were in Amasya (1001 decares, 82 tons) and Kastamonu (817 decares, 66 tons) (Anonymous 2023).

In Turkey, the laws and regulations regulate permits for hemp cultivation, the determination of the provinces, permitted districts, and the procedures to be applied in hemp cultivation. Legislation on hemp includes Law No. 2313 on the Control of Narcotic Substances, the Regulations on the Cultivation and Control of Hemp issued in 1990 and 2016, and Law No. 32154 dated April 5, 2023, on the Forest Law and Certain Laws Amendment. Hemp cultivation for producing fiber, seeds, stalks, and medicines is subject to permission from the Ministry of Agriculture and Forestry.

Hemp production for stalk, fiber, and seed purposes has never been banned in Turkey; this issue has no law or regulation. However, its production can be carried out in a controlled and permitted manner. Currently, authorized hemp cultivation can be carried out in 21 provinces in our country. The permitted provinces are Amasya, Antalya, Bartın, Burdur, Çorum, İzmir, Karabük, Kastamonu, Kayseri, Kütahya, Konya, Malatya, Ordu, Rize, Samsun, Sinop, Sivas, Tokat, Uşak, Yozgat and Zonguldak (Yilmaz and Yazici 2022).

Hemp is a plant that can make significant contributions to the development of our country, and its importance and use have been increasing rapidly both in the world and Turkey in recent years. Hemp-based products have the potential to increase industry and employment, especially in agriculture, by being used in many sectors such as food, feed, cosmetics, health support, construction, energy, and textiles (Yazıcı and Yılmaz, 2021). In the field of agriculture, hemp has an important place, especially in removing weeds from the fields, and it is used as an effective pre-plant in rotation systems, leaving a cleared field for the following crop (Atakisi 1999).

Global warming and climate change negatively impact the environment and agriculture, and water shortage is one of the most critical problems. Lack of sufficient water negatively affects the yield and quality characteristics of the plant. The sustainability of water resources and the increasing demand for limited water resources are among the most critical problems. Considering that 74% of the total water in Turkey is used in the agricultural sector, water saving comes to the forefront in agrarian production (Mirze and Yazici 2023).

Various applications are made to increase the water retention capacity of the soil. Water-retaining polymers are one of these applications, and they contribute to the growth and development of plants in water-scarce regions due to their high water retention capacity (Johnson and Piper 1997; Lobo et al. 2006).

This study aims to determine the effects of different water-retaining polymer doses (0 kg da^{-1}, 2.5 kg da^{-1}, 5 kg da^{-1}, 7.5 kg da^{-1}, 10 kg da^{-1}, 12.5 kg da^{-1}) on fiber and seed yield, some yield components and quality of local hemp genotypes (Narlısaray population and Vezir variety) under Yozgat ecological conditions. Detailed studies on hemp and water-retaining polymers in the agricultural field are limited. This study aims to obtain information about water-retaining polymers, facilitate their use in plant production, popularize them in the hemp sector, test the effectiveness of water-retaining polymers applied at different doses, and transfer the information to farmers.

2. Materials and Methods

2.1. Material

In the experiment, the local hemp genotype Narlısaray population and Vezir variety were supplied by Yozgat Bozok University Hemp Research Institute and used as materials. Narlısaray population is a local hemp genotype from the Vezirköprü district of Samsun province. It is a genotype with a dioecious flower structure, is tall, is suitable for fiber and seed use, and its thousand seed weights range between 17.5 and 18 g. Vezir variety is a variety bred by the Black Sea Agricultural Research Institute; leaf green color intensity; medium, cotyledon shape; narrow, central leaflet length; medium, anthocyanin intensity in male flowers; weak and THC content was observed as very low or absent. The thousand seed weight of the variety is 17.4 grams.

In the study, a water-retaining polymer consisting of organic materials to which a certain amount of straw was added and used. The general purpose of combining the water-retaining polymer with straw is to increase the water retention capacity as much as possible. The water-retaining polymer contains different ratios of Oxygen (O), Hydrogen (H), Carbon (C), Potassium (K) and minerals such as 65-75% cellulose, 15-20% pentazones and hemicellulose, 5-10% lignin, 1-3% protein and wax, 2-10% silica in the straw composition (Yakupoğlu et al., 2019).

2.2. Climate characteristics of the experimental site

Climatic data for 2022 and many years (1929-2022) are presented in Table 1. When Table 1 is examined, the long-term average precipitation for the vegetation period in Yozgat province was 571 mm. In the year when the study started, the average rainfall was calculated as 507.8 mm. In the vegetation period 2022, 63.2 mm less precipitation was realized compared to the long-term average.

The long-term (1929-2021) average temperature in Yozgat province was 9.3 °C. In the growing period of 2022, when the experiment was conducted, the average temperature was 9.5 °C. This data was 0.2 °C higher than the long-term average. The long-term average relative humidity in Yozgat province was 66.2% and 64.2% in the 2022 vegetation period.

2.3. Soil characteristics of the experimental site

The soil properties of the field where the experiment was conducted are shown in Table 2. Amasya Central Research and Application Laboratory analyzed soil samples taken from three different locations and 30 cm depth of the soil. When the soil properties of the experimental area were analyzed, it was observed that it had clay loamy-loamy soil, neutral pH level, low organic matter content, medium salty, slightly calcareous, and very low nitrogen and phosphorus content.

Table 1. Total Precipitation (mm), Average Temperature (°C), and Average Relative Humidity (%) of Yozgat province for many years (1929-2021) and the year of the experiment (2022)

Months	Total Precipitation (mm)		Average Temperature (°C)		Average Relative Moisture (%)	
	2022	Long Years (1929-2021)	2022	Long Years (1929-2021)	2022	Long Years (1929-2021)
January	121.1	68.6	-2.4	-1.7	76.6	77.6
February	48.8	59.7	0.2	0,6	77.1	75.3
Mart	83.3	68.4	-1.5	3.0	73.9	70.9
April	16.4	57.8	11.6	8.5	47.4	64.9
May	50.2	66.1	12.4	13.1	60.2	63.5
June	98.9	45.7	17.5	16.7	64.3	60.7
July	1.2	12.6	18.3	19.6	57.9	56.0
August	7.3	11.4	22.9	19.8	50.9	54.7
September	9.2	18.3	17.4	15.9	48.8	57.1
October	13.9	33.3	10.6	10.8	69.0	64.2
November	23.8	53.4	7.0	5.1	69.5	71.9
December	33.7	75.7	0.8	0.6	75.1	77.7
Tot./Middle.	507.8	571.0	9.5	9.3	64.2	66.2

Table 2. Some chemical and physical properties of the soil in the year the experiment was conducted.

Characteristics	Results	Meaning
Saturation (%)	71.5	Clay loam
pH	7.24	Neutral
Nitrogen kg da^{-1}	2,20	Very Little
Salt (μ^s/cm)	758	No Salt
Lime (%)	0,39	Less Calcareous
Useful Potassium (kg da^{-1})	59.43	Less
Useful Phosphorus (kg da^{-1})	4.14	Very little
Organic Matter (%)	1.24	Less

2.4. Method

The experiment was carried out in the experimental field of Yozgat Bozok University Campus Agricultural Research and Application Center in the vegetation period of 2022. The study was established according to the split-plot experimental design in randomized blocks with three replications as varieties in main plots and water-retaining polymer doses (Control, 2.5 kg da^{-1}, 5 kg da^{-1}, 7.5 kg da^{-1}, 10 kg da^{-1}, 12.5 kg da^{-1}) in sub-plots, totaling 36 sub-plots. Water-retaining polymers were applied to the seedbed during sowing by hand, right next to the seed. Each plot consisted of 6 rows with a row spacing of 20 cm and a length of 5 m. The sowing norm was set at 150 seeds/m². Harvesting was carried out on 20.10.2022, the whole maturity period of the seeds, by cutting the plant from the root collar area with pruning shears. After the harvested plants were left to dry naturally for a certain period (25-30 days), the green parts of the plants were manually removed from the stems. Then, the plant stems were stripped of fiber on 20.12.2022 using the fiber stripping machine (decortication) at Yozgat Hemp Research Institute. The seed blend was made on 17.01.2023 by sieving the seeds from the green parts removed from the stems with the help of sieves.

Statistical analysis of the data obtained from the studies was analyzed using variance analysis according to the Split Plots Experimental Design in Coincidence Blocks. The multiple comparison test LSD (Least Significant Difference) was used to statistically calculate the differences between the means (Efe et al., 2000).

3. Results and Discussion

The results of analysis of variance for plant height, stem diameter, dry stem yield, fiber yield, seed yield, and oil content in different water-retaining polymer doses are given in Table 3 and Table 4.

Table 3. Variance analysis table for plant height, stem diameter, and dry stem yield of male + female plants in hemp in different water-retaining polymer dose applications.

Sources of Variation	Plant Height (cm)			Stem Diameter (mm)			Dry Stem Yield (kg da^{-1})		
	M.S	F Value	P Value	M.S	F Value	P Value	M.S	F Value	P Value
Repetition (A)	1060.69	14.27	0.06	0.79	2.26	0.30	732160	88.39	0.01*
Variety (B)	471.25	6.34	0.12	7.37	21.14	0.04*	81637.5	9.85	0.08
Error 1 (AxB)	74.27	0.34	0.71	0.34	0.71	0.50	8282.98	0.10	0.89
Dose (C)	226.76	1.06	0.41	0.46	0.96	0.46	89437.9	1.16	0.36
B x C	295.10	1.38	0.27	0.57	1.18	0.35	63760.8	0.82	0.54
Error 2	213.54	-	-	0.48	-	-	76944.31	-	-
General	274.89	0.14	-	0.70	0.06	-	110497.21	0.07	-
CV (%)	6.97			7.08			14.94		

* Significant at the 0.05 level, **Significant at the 0.01 level.

When Table 3 is examined, plant height and dry stalk yield values were not statistically significant in the variety, dose, and variety x dose interactions in the water-retaining polymer doses applied in hemp varieties. The effect of the cultivar on average values of stem diameter was found to be significant.

Table 4. Variance analysis table for fiber yield, seed yield and oil ratio of male + female plant height in hemp in different water retaining polymer dose applications.

Sources of Variation	Fiber Yield (kg da^{-1})			Seed Yield (kg da^{-1})			Oil Ratio(%)		
	M.S	F Value	P Value	M.S	F Value	P Value	M.S	F Value	P Value
Repetition (A)	29497.80	30.43	0.03*	23789.2	456.07	0.00**	2.55	0.20	0.83
Variety (B)	5216.05	5.38	0.14	3533.64	67.74	0.01*	0.88	0.06	0.81
Error 1 (AxB)	969.05	0.22	0.79	52.16	0.03	0.96	12.71	4.70	0.02*
Dose (C)	6548.09	1.54	0.22	1783.64	1.20	0.34	3.51	1.30	0.30
B x C	3344.01	0.78	0.57	1212.72	0.82	0.54	1.68	0.62	0.68
Error 2	4248.23	-	-	1475.40	-	-	2.70	-	-
General	5730.70	0.10	-	2734.46	0.01*	-	3.18	0.23	-
CV (%)	17.34			15.80			5.39		

* Significant at the 0.05 level, **Significant at the 0.01 level.

When Table 4 is examined, fiber yield and oil ratio values were not statistically significant in the variety, dose, and variety x dose interaction in the water-retaining polymer doses applied in hemp varieties. Variety effect on seed yield mean values was found to be significant.

As seen in Table 5, the mean values of plant height of hemp varieties varied between 202.65-218.35 cm. The mean values of the Narlısaray population ranged between 194.11 - 224.11 cm, with an average of 213.01 cm, while the mean values of the Vezir variety ranged between 200.16 - 214.03 cm, with an average of 205.78 cm. In water-retaining polymer dose treatments, the longest plant height of the Narlısaray population was 224.11 cm at 5 kg da^{-1} dose, and the shortest was 194.11 cm at 7.5 kg da^{-1} dose. In the Vezir variety, the longest was 214.03 cm at 12.5 kg da^{-1} dose, and the shortest was 200.16 cm at 5 kg da^{-1} dose. According to both varieties' average plant height values, the longest plant height was found at 12.5 kg da^{-1} dose (218.35 cm) and the shortest at 2.5 kg da^{-1} dose (202.65 cm). Yazici et al. (2020) reported that plant height was between 58.8 - 345.0 cm in a study conducted under Tokat conditions. In another study Koçer (2022) conducted with monoic hemp varieties under Tokat conditions, plant height averages were found to be the lowest at 135.42 and the highest at 207.23 cm.

Table 5. Average male + female plant height values and stem diameter of different water-retaining polymer doses in hemp plants.

Doses	Plant Height (cm)			Stem Diameter (mm)		
	Narlısaray	Vezir	Avg.	Narlısaray	Vezir	Avg.
Control	218.71	206.81	212.76	10.66	9.23	9.65
2.5	204.35	200.95	202.65	9.87	8.76	9.31
5	224.11	200.16	212.14	10.39	8.87	9.63
7.5	194.11	211.95	203.03	9.70	9.89	9.80
10	214.13	200.78	207.45	10.05	9.25	9.65
12.5	222.68	214.03	218.35	10.50	9.74	10.12
Avg.	213.01	205.78	209.39	10.19a	9.29b	
LSD (%)	Variety (a): 12.34 Dose (b): 17.53 a x b: 27.81			Variety (a): 0.81 Dose (b): 0.83 a x b: 1.18		

The mean values of stem diameter varied between 9.31 and 10.12 mm. The Narlısaray population ranged between 9.70 - 10.66 mm with an average of 10.19 mm, and in the Vezir variety, 8.76 - 9.89 mm with an average of 9.29 mm. When the stalk diameter data between doses were analyzed, the maximum was 10.66 mm at the control dose, and the minimum was 9.70 mm at the dose of 7.5 kg da^{-1} in the Narlisaray population. In the Vezir variety, the highest stem diameter was determined at the dose of 7.5 kg da^{-1} with 9.89 mm and the lowest stem diameter at the control dose at 9.23 mm. In water-retaining polymer dose applications, stem diameter values were higher (10.12 mm) at 12.5 kg da^{-1} than other doses and the lowest (9.63 mm) at 2.5 kg da^{-1} dose. Şakar (2022) found that the lowest stem diameter was 7.1 mm and the highest was 9.3 mm in a study conducted in Tokat province conditions. Koçer (2022) reported that the stem diameter was 12.39 - 18.05 mm in dioecious male plants and 13.35 - 18.71 mm in female plants.

Table 6. Average dry stem yield and fiber yield values of different water retaining polymer doses in hemp plants.

Doses	Dry Stem Yield (kg da^{-1})			Fiber Yield (kg da^{-1})		
	Narlısaray	Vezir	Avg.	Narlısaray	Vezir	Avg.
Control	1937.22	1949.88	1943.55	400.55	412.77	406.66
2.5	1720.55	1688.88	1704.72	364.44	340.00	352.22
5	2181.11	1816.99	1999.05	480.00	371.11	425.55
7.5	1631.11	1852.44	1741.77	338.88	366.11	352.50
10	1900.00	1707.77	1803.88	385.00	358.88	371.94
12.5	2052.77	1835.33	1944.05	357.77	333.33	345.55
Avg.	1903.79	1808.55		387.77	363.70	
LSD (%)	Variety (a): 130.41 Dose (b): 333.11 a x b: 471.07			Variety (a): 44.59 Dose (b): 78.27 a x b: 110.67		

It was observed that the average dry stem yield of the varieties examined was between 1704.72 - 1999.05 kg da^{-1}. In the Narlısaray population, the average was 1903.79 kg da-1 between 1720.55 - 2181.11 kg da^{-1}, and in the Vezir variety, the average was 1808.55 kg da^{-1} between 1688.88 - 1949.88 kg da^{-1}. Among the doses of water-retaining polymer, the highest dry stalk yield in the Narlisaray population was 2181.11 kg da^{-1} at 5 kg da^{-1} dose, and the lowest was 1631.11 kg da^{-1} at 7.5 kg da^{-1} dose. When the averages of the Vezir variety were examined, the highest dry stalk yield was found in the control dose with 1949.88 kg da^{-1}, and the lowest was found in the 2.5 kg da^{-1} dose with 1688.88 kg da^{-1}. According to the mean values of both varieties, the highest dry stalk yield values were determined at 5 kg da^{-1} dose (1999.05 kg da^{-1}) and the lowest at 2.5 kg da^{-1} dose

(1704.72 kg da^{-1}) (Table 6). In their study, Flajsman et al. (2016) reported that dry stalk yield varied between 8000 – 10000 kg ha^{-1}. Kime (1996), in another study conducted in Canada, found that dry stalk yield was between 2.5 - 3 tons/ha. Meijer et al. (1995) reported that dry stalk yield was between 9.4 - 13.6 tons ha^{-1} in their study conducted between 1987 and 1989.

The average values of fiber yield were found between 345.55-425.55 kg da^{-1}. Fiber yield values of Narlısaray population varied between 338.88 - 480.00 kg da^{-1}, with an average of 387.77 kg da^{-1}, and Vezir variety varied between 333.33 - 412.77 kg da^{-1}, with an average of 363.70 kg da^{-1}. In water-retaining polymer dose applications, the highest fiber yield in the Narlisaray population was 480.0 kg da^{-1} at 5 kg da^{-1} dose, and the lowest was 338.88 kg da^{-1} at 7.5 kg da^{-1} dose. When the Vezir variety was examined, the highest fiber yield was 412.77 kg da^{-1} at the control dose, and the lowest was 333.33 kg da^{-1} at the 12.5 kg da^{-1} dose. When the average of both varieties was examined, the highest fiber yield was found at 5 kg da^{-1} dose (425.55 kg da^{-1}) and the lowest at 2.5 kg da^{-1} dose (352.22 kg da^{-1}). Deleuran and Flengmark (2006) found fiber yield between 170-310 kg da^{-1} in their study on fiber yield. Aksoy (2021) reported that fiber yield was between 11.65 - 242.2 kg da^{-1} in his research conducted under Samsun conditions.

Table 7. Average values of seed yield and oil content of different water-retaining polymer doses in hemp plants.

Doses	Seed Yield (kg da^{-1})			Oil Ratio (%)		
	Narlısaray	Vezir	Avg.	Narlısaray	Vezir	Avg.
Control	233.33	271.66	252.50	29.28	30.43	29.86
2.5	216.66	234.44	225.55	29.84	29.59	29.71
5	272.77	252.77	262.77	31.44	32.07	31.75
7.5	200.55	263.88	232.22	29.20	30.52	29.86
10	221.66	228.88	225.27	30.28	30.85	30.56
12.5	253.33	265.55	259.44	31.29	29.75	30.52
Avg.	233.05b	252.87a		30.22	30.53	
LSD (%)	Variety (a): 10.32			Variety (a): 5.07		
	Dose (b): 46.11			Dose (b): 1.95		
	a x b: 65.22			a x b: 2.78		

The average values of seed yield of the varieties varied between 225.27 - 262.77 kg da^{-1}. Seed yield in the Narlısaray population ranged between 200.55 - 272.77 kg da^{-1} with an average of 233.05 kg da^{-1} and in the Vezir variety between 228.88 - 271.66 kg da^{-1} with an average of 252.87 kg da^{-1}. When the doses applied were examined, the highest dose of 272.77 kg da^{-1} was determined at 5 kg da^{-1} dose with the highest rate of 272.77 kg da^{-1} in the Narlisaray population and the lowest dose of 200.55 kg da^{-1} was determined at 7.5 kg da^{-1} dose. In the Vezir variety, the highest seed yield was 265.55 kg da^{-1} at 12.5 kg da^{-1} dose, and the lowest was 228.88 kg da^{-1} at 10 kg da^{-1} dose. When the mean values of the varieties were examined, the highest seed yield was found at 5 kg da^{-1} dose (262.77 kg da^{-1}) and the lowest at 10 kg da^{-1} dose (225.27 kg da^{-1}) in water-retaining polymer dose applications. In his study, Özdemir (1992) reported that seed yield was between 31.0 - 60.6 kg da^{-1}. Yazici et al. (2022) found that seed yield was 130.90 - 217.86 kg da^{-1} in another study.

The mean values of oil content in different water-retaining polymer treatments in hemp varieties varied between 29.71% and 31.75%. In the Narlısaray population, the oil ratio was found between 29.20 - 31.44%, and in the Vezir variety, between 29.59 - 32.07%. When the applied doses were analyzed, the highest rate of 31.44% was found at 5 kg da^{-1} dose with 31.44% in the Narlisaray population, and the lowest rate was seen at 7.5 kg da^{-1} dose with 29.20%. In the Vezir variety, the highest oil content was 32.07% at 5 kg da^{-1} dose, and the lowest was 29.59% at 2.5 kg da^{-1} dose. When the mean values of both varieties were analyzed, the highest oil content was found at 5 kg da^{-1} dose (31.75%) and the lowest at 2.5 kg da^{-1} dose (29.71%) in water retaining polymer doses applications (Table 7). Höppner and Hartmann (2007) found an average oil content of 23.2% in their study. According to Aksoy (2021), the oil content was between 16.25 - 35.79%.

4. Conclusions

In the study conducted to determine the effect of water-retaining polymer applications on yield and quality traits in hemp (*Cannabis sativa* L.) cultivation under Yozgat conditions, five different water-retaining polymer doses (0, 2.5, 5, 7.5, 10, 12.5 kg da^{-1}) were applied on two different hemp genotypes (Narlısaray and Vezir). The effect of water-retaining polymer doses on the examined traits in hemp plants was statistically insignificant. Still, significant differences were determined in some traits among the varieties. The differences observed between the Narlısaray and Vezir genotypes reveal the varying potential of genetic structures to adapt to environmental conditions. This finding underscores the critical role of genotype selection in hemp cultivation. Moreover, it highlights that while the effects of water-retaining polymers may not always be pronounced, the response of genotypes to environmental conditions can significantly impact yield and quality traits. In the future, examining the effects of water-retaining polymers under longer-term and varied irrigation regimes and focusing on studies comparing the outcomes of genotype selection in different regions will deepen our understanding of this field. In conclusion, in drought-sensitive areas like Yozgat, adopting strategic approaches in the efficient use of water resources and genotype selection can substantially contribute to the sustainability of hemp cultivation.

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References

Aksoy D (2021). Investigation of yield and some quality characteristics of narlısaray population and foreign origin cannabis varieties in Samsun ecological conditions. Master's Thesis, Ondokuz Mayıs University (Unpublished), Türkiye.

Anonymous (2021). Food and Agriculture Organization of the United Nations. https://www.fao.org/faostat/en/#home (access date: 16.01.2023).

Anonymous (2023). Turkish Statistical Institute, https://www.tuik.gov.tr, (access date: 15.06.2023).

Atakisi IK (1999). Fiber Crops Cultivation and Breeding. Trakya University, Tekirdag Faculty of Agriculture Publications. No: 10, Tekirdag, Türkiye. p.121.

Baldini M, Ferfuia C, Piani B, Sepulcri A, Dorigo G, Zuliani F, Danuso, Cattivello C (2018). The performance and potentiality of monoecious hemp (*Cannabis sativa* L.) cultivars as a multipurpose crop. *Agronomy* 8, 162.

Deleuran LC, Flengmark PK (2006). Yield potential of hemp (*Cannabis sativa* L.) cultivars in Denmark. *Journal of Industrial Hemp,* 10(2), 19-31. https://doi.org/10.1300/J237v10n02_03

Efe E, Bek Y, Sahin M (2000). Statistical Methods II. Kahramanmaraş Sütçü İmam University Rectorate, Computer Research and Application Center, Publication No: 10. Kahramanmaraş, Türkiye.

Flajsman M, Jakopic J, Kosmelj K, Kocjanacko D (2016). Morphological and technological characteristics of hemp (*Cannabis sativa* L.) varieties from field trials of Biotechnical faculty in 2016. *Hop Bulletin*, 23, 88-104.

Fleming MP, Clarke RC (1998). Physical evidence for the antiquity of *Cannabis sativa* L. (*Cannabaceae*). *Journal of the International Hemp Association,* 5(2), 80-92.

Gul V (2008). Determination of physical and chemical properties of some hemp seeds in the Black Sea region. Master's Thesis, Ordu University (Unpublished), Türkiye.

Hoppner F, Menge-Hartmann U (2007). Yield and quality of fiber and oil of fourteen hemp cultivars in Northern Germany at two harvest dates. *Landbauforsch,* 57, 219-232.

Johnson MS, Piper CC (1997). Cros-linked, water-storing polymers as aids to drought tolerance of tomatoes in growing media. *Journal of Agronomy and Crop Science,* 178, 23-27.

Kime GG (1996). Personal communication. Hempline, Inc., Ontario, Canada.

Kocer T (2022). Determination of vegetative and yield characteristics of some cannabis (*Cannabis sativa* L.) varieties and populations under Tokat - Kazova conditions. Master's Thesis, Tokat Gaziosmanpaşa University (Unpublished), Türkiye.

Lobo D, Torres D, Gabriels D, Rodriguez N, Rivero D (2006). Effect of organic waste compost and a water absorbent polymeric soil conditioner (hydrogel) on the water use efficiency in a *Caspium annum* (green paper) cultivation. *Agroenviron 2006 Conference*, pp. 453-459.

Meijer WJM, Van der HMG, Werf EW, Mathijssen JM, Van den Brink PWM (1995). Constraints to dry matter production in fiber hemp (*Cannabis sativa* L.). *European Journal of Agronomy,* 4(1), 109-117.

Mirze O, Yazici L (2023). Irrigation in ındustrial hemp (*Cannabis sativa* L.) production. *5th International Haliç Multidisciplinary Scientific Research Congress (Online)*, Ankara, Türkiye, pp. 296-303.

Ozdemir O (1992). The effect of nitrogen and plant density on the yield and some properties of cannabis (*Cannabis sativa* L.). PhD Thesis, Ondokuz Mayıs University (Unpublished), Türkiye.

Ozgul YS, Turkay S, Ustun G (2005). A study on gamma linolenic acid resources and potential of Turkey. *4th GAP Agriculture Congress*, September 21-23, Şanlıurfa, Türkiye, pp. 576-580.

Schultes RE, Hofmann, A. A. (1980). The Botany and Chemistry of Hallucinogens, Charles C. Thomas, Springfield, Illinois, p. 21.

Smith K (2000). Hempseed Oil: A Smart Start. *The Hemp Report,* 2(14), 1488-3988.

Townshend JM, Boleyn JM (2010). Plant density effect on oil seed yield and quality of industrial hemp cv. Fasamo in Canterbury. Agronomy Society of New Zealand Special Publication No. l3 / Grassland Research and Practice Series No. 14.

Yakupoglu T, Gulumser E, Dogrusoz ÇM, Başaran U (2019). Sediment and runoff from water-retaining soil under annual forage crop cultivation. *Journal of Soil Science and Plant Nutrition,* 7(2), 99-109.

Yazici L (2022). Optimizing plant density for fiber and seed production in industrial hemp (*Cannabis sativa* L.). *Journal of King Saud University - Sc*ience 35, 102419.

Yazici L, Yilmaz G (2021). Industrial hemp and its future. *12th International Scientific Research Congress (UBAK)*, Ankara, Türkiye, pp. 362-369.

Yazici L, Yılmaz G, Kocer T, Sakar H (2020). Investigation of some yield characteristics of hemp (*Cannabis sativa* L.) in Tokat ecology. *Journal of International Environmental Application and Science,* 15(2), 104-108.

Yilmaz G, Yazici L (2022). Rising value in the world: industrial hemp (*Cannabis sativa* L.). *Bozok Journal of Agriculture and Natural Sciences,* 1(1), 54-61.

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Socio-Economic and Infrastructural Challenges Affecting the Competitiveness of Nigeria Soybean Production in International Trade: Auto-Regressive Distributive Lag Approach

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HIGHLIGHTS

- The ADR unit root test shows that ARE, RER are stationary at I (0) Level.
- The ADR unit root test shows that SOY, YED, and WOT were stationary at I (1) first difference.
- The ARDL reveals that ARE and YED are significant predictors influencing the competitiveness of soybean production in international trade.
- The speed of adjustment is at the rate of 86%.

Abstract

This research study examined the socio-economic and infrastructural challenges affecting the competitiveness of Nigerian soybean production in international trade: ARDL (Auto-Regressive Distributive Lag Approach). Data of secondary sources were used. The primary and secondary data were utilized for this study. The data covered the period of 2007 to 2022. Data were obtained from FAO, CBN, NBS, World Bank publication, and GHS-P. The econometric tools employed were descriptive statistics, ADF unit root test, and ARDL Model using STATA package. The outcome of ADF unit root test reveals that ARE and RER are stationary at I (0) (level), while SOY, YED, and WOT were stationary at I (1) (First difference). The ARDL employed for co-integration test for the time series data shows that the computed F- statistics of 13.69 is more than the value at upper bound value of 4.09 at the 5% level of significance, this connotes that there is long run association between the regressors in the model The estimated long-run outcome using ARDL show that ARE, YED are the significant

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regressors influencing the competitiveness soybean production in the international market The speed of regulation to where soybean will balance even when there is disequilibrium is at the rate of 86%. Policies that would stimulate the export of soybeans should be carried out, and research activities should be enhanced to improve the quality of the soybeans produced.

Keywords: Socio-economic and infrastructural challenges; competitiveness; Nigerian soybean; production; auto-regressive distributive lag model.

1. Introduction

Soybean (*Glycine max* L.) is a good source of vegetable oil, protein, and amino acids and can be used for soymilk, soy flour, soy dough, soy curd, and animal feed. Hence, soybeans are used for food, feed, oil, and fuel (Mohammed et al. 2016). Soybeans accounted for 35% of the total harvested area in the world devoted to perennial and annual crops. The soybean share in world oil-seed output is over 50% (Yusuf et al. 2022). The derivatives of soybean and soybean seed are the highly traded agricultural produce, which adjudges for about 10% of the total value of the world agricultural market (FAO 2015). The leading producers of soybeans are Brazil, China, the USA, and Argentina. In 2022 and 2021, Nigeria produced 1060000 metric tons and 1166050 metric tonnes of soybean, respectively (FAO 2024). In 2022, the area and yield of soybeans in Nigeria are 1100000 ha and 963.6 kg ha^{-1}, respectively (FAO 2024). In 2022, Nigeria's soybean export, soybean export value, and agricultural export values were 15,300 metric tons, 4,450,000 USD, and 1561.575 million USD respectively (FAO 2024).

Competitiveness at the macroeconomic level is established on the market shares for export (Umar 2020). The competitiveness is estimated as the soybean export value as a percentage of Nigerian agricultural export value. Adopting the model established by Yousif (2015), yield, area, and real exchange rate were the main factors of soybean competitiveness. Umar (2020) documented that agriculture's long-term competitiveness and progress are not associated with short-term regressors such as prices and input costs. For this purpose, the model does not consider price and interest rates. According to Ude et al. (2013) competitiveness can be explained as a comparative concept of the performance or potential of a firm country or sub-unit to sell and supply a commodity in a given market. Competitiveness explains whether a firm could successfully compete in international trade, given existing economic structures and policies (Ude et al. 2013). The comparative advantage can be described as the ability of one nation to produce agricultural produce at a lower opportunity cost of other produce forgone than another nation. Competitiveness as a competitive power is the potential to produce goods and services that meet the test of international trade, raise the welfare level of a country’s citizens, and expand real income. In other words, competitiveness refers to a country’s ability to produce, create, and distribute service products in the international market while raising the returns on its resources. From the perspective of competitiveness at the international level, a country should be able to raise real income and welfare by producing goods and services under fair international trade conditions (Nordin et al. 2008). The major objective is to estimate the socio-economic and infrastructural challenges affecting the competitiveness of Nigerian soybean production in the international market: ARDL approach.

The specific objectives are to (i) examine the summary statistics, e.g., skewness, kurtosis, and Jarque-Bera of the test predictors (ii) determine the stationarity and co-integration test of the test predictors, (iii) evaluate the factors influencing the competitiveness of Nigerian soybean production in international trade, and (iv) estimate the long run relationships of regressors influencing the competitiveness of Nigerian soybean production in the international market.

2. Methodology

This study was conducted in Nigeria. Primary and secondary data were used for this study. The primary data covered the period of 2007 to 2022. The data used were obtained from FAO (Food and Agriculture Organization), CBN (Central Bank of Nigeria), NBS (National Bureau of Statistics), World Bank publication, and (GHS-P) (General Household Survey-Panel) combined with Federal Ministry of Agriculture and Rural

Development. The econometric tools employed were descriptive statistics, ADF (Augmented Dickey-Fuller) unit root test, and ARDL (Auto-Regressive Distributive Lag) Model. Data were analyzed using inferential and descriptive statistics employing the STATA package as follows:

2.1 *Summary statistics*: This involves using skewness, kurtosis, Jarque–Bera, mean, and frequency distribution to have a summary statistic of the data.

2.2 *The ADF model:* The unit root test is carried out to determine the level of integration among predictors under examination. The ADF following Dickey and Fuller (1981) and Dickey and Wayne (1979) model is given as:

$$\Delta Y_t = \pi Y_{t-1} + \sum_{j=1}^{P} \gamma_j \, \Delta Y_{t-1} + \varepsilon_t \tag{1}$$

Were,

Δ = Symbol for the First Difference Operator,
π and γ_j = Parameter Estimates
ε_t = Error Term
Y_t = Time Series Data
P = Proxy for the Maximum Lag Length for the Regressor.

2.3 *The ARDL:* The model follows the work of Pesaran et al. (2001) described the ARDL model as very adjustable as it combines the short and long outcomes in a single equation.

2.4 The Model Specificatiom

The model in its implicit form is given as:

$$\text{SOY} = f(\text{ARE}, \text{YED}, \text{WOT}, \text{RER}, \text{GOP}, \text{CIF}, \text{TRB}, \text{SUP}, \text{ARF}, \text{DIF}) \tag{2}$$

Where,

SOY = The Share of Soybean Export Value as a Percentage of Nigerian Agricultural Export Value (%)
ARE = Area (Ha)
YED = Yield (Kg/ha)
WOT = World Trade in Oil Seed (Metric Tones)
RER = Real Exchange Rate (Naira per Dollar)
GOP = Government Policy (Interest Rate, Percentage)
CIF = Climatic Factors (Ranifall, mm),
TRB = Trade Barrier (Tariff, Naira),
SUP = Substitute Product (Sesame, tons)
ARF = Access Road to Farm (1, Access; 0, Otherwise)
DIF = Distant from Farm to Nearby Market (Km)

The Real Exchange Rate (RER) following Kingu (2014) is calculated as:

$$RER = \frac{CPI_{Nigeria}}{CPI_{USA}} \times NER \tag{3}$$

Where,

$CPI_{Nigeria}$ = Consumer Price Index of Nigeria

CPI_{USA} = Consumer Price Index of United States of America (US)

NER = The Nominal Exchange Rate in Local Currency(₦/$)

2.3 The Auto-Regressive Distributive Lag (ARDL) model is presented as follows:

$$\begin{aligned}\Delta \mathrm{SOY}_t = {} & \alpha_0 + \sum_{i=1}^{k} \alpha_{1i}\Delta SOY_{t-1} + \sum_{i=1}^{k} \alpha_{2i}\Delta ARE_{t-1} + \sum_{i=1}^{k} \alpha_{3i}\Delta YED_{t-1} + \sum_{i=1}^{k} \alpha_{4i}\Delta WOT_{t-1} + \sum_{i=1}^{k} \alpha_{5i}\Delta RER_{t-1} \\ & + \sum_{i=1}^{k} \alpha_{6i}\Delta GOP_{t-1} + \sum_{i=1}^{k} \alpha_{7i}\Delta CIF_{t-1} + \sum_{i=1}^{k} \alpha_{8i}\Delta TRB_{t-1} + \sum_{i=1}^{k} \alpha_{9i}\Delta SUP_{t-1} + \sum_{i=1}^{k} \alpha_{10i}\Delta ARF_{t-1} \\ & + \sum_{i=1}^{k} \alpha_{11i}\Delta DIF_{t-1} + \gamma_1 SOY_{t-1} + \gamma_2 ARE_{t-1} + \gamma_3 YED_{t-1} + \gamma_4 WOT_{t-1} + \gamma_5 RER_{t-1} + \gamma_6 GOP_{t-1} \\ & + \gamma_7 CIF_{t-1} + \gamma_8 TRB_{t-1} + \gamma_9 SUP_{t-1} + \gamma_{10} ARF_{t-1} + \gamma_{11} DIF_{t-1} + \varepsilon_t \end{aligned} \tag{4}$$

Where,

α_0 = Constant Parameter
α_{1i} -α_{5i} = Parameters for Short Run,
γ_1 - γ_5 = Multipliers for Long Run
Δ = Symbol for First Difference
k = Order for Maximum Lag
ε_t = Error Term

The Jarque – Bera statistics is given as:

$$\mathrm{JB} = \frac{N}{6}\left(S^2 + \frac{(K-3)^2}{4}\right) \tag{5}$$

Where,

JB=Jarque – Bera (Number)
S^2 = Skewness (Number)
K = Kurtosis (Number)
N = Number of Observation (Number)

3. Results and Discussion

The summary statistics of factors involved in the competitiveness of Nigerian soybean production in international trade is displayed in Table 1.

3.1. Summary statistics of predictors in determining the competitiveness of Nigeria soybean production in the international market

Table 1 displays the mean estimates of ARE, YED, WOT, SEV, SOY export, and SOY production from 2007 to 2022. The mean forecast of WOT, SOY export, and SOY production are 155,416, 923.9, 18, 940.738, and 732 187 metric tonnes, respectively. The mean estimates of ARE, YED, and SEV are 777, 374.81 ha, 946.5812 kg ha-1, and 8267310 USD, respectively. ARE has highest value of 1,207,740 ha in 2020, while the highest values of YED, WOT, SEV, SOY export, SOY production were 1295.1Kg/ha, 215, 460,396.9 tonnes,14, 251,000 USD, 34, 587.09 tones, and 1,262, 280 tones in the year 2010, 2020, 2020, 2017, and 2020 respectively (Figure 1 and Figure 2). The highest world soybean exports occurred in 2020, estimated at 173.3 million tons (Figure 3). This outcome compares with the highest soybean-producing countries, including Brazil (1st) with an estimated output of 121.2 million tons, followed by the USA (2nd) with an estimated production of 116.2 million tons, and Argentina (3rd) with an estimated output of 43.86 million tons in 2022 (FAO, 2024). The lowest values correspond to 2010 for ARE and SOY production, while those of YED, WOT, SEV, and SOY export are in 2009, 2007, 2019, and 2021 respectively. The kurtosis, skewness, and JB statistics tests were reported to determine the normality of the data. The skewness of zero and kurtosis of 3 signifies that the data are typically distributed. At the same time, the probability of the JB statistics of more than 0.05 connotes the acceptance of the null hypothesis that the data are typically distributed. All the regressors are negatively skewed, with values close to zero and kurtosis close to 3, implying normal distribution. Moreover, the probability figures of JB were more outstanding than 0.05 for all regressors, which signifies that the variables are normally distributed.

Table 1. Summary statistics of predictors in the competitiveness of Nigerian soybean production in international market.

Predictors	ARE (Ha)	YED (100 g Ha^{-1})	WOT (tons)	S0YEV (1000 USD)	SOY Export (tons)	SOY Prod (tons)
Mean	777,374.81	9465.812	155,416,923.9	8267.31	18,940.738	732,187
Maximum	1,207,740	12,951	215, 460,396.9	14, 251	34,587.09	1,262,280
Minimum	281,890	7206	93, 716,241	46	6,184	365,080
Std Deviation	263,318.26	1309.52	40,288,692.64	11113.64	28,723.97	272,090.17
Skewness	-0.03167	-0.043865	-0.03072	-0.03164	-0.03407	-0.04528
Kurtosis	2.25738	2.326381	2.31307	2.320014	2.37009	2.47045
Jarque- Bera	**0.370354**	**0.3076778**	**0.317138101**	**0.3109749**	**0.267651**	0.264312
Sum	12,437,997	151453	2,486,670,782	132277	303051.82	11,714,992
Probability	0.0617246	0.062703	0.064432	0.069274	0.06342	0.06175
Observation	16	16	16	16	16	16

Source: Data Analysis (2024), Source: FAO (2024)
SOYEV- Soyabean Export Value, USD-United States Dollar

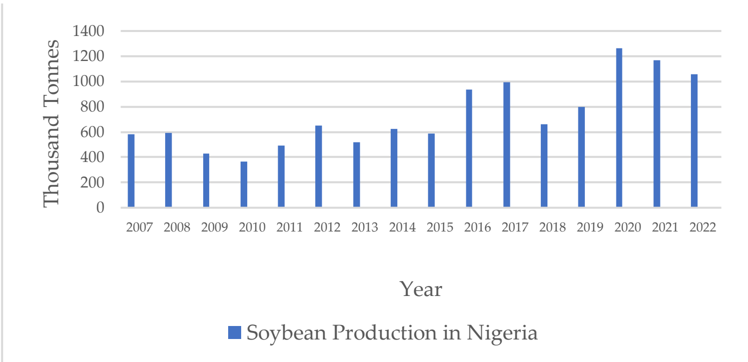

Figure 1. Soybean production (tons) in Nigeria

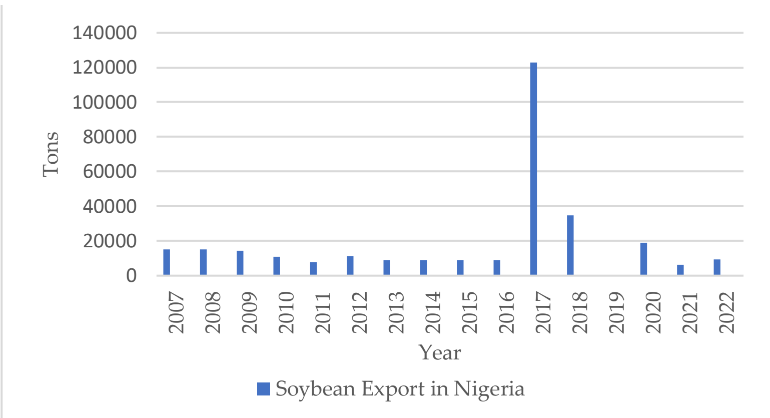

Figure 2. Soybean export (tons) in Nigeria

3.2. The ADF unit root test

The ADF Unit Root test was carried out on the stationarity data, as shown in Table 2. The outcomes display that ARE, RER, GOP, CIF, and TRB are stationary at levels I (0), while SOY, YED, WOT, SUP, ARF, and DIF are stationary at first difference I (1). The regressors have a combined order of integration. This outcome agrees with the outcomes of Umar (2020). This set the stage for employing the Bound test for co-integration analysis and the ARDL technique.

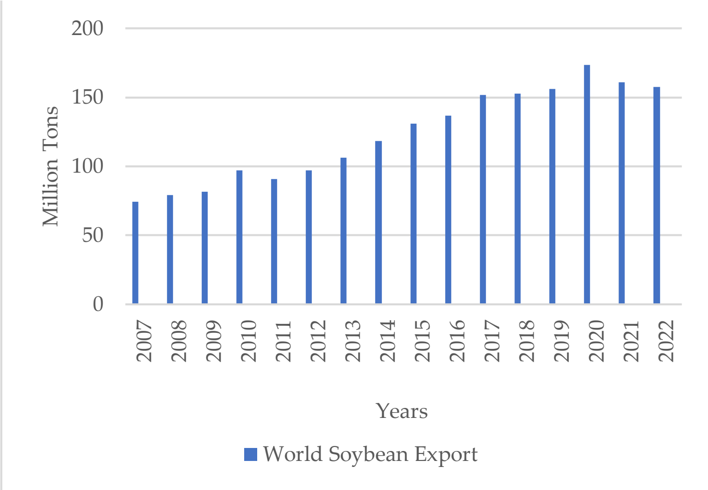

Figure 3. World soybean export in million tons.

Table 2. The ADF stationarity test.

Variables	ADF at Levels	Critical Estimates at 5%	Order of Integration	ADF at 1st Difference	Order of Integration
SOY	- 2.79	-2.96	NS	-4.18	I (1)
ARE	- 3.89	-2.96	S	Na	I (0)
YED	- 1.14	-2.96	NS	-8.09	I (1)
WOT	-1.10	-2.96	NS	-6.63	I (1)
RER	- 5.17	-2.96	S	Na	I (0)
GOP	-5.34	-2.96	S	Na	I (0)
CIF	-4.94	-2.96	S	Na	I (0)
TRB	-4.89	-2.96	S	Na	I (0)
SUP	-1.03	-2.96	NS	-6.74	I (1)
ARF	-1.07	-2.96	NS	-6.02	I (1)
DIF	-1.09	-2.96	NS	-6.79	I (1)

Source: Data Analysis (2024), NS- Not Stationary, Na-Not Available, NS-Stationary.

3.3. The co-integration test

Table 3 displays the bound test for co-integration; the outcome shows the upper and lower bounds for asymptotic or finite sample size and the actual sample size. The bound test revealed the critical estimates of 2.91 and 4.09 for the lower and upper bound, respectively, at the 5% significance level. The estimated F-statistics of 13.69 is more than the upper bound value of 4.09 at the 5% significance level. This outcome shows that the regressors in the model are co-integrated, implying a long-run association between the regressors in the model. This outcome conforms with the outcome of Olojede and Micheal (2020).

Table 3. Bounds test for co-integration result.

Significant Level	I(0) Bound	I(1) Bound	Value
10%	2.47	3.48	F-Statistics = 13.69, K = 4
5%	2.91	4.09	
1%	4.17	5.51	

Source: Data Analysis (2024).

3.4. Factors Determining the competitiveness of Nigeria's soybean in international market

Table 4 displays the regressors influencing the competitiveness of Nigeria's soybean in the international market. The ARE exerts a positive and significant influence on SOY seed export. This agrees with the a priori expectation. A unit increase in ARE cultivated for soybeans will increase SOY export by 0.79. The coefficient of YED is also positive and significant; this connotes that the output of soybean seed can translate to availability for export, which is expected to expand market access to Nigerian soybean seed. The RER has a negative coefficient, which aligns with the a priori expectation. The coefficients of GOP, TRB, SUP, and DIF had negative values and were significantly different from zero in affecting the competitiveness of soybean in international trade. This work is in line with a priori expectations. The coefficients of CIF and ARF had positive values and were significantly different from zero in affecting the competitiveness of soybeans in international trade. This outcome is in line with apriorism expectations. The infrastructure challenges are access roads to farms (ARF) and the distance of farms to nearby market infrastructures (DIF). The error correction parameter conforms to a priori expectation and is statistically significant. The extent of the co-integration term connotes that if there is any departure, the long-run equilibrium is regulated moderately, with about 86% of the disequilibrium being removed in each period. This displays the speed of regulation to where soybeans will balance even when there is disequilibrium at the rate of 86%. The explanatory power of the regressors used in explaining the competitiveness of Nigeria's soybean in international trade is shown by the summary statistics; the R^2 is 0.78 (78%). The F-statistics reveal that the model is statistically significant ($P < 0.05$). The DW calculated is 1.6 which connotes low level of autocorrelation. This outcome is in agreement with the outcomes of Alabi et al. (2022), Obansa et al. (2013), and Umar (2020).

The Estimated Long Run Equation is stated thus:

$$\underset{}{SOY} = \underset{(0.0081)}{0.79ARE} + \underset{(4.17)}{39.51YED} + \underset{(0.007)}{0.006WOT} + \underset{(4.38)}{2.13\,RER} - \underset{(0.026)}{0.162GOP} + \underset{(0.016)}{0.161CIF} - \underset{(0.024)}{0.072TRB} - \underset{(0.022)}{0.111SUP} + \underset{(0.005)}{0.021\,ARF} - \underset{(0.017)}{0.121DI} \quad (6)$$

Table 4. Estimated long run result.

Variable	Coefficient	Standard Error	t- Value
ARE	0.79***	0.081	9.75
YED	39.51***	4.17	9.47
WOT	0.006	0.007	0.86
RER	-2.14	4.38	-0.49
GOP	-0.162***	0.026	-6.23
CIF	0.161***	0.016	10.06
TRB	-0.072**	0.024	-2.99
SUP	-0.111***	0.022	-4.96
ARF	0.021***	0.005	4.07
DIF	-0.121***	0.017	-6.77
C	2.391	561.5	0.004
CointEq (-1)	-0.86***	0.086	-10.00
R^2 = 0.78			
DW = 1.6			
F =74.9**			

Source: Data Analysis (2024), DW-Durbin Watson Statistics
- Significant at 5% Probability Level, *- Significant at 1% of Probability Level

3.5. Diagnostic tests

The outcome of the diagnostic test is presented in Table 5. The outcomes connote that the model passed the test of homoscedasticity, serial correlation, normality (JB), and linearity, given that their probability values are more than 0.05; this connotes the acceptance of the null hypothesis of no serial correlation and presence of homoscedasticity, normal distribution, and no misspecification of function. This, therefore, implies that the stochastic error term is white noise and has a mean of zero and variance that is constant. This makes the estimates reliable and consistent.

Table 5. Summary of diagnostics test.

Test	F-Statistics	Prob (F-Statistics)
No Serial Correlation (Breusch-Godfrey LM Test)	0.9102	0.4001
Homoscedasticity (Breusch-Godfrey)	5.7202	0.7104
Normality (Jarque-Bera)	2.9102	0.2104
Model Specification (Ramsey RESET)	0.6104	0.4143

Source: Data Analysis (2024)

4. Conclusions

This study was executed to determine the socio-economic and infrastructural challenges affecting the competitiveness of Nigeria's soybean production in international trade: auto-regressive distributive lag approach. The primary and secondary data were used for this research. The primary data covered the period 2007 to 2022. The ADF conducted for unit root test revealed that ARE, RER, GOP, CIF, and TRB are stationary at I (0) (level), while SOY, YED, WOT, SUP, ARF, and DIF are stationary at I (1) (first difference). The bound test showed the critical estimates of 2.91 and 4.09 for the lower and upper bound, respectively, at the 5% significance level. The estimated F- statistics of 13.69 is more than the upper bound estimate of 4.09 at the 5% significance level. The estimated long-run outcome using ARDL show that is, YED, GOP, CIF, TRB, SUP, ARF, and DIF is the significant regressors affecting the competitiveness of soybean production in the international market. The speed of regulation to where soybeans will balance even when there is disequilibrium is at the rate of 86%. The explanatory power of the regressors used in explaining the competitiveness of Nigeria's soybean in international trade is shown by the summary statistics; the R^2 is 0.78 (78%). The F-statistic reveals that the model is statistically significant ($P < 0.05$). The DW calculated is 1.6, which connotes a low level of autocorrelation. The aftermath of the diagnostic test connotes the acceptance of the null hypothesis of no serial correlation, presence of homoscedasticity, normal distribution, and no misspecification of function. Based on the outcomes of this research, the following suggestions were made:

(i) Government and private sectors must develop strategies to promote greater access to farm advisory services.

(ii) The government must invest in infrastructures such as roads, markets, and storage investment infrastructures.

(iii) Government and private organizations need to invest in the education of farmers, technical innovations, and information transfer to improve the competitiveness of the producers.

(iv) Policies should be implemented to stimulate the exportation of soybeans. Such policies include the granting of tax holidays, trade barriers, provision of storage facilities, and long-term export credit at concessionary interest rates to exporters of soybeans.

(v) Increasing the productive capacities of producers of soybeans by providing improved seeds, fertilizers, and extension services and providing credit facilities at low interest rates without administrative bottlenecks to producers.

(vi) Research activities should be carried out to improve the quality of soybeans and ensure they are free from pesticides, mycotoxins, contaminations, and aflatoxins.

(vii) The government should be in tune with favorable policies that affect the international market, such as exchange rates, training on soybean production, and capacity building on proper packaging.

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References

Alabi O, Madaki M, Sanusi S, Umar S, Omole E, Olumuyiwa S, David H, Emeghara U, Waziri-Ugwu P, Shaba M (2022). Factors influencing export performance of ginger (*Zinbiger officinale*) in Nigeria. *International Journal of Agriculture, Environment and Food Sciences,* 6(3), 370 – 377. DOI: https://doi.org/10.31015/jaefs.2022.3.5.

Dickey D, Fuller W (1981) Likelihood ratio statistics for autoregressive time series with unit root. *Econometrica,* 49, 1057-1072. http://dx.doi.org/10.2307/1912517.

Dickey D, Wayne A (1979) Distribution of the estimators for autoregressive time series with a unit root. *Journal of the American Statistical Association,* 74,366, 427 – 431.

FAO (2015). Food and Agriculture Organization, Database, Rome, Italy

FAO (2024). Food and Agriculture Organization, Database, Rome, Italy

Kingu J (2014). Determinants of Tanzania agricultural export: Case of cotton lint. *Developing Country Studies*, 4 (1), 91 – 104.

Mohammed A, Al-hassan S, Amegashie D (2016). Technical efficiency of soybean farmers in the Northern Region of Ghana. *ADRRI Journal of Agriculture and Food Sciences,* 11(2), 20 – 38.

Nordin A, Shariff F, Simeh A, Baharim B (2008). Future competitiveness of the Malaysian PKE. *Oil Palm Industry Economics Journal,* 8 (2), 8-16.

Obansa S, Okoroafor O, Aluko O, Eze M (2013). Perceived relationship between exchange rate, interest rate and economic growth in Nigeria: 1970 – 2010. *American Journal of Humanities and Social Sciences,* 1(3), 116 – 124.

Olojede A, Micheal N (2020). Agricultural export and economic growth in Nigeria: A disaggregated analysis. *SSRG International Journal of Economics and Management Studies,* 7(6), 27 – 33.

Pesaran M, Shin Y, Smith R (2001). Bounds testing approaches to the analysis of level relationships. *Journal of Applied Econometrics*, 16(3), 289 – 326.

Ude M, Adeoye B, Yusuf S, Balogun O, Akinlade J (2013). Competitiveness of rice processing and marketing in Ebonyi State: A policy analysis approach. *International Journal of Agricultural Sciences,* 3 (3), 410 – 418.

Umar S (2020). The competitiveness of Nigeria's sesame and soybean export in international trade: 1986 – 1997. PhD Thesis, Department of Agricultural Economics, University of Abuja, (Unpublished) Nigeria

Yusuf H, Omokore D, Olawoye R, Yusuf H, Shuaibu H (2022). Analysis of production efficiency among small-scale soybean farmers in Sabon-Gari Local Government Area of Kaduna State, Nigeria. *International Journal of Agricultural Economic, Management and Development* (IJAEMD), 8, 195 – 205.

Yousif I (2015). Analysis of factors constraining the competitiveness of sesame export in Sudan. Working Paper, No. 1, Agricultural Economics Working Paper Series, Khartoum University.

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Mitigation of Adverse Effects of Drought and Low Temperature Stresses by Seed Priming in Sunflower (*Helianthus annuus* L.) During Germination

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HIGHLIGHTS

- The study evaluated the effect of priming on sunflower germination under drought and low temperatures.
- Drought more severely reduced germination and seedling growth than low temperatures.
- Low temperatures at 15°C decreased shoot and root length by 63% and 59%, respectively.
- Hydration can ameliorate low temperatures and drought-induced damage in sunflower.

Abstract

The study aimed to determine the effects of seed priming on germination and seedling development in sunflowers exposed to drought and low-temperature stresses. The primed seeds (KNO_3, and hydration) of Sanbro MR were evaluated at low temperatures of 15 °C and 18 °C under drought conditions induced by PEG-6000 at the water potentials of 0 (distilled water), −0.2, −0.4, and −0.6 MPa. Unprimed seeds were used as a control, and a temperature of 25 °C was considered optimal. The results showed that germination was negatively affected by both stress factors, although this varies according to priming applications. Drought inhibited germination and seedling growth more than low temperatures. Seedling growth was also more adversely affected by low temperatures compared to germination. Under low temperatures, there was a 63% decrease in shoot length and a 59% decrease in root length. Drought stress resulted in an 85% reduction in shoot length, while roots were reduced by 65%. The primed seeds with KNO_3 gave the highest germination percentage at -6 MPa at 15 °C. However, hydration showed superiority under different drought levels at 18 °C and 25 °C. It was concluded that hydrated seeds could be recommended for increased germination and seedling growth under low temperatures and drought stresses in sunflowers.

Keywords: *Helianthus annuus* L.; hydropriming; potassium nitrate; low temperature; drought

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1. Introduction

Sunflower (*Helianthus annuus* L.) is an annual oilseed crop and is an economically significant commodity in Türkiye because of its substantial role in vegetable oil production. It has an area of 1 million hectares with an average production of 2.5 million tons (TUİK 2024), of which 70% of the sowing area is under rainfed conditions. For this reason, it is exposed to several abiotic stresses such as drought, low and high temperature, salinity, and heavy metals, which affect it from germination to harvest (Ali et al. 2017; Hussain et al. 2018; Haj Sghaier et al. 2023).

The germination process is mainly influenced by moisture, oxygen, and temperature, which are often combined with abiotic stress factors (Jiang et al. 2023). During germination, low temperatures limit water absorption by seeds, leading to the inhibition of germination and seedling growth (Hoppe and Theimer 1997; Chakraborty and Dwivedi 2021; Haj Sghaier et al. 2023). The lack of water restricts the seed's capacity to take in the necessary water and slows down the process of germination by inhibiting the enzymes that are essential for allowing energy for seedling development (Soleimanzadeh 2012; Wen 2015; Haj Sghaier et al. 2023).

Priming promotes germination by activating metabolic activities, enhances osmotic adjustment, strengthens cell membranes, and stimulates the production of protective proteins and antioxidants. As a result, primed seeds show better germination and seedling vigor in drought, salinity, and extreme temperatures (Harris et al. 1999; Kaya et al. 2006; Aswathi et al. 2021; Ghosh et al. 2024; Rehman et al. 2024; Sneha et al. 2024). Seeds can be primed in a variety of media, including water (hydropriming), aerated solutions with low water potential containing polyethylene glycol, or salt solutions ($CaCl_2$, KNO_3, KCl, K_3PO_4, $MgSO_4$, and NaCl) (osmopriming), solid matrix (matripriming), and priming with plant growth regulators and polyamines (Kaya et al. 2024; Rehman et al. 2024). The superiority of primed seeds has been found under drought stress in soybean (*Glycine max* Merril.) (Sintaha et al. 2022), wheat (*Triticum aestivum* L.) (Tabassum et al. 2018), maize (*Zea mays* L.) (Khan et al. 2015), safflower (*Carthamus tinctorius*) (Akbari et al. 2020), and sunflower (*Helianthus annuus* L.) (Kaya et al. 2006). Also, primed seeds have improved the germination performance of maize (*Zea mays* L.) (Guan et al. 2009), rapeseed (*Brassica napus* L.) (Zhu et al. 2021), cotton (*Gossypium hirsutum* L.) (Xia et al. 2023) and sugar beet (*Beta vulgaris* L.) (Kaya and Kulan 2020) under low temperatures. For these reasons, this study was conducted to investigate whether primed sunflower seeds improve germination and early growth under different combinations of low temperatures and drought.

2. Materials and Methods

This study was conducted at Eskişehir Osmangazi University, Seed Science and Technology Laboratory in 2023, Odunpazarı, Eskişehir. The seeds of sunflower hybrid Sanbro MR obtained from Syngenta Seeds Company were used. To create drought stresses, different water osmotic potentials of −0.2, −0.4, and −0.6 MPa were arranged using polyethylene glycol (PEG) 6000 with the formula described by Michel and Kaufmann (1973) for 15 °C, 18 °C, and 25 °C. Distilled water was used as a control (0 MPa).

2.1. Seed priming

Two seed priming were applied to the seeds, as defined by Kaya et al. (2006). For hydration, sunflower seeds were submerged in distilled water at 20 °C for 16 h in the dark. For KNO_3 treatment, the seeds were immersed in a 500 ppm KNO_3 solution at the same temperature and duration and then they were rinsed with tap water. The primed seeds were immediately surface-dried with paper towels and then they were dried to their initial moisture content at room temperature (roughly 22 °C, 45% relative humidity). Primed (hydration and KNO_3) and unprimed (control) seeds were left at room temperature for two days (Kaya et al. 2015).

2.2. Germination test

A germination test was conducted with 200 seeds (4×50) for each treatment to evaluate seed viability according to the ISTA (2003) guidelines. Fifty seeds were inserted into two-layer filter papers wetted with 7 mL of the distilled water for each paper. After filter papers with seeds were rolled, they were placed into a sealed plastic bag to avoid water loss. The packages were incubated at 25 °C in the dark, and seeds with 2 mm radicle were counted every 24 h for 10 d as germinated. To evaluate the speed of germination, mean germination time (MGT) was calculated according to ISTA (2003) rules. MGT= Σ(Dn)/Σn, where n is the seed number germinated on day D and D is the number of days from the beginning of the germination test. On the 10th day, ten seedlings from each treatment were randomly selected to determine the seedling growth traits such as root length (RL), shoot length (SL), seedling fresh weight (SFW), and seedling dry weight (SDW). After the seedling fresh weight was directly weighed, the seedlings were transferred into an oven at 80 °C for 24 hours for the determination of dry weight (Ergin et al. 2021). The root/shoot ratio was determined by dividing the root length by the shoot length. The germination index (GI) was also calculated according to Salehzade et al. (2009) with the following formula 1:

$$\text{GI} = \text{Number of germinated seeds/days of first count} + \ldots + \text{Number of germinated seeds/days of the final count.} \quad (1)$$

Where n is the seed number germinated on day D, and D is the number of days from the beginning of the germination test.

2.3. Statistical analysis

All obtained data were analyzed according to a three factor in a completely randomized design with four replications using the MSTAT-C (Michigan State University, v. 2.10) statistical software. The means were separated by Tukey's test at $p<0.05$ level.

3. Results

In this study, the analysis of variance and main effects of seed priming, temperature, and drought level on germination and early seedling development of sunflower are shown in Table 1. The results showed that temperature, seed priming, drought, and all the combinations of the interactions were found to be significant for germination percentage, mean germination time, and germination index.

At low temperatures, germination percentage only decreased at 15 °C, while decreasing temperature caused a retardation in mean germination time and a reduction in germination index (Table 1). Hydrated seeds gave better results for germination percentage, mean germination time, and germination index than the control and KNO_3 treated seeds. As expected, increased drought stress resulted in increased germination time, while germination percentage and germination index decreased. Previous studies show that germination decreases as drought stress increases (Ahmad et al. 2009) and that priming applications increase germination percentage and germination index (Fatemi 2014; Čanak et al. 2014) are in line with the results of the current study.

Low temperatures caused a reduction in the shoot and root length, seedling fresh weight, and root/shoot ratio in sunflower, while hydration considerably stimulated them. The shoot length was significantly decreased by increasing drought stress. A similar observation was also determined in root length, except for -0.2 MPa PEG. In all treatments, the roots were longer than the shoots, which is proof that shoots are more sensitive than roots under low temperatures and drought stress (Table 2). Root length was not affected by the interaction between temperatures, priming, and drought. This result confirms the findings of Aboutalebian et al. (2021), who reported a reduction in root and shoot growth of canola (*Brassica napus* L.) varieties under drought stress. Similar results were also found in sunflower under drought stress (Nezami et al. 2008; Basit et al. 2024) and rice (*Oryza sativa* L.) under low-temperature stress (Doddagoudar et al. 2023).

Low-temperature stress significantly affected the fresh weight of sunflower seedlings. The fresh weight of sunflowers was significantly reduced at 15 °C. Of priming treatments, the highest seedling fresh weight (174 mg $plant^{-1}$) was recorded from hydration. Drought reduced seedling fresh weight, and a similar trend was also determined for dry weight. There was no significant effect of temperature on dry weight; hydrated seeds produced a lower dry weight of seedlings than the others. The root/shoot ratio decreased with decreasing temperature, but priming treatments helped to increase it significantly. Although they were in the same statistical group, higher values were measured with the application of hydration than with KNO_3. It was determined that under moderate drought stress, the roots developed more rapidly than shoots, which resulted in a higher root-to-shoot ratio than the control (Table 2). Kaya and Kulan (2020) determined that primed sugar beet seeds produced notably heavier seedlings than control. Moghanibashi et al. (2012) stated that lighter seedlings were formed in sunflowers with increasing drought stress levels. Szczerba et al. (2021) reported that seedling weights of soybean varieties at low temperatures were considerably lower compared to optimum temperatures, which is consistent with our results.

Table 1. The main effects of seed priming, temperature, and drought on germination properties of sunflower

	Germination percentage (%)	Mean germination time (day)	Germination index
Temperature (A)			
15 °C	88.1±1.5	4.17±0.16	12.3±0.61
18 °C	92.0±0.9	3.60±0.17	15.0±0.77
25 °C	89.6±2.2	2.69±0.14	21.2±.1.41
Priming (B)			
Control	89.0±2.39	3.72±0.18	14.5±0.87
Hydration	91.9±1.31	3.30±0.19	17.9±1.29
KNO_3	88.8±0.77	3.45±0.16	16.1±1.12
Drought (C)			
0	94.3±0.63	2.20±0.10	25.1±1.41
-0.2 MPa	94.6±0.63	2.98±0.10	17.3±0.61
-0.4 MPa	92.9±0.83	3.76±0.13	13.5±0.53
-0.6 MPa	77.8±2.75	5.02±0.13	8.7±0.37
Analysis of variance			
A	**	**	**
B	**	**	**
A×B	**	**	**
C	**	**	**
A×C	**	**	**
B×C	**	**	**
A×B×C	**	**	**

**: Significance level at p<0.01.

As can be seen in Figure 1A, the root length of the sunflower increased as the temperature increased. At all temperatures, the hydrated seeds developed the longest roots, while the unprimed seeds had the shortest root length. Similarly, the highest root length at all drought stress levels was achieved by hydrated seeds, followed by KNO_3-treated seeds. It increased considerably at -0.2 MPa and then decreased rapidly at higher drought levels, with the lowest reductions occurring in hydration application (Figure 1B). Additionally, increased drought stress had different effects on root length depending on temperature (Figure 1C). The longest root length was obtained at 25 °C under all drought severities, followed by 18 °C and 15 °C. At -0.2 MPa and -0.4 MPa, the root length was longer than that of the unstressed control seeds. This improvement resulted in an increase in root/shoot ratio under these drought stresses. Root growth under stress conditions is directly related to seedling survival (Haj Sghaier et al. 2023), and it can be promoted with priming. Rahimi (2013) demonstrated that seedlings grown from primed seeds developed longer roots under low-temperature stress.

Table 2. The seedling growth parameters of sunflower seeds under different levels of drought and temperatures

	Shoot length (cm)	Root length (cm)	Seedling fresh weight (mg plant^{-1})	Seedling dry weight (mg plant^{-1})	Root/shoot ratio
Temperature (A)					
15 °C	0.70±0.06	3.13±0.27	135±6.4	62.6±0.48	4.27±0.20
18 °C	0.92±0.09	4.77±0.37	148±7.8	61.3±0.58	5.78±0.44
25 °C	1.87±0.28	7.56±0.58	199±16.9	61.4±0.48	7.75±0.72
Priming (B)					
Control	0.96±0.10	3.67±0.35	147±7.8	62.7±0.43	4.41±0.35
Hydration	1.39±0.24	6.28±0.53	174±14.6	60.2±0.58	6.92±0.62
KNO_3	1.14±0.19	5.52±0.52	161±12.4	62.4±0.46	6.46±0.55
Drought (C)					
0	2.70±0.31	5.79±0.26	271 ±14.9	62.3±0.61	2.99±0.26
-0.2 MPa	0.95±0.06	7.89±0.66	159±6.5	62.4±0.50	8.31±0.44
-0.4 MPa	0.61±0.02	4.90±0.52	114±1.6	61.8±0.63	7.61±0.66
-0.6 MPa	0.40±0.02	2.03±0.27	98±0.8	60.6±0.62	4.82±0.60
Analysis of variance					
A	**	**	**	*ns*	**
B	**	**	**	**	**
A×B	**	**	**	*ns*	**
C	**	**	**	*	**
A×C	**	**	**	*	**
B×C	**	**	**	*ns*	**
A×B×C	**	*ns*	**	**	**

*, ** show significance level at p<0.05 and p<0.01, ns: non-significant.

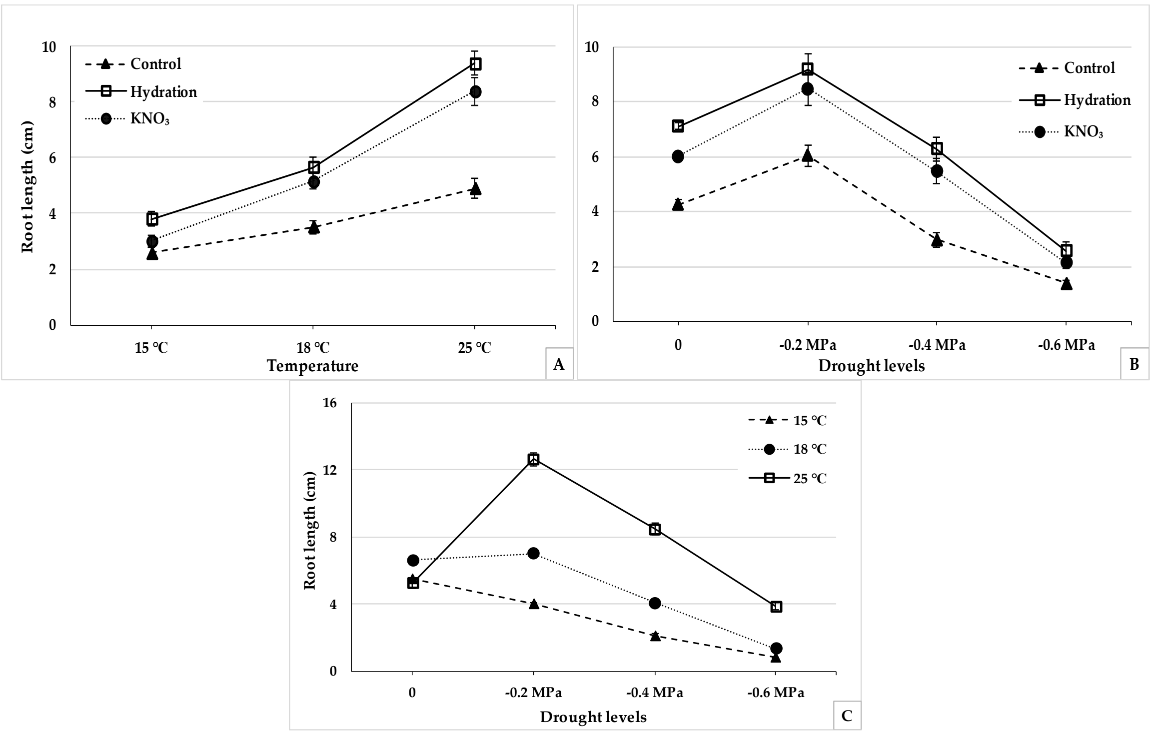

Figure 1. Changes in root length of sunflower under different temperatures (A) and drought levels (B and C)

Table 3. Changes in germination characteristics of sunflower seeds exposed to different seed priming under different drought levels and temperatures

	PEG (MPa)	Germination percentage (%) 15 °C	18 °C	25 °C	Mean germination time (day) 15 °C	18 °C	25 °C
Control	0	99^{a}±0.5	99^{a}±0.5	97^{abc}±1.5	2.98^{f}±0.02	2.31^{jk}±0.05	1.97^{k*}±0.01
	-0.2	97^{ab}±1.0	98^{ab}±0.5	96^{abc}±1.4	3.91^{d}±0.10	3.05^{f}±0.04	2.41^{h-k}±0.04
	-0.4	93^{abc}±2.2	92^{abc}±4.5	94^{abc}±1.4	5.33^{ab}±0.11	4.05^{cd}±0.06	2.88^{f-i}±0.08
	-0.6	73de±6.6	91^{abc}±4.5	42^{f}±5.0	5.83^{a}±0.07	5.42^{ab}±0.06	4.51^{c}±0.09
Hydration	0	93^{abc}±1.5	95^{abc}±1.3	96^{abc}±1.0	2.38^{ijk}±0.03	2.10^{k}±0.08	1.24^{l}±0.04
	-0.2	96^{abc}±1.5	95^{abc}±0.6	98^{a}±1.4	3.25^{ef}±0.05	2.96^{f}±0.03	2.18^{k}±0.05
	-0.4	96^{abc}±1.5	94^{abc}±1.8	98^{a}±1.2	3.99^{d}±0.05	3.64^{de}±0.13	2.82^{f-j}±0.07
	-0.6	67^{e}±5.3	85^{a-d}±4.5	93^{abc}±1.3	5.65^{a}±0.15	5.44^{ab}±0.08	3.94^{d}±0.30
KNO_3	0	89^{abc}±1.7	92^{abc}±1.0	93^{abc}±1.3	2.94^{fg}±0.08	2.44^{g-k}±0.09	1.42^{l}±0.02
	-0.2	88^{a-d}±1.4	93^{abc}±1.0	92^{abc}±1.9	3.73^{de}±0.11	3.10^{f}±0.06	2.25^{k}±0.07
	-0.4	87^{a-d}±3.0	91^{abc}±0.6	92^{abc}±2.3	4.52^{c}±0.06	3.68^{de}±0.07	2.92^{fgh}±0.04
	-0.6	83^{bcd}±3.5	82^{cde}±1.9	86^{a-d}±3.7	5.57^{ab}±0.08	5.06^{b}±0.10	3.78^{d}±0.08

*: Means followed by the same letter(s) are not significant at $p<0.05$.

Germination percentages of primed and unprimed seeds varied with temperatures and drought levels, and it was reduced by decreasing temperatures. At 15 °C, KNO_3-treated seeds germinated lower than the others, but they produced a stable germination percentage under all levels of drought. Moreover, it had the highest germination percentage at -0.6 MPa at 15 °C. An apparent decrease in the germination percentage of unprimed and hydrated seeds was detected at -0.6 MPa. A similar trend was observed at 18 °C (Table 3). It was found that hydrated seeds germinated faster than others at all temperatures and drought levels. Some researchers supporting the results of the current study have reported that priming applications can also alleviate stress-related negative effects on sunflower germination traits (Moghanibashi et al. 2012; Hamidi and Pirasteh-Anosheh 2013; Prayaga et al. 2017). Zhang et al. (2019) determined that germination percentage decreased and mean germination time was prolonged in rapeseed under low-temperature stress.

Table 4. Changes in germination index of sunflower seeds exposed to seed priming treatment under different drought levels and temperatures.

	PEG (MPa)	Germination index 15 °C	18 °C	25 °C
Control	0	17.1^{gh}±0.2	22.4^{cde}±0.5	25.1^{c*}±0.6
	-0.2	12.8^{jkl}±0.3	16.5^{gh}±0.2	20.9^{ef}±0.5
	-0.4	9.0^{mno}±0.3	11.7^{klm}±0.4	17.0^{gh}±0.3
	-0.6	6.4^{o}±0.6	8.5^{no}±0.3	6.9^{o}±0.9
Hydration	0	20.4^{ef}±0.5	24.2^{cd}±1.0	42.3^{a}±0.9
	-0.2	15.4^{hij}±0.3	17.3^{gh}±0.1	23.2^{cde}±0.3
	-0.4	12.4^{jkl}±0.4	13.5^{ijk}±0.6	18.6^{fg}±0.3
	-0.6	7.9^{no}±0.3	8.1^{no}±0.5	12.2^{kl}±0.5
KNO_3	0	16.6^{gh}±0.7	20.7^{ef}±0.5	37.5^{b}±0.4
	-0.2	12.4^{kl}±0.5	16.1^{ghi}±0.3	21.5^{def}±0.8
	-0.4	9.9^{lmn}±0.4	12.9^{jk}±0.2	16.8^{gh}±0.6
	-0.6	7.8^{no}±0.3	8.4^{no}±0.4	12.4^{kl}±0.6

*: Means followed by the same letter(s) are not significant at $p<0.05$.

The germination index decreased with an increase in drought stress at all temperatures. It increased when temperature and drought stress were increased. However, hydrated seeds produced a higher germination index at all levels of drought and temperature. It was determined that priming applications under low temperatures and -0.2 MPa PEG gave higher values than the control (Table 4). The germination index is a property that clearly demonstrates the impacts of temperature and drought stress. It provides valuable insights into the general health and viability of seeds under stress conditions. The efficacy of hydration in inducing low-temperature tolerance is noticeable (Doddagoudar et al. 2023) and an improvement in germination index was determined in canola under drought stress (Heshmat et al. 2011). Furthermore, hydrated seeds produced better results than osmopriming in sunflower (Pahoja et al. 2013), which is consistent with the findings of the present study.

Table 5. Changes in seedling characteristics of sunflower seeds exposed to priming treatments under different drought levels and temperatures.

Priming	PEG (MPa)	Shoot length (cm)			Root/shoot ratio		
		Temperature			Temperature		
		15 °C	18 °C	25 °C	15 °C	18 °C	25 °C
Control	0	1.40^{efg}±0.06	1.89^{de}±0.08	2.68^{c}±0.22	3.30^{klm}±0.24	2.89^{klm}±0.09	1.03^{m}*±0.13
	-0.2	0.74^{jkl}±0.05	0.61^{kl}±0.02	1.32^{f-i}±0.10	4.78^{g-l}±0.26	8.56^{def}±0.73	7.31^{e-i}±0.63
	-0.4	0.45^{l}±0.04	0.57^{kl}±0.02	0.64^{jkl}±0.02	3.10^{klm}±0.49	3.97^{j-m}±0.19	8.27^{def}±0.63
	-0.6	0.31^{l}±0.04	0.44^{l}±0.04	0.50^{l}±0.05	2.72^{lm}±0.24	2.74^{lm}±0.25	4.30^{i-l}±0.51
Hydration	0	1.34^{fgh}±0.08	2.42^{cd}±0.14	6.55^{a}±0.22	4.84^{g-l}±0.33	3.26^{klm}±0.15	1.08^{m}±0.04
	-0.2	0.75^{jkl}±0.03	0.79^{h-l}±0.04	1.85^{ef}±0.04	6.49^{f-j}±0.46	10.69^{bcd}±0.39	7.84^{d-g}±0.72
	-0.4	0.54^{l}±0.03	0.62^{kl}±0.04	0.70^{jkl}±0.02	5.86^{f-k}±0.18	8.41^{def}±0.91	15.07^{a}±0.41
	-0.6	0.31^{l}±0.01	0.38^{l}±0.02	0.42^{l}±0.01	2.71^{lm}±0.19	3.27^{klm}±0.52	13.54^{ab}±1.35
KNO_3	0	1.18^{g-j}±0.07	1.41^{efg}±0.09	5.41^{b}±0.20	4.73^{h-l}±0.41	4.67^{h-l}±0.09	1.10^{m}±0.11
	-0.2	0.63^{kl}±0.02	0.75^{jkl}±0.03	1.10^{g-k}±0.08	5.95^{f-k}±0.14	10.02^{cde}±040	13.15^{ab}±0.92
	-0.4	0.50^{l}±0.02	0.70^{jkl}±0.05	0.77^{i-l}±0.06	3.93^{j-m}±0.15	7.39^{e-h}±1.17	12.49^{abc}±0.65
	-0.6	0.30^{l}±0.01	0.48^{l}±0.02	0.50^{l}±0.02	2.76^{lm}±0.12	3.51^{j-m}±0.55	7.84^{d-g}±0.75

Priming	PEG (MPa)	Seedling fresh weight (mg plant^{-1})			Seedling dry weight (mg plant^{-1})		
		Temperature			Temperature		
		15 °C	18 °C	25 °C	15 °C	18 °C	25 °C
Control	0	208^{cde}±5.9	227^{bcd}±5.7	252^{b}±16.3	65.3^{ab}±1.18	64.0^{ab}±1.15	59.5^{bc}±2.12
	-0.2	131^{h-l}±1.4	140^{hij}±3.1	181^{efg}±10.3	62.5^{ab}±1.11	62.5^{ab}±1.31	62.5^{ab}±0.86
	-0.4	103^{i-l}±1.0	109^{h-l}±0.8	118^{h-l}±7.9	63.3^{ab}±1.21	63.8^{ab}±1.6	61.3^{ab}±2.74
	-0.6	99^{kl}±1.8	101^{i-l}±1.4	97^{kl}±3.1	62.8^{ab}±0.93	62.9^{ab}±0.21	62.0^{ab}±2.12
Hydration	0	224^{bcd}±6.3	257^{b}±7.6	448^{a}±9.8	62.8^{ab}±2.58	63.4^{ab}±1.23	60.3^{abc}±1.97
	-0.2	123^{h-l}±1.9	147^{fgh}±3.0	244^{bc}±16.1	59.9^{bc}±0.67	61.8^{ab}±0.61	62.3^{ab}±0.97
	-0.4	114^{h-l}±2.8	112^{h-l}±2.6	121^{h-l}±2.9	60.5^{ab}±1.58	57.3^{bc}±1.87	60.4^{ab}±2.67
	-0.6	92^{l}±1.4	100^{jkl}±1.7	100^{jkl}±2.8	60.7^{ab}±1.17	51.8^{c}±0.91	60.9^{ab}±1.48
KNO_3	0	187^{def}±6.9	221^{bcd}±5.9	411^{a}±21.5	60.0^{bc}±2.19	63.6^{ab}±2.09	62.0^{ab}±0.81
	-0.2	135^{h-l}±5.1	141^{ghi}±1.7	188de±9.5	68.6^{a}±1.45	60.9^{ab}±1.49	60.3^{ab}±0.46
	-0.4	105^{i-l}±2.7	117^{h-l}±2.2	126^{h-l}±4.9	62.5^{ab}±0.36	62.1^{ab}±0.29	64.8^{ab}±2.06
	-0.6	97^{kl}±2.8	101^{i-l}±1.8	99^{kl}±1.7	62.3^{ab}±0.7	61.3^{ab}±0.61	61.0^{ab}±0.87

*: Means followed by the same letter(s) are not significant at p<0.05.

The shoot lengths of the hydrated seeds were the longest under all of the combinations of temperature and drought stress. Increasing drought severity and decreasing temperature resulted in shortened shoot length, but hydration induced shoot growth. It has also been determined that the shoots of sunflowers are more sensitive to drought than the roots. As the drought levels increased, a rapid and significant drop was determined, particularly in the hydration, as shown in Table 5. In both drought and low-temperature stress, the roots grew more than the shoots. It was determined that hydration had the highest root/shoot ratio in seedlings exposed to a low drought level of -0.2 MPa at 15 °C. Our findings were supported by the results of Ping et al. (2015), who found that the impact of low-temperature stress on the roots of rapeseed is more significant than its impact on the development of shoots. Janmohammadi et al. (2008) also reported that hydropriming promoted the development of maize seedlings under drought-stress conditions.

The seed primings significantly enhanced the shoot fresh weight of the sunflower. However, the seedlings became more sensitive to drought as the temperature increased. As drought severity increased at each temperature level, the fresh weight decreased accordingly. This result is consistent with the findings of Çokkizgin and Bölek (2015), who determined a significant reduction in cotton seedling fresh weight and demonstrated that priming treatments can prevent this growth retardation.

The measurement of dry weight in plants is a valuable indicator of the plant's overall health, growth potential, and ability to adapt to stressful situations. Changes in dry weight may reflect the physiological and biochemical responses of the plant under stress (Chaves et al., 2003). In the present study, seeds primed with KNO_3 under -0.2 MPa PEG at 15 °C produced the highest seedling dry weight (68.6 mg plant^{-1}), while hydrated seeds under -0.6 MPa PEG at 18 °C produced the lowest dry weight (51.8 mg plant^{-1}). Doddagoudar et al. (2023) reported that the seedling dry weight of rice genotypes was significantly influenced after exposure to low temperatures for 28 days.

4. Conclusions

Seed priming is an effective method for enhancing germination characteristics, particularly under stress conditions like drought and low-temperature stress. It has several advantages, including its straightforwardness and the lack of costly equipment and chemicals. The results of this study indicated that seed priming improved sunflower germination and seedling growth under low temperatures and drought stress. Especially, hydration could alleviate the inhibiting effects of low temperatures and drought on the germination and early growth of sunflower seedlings. Besides, sunflower was more susceptible to drought than to low temperatures during the early growth stages. As a result, hydration could be suggested for improving the germination and seedling growth of sunflowers because it does not require any chemicals or sophisticated equipment.

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References

Aboutalebian MA, Mohagheghi A, Niaz SA, Rouhi HR (2012). Influence of hydropriming on seed germination behavior of canola cultivars as affected by saline and drought stresses. *Annals of Biological Research,* 3(11): 5216-5222.

Ahmad S, Ahmad R, Ashraf MY, Ashraf M, Waraich EA (2009). Sunflower (*Helianthus annuus* L.) response to drought stress at germination and seedling growth stages. *Pakistan Journal of Botany,* 41(2): 647-654.

Akbari GA, Heshmati S, Soltani E, Amini Dehaghi M (2020). Influence of seed priming on seed yield, oil content and fatty acid composition of safflower (*Carthamus tinctorius* L.) grown under water deficit. *International Journal of Plant Production,* 14: 245-258.

Ali AS, Elozeiri AA (2017). Metabolic processes during seed germination. In Jimenz-Lopez JC (Eds), *Advances in Seed Biology*. InTech, pp. 141-166. Doi: 10.5772/intechopen.70653

Aswathi KR, Kalaji HM, Puthur JT (2022). Seed priming of plants aiding in drought stress tolerance and faster recovery: a review. *Plant Growth Regulation,* 97(2): 235-253.

Basit A, Ahmad M, Khan UA, Jabeen A, Durrani MU, Kiran R, Khalid A, Niaz M (2024). Evaluation of sunflower hybrids variability in response to drought stress: implications for oil and protein content optimization. *Haya Saudi Journal of Life Science,* 9(5): 188-193.

Čanak P, Jocković M, Ćirić M, Mirosavljević M, Miklić V (2014). Effect of seed priming with various concentrations of KNO_3 on sunflower seed germination parameters in *in vitro* drought conditions. *Research on Crops,* 15(1): 154-158.

Chakraborty P, Dwivedi P (2021). Seed priming and its role in mitigating heat stress responses in crop plants. *Journal of Soil Science and Plant Nutrition,* 21(2): 1718-1734.

Chaves MM, Maroco JP, Pereira JS (2003). Understanding plant responses to drought-from genes to the whole plant. *Functional Plant Biology,* 30(3): 239-264.

Çokkizgin H, Bölek Y (2015). Priming treatments for improvement of germination and emergence of cotton seeds at low temperature. *Plant Breeding and Seed Science,* 71: 121-134.

Doddagoudar SR, Nagaraja M, Lakshmikanth M, Srininvas AG, Shakuntala NM, Hiremath U, Mahanthshivayogayya K (2023). Improving the resilience of rice seedlings to low temperature stress through seed priming. *South African Journal of Botany,* 162: 183-192.

Ergin N, Kulan E, Gözükara MA, Kaya MF, Çetin ŞÖ, Kaya MD (2021). Response of germination and seedling development of cotton to salinity under optimal and suboptimal temperatures. *Kahramanmaraş Sütçü İmam Üniversitesi Tarım ve Doğa Dergisi,* 24(1): 108-115.

Fatemi SN 2014. Germination and seedling growth in primed seeds of sunflower under water stress. *Annual Research & Review in Biology,* 4(23): 3459-3469.

Ghosh S, Bera K, Dutta P, Sadhukhan S (2024). Seed priming and seedling pre-treatment in regulating secondary metabolism for stress tolerance. In Roychoudhury A (Eds), *Biology and Biotechnology of Environmental Stress Tolerance in Plants: Secondary Metabolites in Environmental Stress Tolerance*. Apple Academic Press, pp. 263-304.

Guan YJ, Hu J, Wang XJ, Shao CX (2009). Seed priming with chitosan improves maize germination and seedling growth in relation to physiological changes under low temperature stress. *Journal of Zhejiang University Science B,* 10: 427-433.

Haj Sghaier A, Khaeim H, Tarnawa Á, Kovács GP, Gyuricza C, Kende Z (2023). Germination and seedling development responses of sunflower (*Helianthus annuus* L.) seeds to temperature and different levels of water availability. *Agriculture,* 13(3): 608.

Hamidi R, Pirasteh-Anosheh H (2013). Comparison effect of different seed priming methods on sunflower germination and seedling growth. *International Journal of Agronomy and Plant Production,* 4(6): 1247-1250.

Harris D, Joshi A, Khan PA, Gothkar P, Sodhi PS (1999). On-farm seed priming in semi-arid agriculture: Development and evaluation in maize, rice and chickpea in India using participatory methods. *Experimental Agriculture,* 35(1): 15-29. Doi: 10.1017/S0014479799001027

Heshmat O, Saeed HA, Fardin K (2011). The improvement of seed germination traits in canola (*Brassica napus* L.) as affected by saline and drought stress. *Journal of Agricultural Technology,* 7(3): 611-622.

Hoppe A, Theimer RR (1997). Degradation of oil bodies isolated from cotyledons during germination of rapeseed seedlings. *Journal of Plant Physiology,* 151(4): 471-478.

Hussain M, Farooq S, Hasan W, Ul-Allah S, Tanveer M, Farooq M, Nawaz A (2018). Drought stress in sunflower: Physiological effects and its management through breeding and agronomic alternatives. *Agricultural Water Management,* 201: 152-166.

ISTA (2018). International Rules for Seed Testing. Bassersdorf, Switzerland: International Seed Testing Association.

Janmohammadi M, Dezfuli PM, Sharifzadeh F (2008). Seed invigoration techniques to improve germination and early growth of inbred line of maize under salinity and drought stress. *General and Applied Plant Physiology,* 34(3-4): 215-226.

Jiang F, Lv S, Zhang Z, Chen Q, Mai J, Wan X, Liu P (2023). Integrated metabolomics and transcriptomics analysis during seed germination of waxy corn under low temperature stress. *BMC Plant Biology,* 23(1): 190.

Kaya MD, Bayramin S, Kaya G (2015). The effect of planting geometry and seed priming on sunflower yield under rain-fed conditions. *Fresenius Environmental Bulletin,* 24(11c): 4095-4101.

Kaya MD, Ergin N, Harmancı P, Kulan EG (2024). Seed priming as a method of preservation and restoration of sunflower seeds. *Oilseeds and Fats, Crops and Lipids,* 31: 4.

Kaya MD, Kulan EG (2020). Effective seed priming methods improving germination and emergence of sugar beet under low-temperature stress. *Sugar Tech,* 22: 1086–1091. Doi:10.1007/s12355-020-00848-5

Kaya MD, Okçu G, Atak M, Çıkılı Y, Kolsarıcı Ö (2006) Seed treatments to overcome salt and drought stress during germination in sunflower (*Helianthus annuus* L.). *European Journal of Agronomy,* 24: 291-295.

Khan MB, Hussain M, Raza A, Farooq S, Jabran K (2015). Seed priming with $CaCl_2$ and ridge planting for improved drought resistance in maize. *Turkish Journal of Agriculture and Forestry,* 39(2): 193-203.

Michel BE, Kaufmann MR (1973). The osmotic potential of polyethylene glycol 6000. *Plant Physiology,* 51: 914-916.

Moghanibashi M, Karimmojeni H, Nikneshan P, Behrozi D (2012). Effect of hydropriming on seed germination indices of sunflower (*Helianthus annuus* L.) under salt and drought conditions. *Plant Knowledge Journal,* 1(1): 10-15.

Nezami A, Khazaei HR, Boroumand RZ, Hosseini A (2008). Effects of drought stress and defoliation on sunflower (*Helianthus annuus*) in controlled conditions. *Desert,* 12: 99-104.

Pahoja VM, Siddiqui SH, Narejo MUN, Umrani JH (2013). Response of hydropriming and osmopriming on germination and seedling growth of sunflower (*Helianthus annuus* L.) under salt stress. *International Journal of Agricultural Science and Research,* 3(2): 71-80.

Ping Y, Meng-Zhu X, Zhe Z, Xiao-Hong Z, Li-Yong HU, Zheng-Hua XU (2015). Cluster analysis for different rapeseed varieties on seed germination and seedling growth under low temperature stress. *Research on Crops,* 16(1): 68-77.

Prayaga L, Sarada C, Lakshmamma P (2017). Seed priming for improving germination of sunflower (*Helianthus annuus* L.) at low temperature. *Journal of Oilseeds Research,* 34(2): 93-97

Rahimi A (2013). Seed priming improves the germination performance of cumin (*Cuminum syminum* L.) under temperature and water stress. *Industrial Crops and Products,* 42: 454-460.

Rehman ZU, Haq RU, Ullah S, Iqbal A, Khan AM, Assogba CMA, Awais M (2024). Enhancing growth and yield parameters of Sinapis alba through optimized seed priming techniques. *Agrosystems, Geosciences & Environment,* 7(1): e20466.

Salehzade H, Shishvan MI, Ghiyasi M, Forouzin F, Siyahjani AA (2009). Effect of seed priming on germination and seedling growth of wheat (*Triticum aestivum* L.). *Research Journal of Biological Sciences,* 4: 629-631.

Sintaha M, Man CK, Yung WS, Duan S, Li MW, Lam HM (2022). Drought stress priming improved the drought tolerance of soybean. *Plants,* 11(21): 2954.

Sneha GR, Govindasamy V, Singh PK, Kumar S, Abraham G (2024). Priming of seeds with cyanobacteria improved tolerance in wheat (*Triticum aestivum* L.) during post-germinative drought stress. *Journal of Applied Phycology*, 1-14. Doi: 10.1007/s10811-023-03170-1

Soleimanzadeh H (2012). Response of sunflower (*Helianthus annuus* L.) to selenium application under water stress. *World Applied Science Journal,* 17: 1115-1119.

Szczerba A, Płażek A, Pastuszak J, Kopeć P, Hornyák M, Dubert F (2021). Effect of low temperature on germination, growth, and seed yield of four soybean (*Glycine max* L.) cultivars. *Agronomy* 11(4): 800.

Tabassum T, Farooq M, Ahmad R, Zohaib A, Wahid A, Shahid M (2018). Terminal drought and seed priming improves drought tolerance in wheat. *Physiology and Molecular Biology of Plants* 24: 845-856.

TUİK (2024). Turkish Statistical Institute. https://biruni.tuik.gov.tr/medas/?locale=tr (access date: 15.08.2024).

Wen B (2015). Effects of high temperature and water stress on seed germination of the invasive species Mexican sunflower. *Plos One* 10: e0141567.

Xia J, Hao X, Wang T, Li H, Shi X, Liu Y, Luo H (2023). Seed priming with gibberellin regulates the germination of cotton seeds under low-temperature conditions. *Journal of Plant Growth Regulation* 42(1): 319-334.

Zhang C, Luo T, Liu J, Xian M, Yuan J, Hu L, Xu Z (2019). Evaluation of the low-temperature tolerance of rapeseed genotypes at the germination and seedling emergence stages. *Crop Science* 59(4): 1709-1717.

Zhu ZH, Sami A, Xu QQ, Wu LL, Zheng WY, Chen ZP, Jin XZ, Zhang H, Li Y, Yu Y, Zhou KJ (2021). Effects of seed priming treatments on the germination and development of two rapeseed (*Brassica napus* L.) varieties under the co-influence of low temperature and drought. *Plos One* 16(9): e0257236.

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Research Article

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Salt Stress Sensitivity of Chokeberry (*Aronia melanocarpa* L.) *in vitro* and *in vivo* Conditions

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HIGHLIGHTS

- Results show that the chokeberry plant is sensitive to salty environments.
- Plant fresh and dry weights decreased with increasing salt concentration.
- Increasing salt concentration decreased chlorophyll content.
- It may be recommended not to use saline soil or irrigation waters in chokeberry cultivation.

Abstract

In this study, the reactions of the chokeberry plant, which has attracted attention with its high antioxidant content and importance in human nutrition in recent years, to salt stress *in vitro* and *in vivo* conditions have been investigated. In this context, morphological, physiological, and biochemical reactions of plants at different salt levels *in vitro* and *in vivo* conditions have been studied. Salt concentrations *in vitro* were 1/3 dilute MS, 7/10 dilute MS, MS (control), MS + 1 gL^{-1} NaCl, MS + 3 gL^{-1} NaCl, MS + 6 gL^{-1} NaCl, MS + 8 gL^{-1} NaCl, MS + 9 gL^{-1} NaCl; *in vivo*, it was applied in the form of 25 mm NaCl per week with irrigation water to chokeberry lings planted in 2 liter pots containing peat: pearlite mixture in a ratio of 2:1 *in vivo* condition. The experiment was terminated by determining the salt levels in the soil from the moment the damage to the leaves due to salt stress began. According to the research results, *in vitro* conditions in 1/3 dilute MS, 7/10 dilute MS, MS (control) medium, no damage to explants occurred, MS +1 gL^{-1} NaCl and MS + 3gL^{-1} NaCl doses of shoot tips and leaves browning, MS + 6 gL^{-1} NaCl, MS + 8 gL^{-1} NaCl, MS + 9 gL^{-1} NaCl caused death in the medium. Browning of the shoot tip and leaves has occurred in plants during salt application under *in vivo* conditions. As a result of salt applications, plant height, plant dry weight, root length, chlorophyll content, protein content decreased in parallel with the increase in dose, and there was no change in leaf relative water content and proline content.

Keywords: Chokeberry, Salt stress, *In vitro*, *In vivo*

1. Introduction

Soil salinity is the most important abiotic stress factor after drought in world agriculture and prevents plant growth, especially in arid and semi-arid regions. Today, approximately 45% of the world's agricultural areas

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are exposed to drought stress, and with this drought, salinity occurs in 6% of agricultural areas. Salt stress, which negatively affects the fertility of soils and causes large areas to be left out of agriculture, has become an important problem that limits agricultural production areas today. This stress factor, which constitutes important agricultural constraints, affects plant morphology and anatomy, while also limiting growth and development (D'anna et al. 2003; Keutgen and Keutgen 2003). Salinity affects different physiological and metabolic processes in plants. Influence of these processes causes various symptoms in plants such as decrease in leaf area, increase in leaf thickness and wilting, absorption of leaves, root and shoot necrosis and decrease in stem length. Especially in salt stress, plant nutrient uptake decreases, and ionic imbalances occur in the cells. In addition, the resulting oxidative stress can affect the metabolic activities of the cell and activate antioxidant enzyme activities (Greenway and Munns 1980).

Under abiotic stress conditions, the rate of photosynthesis in plants decreases as a result of the decrease in leaf relative moisture content and leaf water potential (Lawlor 2002). It is known that the main reason for the decrease in photosynthesis rate in saline conditions is due to stomatal limitation (Cornic and Massacci 1996). Under salt stress, the decrease in photosynthetic rate and internal CO_2 concentration due to the closure of stomata prevents photosynthetic metabolism. The closure of stomata during salt stress also negatively affects the nutrition of plants (Oren et al. 1999).

The use of tissue culture methods is increasing day by day in physiological studies on plant stress, as they provide fast and effective results. Tissue culture technique is used advantageously and effectively in the evaluation of selected genotypes and in determining the resistance of fast-growing and developing varieties to some stress conditions such as cold, drought and salinity (Kaya 1988). Nowadays, it is very popular to use abiotic stress resistant plants using tissue culture techniques. Because *in vitro* conditions are more controllable than *in vivo* conditions and intensive work can be done in limited areas (Shiyab et al. 2003).

In a study, “Camarosa” and “Chandler” strawberry cultivars decreased leaf and root fresh weight, leaf area and leaf number, while decreasing stomatal conductance and transpiration rate under saline conditions (Turhan and Eriş 2007). In another study, the responses of four different strawberry genotypes to different NaCl concentrations (0, 25, 50, 75 and 100 mM) were examined under *in vitro* conditions, and it was determined that high salt concentration reduced the multiplication coefficient and affected calcium and sodium concentrations as well as dry weight (Torun et al. 2007). In another study, it was determined that 500, 1000 and 1500mgL^{-1} salt applications inhibited vegetative growth in “Kabarla” and “Gloria” strawberry cultivars plants irrigated with Hoagland nutrient solution, and although dry leaf weight decreased in high dose applications, dry leaf weight increased in low dose applications (Yilmaz and Kina 2008).

In the *in vitro* study using “Sweet Charlie” and “Benihoppe” strawberry varieties, the salt and alkali tolerance indices of the varieties were determined, and it was determined that the rooting rate, average number of roots and root length decreased by limiting the *in vitro* rooting ability in both stress factors (Zhao et al. 2017).

The effects of different NaCl levels (0, 50, 100, 150mM) on “Viking” aronia variety plants in the *in vitro* culture medium were investigated. The ½MS medium compared to the full MS medium performed better at almost all growth parameters at every NaCl concentration. The average highest plant height was recorded in the control treatment (4.77cm) of the ½MS medium; the lowest plant height was recorded in 150mM NaCl treatment (1.67cm) of the MS medium. The lowest leaf area was determined at 150mM NaCl concentration, and it reduced 68.8% in the MS medium and 73% in the ½MS medium. Rooting was highest in the control groups (100%) in both media; a significant gradual decrease occurred in 50 and 100mM NaCl concentration, and rooting did not occur in 150mM NaCl treatment (Nas et al. 2023).

Among the rest of small fruits, chokeberry is one of the most important small fruits in terms of containing the highest percentage of antioxidants. Chokeberry classified as ornamental's shrub as well as decorative autumn coloration that making her very popular (Hirvi and Honkanen 1985). Chokeberry classified between most fruit in terms of antioxidants, where the antioxidants percentage in chokeberry is higher than that is in apple, banana, elderberries and others. The high levels of flavonoids and anthocyanin in aronia is higher than those found in cranberries and five times more. In addition to their containment on an antioxidants and polyphenols and they also contain on mineral and vitamins. As well as the chemicals content in chokeberry it has been alleged that it reduces some disease such as the potential cancer and heart disease. To supply a natural red color in products with poor color stability, chokeberry juice has been increasingly used in the food industry. Commercially, chokeberry is fundamentally used for juice either alone or blended with other fruit juices such as apple or grape. Mainly aronia juice is commercially, used either alone or blended with other juices such as apple and grape. Food coloring, tea, syrup and fruit spread coloring all these uses includes other uses for chokeberry. The uses of aronia juice have different from zone in Europe to other. In Russia apple and chokeberry juices are combined and fermented to producing or giving red wine. Either in Europe the juice often blended with apple juice to give juice a blush in Lithuania have been using chokeberry juice alone or blended with other fruits so as to produce dessert wines. Reports from the Ukraine describe chokeberry as improving the color, tannin level and sugar of grape wines (Smith and Ringenberg 2003). For chokeberry benefits prevent urinary tract infection and weight control, there are others benefits relate in chokeberry fruit such as treat inflammations, hypertension as well as can be very beneficial in cases of arthritis, cardiovascular conditions and other diseases. Chokeberry also contributes to strengthening immunity, blood vessels, lower blood pressure levels, and chokeberry also delays the natural aging process. Therapeutically they show positive effect in the anti-inflammatory and anti-oxidative activity and, also in the treatment several of neoplasms (Kowalczyk et al. 2003).

As a result of the literature review, studies on salinity stress of chokeberry plants under *in vitro* and *in vivo* conditions are quite limited and insufficient. For this reason, the aim of the study was to add new information to the literature and to determine the cultivation limits by measuring the responses of the aronia plant to different levels of salty growing conditions.

In this study, the responses of chokeberry (*Aronia melanocarpa*) plants grown under *in vitro* and *in vivo* conditions to different salt levels were examined in terms of morphological, physiological and biochemical aspects.

2. Materials and Methods

The study was conducted in 2022 in the Plant Biotechnology Laboratory and greenhouses of the Department of Horticulture. In the study, "Viking" chokeberry saplings were used as plant material.

In the experiment, the morphological and biochemical responses of chokeberry saplings propagated *in vitro* to different NaCl doses both *in vitro* and *in vivo* were examined.

2.1. Growing plants under in vitro conditions

For the surface sterilization of cuttings taken from one-year-old shoots, it first started by removing the leaves and leaflets on the shoots. The shoots were divided into 3-4 cm long single-node pieces and left under tap water for 30 minutes. The cuttings taken into the sterile cabinet were first kept in ethyl alcohol (70%) for 1-2 minutes and then rinsed with sterile pure water. It was sterilized for 10-20 minutes in 10% sodium hypochlorite solution containing a few drops of Tween-20. Then, after rinsing with sterile pure water, the sterilized cuttings were transferred to the shoot development medium (Mendi et al. 2003). The prepared cuttings were planted in culture tubes with 10 mL volume media solidified with 8 gL^{-1} agar containing 1.0 mgL^{-1} GA_3, 3% sucrose, MS essential minerals and vitamins. The cultured cuttings were left to develop under 1500-3000 lux white fluorescent light in a growth room at 25±1°C. After 21 days, new shoots were transferred to the medium containing 1.0 mgL^{-1} BA, and when the required number of plants was reached, the plants were transferred to the medium containing IBA (1.0 mgL^{-1}) during the rooting stage.

Shoots developed from cuttings were cultured in MS medium containing 2 mgL^{-1} BAP to ensure reproduction (Ružić et al. 2000). The plants, which were kept in the propagation medium for four weeks, were removed from the culture medium at the end of the 4th week and transferred to the new propagation medium. After the sufficient number of plants was reached, the obtained plants were placed in the rooting medium (Figure 1).

Figure 1. "Viking" chokeberry variety shoot propagation.

2.2. Application of salinity treatments

After reaching a sufficient number of plants, the obtained plants were cultured in MS medium containing 2 mgL^{-1} IBA. Rooted plants after 4 weeks were used for salt applications. For the proliferation treatments, salinity stress was achieved bud shoot tips, from the proliferation medium, which were placed in the standard medium with eight different concentrations of NaCl (1/3 dilute MS, 7/10 dilute MS, MS (control), MS + 1 gL^{-1} NaCl, MS + 3 gL^{-1} NaCl, MS + 6 gL^{-1} NaCl, MS + 8 gL^{-1} NaCl, MS + 9 gL^{-1} NaCl). ECs of these medium were measured and the results are given in Table 1.

Table 1. EC values of *in vitro* environments

Media	EC (mmhos/cm)
1/3 dilute MS	400
7/10 dilute MS	2500
MS (control)	4800
MS+1 gL^{-1} NaCl	6250
MS+3 gL^{-1} NaCl	9000
MS+6 gL^{-1} NaCl	13000
MS+8 gL^{-1} NaCl	15000
MS+9 gL^{-1} NaCl	16500

2.3. Growing plants under in vivo conditions

In the *in vivo* conditions, 25 mM NaCl was applied weekly along with irrigation water to chokeberry saplings planted in 2 liter pots containing a 2:1 peat:perlite mixture. The experiment was concluded by determining the salt levels in the soil from the moment the damage to the leaves due to salt stress began.

In the study, shoot tips of the "Viking" chokeberry variety were used as explants (Figure 3.4). Under *in vitro* culture conditions, aronia plants were treated with 8 different salt concentrations (1/3 dilute MS, 7/10 dilute MS, MS (control), MS + 1 gL^{-1} NaCl, MS + 3 gL^{-1} NaCl, MS + 6 gL^{-1} NaCl, MS + 8 gL^{-1} NaCl, MS + 9gL^{-1} NaCl) were applied.

2.4. Observations and measurements made in the experiment

In determining the salinity index, scores from 1 to 4 are given according to the damages listed below (Sivritepe et al. 2008).

1: No damage to plants,
2: Browning on the shoot tip and leaves,
3: Browning on the entire leaf and stem,
4: Death in plants,

In leaf samples taken from plants in September, photosynthesis efficiency (Arıkan 2017), membrane permeability in leaf discs (Lutts et al. 1996) and leaf relative water content (LRWC) (Kaya and Higgs 2003) were determined in mature leaves using the Li-Cor 600 device, stomatal conductance (Arıkan 2017), chlorophyll-a, chlorophyll-b and total chlorophyll contents in the leaf were determined (Arnon 1949). Protein determination in plants under salt stress was determined according to the "Bradford" (Bradford 1976) method, and proline determination was made spectrophotometrically by the acid-ninhydrin method (Bates et al. 1973).

The fresh weight of roots and plants was determined with the help of precision scales. Dry weights were determined with a precision balance after drying the same samples in an oven at 72ºC for 48 hours (Sanchez et al. 2004).

Plant height was determined by measuring the section from the root collar to the extreme growth point of the plant with the help of a ruler.

At the end of the applications, the areas of the leaf samples taken from the plants were determined using the Adobe Photoshop program (Ipek et al. 2019).

Photosynthetic efficiency (μmol CO_2 $m^{-2}s^{-1}$) was determined during the growing period with the Li-Cor 600 device on a total of 10 leaves of 5 randomly selected plants from each repetition of the applications in the last week of June, July, August and September.

2.5. Experimental design and statistical analysis

The study was carried out under *in vitro* conditions with a total of 240 plants, with 3 replicates in each application and 10 plants in each repetition. In *in vivo* conditions, a total of 60 plants were used in 3 replicates in each application and 10 plants in each repetition. The data obtained from the study were subjected to ANOVA at 5% significance level in the SPSS program, T test and Duncan multiple comparison test was applied to evaluate the differences between the applications.

3. Results

3.1. Effects of salt applications on chokeberry plants in vitro

Explants taken from chokeberry plants were damaged to varying extents by salt applications during their stay in the *in vitro* condition. No damage occurred in explants in 1/3 dilute MS, 7/10 dilute MS and MS (control) medium, browning of shoot tips and leaves at MS + 1 gL^{-1} NaCl and MS + 3 gL^{-1} NaCl doses, MS + 6 gL^{-1} NaCl, deaths occurred in, MS + 8 gL^{-1} NaCl, MS + 9 gL^{-1} NaCl medium (Figure 2).

Figure 2. NaCl application to chokeberry *in vitro* conditions, from left to right 1/3 MS, 7/10 MS, MS, 1 gL^{-1} NaCl, 3 gL^{-1} NaCl, 6 gL^{-1} NaCl, 8 gL^{-1} NaCl, 9 gL^{-1} NaCl

The effects of salt applications on plant height, plant fresh and dry weights were found to be statistically significant. Applications with the longest plant height; It occurred in 7/10 dilute MS medium (4.40 cm), followed by 1/3 dilute MS (3.76 cm) and control applications (3.68 cm). After this application, plant heights decreased with the increase in salt concentration (Table 2).

Table 2. Effects of salt applications on plant height, plant fresh and dry weight, root length, leaf number and leaf area.

Treatments	Plant Height (cm)	Plant fresh weight (g)	Plant dry weight (g)	Root length (mm)	Leaf number	Leaf area (cm^2)
1/3 dilute MS	3.76 b*	0.286 a	0.040 a	15.21 a	11.3 a	2.96 c
7/10 dilute MS	4.40 a	0.282 a	0.044 a	13.72 a	12.5 a	4.42 b
MS (control)	3.68 b	0.242 a	0.039 a	11.92 a	13.0 a	4.97 b
MS+1 gL^{-1} NaCl	3.09 c	0.229 a	0.036 a	14.88 a	11.4 a	5.57 a
MS+3 gL^{-1} NaCl	1.80 d	0.081 b	0.015 b	1.85 b	7.3 b	2.65 c

*Means separation within the column by Duncan's multiple range test and means marked with different letters (a, b, c...) indicate a significant differences $P < 0.05$

There was no difference in terms of plant fresh and dry weight between 1/3 dilute MS, 7/10 dilute MS, MS and MS + 1 gL^{-1} NaCl medium, but there was a significant decrease in the MS + 3 gL^{-1} NaCl dose. The effects of the treatments on root length, number and area of leaves were also found to be statistically significant. There was no difference in root length and number of leaves between 1/3 dilute MS, 7/10 dilute MS, MS and MS + 1 gL^{-1} NaCl medium, and a significant decrease occurred at the MS + 3 gL^{-1} NaCl dose. The maximum leaf area was determined in MS + 1 gL^{-1} NaCl medium, followed by MS and 7/10 MS medium, and the least occurred in 1/3 dilute MS and MS + 3 gL^{-1} NaCl dose (Table 2).

Significant effects of the applications on chlorophyll contents have also been determined. While total chlorophyll contents were close to each other in 1/3 MS, 7/10 MS and MS medium, chlorophyll contents decreased significantly in parallel with the increase in salt dose. There was no difference in chlorophyll-a contents in 1/3 MS, 7/10 MS, MS and MS + 1 gL^{-1} NaCl medium, and an increase occurred at the MS + 3 gL^{-1} NaCl dose. On the other hand, no difference was found in chlorophyll-b content between 1/3 MS, 7/10 MS, MS and MS + 1 gL^{-1} NaCl medium, and a significant decrease was determined at the MS + 3 gL^{-1} NaCl dose (Table 3).

Table 3. Effects of salt applications on chlorophyll, protein and proline contents

Treatments	Total chlorophyll (gL^{-1})	Chlorophyll-a (gL^{-1})	Chlorophyll-b (gL^{-1})	Protein* (μg protein g^{-1} FW)	Prolin (μg prolin g^{-1} FW)
1/3 dilute MS	117.46 a*	39.69 b	62.02 a	91.55 b	20.94
7/10 dilute MS	117.49 a	39.70 b	62.03 a	111.31 a	19.36
MS (control)	117.60 a	39.75 b	62.11 a	113.66 a	23.82
MS+1 gL^{-1} NaCl	82.26 b	39.73 b	62.09 a	106.17 ab	23.32
MS+3 gL^{-1} NaCl	71.85 c	43.87 a	29.48 b	14.93 c	20.42
					N.S.**

*Means separation within the column by Duncan's multiple range test and means marked with different letters (a, b, c...) indicate a significant differences $P < 0.05$.
**N.S.: Non-significant

The effects of salt applications on protein content were found to be statistically significant, while their effects on proline content were found to be insignificant. While protein contents were close to each other in 1/3 MS, 7/10 MS, MS and MS + 1 gL^{-1} NaCl medium, a significant decrease occurred at MS + 3 gL^{-1} NaCl dose (Table 3).

3.2. Effects of salt applications on aronia plants in vivo

As a result of salt application *in vivo*, browning of shoot tips and leaves occurred in plants (Figure 3). The effect of salt applications on plant height *in vivo* was found to be statistically insignificant. With salt application, stem diameter, root length, leaf area, plant fresh and dry weight, and root fresh and dry weight decreased significantly compared to the control (Table 4).

Figure 3. Effects of salt applications on chokeberry plants *in vivo*

Table 4. Effects of salt applications on some morphological features *in vivo.*

Treatments	Plant height (cm)	Stem diameter (mm)	Root length (mm)	Leaf area (cm^2)	Plant fresh weight (g)	Plant dry weight (g)	Root fresh weight (g)	Root dry weight (g)
Control	52.62	7.13 a*	41.95 a	4.63 a	33.76 a	17.92 a	33.00 a	19.14 a
Salt	51.25 N.S**	6.11 b	26.55 b	3.61b	16.23 b	8.95 b	15.44 b	6.90 b

*Means separation within the column by t test and means marked with different letters (a, b, c...) indicate a significant differences $P < 0.05$.

**N.S.: Non-significant

Table 5. Effects of salt applications on some physiological characteristics *in vivo*

Treatments	Membrane permeability	LRWC (%)	Protein (μg g^{-1} FW)	Prolin (μg g^{-1} FW)	Total chlorophyll (gL^{-1})	Chlorophyll-a (gl^{-1})	Chlorophyll-b (gl^{-1})
Control	155.4 b*	61.70 a	18.37 a	9.21 b	88.23 a	29.49	39.03 a
Salt	226.2 a	67.37 a N.S.**	12.61 b	13.06 a	80.64 b	30.13 N.S.**	29.49 b

	Photosynthesis efficiency (μmol CO_2 $m^{-2}s^{-1}$)	Stoma conductance (mmol $m^{-2}s^{-1}$)
Control	0.379 a	0.077 b
Salt	0.238 b	0.111a

*Means separation within the column by t test and means marked with different letters (a, b, c...) indicate a significant differences $P < 0.05$.

**N.S.: Non-significant

The effects of the treatments on leaf relative water content and chlorophyll a content were found to be statistically insignificant, but their effects on membrane permeability, protein, proline, total chlorophyll and chlorophyll-b, photosynthesis efficiency and stomatal conductance were found to be significant (Table 5). Salt application increased membrane permeability and stomatal conductance compared to the control, and decreased protein, proline, total chlorophyll and chlorophyll-b content and photosynthesis efficiency compared to the control.

4. Discussion

Salt applications have harmful effects on plants both *in vitro* and *in vivo*. This shows that the chokeberry plant is sensitive to salty environments. There is no information in the literature about the sensitivity of the chokeberry plant to salt. In this respect, the results obtained are valuable as they are the first data on the subject.

Salt applications negatively affected the underground and above-ground development of plants. According to Özelçi (2020), salinity shrinks the main stems, reduces the formation of side branches and causes the death of newly formed juicy branches, and also tries to inhibit cambium activity, which increases its concentration in the middle part. In addition, the applications also reduced total chlorophyll, photosynthetic activity and protein contents. Similar effects of salt on cultivated plants have been determined in many studies. In a study conducted under *in vitro* conditions on "Myrobolan 29C" rootstock, it was determined that rooting speed, number of roots, root length, plant height, fresh plant weight, dry plant weight and chlorophyll content decreased significantly with the increase in salt concentration (Ipek et al. 2019). These results show that environmental salinity reduces water potential and the ability of plants to take up water. This rapidly reduces the rate of cell expansion in growing tissues (Munns 2011). In another study, it was determined that salt applications prevented vegetative growth in "Kabarla"and "Gloria" strawberry varieties irrigated with Hoagland nutrient solution, and dry leaf weight decreased in high dose applications (Yilmaz and Kina 2008). In another study, in an *in vitro* study using "Sweet Charlie" and "Benihoppe" strawberry varieties, the salt and alkali tolerance indices of the varieties were determined and it was determined that the rooting rate, average number of roots and root length decreased by limiting the *in vitro* rooting ability in both stress factors (Zhao et al. 2017). Similar effects of salt applications have been detected on lemon (Sharma et al. 2013), *Aloe vera* (Moghbeli et al. 2012) and citrus rootstocks (Bahmani et al. 2012).

The fact that salt reduces the development of above- and below-ground organs may be due to the inhibitory effects of salt on metabolic activities related to cell division, differentiation and elongation. Additionally, salinity's reduction of endogenous auxin levels (Khan et al. 1976) may be another effective factor.

Plant fresh and dry weights decreased with increasing salt concentration. Similar results were obtained in studies conducted on the subject in different plant species (İpek et al. 2019; Moghbeli et al. 2012; Sharma et al. 2013; Ghaleb et al. 2010). This result is explained by the fact that high NaCl levels inhibit leaf expansion, largely due to inhibition of cell division rather than cell expansion (Chartzoulakis and Klapaki 2000).

Increasing salt concentration decreased chlorophyll content. Chartzoulakis and Klapaki (2000) on quince, (Sivritepe et al. 2008) on grapes, Harb et al. (2002) on banana; similar results were obtained by İpek et al. (2019) on "Myrobolan 29C" rootstock. These results can be explained as chlorophyll biosynthesis is inhibited by salt application. The depressive effect of stress conditions on the absorption of some ions involved in chlorophyll formation, such as Mg and Fe, causes chlorophyll suppression in leaves and/or an increase in some growth inhibitors. Ethylene or abscisic acid production (Hanafy Ahmed et al. 2002), which increases aging that may occur in case of salt stress, is also effective here.

The negative effects of salt applications on chlorophyll content also negatively affect all processes related to photosynthesis. In this case, the negative impact of characteristics such as number of leaves, leaf area, underground and above-ground growth and protein content can be attributed to this. Salinity affects different physiological and metabolic processes in plants. Affecting these processes causes various symptoms in plants such as decrease in leaf area, increase in leaf thickness and wilting, absorption of leaves, root and shoot necrosis and decrease in stem length (Parida and Das 2005).

The effects of salt applications in *in vitro* and *in vivo* conditions are generally parallel. Salt applications significantly reduced plant fresh weight, plant dry weight, leaf area, total chlorophyll, chlorophyll-b and protein contents in both environments. Salt applications significantly reduced plant height *in vitro*, while a decrease occurred *in vivo*, but no statistical difference was detected. The highest salt concentration increased chlorophyll-a content *in vitro* compared to the control, while it increased *in vivo*, but no statistical difference was detected. While the applications did not have significant effects on proline content *in vitro*, they significantly increased *in vivo*.

5. Conclusions

As a result of the data obtained, varying degrees of damage occurred to chokeberry plants from salt applications *in vitro*. While there was no damage to the explants in 1/3 dilute MS, 7/10 dilute MS, MS (control) medium, browning of the shoot tips and leaves was observed at the doses of MS + 1 gL^{-1} NaCl and MS + 3 gL^{-1} NaCl, and MS + 6 gL^{-1} NaCl, deaths occurred in plants in, MS + 8 gL^{-1} NaCl, MS + 9 gL^{-1} NaCl medium.

Salt applications caused harmful effects on chokeberry both *in vitro* and *in vivo*. From this perspective, it can be said that the aronia plant is sensitive to salty growing environments. In this respect, it may be recommended not to use saline soil or irrigation waters with high salt content in chokeberry cultivation, or to cultivate it by reclaiming the soil and irrigation water of the land where it will be grown.

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References

Arnon DI (1949). Copper enzymes in isolated chloroplasts. Polyphenoloxidase in Beta vulgaris. *Plant Physiology*, 24: 1-10.

Arıkan Ş (2017). Faydalı rizobakteri uygulamalarının tuzlu toprak şartlarında elma ve kirazda etkileri. Yüksek Lisans Tezi, Selçuk Üniversitesi, Konya-Türkiye.

Bahmani R, Gholami M, Mozafari AA, Alivaisi R (2012). Effects of salinity on in vitro shoot proliferation and rooting of apple rootstock MM 106. *World Applied Sciences Journal,* 17: 292-295.

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

Bradford MM (1976). A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. *Analytical Biochemistry*, 72: 248-254.

Chartzoulakis K, Klapaki G (2000). Response of two greenhouse pepper hybrids to NaCl salinity during different growth stages. *Scientia Horticulturae*, 86:247-260.

Cornic G, Massacci A (1996). Leaf Photosynthesis Under Drought Stress, In Photosynthesis and the Environment. Dordrecht: Springer Netherlands, 347-366.

D'anna F, Incalcaterra G, Moncada A, Miceli A (2003). Effects of different electrical conductivity levels on strawberry grown in soilless culture. In: *International Symposium on Managing Greenhouse Crops in Saline Environment,* 355-360.

Ghaleb WS, Sawwan JS, Akash MW, Al-Abdallat AM (2010). In vitro response of two citrus rootstocks to salt stress. *International Journal of Fruit Science,* 10: 40-53.

Greenway H, Munns R (1980). Mechanisms of salt tolerance in nonhalophytes. *Annual Review of Plant Physiology,* 31: 149-190.

Hanafy Ahmed AH, Hassan HM, Gad MMA, Amin MA (2002). Micropropagation of Myrtus communis and increasing its tolerance to salinity by using polyamines. In: *2nd Congress of Recent Technologies in Agriculture*, Cairo Univ., Fac., of Agric., Giza.

Harb EM, El–Shihy OM, Ahmed AH, Mualla NA (2002). Using plant tissue culture technique for rapid propagation and salt tolerance in banana plant. In: *Proceedings of the 2 nd Congress on Recent Technologies in Agriculture*, Faculty of Agriculture, Cairo University, Giza, 28-30.

Hirvi T, Honkanen E (1985). Analysis of the volatile constituents of black chokeberry (*Aronia melanocarpa*). *Journal of the Science of Food and Agriculture,* 36: 808-810.

İpek M, Arıkan Ş, Pırlak L, Eşitken A (2019). Phenological, morphological and molecular characterization of some promising walnut (*Juglans regia* L.) genotypes in Konya. *Erwerbs-Obstbau,* 61: 149-156.

Kaya Z (1988). Doku kültürünün orman ağaçları ıslah çalışmalarındaki yeri. *Orman Mühendisliği Dergisi* 25: 12-19.

Kaya C, Higgs D (2003). Supplementary potassium nitrate improves salt tolerance in bell pepper plants. *Journal of Plant Nutrition,* 26: 1367-1382.

Keutgen AJ, Keutgen N (2003). Influence of NaCl salinity stress on fruit quality in strawberry. In: *International Symposium on Managing Greenhouse Crops in Saline Environment*, 155-157.

Khan MI, Khan MA, Khizar T (1976). Plant growth regulators from species differing in salt tolerance as affected by soil salinity. *Plant and Soil*, 45: 267-271.

Kowalczyk E, Krzesiñski P, Kura M, Szmigiel B, Blaszczyk J (2003). Anthocyanins in medicine. *Polish Journal of Pharmacology,* 55: 699-702.

Lawlor DW (2002). Limitation of photosynthesis in water stressed leaves: stomata vs. metabolism and the role of ATP. *Annals of Botany,* 89: 1-15.

Lutts S, Kinet JM, Bouharmont J (1996). Effects of salt stress on growth, mineral nutrition and proline accumulation in relation to osmotic adjustment in Rice 118 (*Oryza sativa* L.) cultivars differing in salinity resistance. *Plant Growth Regulation,* 19: 207-218.

Mendi YY, İpek M, Kaçan H, Çuruk S, Sarı N, Çetiner S, Gaba V (2003). A Histological analysis of regeneration in watermelon. *Journal of Plant Biochemistry & Biotechnology,* 12: 147-150.

Moghbeli E, Fathollahi S, Salari H, Ahmadi G, Saliqehdar F, Safari A, Grouh MSH (2012). Effects of salinity stress on growth and yield of Aloe vera L. *Journal of Medicinal Plants Research,* 6: 3272-3277.

Munns R (2011). Plant adaptations to salt and water stress: differences and commonalities. *Advances in Botanical Research*, 57: 1-32.

Nas Z, Eşitken A, Pırlak L (2023). The responses of 'Viking' aronia variety to salinity stress under in vitro conditions. *Erwerbs Obstbau,* 65: 2547-2552.

Oren R, Sperry JS, Katul GG, Pataki DE, Ewers BE, Phillips N, Schäfer KVR (1999). Survey and synthesis of intra-and interspecific variation in stomatal sensitivity to vapour pressure deficit. *Plant, Cell & Environment,* 22: 1515-1526.

Özelçi D (2020). In vitro ortamda kuraklık stresine maruz bırakılan *Morus nigra* L. (Karadut)'da melatoninin etkisinin biyokimyasal ve fizyolojik cevaplarla değerlendirilmesi, MSc Thesis, İnönü Üniversitesi (Unpublished), Türkiye.

Parida AK, Das AB (2005). Salt tolerance and salinity effects on plants: a review. *Ecotoxicology and Environmental Safety,* 60: 324-349.

Ruzic D, Saric M, Cerovic R, Culafic L (2000). Relationship between the concentration of macroelements, their uptake and multiplication of cherry rootstock Gisela 5 *in vitro. Plant Cell, Tissue and Organ Culture*, 63: 9-14.

Sanchez FJ, De Andres EF, Tenorio JL, Ayerbe L (2004). Growth of epicotyls, turgor maintenance and osmotic adjustment in pea plants (*Pisum sativum* L.) subjected to water stress. *Field Crops Research,* 86: 81-90.

Sharma LK, Kaushal M, Bali SK, Choudhary OP (2013). Evaluation of rough lemon (*Citrus jambhiri* Lush.) as rootstock for salinity tolerance at seedling stage under in vitro conditions. *African Journal of Biotechnology,* 12: 6267-6275.

Shiyab SM, Shibli RA, Mohammad MM (2003). Influence of sodium chloride salt stress on growth and nutrient acquisition of sour orange in vitro. *Journal of Plant Nutrition,* 26: 985-996.

Sivritepe N, Erturk U, Yerlikaya C, Turkan I, Bor M, Ozdemir F (2008). Response of the cherry rootstock to water stress induced in vitro. *Biologia Plantarum,* 52: 573-576.

Smith D, Ringenberg C (2003). NF581 Aronia Berries. Historical Materials from University of Nebraska-Lincoln Extension, 40.

Torun AA, Kaçar-Aka Y, Kıllı O, Erdem H, Yardım P, Serçe S, İnan S (2007). Bazı *Fragaria chiloensis* genotiplerinin değişik NaCl konsantrasyonlarına *in vitro* çoğaltma sırasındaki tepkileri. *Derim,* 24: 27-33.

Turhan E, Eriş A (2007). Growth and stomatal behaviour of two strawberry cultivars under long-term salinity stress. *Turkish Journal of Agriculture and Forestry,* 31: 55-61.

Yilmaz H, Kina A (2008). The influence of NaCl salinity on some vegetative and chemical changes of strawberries (*Fragaria x ananassa* L.). *African Journal of Biotechnology,* 7: 2221-2227.

Zhao CB, Zhang H, Xu XY, Cao Y, Zheng MZ, Liu JS, Wu F (2017). Effect of acetylation and succinylation on physicochemical properties and structural characteristics of oat protein isolate. *Process Biochemistry,* 57: 117-123.

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Research Article

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The Effect of Rootstocks and Berry Heterogeneity on the Phytochemical Properties in *Vitis vinifera* L. Papazkarası Grapes

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HIGHLIGHTS

- A total of; 144 vines, 1440 clusters and 7200 berries were evaluated in this study.
- The highest anthocyanin and phenolic concentrations were found in berries with diameters ≤12.00mm.
- None of the berry size groups or rootstocks reached full ripeness.
- A reduction in berry size may improve grape quality, offering potential benefits for viticultural practices
- Wines from berry size groups with better must quality are expected to yield clearer sensory results.

Abstract

There are differences in development and composition between the clusters on the vines and the grape berries on the clusters. Therefore, grouping berries by size in wine grape varieties can help better manage the composition of the wine to be produced. The aim of this research is to reveal the effect of berry heterogeneity on primary and secondary metabolites in the cv. Papazkarası. The Papazkarası vines are grafted onto the rootstocks 1103P, 110R, and 420A and are trained in the double Cordon Royat system. The clusters harvested from each rootstock were separated into individual berries. These berries were then grouped by size using sieves. The size groups were: ≤12.00 mm; 12.01-14.00 mm; 14.01-16.00 mm; 16.01-18.00 mm; and ≥18.00 mm, forming five groups. A control group was created by taking berries from each size group. In terms of primary metabolites in the must, the Papazkarası/1103P combination and the ≥18.00 mm size group stood out. However, the maturity indices, Brix, and % alcohol criteria were found to be insufficient for all rootstocks and berry sizes. Regarding secondary metabolites, it was found that the ≤12.00 mm and 12.01-14.00 mm size groups had high values in all graft combinations. The size group with the highest total anthocyanin and total phenolic content was the ≤12.00 mm group. Based on these results, berry size reduction practices will improve quality. The quality of the wines obtained from different size groups should also be supported by sensory analyses. Sorting the berries by size will positively impact grape and thus wine quality.

Keywords: Berry size; wine grape; Papazkarası; autochthonous variety; secondary metabolites

1. Introduction

Berry size is one of the most important factors affecting grape and wine quality (Champagnol 1998; Gil et al. 2015; Melo et al. 2015). Significant differences in composition and development exist between the clusters

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on the vine (Nicolle et al. 2023) and between the berries within a cluster (Gouthu et al. 2014). The phenolic compounds in berry composition and their ratios are controlled by genetic structure, which is a characteristic of the species and variety (Cantürk and Kunter 2019). Additionally, it has been emphasized that the amount of these compounds is shaped by terroir factors (climate, soil, berry ripening stage, cultural practices, etc.) (Kennedy 2008; Kizildeniz et al. 2015; Shi et al. 2016; Foroni et al. 2017; Ribéreau-Gayon et al. 2021). Particularly in red grape varieties, attention should be paid not only to sugar content and acidity values during ripening but also to phenolic compounds and their extractability, as they directly affect the organoleptic properties of the wine (Ramos et al. 2024). Furthermore, along with phenolic compounds in the chemical composition of berries, organic acids also play a significant role in improving the sensory properties of wines depending on the degree of berry ripeness (Silva and Queiroz 2016).

Berry size has an impact on quality, though there are differing opinions on the matter. Matthews and Nuzzo (2007) reported that smaller berries are preferred because they have a higher surface-to-volume ratio, allowing for more compounds to transfer from the skin to the must, while larger berries are less favored due to dilution of their composition. However, Roby and Matthews (2004) argued that the skin-seed to pulp ratio does not change based on berry size, and since the skin grows along with the berry, the surface-to-volume ratio does not always determine the amount of soluble substances that can be extracted from the skin (Matthews and Nuzzo 2007; Barbagallo et al. 2011). They suggested that differences in berry size can lead to changes in composition, making it more challenging to control the ripening process (Barbagallo et al. 2011).

Grapes, which contain numerous secondary metabolites, are rich in polyphenols. The main polyphenols include anthocyanins, flavonols, and stilbenes-three classes of compounds that play a crucial role in vine metabolism and exhibit unique characteristics (Flamini et al. 2013). Among polyphenols, tannins are also present in both the seeds and skin (Ramos et al. 2024). The phenolic composition is greatly influenced by grape varieties, environmental conditions, and cultural practices (He and Giusti 2010). Arozarena et al. (2002) reported that the composition of anthocyanins in grape berries is controlled by genetic factors and varies according to variety. Zhang et al. (2021) emphasized that environmental factors such as temperature, sunlight, soil, and cultural practices affect these ratios. It has also been noted that temperature plays a crucial role in the formation of phenolic compounds (Spayd et al. 2002). However, the phenolic potential of grapes is mainly determined by the phenolic composition of the skin -anthocyanins, flavonols, and tannins- and the ability of these compounds to be extracted from the must (Schwarz et al. 2005; Casassa and Habertson 2014).

When the local cv. Papazkarası was grown at an altitude of 1030 m in Tunceli province, it was found that the total phenolic content in the skin and seeds was 21323 mg GAE kg^{-1}, and the total anthocyanin content was 816.1 mg kg^{-1} (Sanyürek et al. 2018). Özdemir (2017) reported that the Papazkarası grape variety from Kırklareli had a Titratable Acidity (TA) of 6.67 g L^{-1}, pH of 3.18, and Total Soluble Solids (TSS) of 19°Brix. Faikoğlu (2014) recorded that the TSS of the hardaliye (a traditional fermented beverage) made from the cv. Papazkarası was 5.2 g 100 mL^{-1}, pH 3.37, and TA 1.167 g 100 mL^{-1}. Using the cv. Papazkarası from five different vineyards, Erseç and Demirci (2023) produced wine via spontaneous fermentation and identified nine different strains through DNA sequencing.

Additionally, rootstocks influence grape quality by altering various physiological responses, such as managing the vigor of grafted scions, changing the hydraulic capacity of the root system, and controlling stomatal closure to reduce water loss during water stress (Gambetta et al. 2012; Marguerit et al. 2012). Besides contributing to disease control, rootstocks also affect grape quality by altering phytochemical components (Chen et al. 2024). Blank et al. (2022) stated that rootstocks are a powerful tool in managing grape phenolic components; for instance, the total tannin ratio of a variety grafted onto SO4 rootstock was 15% higher than that grafted onto Riparia gloire rootstock. However, Wang et al. (2019) found that rootstocks had little effect on the phenolic components of grapes. Different rootstocks have been shown to alter the growth and development of varieties, as well as their grape quality and resistance to stress factors (Ulaş et al. 2014; Ausari et al. 2024). Rootstocks grafted onto different grape varieties such as Syrah (Walker 2019), Cabernet Sauvignon (Miele and Rizzon 2019), and Pinot Noir (Harbertson and Keller 2012) have been suggested to cause changes in pH and TA content. Moreover, it has been found that the total phenolic content, total tannin, and flavan-3-ol accumulation of Cabernet Sauvignon grapevines grafted onto rootstocks such as SO4 and 1103P are affected

(Koundouras et al., 2009). Rootstocks can change the grape ripening rate and composition, such as TSS and TA (Stevens et al. 2008; Koundouras et al. 2009). Harbertson and Keller (2012) grafted Merlot, Syrah, and Chardonnay varieties onto five different rootstocks and reported that rootstocks did not affect anthocyanin and tannin levels. All measured grape and wine components, except TSS, changed significantly with the grafted variety.

Barbagallo et al. (2011) categorized grapes from the Syrah/99R graft combination into four groups based on their weight: 1. ≤1.50 g; 2. 1.51-2.00 g; 3. 2.01-2.50 g; 4. ≥2.50 g. As a result, they concluded that reducing variability in berry weight and size is important for improving wine quality. Melo et al. (2015) separated Syrah grape berries into three groups by diameter: small (<13 mm), medium (13-14 mm), and large (>14 mm), and found that smaller berries had a higher numerical density, while larger berries had a lower density. Chen et al. (2018) categorized Cabernet Sauvignon grape berries into three different size groups: small (0.75 g), medium (0.76-1.25 g), and large (>1.25 g), and reported that more than 50% of the berry population belonged to the medium group. They found that the physicochemical and biochemical parameters of the berries were significantly affected by berry size. Ünlüsoy (2019) determined that the 8mm-10mm berry size group of the Merlot grape variety had the highest TA, total phenolic content, total anthocyanin, total tannin, and total antioxidant values. They also stated that the best quality berry sizes were 8-10 mm and 10-12 mm.

The aim of this research is to reveal the changes in primary and secondary metabolites in the must of Papazkarası grape variety grafted onto three different rootstocks according to different berry size groups. Another goal is to determine which berry size should be used to improve the phytochemical composition of grape juice to enhance quality, based on the idea that achieving quality in the wine industry depends on grape quality.

2. Materials and Methods

2.1. Location and Plant Material

The trial was conducted in the vineyards of Irem Çamlıca Vineyards and Winery Ltd. Co. in Kırklareli (41°61'23.26"N, 27°61'89"E, 304 m altitude), where Papazkarası vines trained in the Double Cordon Royat trellising system were used as plant material. The 10-year-old Papazkarası vines were grafted onto 1103P, 110R, and 420A rootstocks. The planting distance was 2×1 m with 500 vines da^{-1}, and no irrigation or fertilization was applied.

The cv. Papazkarası is native to Türkiye. As an indigenous variety, it is commonly cultivated in the Thrace Region (Korkutal et al. 2019). It ripens in mid-October, its berries are medium-sized, and its clusters are compact. It is commonly used for both winemaking and table consumption. When processed into wine, it exhibits medium to low tannin content, high acidity, and high aroma. It is registered in the VIVC catalog under number 8923 as a hybrid of Alba Imputotato x Prokupac, originating from Türkiye (VIVC 2024).

The rootstocks used are: 1103P (Berlandieri Resseguier No:2 x Rupestris du Lot hybrid), 110R (Berlandieri Resseguier No:2 x Rupestris 110 Richter hybrid), and 420A (Berlandieri x Riparia 420A Millardet et de Grasset hybrid). The 1103P rootstock is highly resistant to the root form of phylloxera and shows 17% resistance to "active" limestone. It adapts well to dry conditions and is particularly suited to acidic soils, with fairly good tolerance to chlorides. The 110R rootstock is even more resistant to the root form of phylloxera. It also shows 17% resistance to "active" limestone and adapts very well to drought. It is especially suitable for dry, poor, stony, and schist soils. The 420A rootstock, like the other rootstocks, is highly resistant to the root form of phylloxera, with 20% resistance to "active" limestone. This rootstock is productive and well-adapted to deep, clay-limestone soils, but it has a low capacity for absorbing K from the soil (Plantgrape 2024).

2.2. Trial Design and Statistical Analysis

The trial, established with 144 vines in a Randomized Block Design, involved examining a total of 1440 clusters and 7200 berries across three rootstock groups, with 480 clusters from each rootstock. Berries were grouped by diameter using sieves. A control sample was created selecting berries from each size group, resulting in six berry size groups:

- E0 (Control): Mixed-sized berries,
- E1: Berries with a diameter less than Ø 12 mm,
- E2: Berries with a diameter between Ø 12.01-14.00 mm,
- E3: Berries with a diameter between Ø 14.01-16.00 mm,
- E4: Berries with a diameter between Ø 16.01-18.00 mm,
- E5: Berries with a diameter ≥ 18.01 mm.

The measurement results of grape berry samples collected at harvest were analyzed using the MSTAT-C statistical software package, and the differences between berry sizes and rootstocks were determined using the Least Significant Difference (LSD) test.

2.3. Analysis of Primer and Seconder Metabolites

Changes in Total Soluble Solids (TSS), Titratable Acidity (TA), and pH were monitored between veraison and harvest. Basic must analyses were performed on berry samples taken from each size group, including the control. The TSS (°Brix) of the juice from the six size groups was measured using a hand refractometer, the alcohol content (%) was determined using the Blouin and Guimberteau (2000) method, the TA value (g-tartaric acid L^{-1}) was determined using the titrimetric method, and the pH was determined according to Cemeroğlu (2015). Maturity Indices, such as °Brix/TA and pH^2×°Brix, were calculated according to Blouin and Guimberteau (2000).

From each size group, 100 berry samples were taken to determine anthocyanin, tannin, total phenolic content, total polyphenol index (TPI), etc. To determine the total anthocyanin content (mg kg^{-1}) from secondary metabolites, a puree of crushed grape berries was transferred into glass bottles (100ml), and an acidified methanol solution (2% HCl) was added to make up the volume to 100ml. After being left in the dark for one day, the extract was filtered, transferred into plastic tubes, and stored in a deep freezer (-18°C). Total monomeric anthocyanins were determined using the pH Differential Method. In this method, the difference between absorbance values measured at pH 1.0 and pH 4.5 is directly proportional to anthocyanin concentration. pH 1.0 (0.025 M KCl buffer solution) and pH 4.5 (0.4 M NaOAc buffer solution) were used as buffers. Absorbance readings were taken with a spectrophotometer at wavelengths of 520-720 nm. To determine the total phenolic content (mg kg^{-1}), 75 ml of distilled water + 1ml of extract was added to a volumetric flask. After adding 5 ml of Folin-Ciocalteu reagent and shaking, the mixture was left to stand for 3 min, followed by the addition of 10 ml of saturated Na_2CO_3 solution and 9ml of distilled water, making up the volume to 100 ml. After 60 min, the absorbance was read at 720nm using a spectrophotometer (Cemeroğlu 2015). To determine the total tannin content (mg kg^{-1}), 5ml of methanol + 1ml of extract was added. From this solution, 100 µl was taken, 500 µl of Folin-Ciocalteu reagent was added, followed by 1ml of Na_2CO_3, and the volume was made up to 10 ml with distilled water, with absorbance readings taken at 750 nm using a spectrophotometer (Mohammed and Manan 2015). To determine the Total Polyphenol Index (TPI), 1ml of must was diluted with 50 ml of distilled water, centrifuged at 8000 rpm for 10 min, and absorbance was read at 280 nm using a spectrophotometer. The dilution factor was multiplied by the absorbance value for the final calculation (INRA 2007).

3. Results and Discussion

3.1. Grouping of Berries According to Diameter (berries)

The berries from the clusters of each rootstock group were separated according to diameter (Figure 1). Accordingly, the number of berries in clusters from the 1103P rootstock was concentrated in the E3 group (42.63 berries), with the fewest berries found in the E1 (5.83 berries) and E5 (8.48 berries) groups. For the 110R rootstock, the highest number of berries was in the E3 group (40.77 berries), and the lowest number was in the E5 group (3.23 berries). For the 420A rootstock, the group with the highest number of berries was E3 (51.25 berries), although the E2 group (40.54 berries) also had a high berry count. The E5 group (2.50 berries) had the lowest value.

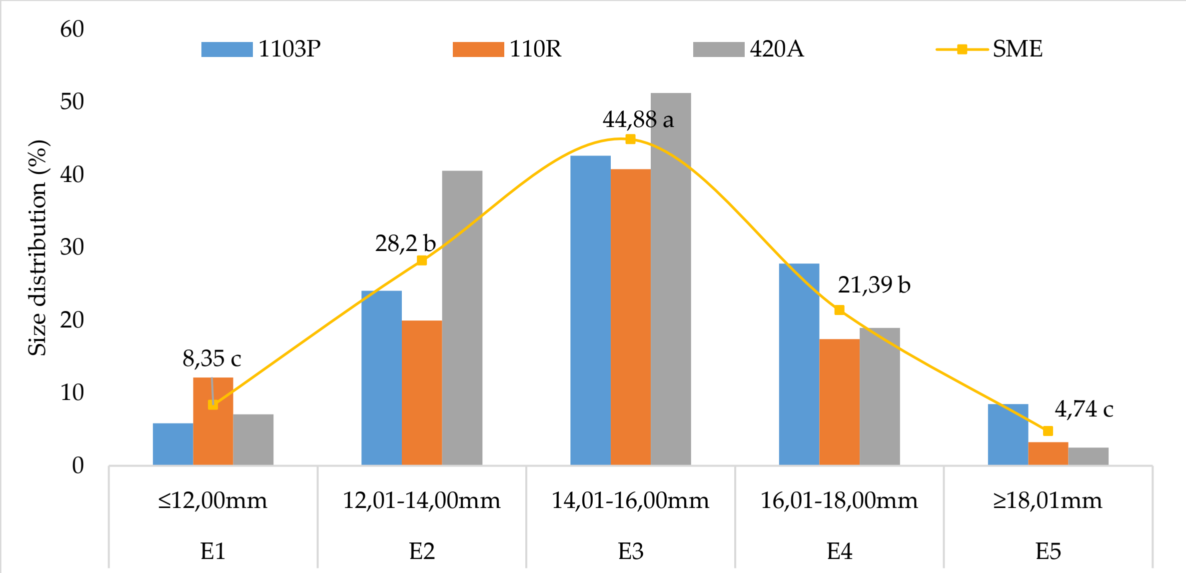

Figure 1. Size distribution of berries in the cluster [RME=Rootstock Main Effect, SME=Size Main Effect]

(RME $LSD_{0.01}$ = 8.66; SME $LSD_{0.01}$ = 5.74; Rootstock x Size Interaction $LSD_{0.01}$ = 4.39)

When examining the size distribution of the berries, the 1103P rootstock was found to be more effective in larger berries, while the 420A rootstock was more effective in smaller berries. This difference is thought to be due to the timing of pollination (Pisciotta et al. 2012; Tarter and Keuter 2005).

3.2. Proportional Distribution of Berry Size Groups (%)

In all rootstocks, berry sizes were concentrated in the E3 group. The proportion of the E2 size group in the 420A rootstock (33.09%) was higher than the E3 size group in the other rootstocks. In the 1103P rootstock, the proportion of the E4 size group (25.51%) was higher than that of the other rootstocks. Overall, 85-90% of the berries in the clusters fell within the 12.01-18.00 mm (E2, E3, and E4) size range (Figure 2).

3.3. Changes in Must Composition Between Veraison and Harvest

For wine grape varieties to reach harvest maturity, the desired values for TSS (20-25°Brix), pH (3.2-3.5), and TA (3-9 g L^{-1}) are considered (Blouin and Guimberteau 2000). The TSS value obtained from the study was 20.41°Brix, and the TA value was 7.13 g L^{-1}, which fall within the specified ranges. In summary, as expected, TSS increased and TA decreased by harvest time. However, the pH value remained almost unchanged between days of the year (DOY) 252 and 283, with a pH of 2.81 at harvest (Figure 3).

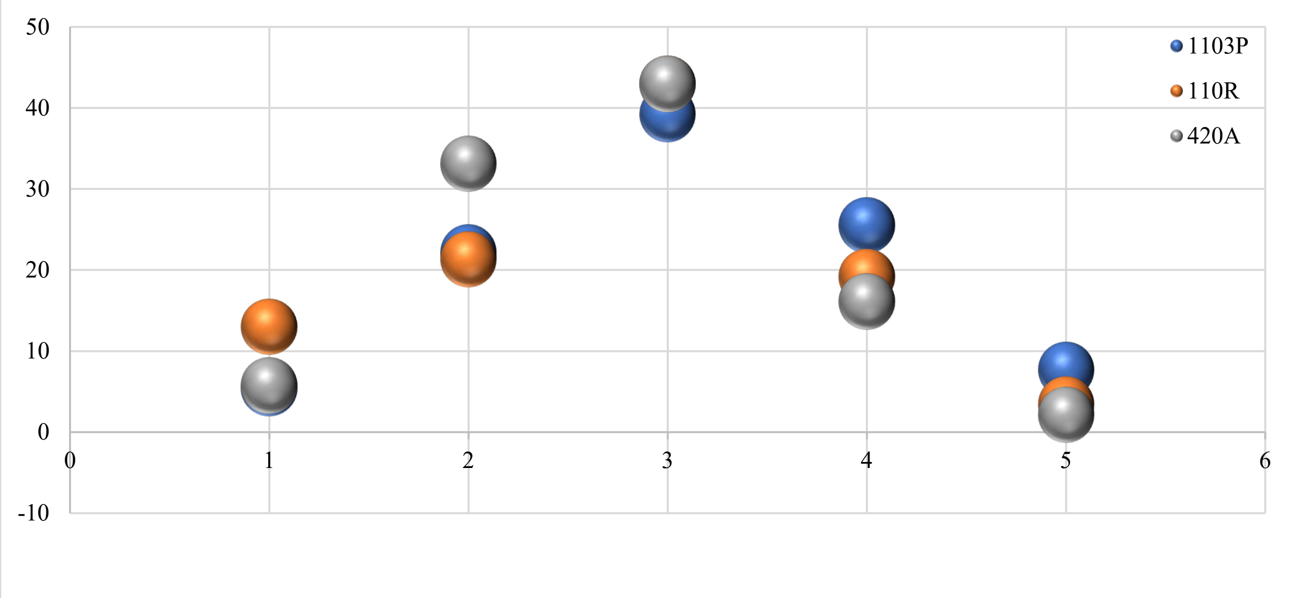

Figure 2. Proportional distribution of berry sizes in 100 berries

(SME $LSD_{0,01}$=10,71; Rootstock x Size Interaction $LSD_{0,01}$=8,18)

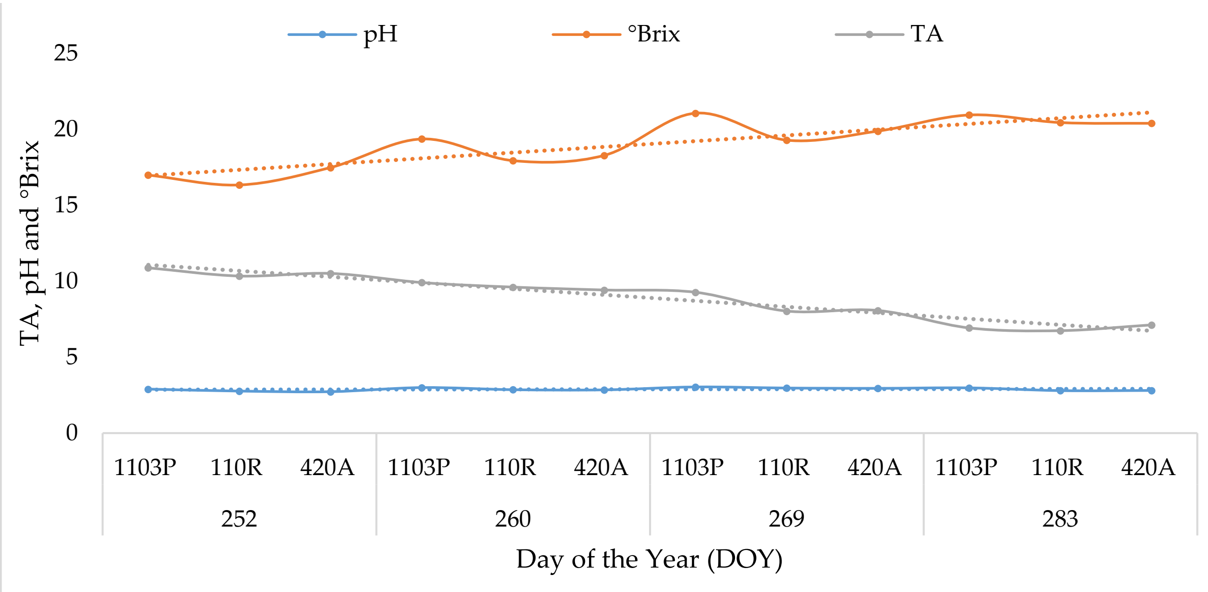

Figure 3. Weekly changes in must composition between veraison and harvest

3.4. Total Soluble Solids (TSS) (°Brix)

The factor that statistically significantly affected the TSS value in the cv. Papazkarası was the Size Main Effect (SME). When berry size was E5 (≥18.00 mm), the TSS value reached 21.76°Brix, and it was found that only this value was very close to the optimal range for wine grape varieties, 22-24°Brix (Cox 2015), while other berry sizes did not reach the optimum value (Table 1). The finding from Ünlüsoy (2019) that the 14-16mm berry size group in the Merlot/1103P combination gave the highest TSS value (21.51°Brix) aligns with the results of this study. Additionally, the TSS value of cv. Papazkarası grafted onto the 110R rootstock in the 2015 harvest season was found to be 19.00°Brix (Özdemir 2017), which is very close to the results of this study (20.46°Brix). Moreover, the findings are similar to those of Barbagallo et al. (2011), who reported a positive correlation between berry size and TSS. However, the findings contradict those of Chen et al. (2018) and Melo et al. (2015), who found a negative correlation between berry size and TSS. This difference is thought to be due to factors such as vineyard location, grape variety, etc. The finding that the E4 and E5 groups had higher TSS values than the control is also consistent with Bahar et al. (2024). The Rootstock Main Effect (RME) was not statistically significant, and these values were quite close. This finding contradicts the results of Korkutal et al. (2023), who stated that rootstocks affect TSS values, and this discrepancy is thought to be due to the characteristics of the location where the research was conducted.

Table 1. TSS, Alcohol, TA, pH, °Brix/TA, and pH²×°Brix Values

	Rootstock	E0 (K)	E1	E2	E3	E4	E5	RME
TSS (°Brix)	1103P	20.78	20.30	20.40	20.68	21.13	22.45	20.96
	110R	20.73	19.90	20.18	20.40	20.48	21.10	20.46
	420A	20.18	19.70	20.20	20.25	20.38	21.73	20.41
SME		20.56 b	19.97 b	20.26 b	20.44 b	20.66 b	21.76 a	
SME $LSD_{0.01}$ = 1.91								
Alcohol (%)	1103P	11.94	11.60	11.67	11.86	12.17	13.08	12.05
	110R	11.90	11.34	11.52	11.67	11.73	12.15	11.72
	420A	11.52	11.21	11.54	11.57	11.65	12.58	11.68
SME		11.79	11.38	11.58	11.70	11.85	12.60	
NS								
TA ($g\ L^{-1}$)	1103P	8.12	7.09	6.90	6.81	6.54	6,11	6.93
	110R	6.79	7.44	7.13	6.90	6.42	5.83	6.75
	420A	6.43	8.29	7.58	7.40	7.22	5.84	7.13
SME		7.11 a	7.61 a	7.20 a	7.04 a	6.73 ab	5.93 b	
SME $LSD_{0.01}$=2.67								
pH	1103P	3.01	2.77	2.86	2.95	3.11	3.17	2.98 a
	110R	2.78	2.68	2.78	2.81	2.87	2.89	2.80 b
	420A	2.73	2.78	2.75	2.79	2.86	2.94	2.81 ab
SME		2.84 ab	2.74 b	2.80 b	2.85 ab	2.94 ab	3.00 a	
RME $LSD_{0.01}$=0.38; SME $LSD_{0.01}$=0.38								
°Brix/TA	1103P	2.56	2.86	2.96	3.04	3.21	3.67	3.05
	110R	3.05	2.67	2.83	2.96	3.19	3.62	3.05
	420A	3.14	2.38	2.66	2.74	2.82	3.72	2.91
SME		2.92 b	2.64 b	2.82 b	2.91 b	3.07 ab	3.67 a	
SME $LSD_{0.01}$= 1.05								
pH²×°Brix	1103P	187.52	155.48	166.57	180.23	202.79	225.24	186.30 a
	110R	159.60	142.40	155.64	161.37	168.36	175.92	160.55 b
	420A	149.81	152.25	153.04	157.31	166.37	187.53	161.05 ab
SME		165.64 ab	150.04 b	158.42 b	166.30 ab	179.17 ab	196.23 a	
RME $LSD_{0.01}$=51.99; SME $LSD_{0,01}$=67.76								

3.5. % Alcohol

In the cv. Papazkarası, the effects of the Rootstock Main Effect (RME) and the berry Size Main Effect (SME) on alcohol percentage were not statistically significant (Table 1). It is known that as the TSS in grapes increases, the alcohol level in wine will also increase to a certain level (Feifel et al. 2024). In the study, a numerically high alcohol value was observed in the 1103P x E5 interaction (13.08%), which was thought to be due to the 22.45°Brix berry size of E5. This value was determined to be low based on Cox's (2015) finding that wine grape varieties should have a 22-24°Brix value. Other size groups did not reach the desired alcohol level. For producing high-quality wine, the stage with the highest sugar content and consequently the highest alcohol level is the harvest stage (Conde et al., 2007). However, the findings of the study revealed that the desired full maturity was not achieved at harvest, contrary to the researchers' expectations. On the other hand, similar to the findings of Ausari et al. (2024), it was concluded that the effects of rootstocks on alcohol content were not statistically significant.

3.6. Titratable Acidity (TA) (g-tartaric acid L^{-1})

In terms of titratable acidity (TA), the berry Size Main Effect (SME, $LSD_{0.01}$) was statistically significant (Table 1). The highest TA values were obtained from berry sizes E0, E1, E2, and E3 (7.04-7.61 g L^{-1}). This was followed by E4 with 6.73 g L^{-1}. However, the finding from Cox (2015) that this value should be 0.60-0.8g mL^{-1} for red wine grape varieties does not match with the E5 (5.93 g L^{-1}) size. On the other hand, it was observed that TA decreases as berry size increases. This finding was found to be consistent with the results reported by Chen et al. (2018) and Ünlüsoy (2019), which indicate that smaller berries have higher TA values. However, the results contradict the findings of Melo et al. (2015), who reported that TA is not affected by berry size. This difference is thought to be due to varietal differences. Additionally, the finding that the TA value of Papazkarası/110R vines in the vineyard where the study was conducted was 6.67 g L^{-1} in the 2015 harvest is similar to the study results (6.75 g L^{-1}) of Özdemir (2017).

3.7. pH

In the cv. Papazkarası, both the RME and the SME were found to be significant (Table 1). When examining the effect of rootstocks, the highest pH value was found in the 1103P rootstock (2.98), followed by the 420A rootstock (2.81) and the 110R rootstock (2.80). The pH value of 2.80 recorded for the 110R rootstock of the Papazkarası grape variety in 2021 from the same vineyard is thought to differ from the 3.18 pH value reported by Özdemir (2017) in 2015 due to differences in the trial years. According to the SME, the E5 size had the highest pH value (3.00). E0, E3, and E4 sizes were in the same significance group, while E1 (2.74) and E2 (2.80) sizes were in the last significance group. In general, there is a positive correlation between berry size increase and pH increase (Chen et al. 2018). However, the study did not reach Cox's (2015) finding that the optimum pH value for red varieties is 3.4. Since higher pH values can negatively affect wine quality by promoting unwanted bacterial growth, as reported by Cox, it is thought that the obtained values will not negatively impact quality.

3.8. Maturity Indices

°Brix/TA

The berry Size Main Effect was significant for °Brix/TA (Table 1). In terms of SME, the E5 group (3.67) stood out. It was found that the E4 group (3.07) followed the E5 group, and the other berry size groups were in the last significance group. Dardeniz and Kısmalı (2002) provided guidance on measures to be taken in regions with ripening issues, based on their finding that they achieved 5-9 days earlier ripening with 30-60% cluster thinning.

pH^2x°Brix

Among the ripening indices, both RME and SME were found to be significant for $pH^2\times$°Brix (Table 1). Among the rootstocks, 1103P had the highest value (186.30), 110R had the lowest (160.55), and 420A was between the two (161.05). The E5 size group (196.23) had the highest value, while E0, E3, and E4 (165.64-179.17) sizes were in the middle, and E1 and E2 (150.04-158.42) had the lowest values. Contrary to Blouin and Guimberteau's (2000) definition that full ripeness is achieved when this index exceeds 260°Brix, it can be said that none of the berry size groups or rootstocks reached full ripeness.

3.9. Secondary Metabolites

Total Anthocyanin Content (mg kg^{-1})

Total anthocyanin content was influenced by both rootstock and berry size (Table 2). The 110R rootstock had the highest total anthocyanin content (852.70 mg kg^{-1}), followed by the 420A rootstock (744.88 mg kg^{-1}), with the 1103P rootstock (565.43 mg kg^{-1}) in the last group. This finding is similar to Blank et al. (2022), who reported that the 110R rootstock had a higher anthocyanin concentration than the 125AA rootstock. The lowest total anthocyanin content was found in berries of size E5 (535.94 mg kg^{-1}), while the highest value was obtained from berries of size E1 (973.30 mg kg^{-1}). There was no difference between the control (E0), E3, and E4 berry sizes. The finding that small berries (<13 mm) had higher anthocyanin values than other sizes is consistent with Melo et al. (2015) and Ünlüsoy (2019). When the total anthocyanin content in grape berry skin is expressed in mg kg^{-1}, Barbagallo et al. (2011) stated that increasing berry weight reduces anthocyanin levels. The findings of this study are similar; as berry size increased (≥18.01 mm = 535.94 mg kg^{-1}), the anthocyanin content decreased. Bahar and Kurt (2015) reported that increasing the number of berries per cluster and reducing berry size led to an increase in anthocyanin content. A similar finding was obtained in this study, as smaller berries (12.00 mm, 12.01–14.00 mm, and 14.01–16.00 mm) had higher anthocyanin levels. Sanyürek et al. (2018) determined that the total anthocyanin content of the Papazkarası grape variety grown under Tunceli conditions was 816.1 mg kg^{-1}. This finding was found to be nearly identical to the anthocyanin content of E2-sized berries in the study (833.92 mg kg^{-1}).

Table 2. Total anthocyanins, Total phenolic content, Total tannins, and TPI values

	Rootstock	E0 (K)	E1	E2	E3	E4	E5	RME
T. Anthocyanins (mg kg^{-1})	1103P	548.20	828.11	680.61	550.53	444.84	340.30	565.43 b
	110R	731.33	1145.58	1025.95	871.47	717.39	624.47	852.70 a
	420A	658.54	946.19	795.21	780.11	646.15	643.06	744.88 ab
SME		646.02 bc	973.30a	833.92 ab	734.04 bc	602.79 bc	535.94 c	
SME $LSD_{0.01}$ =3.99; RME $LSD_{0.01}$=3.99								
T. Phenolic Content (mg kg^{-1})	1103P	5092.98	9512.10	6823.64	6473.71	4773.05	3110.38	5964.31 b
	110R	6867.63	10514.90	9050.19	6617.34	6801.97	6328.73	7696.79 a
	420A	5666.87	8848.23	7422.52	5609.88	5331.93	4663.07	6257.08ab
SME		5875.82 bc	9625.07 a	7765.45 b	6233.64 bc	5635.65 bc	4700.73 c	
SME $LSD_{0.01}$ =3.57; RME $LSD_{0.01}$=3.57								
T. Tannins (mg kg^{-1})	1103P	3400.1421	4844.7273	3992.7351	3085.9560	3830.6106	3022.2243	3696.07
	110R	3439.2756	3624.8802	4077.7107	3091.5465	2334.5928	1990.2180	3093.04
	420A	2988.6813	4031.8686	3950.2473	3334.1742	2993.1537	2900.3514	3366.41
SME		3276.03 abc	4167.16 a	4006.90 ab	3170.56 ab	3052.79 c	2637.60 c	
SME $LSD_{0.05}$=1.59								
TPI	1103P	12.87	23.68	14.53	13.45	11.60	8.25	14.06 b
	110R	13.80	25.76	18.42	16.16	15.03	9.58	16.46 a
	420A	14.38	22.21	15.80	12.23	13.46	11.31	14.90 ab
SME		13.68 bc	23.88 a	16.25 b	13.94 b	13.37 bc	9.71 c	
RME $LSD_{0.01}$=4.30; SME $LSD_{0.01}$=8.94								

Total Phenolic Content (mg kg^{-1})

The Rootstock Main Effect was statistically significant, and the 110R rootstock had the highest total phenolic content (7696.79 mg kg^{-1}) (Table 2). This was followed by the 420A and 1103P rootstocks. According to SME, the phenolic content was also statistically significant. The highest total phenolic content was found in E1 size berries (9625.07 mg kg^{-1}). The lowest amount was recorded in the E5 size group (4700.73 mg kg^{-1}), while no difference was found between the E3 and E4 size groups. This finding aligns with Ünlüsoy (2019), who reported the highest phenolic content in the smallest berry size group (8–10 mm). Similarly, in this study, the berry size group E5 had the lowest phenolic content. However, the results contradict the findings of Melo et al. (2015), who reported that berry size did not affect phenolic content. This discrepancy is thought to be due to differences in rootstock, variety, and location.

Total Tannin Content (mg kg^{-1})

Berry size significantly affected total tannin content in the cv. Papazkarası (Table 2). The highest total tannin content was found in the smallest berry size group (E1=4167.16 mg kg^{-1}). No difference was found between E2, E3, E4, and E5 berry sizes in terms of tannin content. As expected, the E0 size (3276.03 mg kg^{-1}) had a total tannin content value between E2, E3, E4, and E5. The smallest tannin values were found in the E4 (3052.79 mg kg^{-1}) and E5 (2637.60 mg kg^{-1}) berry sizes. The findings are consistent with Ünlüsoy (2019). In short, as berry size decreased, tannin content increased. The finding that the tannin content in the 110R rootstock was lower than in other rootstocks is consistent with the findings of Ausari et al. (2024).

Total Polyphenol Index (TPI)

Berry size and rootstock types had significant effects on the Total Polyphenol Index (TPI), which indicates the degree of phenolic maturity in grapes (Table 2). The 110R rootstock had the highest TPI value (16.46), while the 1103P rootstock had the lowest (14.06). The 420A rootstock had a value between these two rootstocks (14.90). According to the SME, E1-sized berries had the highest TPI value (23.88), while the E5 size had the lowest TPI value (9.71). The other berry size groups and the control group were between these two values.

4. Conclusions

The three different rootstocks onto which the cv. Papazkarası was grafted were generally insufficient in terms of the criteria examined in this terroir and ripeness. The 1103P rootstock had a high °Brix value, reduced titratable acidity, increased pH, and achieved the highest alcohol content, but it did not reach the desired level. The pH^2×°Brix should be 260, but it was 186.03, indicating that full ripeness was not achieved. The 1103P rootstock exhibited higher total tannin content compared to the other rootstocks. It should be noted that a high total tannin content can result in an astringent taste in the wine. The 110R rootstock had moderate sugar accumulation, low pH, and TA values, and the alcohol content was also recorded as moderate. This rootstock had the lowest ripeness index value. Full ripeness was not achieved with this rootstock either, and small and late-ripening berries negatively affected the quality of the must. Although small berries increased phenolic components, they reduced the total tannin content. The 420A rootstock, despite having the lowest sugar content, did not show a significant difference compared to the other rootstocks. Due to the sugar accumulation, the pH value was low, and the acid content was high. Although this rootstock did not reach the required level of ripeness, its homogeneous berries show promise in terms of must quality. However, studies with different rootstocks need to be continued.

For all rootstocks, the highest number of berries was observed in the size range of 12.01 mm to 18.00 mm (E2, E3, E4, and E0). Although these berries were not of high quality, they are considered suitable for winemaking. The other size groups, E1 and E5 (≤12.00 mm and ≥18.00 mm), were rich in primary and secondary metabolites but did not reach sufficient berry numbers. The highest levels of anthocyanins and phenolic compounds were recorded in berries sized ≤12.00 mm. These findings suggest that reducing berry size could improve quality. It is believed that sensory evaluation of wines produced from the berry size groups that improve must quality will provide more definitive results.

To enhance the phytochemical composition, efforts should be made to reduce berry sizes through various techniques and to increase ripeness, focusing on methods that raise alcohol content. Sorting berries by size manually is a long and costly process but using mechanization could speed up this process and reduce costs. It is anticipated that this method will be beneficial in improving grape and, consequently, wine quality.

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References

Arozarena I, Ayestarán B, Cantalejo M, Navarro M, Vera M, Abril I, Casp A (2002). Anthocyanin composition of Tempranillo, Garnacha and Cabernet Sauvignon grapes from high and low-quality vineyards over two years. *European Food Research and Technology,* 214(4): 303-309. https://doi.org/10.1007/S00217-001-0471-0

Ausari PK, Gurjar PKS, Somkuwar RG, Naruka IS, Sharma AK, Gharate PS (2024). Effect of rootstocks on yield and wine quality of Sauvignon Blanc variety. *Plant Archives,* 24(1): 1477-1482. https://doi.org/10.51470/PLANTARCHIVES.2024.v24.no.1.207

Bahar E, Kurt C (2015). Farklı toprak işleme ve yaprak alanı/ ürün miktarının Syrah üzüm çeşidinin fizyolojisi, morfolojisi ve üzüm bileşimi üzerine etkileri: II. Şıra özellikleri üzerine etkileri. *Selçuk Tarım ve Gıda Bilimleri Dergisi-A,* 27(Özel Sayı): 316-332.

Bahar E, Korkutal I, Uzun M (2024). Effects of different water stress levels, heterogeneity, and location on berry phytochemical properties in an organic and conventional vineyard (*Vitis vinifera* cv. Cabernet-Sauvignon). *KSU Journal of Agriculture and Nature,* 27(5): 1042-1054. https://doi.org/10.18016/ksutarimdoga.1333996

Barbagallo MG, Guidoni S, Hunter JJ (2011). Berry size qualitative characteristics of *Vitis vinifera* L. cv. Syrah. *South African Journal of Enology and Viticulture,* 32(1): 129-136. https://doi.org/10.21548/32-1-1372

Blank M, Samer S, Stoll M (2022). Grapevine rootstock genotypes influences berry and wine phenolic composition (*Vitis vinifera* L. cv. Pinot noir). *OENO One* 56(1): 133–144. https://doi.org/10.20870/oeno-one.2022.56.1.4459

Blouin J, Guimberteau G (2000). Maturation et Maturite des Raisins. Feret, Bordeaux, France. p.168.

Cantürk S, Kunter B (2019). Üzümlerde aroma bileşikleri. In: Cantürk S, Kunter B (Eds), *Current Research and Assesments for Agricultural Sciences.* 1st Edition, IVPE, Cetinje-Montenegro. pp.19-28.

Casassa LF, Harbertson JF (2014). Extraction, evolution, and sensory impact of phenolic compounds during red wine maceration. *Annual Review of Food Science and Technology,* 5: 83-109. https://doi.org/10.1146/annurev-food-030713-092438

Casassa LF, Keller M, Harbertson JF (2015). Regulated deficit irrigation alters anthocyanins, tannins and sensory properties of Cabernet Sauvignon grapes and wines. *Molecules,* 20(5): 7820-7844. https://doi.org/10.3390/molecules20057820

Cemeroğlu B (2015). Gıda Analizleri. Bizim Büro Basımevi, Ankara, Türkiye. p.480.

Champagnol F (1998). Critéres de Qualitié de la Vendange. in: Flanzy, C. (Ed) Oenologie, Fondements Scientifiques et Technologiques. Lavoisier Tec & Doc, Paris, France. p.1311.

Chen KW, He F, Wang XY, Liu X, Duan QC, Wang J (2018) Influences of berry size on fruit composition and wine quality of *Vitis vinifera* L. cv. 'Cabernet Sauvignon' grapes, *South African Society for Enology and Viticulture,* 39(1): 67-76. https://doi.org/10.21548/39-1-2439

Chen Y, Liang Z, Krstic M, Clingeleffer P, Howell K, Chen D, Zhang P (2024) The influences of rootstock on the performance of Pinot Noir (*Vitis vinifera* L.): Berry and wine composition. *Australian Journal of Grape and Wine Research,* 7586202: 19. https://doi.org/10.1155/2024/7586202

Conde C, Silva P, Fontes N, Dias ACP, Tavares RM, Sousa MJ, Agasse A, Delrot S, Gerós H (2007). Biochemical changes throughout grape berry development and fruit and wine quality. *Food,* 1(1): 1-22.

Cox J (2015). From Vines to Wines, 5th Edition: The Complete Guide to Growing Grapes and Making Your Own Wine. Storey Publishing, North Adams, Massachusetts, USA, p.264.

Dardeniz A, Kısmalı İ (2002). Amasya ve Cardinal üzüm çeşitlerinde farklı ürün yüklerinin üzüm ve çubuk verimi ile kalitesine etkileri üzerine araştırmalar. *Ege Üniversitesi Ziraat Fakültesi Dergisi,* 39(1): 9-16.

Erseç Ç, Demirci AŞ (2023). Isolation, identification and determination of *Saccharomyces cerevisiae* yeast species from the wines made by spontaneous fermentation using Papazkarası grapes from Thrace Region. *Journal of Tekirdag Agricultural Faculty*, 20(1): 155-165. https://doi.org/10.33462/jotaf.1124694

Faikoğlu F (2014). Adakarası, Papazkarası, Kalecik karası üzüm çeşitleri kullanılarak üretilen hardaliyelerin kalitesinin ve duyusal özelliklerinin araştırılması. MSc Thesis, Uludağ University (Unpublished), Türkiye.

Feifel S, Weilack I, Markusevics E, Zimmermann D, Wegmann-Herr P, Weber F, Richling E, Durner D (2024). Influence of potential alcohol in grapes on phenolic and sensory characteristics of red wine. *Journal of Agricultural and Food Chemistry,* 72(22): 12725-12737. https://doi.org/10.1021/acs.jafc.4c01035

Flamini R (2013). Mass spectrometry in grape and wine chemistry. Part I: Polyphenols. *Mass Spectrometry Reviews,* 22: 218-250. https://doi.org/10.1002/mas.10052

Foroni F, Vignando M, Aiello M, Parma V, Paoletti MG, Squartini A, Rumiati RI (2017). The smell of terroir! Olfactory discrimination between wines of different grape variety and different terroir. *Food Quality and Preference,* 58: 18-23. https://doi.org/10.1016/j.foodqual.2016.12.012

Gambetta G, Manuck C, Drucker S, Shaghasi T, Fort K, Matthews M, Walker MA, McElrone AJ, McElrone A (2012). The relationship between root hydraulics and scion vigour across Vitis rootstocks: what role do root aquaporins play? *Journal of Experimental Botany,* 63(18): 6445-6455. https://doi.org/10.1093/jxb/ers312, 2-s2.0-84869987523

Gil M, Pascual O, Gómez-Alonso S, García-Romero E, Hermosín-Gutiérrez I, Zamora F, Canals JM (2015). Influence of berry size on red wine colour and composition. *Australian Journal of Grape and Wine Research,* 21(2): 200-212. https://doi.org/10.1111/ajgw.12123

Gouthu S, O'Neil ST, Di Y, Ansarolia M, Megraw M, Deluc LG (2014). A comparative study of ripening among berries of the grape cluster reveals an altered transcriptional programme and enhanced ripening rate in delayed berries. *Journal of Experimental Botany,* 65(20): 5889-902. https://doi.org/10.1093/jxb/eru329

Harbertson JF, Keller M (2012). Rootstock effects on deficit-irrigated winegrapes in a dry climate: grape and wine composition, *American Journal of Enology and Viticulture,* 63(1): 40-48. https://doi.org/10.5344/ajev.2011.11079

He J, Giusti MM (2010). Anthocyanins: Natural colorants with health-promoting properties. *Annual Review of Food Science and Technology,* 1: 163-187. https://doi.org/10.1146/annurev.food.080708.100754

INRA (2007). Determination d'Anthocyanes En Echantillons De Raisin. Mode Operatiore. Ref: MO-LAB-23. Version: 1, Septembre 2007. UE Pech Rouge, France. p.2.

Kennedy AJ (2008) Grape and wine phenolics: Observations and recent findings. *Ciencia e Investigacion Agraria,* 35(2): 107-120. https://doi.org/10.4067/S0718-16202008000200001

Kizildeniz T, Mekni I, Santesteban H, Pascual I, Morales F, Irigoyen JJ (2015). Effects of climate change including elevated CO_2 concentration, temperature and water deficit on growth, water status, and yield quality of grapevine (*Vitis vinifera* L.) cultivars. *Agricultural Water Management,* 159: 155-164. https://doi.org/10.1016/j.agwat.2015.06.015

Korkutal İ, Bahar E, Güvemli Dündar D (2019) Edirne ili Uzunköprü ilçesi bağcılık yapısının incelenmesi. *ÇOMÜ Ziraat Fakültesi Dergisi,* 7(1): 127-136. https://doi.org/10.33202/comuagri.457451

Korkutal İ, Bahar E, Koskosoğlu B (2023). Cabernet Franc üzüm çeşidinin fitokimyasal özelliklerine eğimdeki konumun, anacın ve salkım seyreltmenin etkisi. *Mustafa Kemal Üniversitesi Tarım Bilimleri Dergisi,* 28(3): 536-556. https://doi.org/10.37908/mkutbd.1231642

Koundouras S, Hatzidimitriou E, Karamolegkou M, Dimopoulou E, Kallithraka S, Tsialtas JT, Zioziou E, Nikolaou N, Kotseridis Y (2009). Irrigation and rootstock effects on the phenolic concentration and aroma potential of *Vitis vinifera* L. cv. Cabernet Sauvignon grapes. *Journal of Agricultural and Food Chemistry,* 57(17): 7805-7813. https://doi.org/10.1021/jf901063a

Marguerit E, Brendel O, Lebon E, Van Leeuwen C, Ollat N (2012). Rootstock control of scion transpiration and its acclimation to water deficit are controlled by different genes. *New Phytologist,* 194(2): 416-429. https://doi.org/10.1111/j.1469-8137.2012.04059.x

Matthews MA, Nuzzo V (2007). Berry size and yield paradigms on grape and wine quality. *Acta Horticulturae,* 754*:* 423-436. https://doi.org/10.17660/ActaHortic.2007.754.56

Melo MS, Schultz HR, Volschenk CG, Hunter JJ (2015). Berry size variation of *Vitis vinifera* L. cv. Syrah: Morphological dimensions, berry composition and wine quality. *South African Journal of Enology and Viticulture,* 36(1): 1-10. https://doi.org/10.21548/36-1-931

Miele A, Rizzon LA (2019). Rootstock-scion interaction: 3. Effect on the composition of Cabernet Sauvignon wine, *Revista Brasileira de Fruticultura,* 41(1): e-642. https://doi.org/10.1590/0100-29452019642

Mohammed S, Manan AF (2015) Analysis of total phenolics, tannins and flavonoids from *Moringa oleifera* seed extract. *Journal of Chemical and Pharmaceutical Research, 7*(1): 132-135.

Nicolle P, Barthe C, Dorais M, Dubé G, Angers P, Pedneault K (2023). Impact of cluster thinning and harvest date on berry volatile composition and sensory profile of *Vitis* sp. Seyval blanc and Vandal-Cliche. *OENO One,* 57(4). https://doi.org/10.20870/oeno-one.2023.57.4.7719

Özdemir S (2017). Farklı maya suşlarının Narince ve Papazkarası (*Vitis vinifera*) üzüm çeşitlerinden üretilen şarapların kaliteleri üzerine etkileri. MSc.Thesis, Tekirdag Namık Kemal University (Unpublished), Türkiye.

Pisciotta A, Abruzzo F, Barbagallo MG, Santangelo T, di Lorenzo R (2012). Ulteriori approfondimenti degli effetti della dimensione degli acini sulla qualità dell'uva nella cv. Cabernet Sauvignon. *Italus Hortus,* 3(1): 82-88.

Plantgrape (2024). Rootstocks. https://www.plantgrape.fr/en/varieties/rootstock-varieties (access date: 26.09.2024)

Ramos MC, Jara, MAI, Rosillo L, Salinas MR (2024). Effect of temperature and water availability on grape phenolic compounds and their extractability in Merlot grown in a warm area. *Scientia Horticulturae,* 337: 113475. https://doi.org/10.1016/j.scienta.2024.113475

Ribéreau-Gayon P, Glories Y, Maujean A, Dubourdieau D (2021). Handbook of Enology, Vol 2: The Chemistry of Wine and Stabilization and Treatments (3rd Edition). John Wiley and Sons, West Sussex, England. p.441.

Roby G, Matthews MA (2004). Relative proportions of seed, skin and flesh, in ripe berries from Cabernet Sauvignon grapevines grown in a vineyard either well irrigated or under water deficit. *Australian Journal of Grape and Wine Research,* 10: 74-82. https://doi.org/10.1111/j.1755-0238.2004.tb00009.x

Sanyürek KN, Tahmaz H, Çakır A, Söylemezoğlu G (2018). Tunceli ilinde yetiştirilen bazı şaraplık üzüm çeşitlerinde antioksidan aktivitenin ve fenolik bileşiklerin belirlenmesi. *Türk Tarım ve Doğa Bilimleri Dergisi,* 5(4): 551–555. https://doi.org/10.30910/turkjans.471340

Schwarz M, Picazo-Bacete JJ, Winterhalter P, Hermosín-Gutiérrez I (2005). Effect of copigments and grape cultivar on the color of red wines fermented after the addition of copigments. *Journal of Agricultural Food and Chemistry,* 53: 8372-8381. https://doi.org/10.1021/jf051005o

Shi PB, Yue TX, Ai LL, Cheng YF, Meng JF, Li MH, Zhang ZW (2016). Phenolic compound profiles in grape skins of Cabernet Sauvignon, Merlot, Syrah and Marselan cultivated in the Shacheng area (China). *South African Journal of Enology and Viticulture,* 37(2): 132-138. http://www.scielo.org.za/scielo.php?script=sci_arttext&pid=S2224-79042016000200008&lng=en&nrm=iso

Silva LR, Queiroz M (2016). Bioactive compounds of red grapes from Dão region (Portugal): evaluation of phenolic and organic profile. *Asian Pacific Journal of Tropical Biomedicine,* 6(4): 315-321. https://doi.org/10.1016/j.apjtb.2015.12.015

Spayd SE, Tarara JM, Mee DL, Ferguson JC (2002). Separation of sunlight and temperature effects on the composition of *Vitis vinifera* cv. Merlot berries. *American Journal of Enology and Viticulture,* 53: 171-182. https://doi.org/10.5344/ajev.2002.53.3.171

Stevens RM, Pech JM, Gibberd MR, Walker RR, Jones JA, Taylor J, Nicholas PR (2008). The effect of reduced irrigation on growth, yield, ripening rates and water relations of Chardonnay vines grafted on five rootstocks. *Australian Journal of Grape and Wine Research,* 14: 177-190. https://doi.org/10.1111/j.1755-0238.2008.00018.x

Tarter ME, Keuter SE, (2005) Effect of rachis position on size and maturity of Cabernet Sauvignon berries. *American Journal of Enology and Viticulture,* 56(1): 86-89. Doi:10.5344/ajev.2005.56.1.86

Ulaş S, Güler A, Candemir A (2014). Effect of rootstocks on different physiological parameters in some grape cultivars. *Turkish Journal of Agricultural and Natural Sciences,* 1(Special Issue-1): 1097-1100.

Ünlüsoy S (2019). Merlot üzüm çeşidinde farklı toprak işleme ve salkım seyreltme uygulamalarının tane heterojenitesi ve bileşimi üzerine etkileri. MSc Thesis, Tekirdag Namık Kemal University (Unpublished), Türkiye.

VIVC (2024). Papazkarası üzüm çeşidi. https://www.vivc.de/index.php?r=passport%2Fview&id=8923 (acces date: 26.09.2024)

Walker RR, Blackmore DH, Clingeleffer PR, Holt H, Pearson W, Francis I (2019). Effect of rootstock on yield, grape composition and wine sensory attributes of Shiraz grown in a moderately saline environment. *Australian Journal of Grape and Wine Research,* 25(4): 414-429. https://doi.org/10.1111/ajgw.12409

Wang Y, Chen WK, Xiao-tong G, He L, Yang XH, He F, Duan, CQ, Wang J (2019). Rootstock-mediated effects on Cabernet-Sauvignon performance: Vine growth, berry ripening, flavonoids, and aromatic profiles. *International Journal of Molecular Sciences,* 20: 401. https://doi.org/10.3390/ijms20020401

Zhang K, Zhang ZZ, Yuan L, Gao XT, Li Q (2021). Difference and characteristics of anthocyanin from Cabernet Sauvignon and Merlot cultivated at five regions in Xinjiang. *Food Science and Technology,* 41(1): 72-80. https://doi.org/10.1590/fst.11020

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Investigation of the Potential of Growing Some Early-Season Soybean Genotypes as Alternative Crops Under the Ecological Conditions of Sivas

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HIGHLIGHTS

- In this study, twenty-five soybean genotypes and five registered soybean varieties (Arısoy, Traksoy, Samsoy, Soyanam and Ataem-7) were used as materials.
- With this study, it is thought that Arısoy, Traksoy, Samsoy, Soyanam, Ataem-7 varieties and ÜNV-7, ÜNV-11, ÜNV-20 genotypes can be successfully grown in Sivas climate conditions.
- However, in order to reach definitive conclusions, further studies are needed in different climate conditions and for many years.

Abstract

Soybean is of strategic importance among oilseed crops due to its versatile uses and its ability to meet the growing demand for vegetable oils driven by the rapidly increasing global population. In Türkiye, soybean production remains far below the required levels to meet domestic demand, resulting in a growing reliance on imports each year. However, Türkiye has the potential for soybean production, given its favorable climate and soil conditions. To increase production, the development of local cultivars and the expansion of planting areas are essential. Specifically, including soybean in crop rotation systems in regions such as Central Anatolia, where the crop can be easily grown and developing new high-yield, high-quality varieties suited to these areas, will be crucial steps in boosting domestic production. This study aimed to evaluate the agro-morphological characteristics of several early-maturing soybean genotypes in the Sivas ecological conditions during the summer growing season and to explore soybean as an alternative crop. In this study, twenty-five soybean genotypes and five registered soybean varieties (Arısoy, Traksoy, Samsoy, Soyanam and Ataem-7) were used as materials. The study was conducted over one year during the 2023 soybean growing season at the Agricultural R&D Center trial field of Sivas University of Science and Technology. The field trial was set up according to a randomized block design with three replications. The obtained data showed that the days to first flowering ranged from 74.67 to 82.33 day and the days to maturity varied between 158 and 160 day. Additionally, the number of lateral branches ranged from 2.47 to 7.34 piece, the first pod height varied from 8.14 to 23.80 cm and the number of pods per plant ranged from 41.40 to 155 pods plant^{-1}. The number of seeds per pod ranged from 2.47 to 3.11 piece and the pod weight varied from 0.10 to 0.36 g. The 100-seed weight was determined to be an average of 7.14 g. Significant statistical differences at the 5% level were observed

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for plant height, first pod height, number of pods per plant, number of seeds per pod, number of lateral branches, pod length, pod width, pod weight, and 100-seed weight. Based on these findings, it can be concluded that the varieties Arısoy, Traksoy, Samsoy, Soyanam, Ataem-7 and the genotypes ÜNV-7, ÜNV-11, ÜNV-20 can be successfully cultivated in the climatic conditions of Sivas. However, it is emphasized that further studies over multiple years and in different climatic conditions are necessary to draw definitive conclusions.

Keywords: *Glycine max* L.; agro-morphological characteristics; adaption; breeding

1. Introduction

Soybean (*Glycine max* L.) is a significant livelihood source in many countries, particularly in China and Korea and stands out as an oilseed crop with extensive global applications (Bakal et al. 2017). It is a nutrient-rich source, with high protein and fat content. In kitchens, soybeans are used in various forms, such as soy milk, soy sauce, soy flour and soybean oil. Additionally, due to its high fiber content, it facilitates digestion, supports heart health and is commonly preferred in diets. Furthermore, soybeans have been shown to improve metabolism and reduce the risk of diabetes. Soybean holds a prominent place in the oilseed category globally, with its versatility and the growing demand for plant-based oils being key factors in this importance (Arıoğlu et al. 2012; Güngör and Üstün 2015; Boerema et al. 2016; Nadeem et al. 2021a). In Türkiye, however, soybean production is insufficient to meet domestic demand and this gap is filled by increasing imports each year. Despite having the necessary climate and soil conditions for soybean production, insufficient domestic cultivation has led to higher imports. This gap could be closed by promoting soybean planting and developing locally adapted high-yield, high-quality varieties. To increase domestic production, it is necessary to include soybean in crop rotation in regions like Central Anatolia, where the crop can be grown easily and to develop varieties that perform well in these areas. Genotype, environment and genotype × environment interactions significantly affect yield and quality in soybeans (Erbil and Gür 2017; Gül and Arslanoğlu 2020; Okcu 2020; Nadeem et al. 2021b). Due to the rapid increase in global population and demand for plant oils, soybean cultivation has gained considerable importance. In 2010, global production of oilseeds was 832 million tons, rising to 1.102 billion tons by 2019. Soybean's extensive use has made it the second-largest oilseed crop after palm, with 3.409 million tons produced. Soybean is the leading oilseed in terms of planted area (11.525 million hectares) and is second in terms of production (3.047 million tons). According to 2019 data, approximately 1.205 million hectares, or 37.37% of the global oilseed planting area, is covered by soybeans (FAO 2022).

This study aims to evaluate the agro-morphological characteristics of some early-maturing soybean genotypes during the summer growing season under the ecological conditions of Sivas Province. The goal is to explore the potential of soybean as an alternative crop. This research is expected to make significant contributions to expanding soybean cultivation areas in Türkiye and to the economy of local farmers.

2. Materials and Methods

In this study, twenty-five soybean genotypes and five registered soybean varieties (Arısoy, Traksoy, Samsoy, Soyanam and Ataem-7) were used as materials. The study was conducted over one year during the 2023 soybean growing season at the Agricultural R&D Center trial field of Sivas University of Science and Technology. The field trial was set up according to a randomized complete block design with three replications. In the trial, each genotype was planted in 5-meter plots with 4 rows, 70 cm row spacing and 10 cm plant spacing. Along with sowing, 6 kg da^{-1} of nitrogen (N) and 8 kg da^{-1} of phosphorus (P_2O_5) were applied. The planting was carried out on May 16, 2023, taking into account the climatic conditions of the region. During the growing season, weed control, irrigation and all necessary maintenance operations were performed as required based on climatic conditions. A drip irrigation system was used for irrigation. The soil properties of the trial field at the Agricultural R&D Center of Sivas University of Science and Technology are presented in Table 1.

The soil at the location where the study was conducted is a silty clay loam with a pH value of 7.28. It is characterized by low organic matter content (1.7%), high potassium levels (93.59 kg da^{-1}), low phosphorus

(P_2O_5), lime content (19.6) and low salt content (0.33%). During the study, there were no issues with groundwater and the land was adequately drained (Table 1).

Table 1. The soil characteristics of the experimental site at the Sivas Science and Technology University, Agricultural Research and Development Center

Depth	Texture	pH	Lime (% CaCO3)	Salt (%)	Phosphorus (P_2O_5 kg da^{-1})	Potassium (K_2O kg da^{-1})	Organic Matter Content (%)
0-30 cm	Silty clay loam	7.28	19.6	0.33	3.40	93.59	1.7

Table 2. The climate data for Sivas province during the 2023 growing season, along with long-term climate data, are presented*

	Total Precipitation (mm)		Average Temperature (˚C)		Average Relative Humidity (%)	
Months	**2023**	**Long Term**	**2023**	**Long Term**	**2023**	**Long Term**
April	74.8	33.7	9.1	8.9	92.8	62.3
May	56.4	54.7	13.0	13.5	93.6	61.1
June	51.4	43.4	17.3	17.0	95.3	58.3
July	3.0	6.2	20.1	20.0	82.8	54.0
August	3.6	4.5	23.4	20.3	76.6	53.0
September	4.3	17.8	19.2	16.3	72.3	62.0
October	7.6	36.8	18.4	10.9	74.5	64.0
Total/Average	201.1	197.1	17.21	15.27	83.99	59.24

* Sivas Provincial Meteorology Directorate

The climatic data for Sivas Province during the 2023 growing season, as well as long-term averages, are presented in Table 2. Sivas has a continental climate characterized by hot and dry summers and cold, snowy winters. The basic climatic values, such as total precipitation, average temperature and average relative humidity for the study period, are shown in Table 2. During the trial months, the total precipitation was lowest in July (3.0 mm) and highest in April (74.8 mm). In 2023, the lowest average temperature occurred in April (9.1 °C), while the highest average temperature was observed in August (23.4 °C). The lowest average relative humidity was recorded in September (72.3%) and the highest average relative humidity occurred in June (95.3%). In this study, several morphological characteristics of soybean genotypes and varieties were investigated, including days to flowering (50%), days to maturity, plant height, first pod height, number of pods per plant, number of seeds per pod, number of branches, pod length, pod width, pod weight and 100-seed weight. The obtained data were subjected to analysis of variance using the MSTATC statistical software and the differences between means were grouped using the Least Significant Difference (LSD) test ($p \le 0.05$).

3. Results and Discussion

The data for the first flowering day count, days to maturity, plant height, number of branches, first pod height and number of pods per plant for the soybean genotypes and varieties studied are presented in Table 3.

Upon examining Table 3, it is observed that the number of days to first flowering in the soybean genotypes and varieties ranged from 74.67 to 82.33 days, with an average of 79.55 days. The difference in the number of days to first flowering among the samples was found to be statistically significant at the 5% level (Table 3). One of the most important factors affecting the growth and development of soybeans is day length. Soybean is a short-day plant and as the day length increases, the time to begin flowering also extends. Late-maturing soybean varieties are more sensitive to day length and tend to produce more flowers under long-day conditions. Ünal (2007) reported that the flowering period for the soybean lines obtained through hybridization ranged from 35.00 to 45.00 days. Other similar studies reported that the days to flowering ranged from 79.47 to 80.72 days (Hızlı et al. 2023), from 38.9 to 42.7 days in 2018 and from 37.7 to 40.7 days in 2019 (Erbil 2020). The results of our study on flowering time are similar to some of the studies, while differing in others. These differences may be due to factors such as sowing time, day length, genetic structure, varieties used, growing regions and climatic conditions (Erbil 2020; Zhang et al. 2001).

Table 3. The data for the soybean genotypes and varieties, including the first flowering day (days), days to maturity (days), plant height (cm), first pod height (cm), number of sub-branches (piece) and number of pods per plant (pods $plant^{-1}$), as well as the groups formed based on the results of the analysis of variance, are presented.

Varieties	First Flowering (day)	Number of Days to Maturity (day)	Plant Height (cm)	Number of Sub-Branches (piece)	First Pod Height (cm)	Number of Pods Per Plant (pods $plant^{-1}$)
Arısoy	79.00 a-c	158.00	113.3 b-c	3.96 h-k	23.80 a	41.40 m
Traksoy	74.67 c	158.00	90.2 g-j	3.27 k-l	18.40 c-g	59.93 k-l
Samsoy	77.33 a-c	158.00	87.20 h-l	4.87 e-ı	19.00 b-f	155.5 a
Soyanam	76.00 b-c	158.00	95.20 f-h	4.57 f-k	14.00 h-m	91.07 e-h
Ataem-7	82.00 a	158.00	103.7 c-f	3.40 j-l	17.73 c-h	62.00 j-l
4	82.00 a	158.00	111.5 b-d	4.38 f-k	18.27 c-g	65.87 j-l
5	77.67 a-c	159.00	110.3 c-d	4.20 g-k	19.80 b-e	71.93 ı-l
6	81.33 a-b	159.00	82.07 j-m	4.73 e-j	15.05 g-l	69.78 j-l
7	82.00 a	160.00	90.20 g-j	4.73 e-j	17.73 c-h	73.13 ı-k
8	82.33 a	159.00	103.0 d-f	3.93 h-k	18.27 c-g	87.27 f-ı
9	82.00 a	158.00	107.2 c-e	3.65 ı-l	13.27 j-m	69.33 j-l
10	82.33 a	160.00	98.67 e-g	4.33 f-k	21.20 a-d	58.80 k-l
11	77.67 a-c	159.00	128.5 a	3.48 j-l	22.47 a-b	61.27 j-l
12	75.67 b-c	158.00	121.1 a-b	2.47 l	21.47 a-c	41.40 m
ÜNV-2	82.00 a	159.00	88.11 h-l	7.34 a	17.61 d-h	75.67 h-j
ÜNV-3	79.00 a-c	159.00	83.13 j-m	4.40 f-k	8.67 n-o	104.1 c-e
ÜNV-4	82.33 a	158.00	89.80 g-k	6.60 a-b	11.43 l-o	108.1 c
ÜNV-5	80.00 a-c	160.00	94.47 f-ı	3.64 ı-l	16.80 e-j	76.42 g-j
ÜNV-6	78.33 a-c	159.00	80.13 k-n	6.27 a-d	13.80 ı-m	57.40 l
ÜNV-7	81.00 a-b	158.00	93.85 f-ı	5.43 b-g	14.28 h-m	107.0 c-d
ÜNV-8	82.00 a	159.00	79.80 l-n	5.27 b-h	13.07 j-m	91.60 d-g
ÜNV-11	76.67 a-c	159.00	84.82 ı-m	6.62 a-b	14.51 h-m	99.09 c-f
ÜNV-12	78.33 a-c	158.00	83.87 j-m	4.93 d-ı	11.00 m-o	98.13 c-f
ÜNV-13	79.67 a-c	160.00	68.33 o	5.00 c-ı	17.50 d-ı	105.2 c-e
ÜNV-15	75.67 b-c	158.00	80.22 k-n	4.94 d-ı	19.33 b-f	131.2 b
ÜNV-16	80.00 a-c	159.00	75.08 m-o	5.65 b-f	12.35 k-n	65.13 j-l
ÜNV-17	81.33 a-b	158.00	76.75 m-o	7.25 a	15.17 g-l	112.1 c
ÜNV-18	79.67 a-c	158.00	75.74 m-o	5.98 a-e	14.30 h-m	76.03 g-j
ÜNV-19	79.33 a-c	159.00	71.97 n-o	5.47 b-g	8.14 o	58.0 k-l
ÜNV-20	79.33 a-c	158.00	87.20 h-l	6.33 a-c	15.61 f-k	103.9 c-e
Average	79.55	158.63	91.85	4.90	16.13	85.16
F value	1.92*	NS	17.92**	6.27**	8.45**	24.39**
Mean square	8.512	0.00	37.201	0.711	5.270	92.086
LSD (%)	4.768	-	9.969	1.378	3.752	15.68

*: $p<0.05$, **: $p<0.01$

The number of days to maturity ranged from 158 to 160 days. There was no statistically significant difference between the varieties and genotypes regarding the number of days to maturity. ÜNV-10, ÜNV-5 and ÜNV-13 were identified as the genotypes that reached the latest harvest maturity (Table 3). In a study by Erbil (2020), the physiological maturity period ranged from 119.2 to 135 days, while Malik et al. (2011) reported the average physiological maturity period as 101.18 days. The findings in our study are partially similar to those reported in other studies.

The average plant height of the soybean genotypes and varieties used in the study was determined to be 91.85 cm. The lowest plant height was found in the ÜNV-13 genotype at 68.33 cm, while the highest plant height was recorded in genotype 11 at 128.5 cm (Table 3). The difference in plant height values was statistically significant at the 1% level (Table 3). Similar studies have reported plant heights ranging from 64 to 118 cm (Bakoğlu and Ayçiçek 2005), from 71.3 to 121.6 cm (Erbil 2020) and from 41.17 to 57.50 cm (Mert and İlker 2016). Özer (2021) found plant heights of 65.58 to 74.50 cm in the first year and 37.67 to 48.08 cm in the second year of his study.

Table 4. The data for the soybean genotypes and varieties, including the number of seeds per pod (seeds), pod weight (g), pod length (cm), pod width (cm) and 100-seed weight (g), as well as the groups formed based on the results of the analysis of variance, are presented.

Varieties	The Number of Seeds Per Pod (piece)	Pod Weight (g)	Pod Length (cm)	Pod Width (cm)	100-Seed Weight (g)
Arısoy	3.07 a-c	0.29 a-f	4.67 a-c	0.91 a-d	8.34 c-g
Traksoy	3.13 a	0.36 a	4.88 a-b	0.93 a-d	11.33 a
Samsoy	2.93 a-d	0.32 a-c	4.45 a-d	0.90 a-d	8.37 b-g
Soyanam	2.93 a-d	0.29 a-f	4.37 a-e	0.89 a-d	8.39 b-g
Ataem-7	3.00 a-c	0.34 a-b	4.99 a	1.07 a-b	8.45 b-f
4	2.84 c-d	0.23 f-ı	4.72 a-c	1.07 a-b	4.02 m
5	2.95 a-d	0.23 f-ı	4.60 a-d	1.01 a-c	6.34 h-l
6	2.98 a-d	0.21 g-ı	3.67 e-f	0.81 b-e	5.28 j-m
7	3.04 a-c	0.24 d-ı	4.63 a-d	0.94 a-d	6.53 h-k
8	2.99 a-c	0.20 ı	4.23 b-f	0.87 a-d	6.82 g-j
9	2.83 c-d	0.21 h-ı	4.60 a-d	0.95 a-d	4.29 m
10	2.88 b-d	0.20 ı	4.45 a-d	0.92 a-d	4.35 m
11	3.05 a-c	0.29 a-f	4.24 a-f	0.95 a-d	3.94 m
12	2.87 b-d	0.28 b-h	4.67 a-c	1.04 a-b	4.24 m
ÜNV-2	2.74 d	0.20 ı	2.11 h-ı	0.46 g-h	8.14 d-g
ÜNV-3	2.87 b-d	0.26 c-ı	4.29 a-f	0.86 a-d	11.37 a
ÜNV-4	2.89 b-d	0.23 e-ı	4.12 c-f	0.86 a-d	6.06 ı-l
ÜNV-5	2.92 a-d	0.28 b-g	4.12 c-f	0.87 a-d	8.46 b-f
ÜNV-6	2.92 a-d	0.26 c-ı	4.69 a-c	0.75 c-f	6.48 h-l
ÜNV-7	3.01 a-c	0.34 a-b	4.22 b-f	0.88 a-d	9.92 a-b
ÜNV-8	3.01 a-c	0.25 d-ı	4.54 a-d	0.86 a-d	9.00 b-e
ÜNV-11	3.04 a-c	0.30 a-e	3.88 d-f	0.94 a-d	9.85 a-c
ÜNV-12	2.95 a-d	0.28 b-h	4.71 a-c	0.88 a-d	7.55 e-ı
ÜNV-13	2.47 e	0.10 j	2.86 g-h	0.54 e-h	4.41 m
ÜNV-15	2.87 b-d	0.22 g-ı	2.17 h-ı	0.42 h	7.06 f-ı
ÜNV-16	3.00 a-c	0.34 a-b	4.47 a-d	1.10 a	9.61 b-d
ÜNV-17	2.97 a-d	0.25 d-ı	1.97 ı	0.51 f-h	5.16 k-m
ÜNV-18	3.00 a-c	0.31 a-d	4.16 b-f	0.70 d-g	7.79 e-h
ÜNV-19	3.11 a-b	0.25 d-ı	2.23 h-ı	0.54 e-h	4.93 l-m
ÜNV-20	2.97 a-d	0.24 e-ı	3.55 f-g	0.73 d-g	7.67 e-h
Average	2.94	0.26	4.04	0.83	7.14
F value	2.12**	5.58**	3.50**	10.82**	15.25**
Mean square	0.022	0.002	0.029	0.214	0.928
LSD (%)	0.24	0.07309	0.7561	0.2783	1.574

*: p<0.05, **: p<0.01

In this study, the number of sub-branches, first pod height and number of pods per plant ranged from 2.47 to 7.34 piece, 8.14 to 23.80 cm and 41.40 to 155 (pods plant^{-1}), respectively (Table 3). The differences among these three traits were statistically significant at the 1% level (Table 3). Bakoğlu and Ayçiçek (2005) reported that the first pod height ranged from 12 to 31 cm, with an average of 18.57 cm; the number of branches ranged from 1 to 3, with an average of 1.93; and the number of pods per plant ranged from 21 to 76, with an average of 44.30. Mert and İlker (2016) reported that the number of pods per plant ranged from 36.33 to 48.33. Hızlı et al. (2023) observed the first pod height to be an average of 5.37 cm and the number of branches to be 4.02. Erbil (2020) reported that the first pod height ranged from 3.1 to 8 cm in 2018, while Civelek (2006) found first pod height values between 3.95 and 8.81 cm in a study conducted in Samsun. Arslan (2007) reported that the first pod height ranged from 10.3 to 22.8 cm in the main crop and from 4.3 to 9.3 cm in the second crop.

Upon examining Table 4, the number of seeds per pod and pod weight ranged from 2.47 to 3.11 seeds per pod and from 0.10 to 0.36 g, respectively. Bakoğlu and Ayçiçek (2005) found the number of seeds per pod to range from 2 to 3.90. The average pod length and pod width of the soybean genotypes and varieties were determined to be 4.04 cm and 0.83 cm, respectively (Table 3). The differences among the number of seeds per pod, pod weight, pod length and pod width were statistically significant at the 1% level (Table 4).

In this study, the average 100-seed weight was determined to be 7.14 g. The lowest 100-seed weight was observed in genotype number 11, with a value of 3.94 g, while the highest 100-seed weight was found in genotype ÜNV-3, with a value of 11.37 g (Table 4). This difference between genotypes was found to be statistically significant at the 1% level. In a study by Erbil (2020), the thousand-seed weight of soybean genotypes was determined to be 155.89 g in 2015 and 155.53 g in 2016. Bakal et al. (2021) reported that the 1000-seed weight of soybean varieties ranged from 160.0 to 155.2 g under different treatments. Bakoğlu and Ayçiçek (2005) found that the 100-seed weight ranged from 6 to 17 g. Research has emphasized that seed weight can be influenced by various factors such as genotype, environmental conditions, sowing time and cultural practices and it is one of the most important indicators of yield. Furthermore, it has been stated that seed size can vary significantly among genotypes.

4. Conclusions

The results obtained from this study indicate that the expansion of soybean cultivation areas could provide significant economic benefits for regional farmers. Accordingly, it has been concluded that the varieties Arısoy, Traksoy, Samsoy, Soyanam, Ataem-7, along with genotypes ÜNV-7, ÜNV-11 and ÜNV-20, can be successfully cultivated under the climatic conditions of Sivas province. Both field and laboratory studies have shown that these varieties and genotypes are well-suited to the agroecological conditions of Sivas. However, to make a definitive evaluation and obtain long-term results, further studies under varying climatic conditions over different years are necessary. Such long-term and comprehensive research is crucial for ensuring the sustainability of soybean production and for the development of regional agricultural policies. In this context, it is believed that expanding soybean production areas more widely is necessary to increase domestic production, improve production efficiency and contribute to the local economy.

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References

Arıoğlu H, Özyurtseven S, Güllüoğlu L (2012). Determination of oil yield and fatty acid contents of some soybean [*Glycine max* (L.) Merr] varieties grown under second crop conditions-II. *Çukurova University Journal of Agriculture*, 27(2): 1-10.

Arslan D (2007). Effects of selections made at main and second crop planting times on yield and yield components in soybean (*Glycine max* (L.) Merll). Master Thesis, On Dokuz Mayıs University, Samsun, Türkiye.

Bakal H, Bağırkan Ö, Arıoğlu H (2021). Effects of mechanical damage at the top growth point in different development periods of soybean plants grown under second crop conditions on seed yield and some agronomic traits. *Mustafa Kemal University Journal of Agricultural Sciences*, 26(1): 117-127.

Bakal H, Gulluoglu L, Bihter O, Arioglu, H (2017). The effect of growing seasons on some agronomic and quality characteristics of soybean varieties in Mediterranean Region in Türkiye. *Turkish Journal of Field Crops*, 22(2): 187-196.

Bakoğlu A, Ayçiçek M (2005). Agricultural characteristics and seed yield of soybean (*Glysine max*. L.) in Elazığ conditions. *Fırat University Journal of Science and Engineering Sciences*, 17(1): 52-58.

Boerema A, Peeters A, Swolfs S, Vandevenne F, Jacobs S, Staes J, Meire P (2016). Soybean trade: balancing environmental and socio-economic impacts of an intercontinental market. *Plos One*, 11(5): e0155222.

Civelek T (2006). Effect of foliar iron application on yield, yield components and important quality traits in some soybean (*Glycine max* L.) cultivars. Master Thesis, Ondokuz Mayıs University, Samsun, Türkiye.

Erbil E (2020). Evaluation of yield and quality traits of some advanced soybean (*Glycine max*. L.) lines in Şanlıurfa second crop conditions. *ISPEC Journal of Agricultural Sciences*, 4(2): 272-284.

Erbil E, Gür MA (2017). Investigation of performance of some advanced soybean (*Glycine max*. L.) lines in Şanlıurfa second crop conditions in terms of yield traits using physiological and morphological parameters. *Harran Journal of Agriculture and Food Sciences*, 21(4): 480-493.

FAO, 2022. Food and agriculture organization. https://www.fao.org/statistics/en/ (access date: 06.10.2024).

Gül S, Arslanoğlu F (2020). The influence of organic fertilizer applications on seed yield and some quality properties of soybean grown as second crop. *Uşak University Journal of Science and Nature Sciences*, 4(2): 114-126.

Güngör H, Üstün A (2015). The effect of two different row spacings on yield and some yield components in some soybean (*Glycine max*. (L.) Merill) genotypes in Konya ecology. *Gaziosmanpaşa University Journal of Agriculture Faculty*, 32(2): 100-106.

Hızlı H, Çubukçu P, Şahar AK (2023). Path analysis, genetic variability and correlation studies of related characters for forage soybean (*Glycine max* (L.) Merill). *Osmaniye Korkut Ata University Journal of Science Institute*, 6(2): 1513-1528.

Malik MFA, Ashraf M, Qureshi AS, Khan MR (2011). Investigation and comparison of some morphological traits of the soybean populations using cluster analysis. *Pakistan Journal of Botany*, 43(2): 1249-1255.

Mert M, İlker E (2016). Studies on the adaptation of some soybean (*Glycine max* (L.) Merill) lines and varieties to Aksaray region under main crop conditions. *Journal of Field Crops Central Research Institute*, 25(2): 176-181.

Nadeem MA, Habyarimana E, Karaköy T, Baloch FS (2021b). Genetic dissection of days to flowering via genome-wide association studies in Turkish common bean germplasm. *Physiology and Molecular Biology of Plants*, 27(7): 1609-1622.

Nadeem MA, Yeken MZ, Shahid MQ, Habyarimana E, Yılmaz H, Alsaleh A, Hatipoğlu R, Çilesiz Y, Khawar KM, Ludidi N, Ercişli S, Aasim M, Karaköy T, Baloch FS (2021a). Common bean as a potential crop for future food security: an overview of past, current and future contributions in genomics, transcriptomics, transgenics and proteomics. *Biotechnology & Biotechnological Equipment*, 35(1): 759-787.

Okcu M (2020). Effects of different sowing times on some agronomic traits in soybean varieties. *Gümüşhane University Journal of Science Institute*, 10(4): 972-982.

Özer N (2021). Determination of hay yield and some plant traits of soybean (*Glycine max* L.) harvested in different phenological periods. Master's Thesis, Tekirdağ Namık Kemal University, Tekirdağ, Türkkiye.

Ünal İ (2007). Determination of some agronomic traits of soybean (*Glycine max* (L.) Merr.) lines obtained by hybridization. Master's Thesis, Selçuk University, Konya, Türkiye.

Zhang L, Wang R, Hesketh JD (2001). Effects of photoperiod on growth and development of soybean floral bud in different maturity. *Agronomy Journal*, 93: 944-948.

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Research Article

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Assessing Factors Influencing the Adoption of Orchard Management Practices among Walnut Growers: A Study of Exotic and Native Jumlish Varieties of Nepal

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HIGHLIGHTS

- Most commercial walnut growers are unaware of good orchard management practice.
- This study aimed to examine the key factors that affect the adoption of orchard management practices.
- Education and area under walnut cultivation substantially impact the adoption rate.
- Locally grafted Jumlish walnut has proven to be more adaptable to Nepal's agro-climatic conditions than several exotic types.

Abstract

Walnut, a high-value crop with significant global demand, plays a vital role in enhancing the agricultural economy of the western high hills of Nepal. Sustainable orchard management practices are crucial to improving plant health and farm productivity. A survey conducted from February to July 2023 assessed the management practices of walnut orchards and evaluated the agro-climatic suitability of exotic walnut varieties in Rukum-East, Nepal. Using a simple random sampling method, 120 commercial walnut farmers participated in the study, with data collected through pre-tested, semi-structured questionnaires. The analysis utilized descriptive statistical tools, t-tests, and logistic regression models. Results revealed that farmers cultivated an average of 6.79 ropani of land with walnuts and had 2.22 years of orchard management experience. Exotic varieties such as Chandler (US origin), Franquette, and Fernette (French origin) were grown by 64.2% of farmers, while 35.8% relied on locally grafted Jumlish varieties. Globally relevant insights emerged, showing that education and cultivated area influenced orchard sanitation, and factors like land direction, road access, irrigation, and the use of Bordeaux paste significantly reduced dieback issues. Locally grafted varieties demonstrated superior growth performance and agro-climatic adaptability compared to the exotic varieties. This study highlights the global importance of integrating modern orchard care, subsidized quality sapling distribution, irrigation infrastructure, and grafting techniques to reduce sapling mortality. These findings contribute to the worldwide discourse on sustainable walnut cultivation in diverse agro-climatic contexts, promoting practices applicable to other regions with similar challenges.

Keywords: *Juglans regia;* Dieback; Hartley and Payne; Subsidy; Training and Extension

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1. Introduction

Horticultural commodities contribute 14% to the total agricultural GDP (AGDP), while fruits alone contribute 7.04% (MoALD 2021). With undulating slopes, the hilly and high-hill topography shows promise for horticultural crops such as apple, walnut, peach, and citrus. *Juglans* is a genus within the Juglandaceae family, and its seeds are commonly recognized as walnuts (Jahanban et al. 2019). The Persian walnut tree, also called English walnut or *Juglans regia* L., is a widespread monoecious species belonging to the order Fagales (Bernard et al. 2018). Walnuts exhibit adaptability to temperate climates worldwide (Mir and Kumar 2011) and have historical significance in traditional medicine and religious practices in Nepal (Aryal et al. 2009). The horticulture sector and private nurseries produce over 335,000 non-grafted walnut saplings and around 30,000 grafted saplings annually (NCFD 2020). Nepal's total walnut cultivation area is 5988 hectares, with a national average production of 10,895 MT and a yield of 4.46 MT/ha (MOALD 2022). Rukum-East, covering 502 hectares, dedicates 355 hectares to walnut cultivation, producing 1398 MT with a productivity of 3.94 MT/ha, below the national average (MOALD 2022).

The walnut species in Nepal is *Juglans regia* L., locally referred to as 'Okhar' (Thapa et al. 2021). It is grown in temperate zones (Dhakal et al. 2003) as well as in the country's mountainous regions at elevations of 1350–3000 meters above sea level (Khanal et al. 2023). The local hard-shelled walnut (*Hade*), abundantly found in the subtropical and temperate forests of Nepal, has thrived for centuries and serves as an excellent rootstock, well-adapted to the agro-ecological, climatic, and soil conditions of the region (Chalise et al. 2021). Some exotic thin-shelled varieties, such as 'Hartley', 'Payne', and 'Ashley', grafted onto these native rootstocks, are gaining popularity in commercial farming due to their significant economic value. They are collectively called the Jumlish varieties, as most of the breeding research related to walnuts is conducted at the Horticultural Research Station in Rajikot, Jumla (Sharma et al. 2022). Recently some exotic varieties such as Chandler, Franquette, Fernor, and Fernett, are been commercially cultivated in Nepal. Traditional rootstocks, *Juglans regia* (English walnut), native to their respective countries, have been used for these exotic varieties, which have been widely cultivated with financial aid and subsidy programs from governmental bodies. This has raised concerns regarding their climatic and soil adaptability in Nepal. The tall Persian walnut tree, with an average height of 25-35 meters and a wide trunk, bears green fruits with semi-fleshy coverings and brown, uneven nuts. Walnut serves as a prime example of the underutilization of available plant genetic resources. Variation plays a vital role in identifying superior seedlings with desirable traits, such as high-quality nuts and adaptability to local climatic and soil conditions, to support the potential expansion of walnut cultivation in the country. A heterogeneous population enables us to choose the best walnut genotypes, which is an easy and rapid way to improve the variety because breeding takes a long time for fruit crops, particularly walnuts with lengthy juvenile periods.

Walnuts are one of the main fruits that can bring in money and create jobs in the mountain districts of western Nepal. Regarding climate appropriateness, walnut cultivation has a comparative and competitive advantage and a lot of potential for export. Nepal is obliged to import more and more walnuts from foreign countries for commercial cultivation. Farmers cannot provide the best management practices to ensure walnut saplings' higher survival after establishing orchards. There are no regular orchards of walnuts in the country because the existing plants are of seedling origin. Lack of quality planting materials and mineral fertilizers are the main constraints to walnut cultivation in Nepal. Till now, there was no registered variety of walnuts in the country, and no walnut breeding program was performed in the past. Nepal Agriculture Research Council (NARC) is the sole agent responsible for fruit research. Several studies have been done in foreign countries to identify and introduce the superior walnut genotypes. Still, in Nepal, the first initiation of walnut research was to identify potential genotypes. In the Rukum-East district, farmers encounter challenges such as a low walnut sapling survival rate, heightened disease and pest prevalence, and a lack of practical information and technological knowledge in production (Thapa and Dhimal 2017). Additionally, nursery farmers face issues like land fragmentation, subsistence farming, inadequate fencing around orchards, scarcity of scion, transportation challenges, traditional cultivation methods, negligence in nursery management, and limited availability of disease-free saplings (Asiedu et al. 2012).

The walnut zone, Prime Minister Agriculture Modernization Project (PMAMP) of Rukum-East district, has been focusing on walnut cultivation and now has a production area of 355 ha (MoALD 2022). In Nepal, some agricultural innovation is already taking place. Enhanced agroforestry has improved livelihoods on the hillsides of Nepal as part of intricate farming systems (Pandit, 2014). According to Barrueto et al. (2018), women, poor people, and farmers without land lack the resources, risk-taking ability, land, or expertise necessary to weigh the advantages and disadvantages of this crop. Land size was one apparent determinant for whether a farmer grows trees, as stated by Alan Oli et al. (2015), and farm size influences the agricultural innovation (Aase et al. 2013). However, until now, no study or research has been conducted to find the condition of walnut-growing orchards, along with the potential and constraints regarding their establishments. In addition, there hasn't been adequate research regarding the suitability of grafted varieties of walnuts in Rukum-East. So, a study is required to assess the factors responsible for inhibiting dieback incidents in the walnut orchard, along with factors influencing the adoption of good orchard management practices. This study will sort out the various factors prevailing in the area, leading to hindrances in production. This study aims to determine the actual cause of the problems concerning walnut cultivation and management practices.

We conducted a semi-structured household survey in walnut orchards to identify factors influencing dieback and the adoption of good orchard management practices (GAPs). Quantitative data from closed questions were analyzed using statistical and logistic regression methods to determine drivers of GAP adoption. Insights from open-ended responses provided additional context on the agro-climatic suitability and growth performance of two walnut types. We developed recommendations for future cultivators, development partners, and policymakers based on these findings.

Adoption is defined as integrating new technology into existing practices, typically involving a phase of "trying" and varying degrees of acceptance (Loevinsohn et al. 2013). The two primary dimensions of adoption are adoption intensity, measuring how swiftly farmers embrace innovations, and adoption rate, indicating the extent to which a specific technology is used over time (Mwangi and Kariuki 2015). Various economic, social, and technical factors, coupled with the risk-taking mindset of farmers, significantly influence the adoption of agricultural production technologies and orchard management methods in countries like Nepal. Age is considered a factor in determining acceptance of modern technology and adoption of improved practices (Dhraief et al. 2019). One of the key factors influencing the adoption of new technology is gender. Men who head households are likelier to adopt and apply modern technologies, whereas women are likelier to use indigenous knowledge (Markovic 2021). Male-headed households are more equipped to use innovation because they have access to more knowledge than female-headed households, which are more susceptible to cultural norms and freedom (Melesse 2018). Higher education impacts respondents' attitudes and ideas, making them more open-minded, rational, and able to recognize the benefits of new technologies (Mwangi and Kariuki 2015). Larger households can relax the labor constraints necessary when introducing new technologies, which determines the adoption process Alan (Mwangi and Kariuki 2015). The size of a farm is a sign of affluence and possibly a proxy for social standing and communal influence (Zewdie 2021). A critical factor in adopting new technology is the availability and accessibility of extension services (Mwangi and Kariuki 2015). Information obtained through extension services makes a new technology's performance less uncertain, influencing how an individual embraces it.

2. Materials and Methods

2.1. Study Site

Rukum-East stands out as a prominent tourist destination in Nepal's mountainous region, celebrated for its 52 lakes and 53 peaks, covering an area of 1161.13 km^2 and housing a population of 57,962 as per the 2021 Nepal census. The region comprises three rural municipalities: Putha Uttarganga, Sisne, and Bhume. The research focused on the rural municipalities of Bhume (wards no. 2, 6, 7), Putha Uttarganga (wards no. 2, 5, 6, 10, 11), and Sisne (wards no. 2, 3, 4) due to their inclusion in the commercial walnut zone under PMAMP, Rukum-East. The selection of this study area was purposeful, considering factors such as the extent of walnut production, the number of walnut-cultivating farmers, the production of walnut seedlings and saplings, accessibility, and suitability.

The survey encompassed a diverse range of orchards, including newly established and old orchards, both registered and unregistered, and small—and large-scale orchards. It also included insights from new farmers interested in walnut cultivation. The study delved into farmers' perspectives regarding knowledge, adoption rates, and the performance of exotic walnuts within the command zone of the walnut orchard.

2.2. Research Design and Field Survey

A preliminary field visit was conducted to gather data on the research survey's socioeconomic, institutional, and geographic aspects. Following identifying issues and assessing their impact on knowledge levels and sapling growth, a thorough review of literature, methods, and methodology was undertaken to align the research topic with the study's location and objectives. Both primary and secondary data were collected. A semi-structured questionnaire was pre-tested with ten respondents from a nearby location to ensure the effectiveness of the household survey.

2.3. Sampling Procedure and Determination of Sample Size

Farmers from the study site were selected as respondents. The sample population consisted of farmers in the rural municipalities of Bhume, Putha Uttarganga, and Sisne in Rukum-East who now cultivate walnuts, were new producers, and were already familiar with the crop. The sampling frame was the list of those farmers. Simple random sampling without replacement was followed. For this, a lottery system of sampling procedures was applied. The sample size was estimated using the simplified formula given by Taro Yamane (Naing 2003).

The formula used for sample size determination:

$$n= \frac{N}{1+Ne^2}, \tag{1}$$

Where, n = Sample size

N=Population size

e = Sampling error

The yearly progress report of PMAMP, walnut zone, Rukum-East, provided all the data regarding 180 walnut growers. This sampling frame of 180 commercial walnut farmers was used to select the respondents with a sample size of 124.13 (rounding it to 120). Using Taro Yamane's formula, the sample size was determined with a 5% margin of error and a 95% confidence level. The walnut farmers were assigned numbers on individual slips of paper that were all the same size, shape, and color. They are folded and mixed up in a small box. The number of slips was chosen using a blindfold selection process to get the appropriate sample size.

2.4. Data Collection and Data Types

The data was gathered through personal interviews, where respondents were asked questions according to a structured interview schedule and checklist to obtain the necessary information. The key informant interview (KII) was carried out primarily to triangulate data and information obtained from scheduled interviews and secondarily to obtain additional qualitative information. Primary data were obtained from a questionnaire survey, focus group discussion (FGD), and field observation. Secondary data were obtained from the annual reports of PMAMP-PIU, Rukum-East, publications of horticultural centers, journal articles, and research publications.

2.5. Data analysis Technique

The collected primary data was coded, entered, and cleaned before being prepared for analysis. Various analytical tools were used for this process. Microsoft Excel (2010) and SPSS (version 25) were utilized to analyze the data. Descriptive statistics, such as frequency, percentage, and mean, were calculated to determine the distribution of study variables. Inferential statistics, including chi-square (χ^2) tests and independent sample t-tests, were applied to check for statistically significant differences. Additionally, MS Excel was used to create problem indices, pie charts, and bar diagrams.

2.5.1. Force ranking method

Forced rank scaling was used to index the problems faced by the walnut growers in Rukum-East. The ranking of the attitudes of the farmers regarding pull factors for walnut cultivation was also calculated using the same formula (Chaudhary et al. 2023). The intensity of problems and ranking of attitudes were scaled from 1 to 7, and index values of 0.14, 0.28, 0.42, 0.57, 0.71, 0.85, and 1.00 were assigned. The index was computed by the following formula:

$$Iimp = \sum (S_i * F_i/N), \quad (2)$$

Where, Iimp = Index of importance

$\sum$ = Summation S_i = I^{th} value of scale

F_i = frequency of I^{th} importance given by the respondents

N = Respondent's number

2.5.2. Binary logistic regression model

A binary logistic regression model was used to analyze the influencing factors in dieback inhibition and adoption of improved orchard management practices among walnut growers through socio-demographic attributes. Adoption of improved practices was taken as a dependent variable, and gender, age, education, ethnicity, active members, family income, occupation, and area under walnut cultivation were taken as independent or predictor variables.

Adoption of improved orchard management practices among walnut growers = f (gender, age, education, ethnicity, active working members, family income, occupation, area under walnut cultivation).

In this study, the dependent variable was the farmers' decision to adopt or not adopt orchard management practices, coded as 1 for adoption and 0 for non-adoption. The logistic model was used to predict the logit of this dependent variable based on various independent variables.

Also, influencing factors for suppression of dieback incidence in the walnut orchard = f (types of land, years of experience, time spent in walnut orchard, direction of land, distance to office, soil amendments, road access to orchard, irrigation facility, quality saplings, farmer's group, access to inputs, bordeaux paste, orchard sanitation, training and extension services, and subsidies).

The dependent variable for this study was dieback incidents in walnut orchards or not, with a value of 1 (if there is a dieback incident) and 0 (if there is no such incident). The logistic model predicts the logit of the dependent variable (dieback incident) from independent variables. The list of variables used in the model and their description are given in (Table 1) and (Table 2).

The likelihood of farmers being adopted the improved practices is predicted by odds (y=1); that is the ratio of the probability that Y equals to 1 to the probability that Y does not equal to 1 is shown below in Eq. (1):

$$Odd\ Y = P(Y=1)/ (1-P(Y=1), \quad (3)$$

The binary logistic regression model is specified as follow:

The logit (Y) is given by natural log of odds as shown below in Eq. (2):

$$In\ [p\ (Yi=1)/ (1-p\ (Yi=1))] = log\ odds = Logit\ (Y), \quad (4)$$

This can be expanded as the logistic transformation of the probability of the adoption of the practices and dieback incident suppression and is shown in Eq. (3) below:

$$\text{Logit (Y)} = \propto + \sum \beta 1X1 + \sum \beta 2X2 + \ldots + \sum \beta nXn + \varepsilon i, \quad (5)$$

Where Y= dependent variables (Adoption) with 1= adopted and 0 = not adopted;

$\propto$= intercept

εi= error index

$\beta 1, \ldots, \beta n$= coefficient of independent variables

X1… Xn = the independent variables

P (p) = probability of adopting the improved management practices in the walnut orchard

1-P = probability of not adopting the improved management practices in the walnut orchard

Ln = natural log

Table 1. Overview of the variables utilized in the binary logistic regression model to assess the adoption of improved management practices among commercial walnut growers

Variable	Type	Description	Value
Dependent variable			
Adoption of improved POPs (Y)	Dummy	Farmers has adopted the improved POPs or not	1 if adopted, 0 if not adopted
Independent variables			
Age (X_1)	Continuous	Age of the respondent	Years
Gender (X_2)	Dummy	Gender of the respondent	1 if male, 0 if female
Education (X_3)	Dummy	Education status of respondent	1 if educated, 0 if illiterate
Ethnicity (X_4)	Dummy	Ethnicity of the respondent	1 if janajati, 0 if other
Occupation (X_5)	Dummy	Occupation of the respondent	1 if agriculture, 0 if other
Active working members (X_6)	Continuous	Actively working family members of respondent	Person
Annual income (X_7)	Continuous	Annual income of respondent	Amount
Area under walnut cultivation (X_8)	Continuous	Total land size for walnut orchard	Ha

2.5.2. Independent sample t-test and chi-squar

The independent sample t-test was employed to compare the means of two unrelated groups—specifically, the growth performance and agro-climatic suitability of exotic and Jumlish walnut varieties. This test aims to determine whether there is a significant difference between the two samples. A chi-square test was also conducted to assess the significant differences in agro-climatic suitability variables between the exotic and Jumlish walnuts.

$$x^2 = \sum \frac{(Oij - Eij)^2}{Eij}, \quad (6)$$

Where,

χ^2 = chi-square,

O_{ij} = observed frequency of each ijth term,

E_{ij} = indicates the expected frequency of ijth term,

i = 1, 2, 3 . . . r, j = 1, 2, 3 . . . k.

This was evaluated at a probability level of 0.05 across various degrees of freedom.

Table 2. Description of variables used in the binary logistic regression model for possible influencing factors for inhibition of dieback incident in the walnut orchard

Variable	Type	Description	Value
Dependent variable			
Dieback incident (Y)	Dummy	Dieback incident seen or not	1 if seen, 0 if not seen
Independent variables			
Land types (X_1)	Dummy	Land types of orchard	1 if lowland, 0 if not
Years of experience (X_2)	Continuous	No. of year of experience of orchard owner	Years
Time spend in orchard (X_3)	Continuous	Average time spend in orchard per week	Hours
Slope of land (X_4)	Dummy	Types of slope of walnut orchard	1 if steep slope, 0 otherwise
Direction of land(X_5)	Dummy	Orientation of land of walnut orchard	1 if South-West, 0 otherwise
Distance to zone office (X_6)	Continuous	Average distance of orchard owner to zone office	Km
Soil amendments (X_7)	Dummy	Soil testing of orchard	1 if No, 0 otherwise
Road access to orchard (X_8)	Dummy	Access of road to walnut orchard	1 if No, 0 otherwise
Irrigation facility (X_9)	Dummy	Provision of irrigation in orchard	1 if No, 0 otherwise
Quality saplings (X_{10})	Dummy	Access to quality saplings	1 if No, 0 otherwise
Farmer's group (X_{11})	Dummy	Involvement in farmer's group	1 if No, 0 otherwise
Access to inputs (X_{12})	Dummy	Access to inputs and services	1 if No, 0 otherwise
Bordeaux paste (X_{13})	Dummy	Bordeaux paste/spray in walnut orchard	1 if No, 0 otherwise
Orchard sanitation (X_{14})	Dummy	Sanitation maintenance of orchard	1 if No, 0 otherwise
Trainings and extension services (X_{15})	Dummy	Access to trainings and extension services	1 if not received, 0 otherwise
Subsidies (X_{16})	Dummy	Subsidies received by orchard owners	1 if No, 0 otherwise

3. Results and Discussion

3.1. Different Management Practices Adopted by Orchard Owners in Rukum-East, Nepal

Among the sampled respondents, 60% of orchard owners were observed using farmyard manure as a fertilizer source, while 42.5% ensured timely irrigation for their plants. Additionally, 48.3% of farmers practiced intercropping in their orchards, 66.7% implemented mulching, and 26.7% performed pruning on time. Furthermore, 76.5% of walnut orchard owners maintained proper plant spacing, and 37.5% and 70.8% of farmers adopted disease and insect management practices. About 33.3% utilized fencing around their orchards, predominantly employing wired and stone fences. Moreover, 42.5% actively participated in bio-pesticide preparation, and 78.3% had dug pits for planting walnut saplings. About 60% of respondents engaged in weeding in their orchards, and 31.7% conducted soil amendment practices such as testing and treatments.

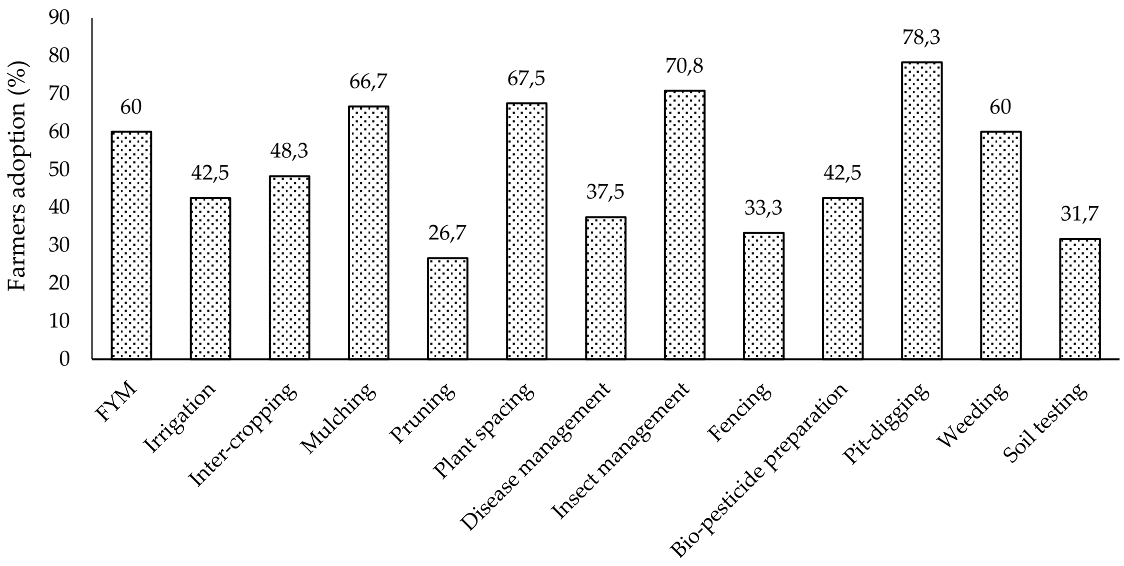

Figure 1. Different management practices adopted by orchard owners in Rukum-East (Source: Field survey, 2023)

3.2. Trainings Received by the Respondents of Rukum-East, Nepal

In the given figure, 25% of respondents from the sampled households had received training on grafting. Similarly, 5% had walnut and pruning training, 4% on bio-pesticide preparation, and 9% on disease and insect/pest management. Additionally, 13% had training on general walnut cultivation practices, 18% on nursery establishments, and 26% had not received any training.

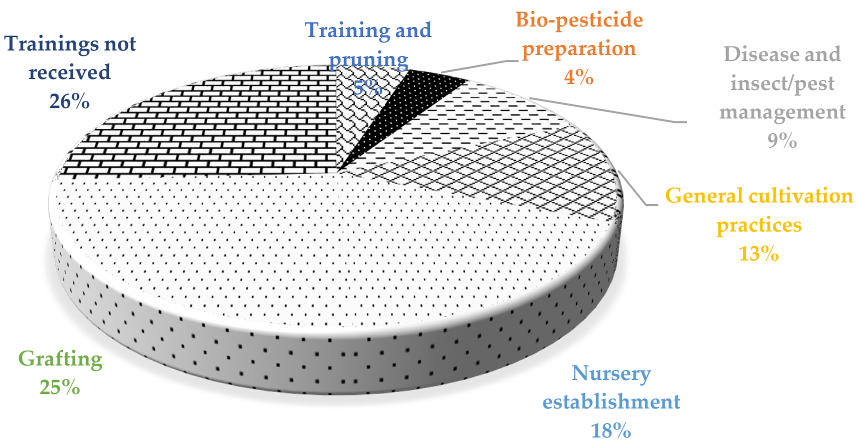

Figure 2. Different trainings received by the respondents (Source: Field survey, 2023)

3.3. Disease/pests and İnsect İncidence in the Walnut Orchard of Rukum-East, Nepal

The results in (Figure 3) showed that the highest % of dieback disease incidents, 32%, were observed in most walnut orchards in the study site, followed by root rot affecting 24% of the walnut orchards. Additionally, 21% of walnut orchards experienced trunk borer infestation, while 16% reported anthracnose incidence. Weevil incidents affected 4% of walnut orchards, and 3% did not report any disease or insect pest incidence.

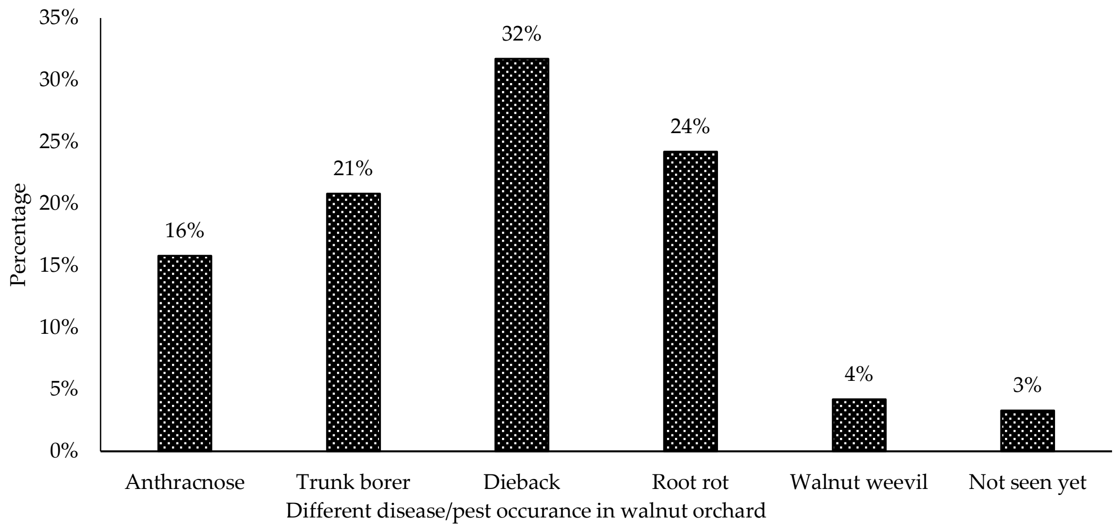

Figure 3. Disease/pests and insect incidence in the study area (Source: Field survey, 2023)

3.4. Influence of Socio-Demographic Characteristics on the Adoption of Orchard Management Practices

The study revealed that the adoption of good orchard management practices was positively influenced by education, ethnicity, and the area under walnut cultivation. Among the independent variables, annual family income had a significant but negative influence at the 5% significance level. Similarly, age was negatively influenced by adopting good orchard management practices at the 1% significance level. For a one-unit increase in gender and age, the log odds of adoption decrease by 0.15 and 0.81, respectively. Gender plays a significant role in technology adoption because the household head, typically a male, holds more access to and control over essential production resources than females, largely due to prevailing socio-cultural values and norms (Mesfin 2005; Omonona et al. 2006). Women are the primary cultivators and caretakers of the home garden, and they are more likely to collaborate with men in cultivating the property adjacent to the home garden (Gebre et al. 2019). Since men head most households in Rukum-East, the adoption of orchard management practices is negatively influenced by gender. Research has shown a negative correlation between age and technology adoption, as explained by Barrera et al. (2005) and Adesina and Zinnah (1993). This phenomenon occurs because older farmers tend to become more risk-averse and less interested in long-term farm investments. In contrast, younger farmers generally exhibit lower risk aversion and are more inclined to experiment with new technologies.

Education and the area under walnut cultivation have a positive influence and a significant effect at 1% and 5%, respectively. Therefore, for a unit increase in education and the area under walnut cultivation, the log odds ratio of adopting management practices increases by 13.16 and 1.30, respectively. The education level of a farmer is considered to have a favorable influence on the decision to adopt new technology. Farmer education enhances the ability to obtain, understand, and apply information about adopting new technology (Lavison 2013; Mignouna et al. 2011; Namara et al. 2003). This is because higher education shapes respondents' attitudes and beliefs, making them more receptive, rational, and capable of evaluating the advantages of adopting new technologies. Ahmed and Bagchi (2004) reported a positive interaction between farm size and agricultural technology adoption. Farmers with larger farm sizes are more likely to accept new technology because they can afford to devote a portion of their land to experimenting with the latest technologies, unlike farmers with smaller farm sizes (Uaiene 2011).

The model's chi-square (χ^2) value of 34.43 and the log-likelihood ratio of -23.945721 suggest that all the variables included in the model have a significant impact on the likelihood of adopting orchard management practices for walnuts. The pseudo-R^2 value of 0.4183 means that about 41.83% of the decision to adopt good orchard management practices is governed by the tabulated variables, i.e., the model fits 41.83% of the given data.

Table 3. Influence of socio-demographic characteristics on the adoption of orchard management practices

Socio-demographic characters	Coefficient	Odds ratio	Std. Err.	Z	p>lzl
Gender (Female=1)	-1.87	0.15	0.12	-2.27	0.02**
Age (Years)	-0.20	0.81	0.60	-2.75	0.006***
Education (Yes=1)	2.57	13.16	12.61	2.69	0.007***
Ethnicity (Janajati=1)	0.09	1.09	0.84	0.12	0.90
Annual income	-0.61	0.54	0.58	-0.57	0.56
Occupation (Agriculture=1)	-0.78	0.45	0.56	-0.63	0.52
Active members	-0.22	0.80	0.161	-1.10	0.27
Area under walnut cultivation (Ha)	0.26	1.30	0.163	2.08	0.03**
Constant	19.63	4.00	13.52	1.45	0.14
Summary statistics					
Number of observations	120				
LR chi^2 (10)	34.43				
Prob>chi^2	0.0000				
Pseudo R^2	0.4183				
Log likelihood	-23.945721				

Note: The symbols *, **, and *** represent significance levels of 10%, 5%, and 1%, respectively.

3.5. Mean Comparison for the Agro-Climatic Suitability and Growth Performance among two Walnut Types in Rukum-East, Nepal

The independent sample t-test was employed to compare two sample means from unrelated groups, specifically focusing on the growth performance and agro-climatic suitability of exotic and Jumlish walnut varieties. This test aimed to discern any differences between the two samples. According to the results presented in (Table 4), the height of both plants, survival percentage, number of branches, and fruiting years exhibited statistical significance at the 1% level. Additionally, for the categorical variables, such as tolerance to diseases and pests, as well as the ability to cope with abiotic stress, a high level of significance at the 1% level was observed. These findings signify that the mean differences between the two varieties in various growth parameters are highly significant. Notably, Jumlish walnut demonstrated superior adaptability compared to various exotic walnut varieties. This implies that Jumlish walnut outperformed exotic varieties regarding key growth parameters. The statistical significance underscores the robustness of these differences, indicating a noteworthy advantage for Jumlish walnut in terms of height, survival percentage, number of branches, fruiting years, and resilience to diseases, pests, and abiotic stress.

3.6. Influencing Factors for Dieback Incident among Walnut Orchard Owners of Rukum-East, Nepal

Various factors, such as the orientation of the land, motor road accessibility, subsidies, and management practices like time spent in orchards, irrigation facilities, and the application of Bordeaux paste, influence the occurrence of dieback in walnut orchards in Rukum-East. The results of a binary logistic regression model, as presented in (Table 5), elucidate the impact of these independent variables on dieback incidence. Sixteen independent variables were utilized for the regression analysis, and six were deemed statistically significant among them. The time dedicated to orchard management exhibited a significant effect at the 5% significance level. Additionally, the orientation of the land, road access to the orchard, availability of irrigation facilities, application of Bordeaux paste, and the presence of subsidies were found to have significant effects at the 5%, 1%, 5%, 5%, and 10% levels of significance, respectively.

The findings suggest that for each additional unit of time spent in the orchard, there is a corresponding 2% reduction in the likelihood of experiencing dieback incidents. The log odds of an increase in dieback incidents decrease by 1.46. This could be attributed to higher adoption and awareness of management practices aimed at mitigating dieback. Notably, ignorance of orchard management and poor practices strongly correlates with a decline in orchard productivity (Poudel et al., 2022). The probability of experiencing a dieback incident decreases by 18.7% with a one-unit increase in the direction of land facing northeast. The log odds of a reduction in dieback incidence increase by 0.20. Walnuts thrive in well-drained, fertile soils on lower north- and east-facing slopes. They do not fare well on steep slopes facing south or west due to the excessive heat and dry conditions. Midday water potentials were similarly higher on south-facing slopes, but plant frequency and groundcover were more abundant on north-facing slopes (Ulrich et al. 2008). A similar outcome was observed by Poudel et al. (2022) in a citrus orchard, where they noted that south and east-facing slopes receive more sunlight than north-facing slopes, leading to increased moisture loss and hindered growth.

Table 4. Independent sample t-test for mean comparison between two walnut types

Continuous variables	**Mean value**	**S.D**		**S.E**	**t-value**	**p-value**
Height of Plant						
• Jumla	97.21	31.67		4.23	5.574***	0.0000
• Exotic	61.78	35.37		4.42		
Survival % of plant						
• Jumla	75	17.74		2.37	6.936***	0.0000
• Exotic	49.37	22.10		2.76		
No. of branches						
• Jumla	3.01	0.94		0.126	6.671***	0.0000
• Exotic	1.82	1.00		0.125		
Fruiting year						
• Jumla	0.821	0.38		0.051	6.198***	0.0000
• Exotic	0.328	0.47		0.059		
Categorical variables	**Overall (120)**	**Farmers category**		**Chi-square value**	**p-value**	
Tolerant to Pest/Diseases		Yes	No			
• Jumla	56	51	5	62.642***	0.000	
• Exotic	64	12	52			
Ability to cope abiotic stress						
• Jumla	56	50	6	67.619***	0.000	
• Exotic	64	9	55			

Note: The symbols *, **, and *** represent significance levels of 10%, 5%, and 1%, respectively.

Through the marginal effect after logistic regression, it is predicted that with a one-unit increase in road access to orchards and irrigation facilities, the probability of experiencing dieback incidents decreases by 2.3% and 2.1%, respectively. Similarly, the log odds of suppressing dieback incidence increase by 0.14 and 0.16. The absence of irrigation and erratic rainfall can adversely impact orchard yield and production, creating opportunities for various diseases and pests (Dorji et al. 2016). Irrigation is vital in promoting more significant root growth, canopy development, fruit yield, and quality, making it a crucial tool in the battle against diseases and pests (Levy 1998). Similarly, easy road access to orchards enables farmers to efficiently implement improved management practices against diseases and pests in a shorter time. Infrastructure enhancements, including roads, canals, and transportation systems, should be prioritized to mitigate orchard losses (Hussen and Yimer 2013). Dieback incidents were negatively influenced by the application of Bordeaux paste at the 5% significance level. The results demonstrate that with a one-unit increase in the application of Bordeaux paste, the probability of experiencing a dieback incident decreases by 8.7%. The log odds of dieback incidence decrease by 0.48. Applying the Bordeaux mixture significantly and positively impacts disease suppression and orchard productivity. The components of the Bordeaux mixture act as fungicides and bactericides, effectively managing various walnut diseases such as stem cankers and dieback (Sharma et al., 2012).

In this context, the provision of subsidies negatively influences the incidence of dieback. According to the marginal effect after logistic regression, it is predicted that subsidies for quality saplings are 0.2% less likely to contribute to dieback incidents in walnut orchards, a significance observed at the 10% level. Adopting recently released agricultural technologies is promoted through subsidies (Kattel et al. 2020; Subedi et al. 2019). Subsidies on products and services enable farmers to acquire quality-grafted saplings at more affordable rates, ultimately reducing the likelihood of dieback incidents in their orchards.

The model's chi-square (χ^2) value of 28.86, along with a log-likelihood ratio of -45.61975, suggests that all variables included in the model significantly affect the likelihood of dieback occurrence. The pseudo-R^2 value of 0.2403 means that about 24.03% of dieback incident decisions are governed by the tabulated variables, i.e., the model fits 24.03% of the given data.

Table 5. Logistic regression analysis and marginal effect after logistic analysis of dieback incident with different factors causing incident of dieback

Dieback Score	Odds ratio	Std. Err.	Z	p>lzl	dy/dx	p>lzl
Land types	3.44	3.94	1.08	0.27	1.48	0.27
Years of experience	0.59	0.20	-1.49	0.13	-0.062	0.12
Time spend in orchard	1.46	0.26	2.14	0.03**	0.046	0.02**
Slope of land	1.72	1.43	0.65	0.51	0.065	0.51
Direction of Land	0.20	0.13	-2.51	0.01**	-0.187	0.006**
Distance to office	0.99	0.020	-1.38	0.70	-0.000	0.70
Soil amendments	1.00	0.89	0.01	0.99	0.001	0.99
Road access to orchard	0.14	0.10	-2.80	0.00***	-0.230	0.00***
Irrigation facility	0.16	0.14	-2.10	0.03**	-0.215	0.02**
Quality saplings	3.22	2.38	1.58	0.11	0.140	0.10
Farmers group	0.59	0.55	-0.56	0.57	-0.619	0.57
Access to inputs	0.76	0.76	-0.27	0.78	-0.032	0.78
Bordeaux paste	0.48	0.51	-0.68	0.05**	-0.087	0.49
Orchard sanitation	1.70	1.45	0.63	0.53	0.064	0.52
Training	1.45	1.38	0.39	0.69	0.045	0.69
Subsidies	0.98	0.01	-1.74	0.08*	-0.002	0.07*
Constant	47.51	130.74	1.40	0.16	-	-
Summary statistics						
Number of observations	120					
LR chi^2 (14)	28.86					
Prob>chi^2	0.0249					
Pseudo R^2	0.2403					
Log likelihood	-45.61975					

Note: The symbols *, **, and *** represent significance levels of 10%, 5%, and 1%, respectively.

3.7. Ranking of the Major Constraints Faced by the Walnut Growers in Rukum-East, Nepal

According to reports from farmers, the successful establishment of walnut orchards faces several constraints. When ranked using an indexing technique, the high mortality of imported walnuts emerged as the primary constraint, recognized by the majority of respondents. The index value for this issue was the highest (0.75), making it the most serious challenge in the research site, as depicted in (Table 6) below. Irrigation facilities and shortages of storage tanks were ranked second with an index value of 0.63, followed by fencing constraints with an index value of 0.499, placing them in third place. Constraints such as fencing (0.499), nurseries and quality-grafted scions (0.4932), effective and practical training (0.497), insect/pest and disease severity (0.479), and high labor costs (0.43) were ranked 3rd, 4th, 5th, 6th, and 7th, respectively.

3.8. Ranking of the Major Attraction Factors by the Walnut Growers in Rukum-East, Nepal

Among the sampled respondents, the primary attraction for walnut cultivation was marginal land utilization, followed by agro-climatic suitability for successful orchard establishment. The index value for marginal land utilization was the highest (0.77) and ranked as the most appealing aspect in the research site, as shown in (Table 7) below. As reported by the farmers, the second most important factor for establishing a walnut orchard is the agro-climatic suitability of the region, with an index value of 0.62. Government support and prioritization are ranked third, with an index of 0.59. The unsuitability of other crops is ranked fourth with an index value of 0.54. Similarly, storage possibilities, marketing, and lower risks for final production rank 5th and 6th, respectively.

Table 6. Ranking of the major problems faced by the walnut growers in the study area

Constraints of walnut production	Index	Ranking
High mortality of exotic walnut varieties	0.75	I
Irrigation facility and shortage of storage tanks	0.63	II
Fencing	0.499	III
Nurseries and quality-grafted scions	0.492	V
Effective and practical trainings	0.497	IV
Insect/Pest and disease severity	0.479	VI
High labor cost	0.43	VII

Source: Field survey, (2023)

Table 7. Ranking of the attitudes of the major attraction factors for the walnut growers in the study area

Attraction factor for walnut production	Index	Ranking
Marginal land utilization	0.77	I
Agro-climatic suitability	0.62	II
Storage possibilities & marketing	0.50	V
Government support & prioritization	0.59	III
Unsuitability of other crops	0.54	IV
Less risk for final production	0.43	VI

Source: Field survey, (2023)

4. Limitations

While the study's findings may assist dispersed farmers nationwide, the ability to generalize about the overall agricultural situation in the country is hindered by its narrow focus on a single region. The study's limitations, including a restricted period and financial constraints, resulted in limited information acquisition. Despite these constraints, the study remains a valuable resource for future research academics. This study will examine the various factors prevailing in the area that lead to hindrances in production. This study aims to determine the actual cause of the problems concerning walnut cultivation and management practices.

5. Conclusion and Policy Recommendations

5.1. Conclusion

Walnut cultivation is highly income-generating if orchards are appropriately managed. The insights from this study extend beyond Nepal and are highly relevant to other mountainous and subtropical regions where walnuts are cultivated. Farmers face similar challenges in adopting sustainable orchard management practices in many walnut-growing areas, such as parts of India, Turkey, Iran, and Central Asia. Factors like gender, age, education, landholding size, and access to essential resources such as irrigation, roads, and subsidies play a key role in shaping their decisions. Recognizing these influences is crucial for developing extension programs that support farmers in improving productivity, maintaining quality standards, and overcoming trade barriers.

Furthermore, the study emphasizes the need for climate-resilient, high-yielding walnut varieties and sustainable farming techniques to ensure long-term orchard health. Common issues, including limited time spent in orchards, improper land orientation, and the prevalence of dieback due to poor management, affect walnut production globally. By addressing these challenges and tailoring extension programs to local conditions, policymakers and agricultural experts can promote best practices that enhance the sustainability and resilience of walnut farming across diverse agro-climatic regions.

5.2. Policy Recommendation

The major problems like dieback and root rot disease, and trunk borer should be prioritized by the farmers. The government and the concerned projects should focus on improving knowledge of cultivation techniques, training on several varieties and their performance in their region, post-harvest aspects, and marketing the soft-shelled walnuts. Further studies on economic efficiency, technical efficiency, and value-chain analysis of the farmers adopting improved management practices than non-adopters can be recommended. The government should motivate growers by promoting certified planting materials that are climate-suited and high-yielding and training in managing the farm. The growth performance of exotic walnuts should be further verified at different altitude levels, especially at higher altitudes, before mass distribution to farmers. Adaptation and the production potential of Jumlish walnuts should be checked and verified.

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References

Adesina AA, Zinnah MM (1993). Technology characteristics, farmers' perceptions and adoption decisions: A Tobit model application in Sierra Leone. *Agricultural Economics, 9*(4), 297–311.

Ahmed S, Bagchi KK (2004). Factors and constraints for adopting new agricultural technology in Assam with special reference to Nalbari district: An empirical study. *Journal of Contemporary Indian Policy, 35*, 60–68.

Aryal K, Berg Å, Ogle B (2009). Uncultivated plants and livelihood support–a case study from the Chepang people of Nepal. *Ethnobotany Research and Applications, 7*, 409–422.

Asiedu JBK, Owusu-Sekyere JD, Taah KJ, Van Der Puije GC, Ocloo E (2012). The nursery industry in Ghana: Prospects and challenges. *ARPN Journal of Agricultural and Biological Science, 7*(6), 443–453.

Barrera V, Norton GW, Alwang JR, Mauceri M (2005). Adoption of integrated pest management technologies: A case study of potato farmers in Carchi, Ecuador (No. 378-2016-21506).

Bernard A, Lheureux F, Dirlewanger E (2018). Walnut: Past and future of genetic improvement. *Tree Genetics & Genomes, 14*(1), 1.

Papademetriou MK, Herath EM (1999). Deciduous fruit production in Asia and the Pacific. RAP Publication, Bangkok, Thailand, p:37.

Dhakal LP, Shrestha KR, Shrestha KB, Lilleso JPB (2003). Tree planting zones for the benefits to the small holders in Nepal. *World Agroforestry Centre,* 1–18.

Dhraief MZ, Bedhiaf S, Dhehibi B, Oueslati-Zlaoui M, Jebali O, Ben-Youssef S (2019). Factors affecting innovative technologies adoption by livestock holders in arid area of Tunisia. *New Medit: Mediterranean Journal of Economics, Agriculture and Environment= Revue Méditerranéenne D'Economie Agriculture et Environment*, *4*.

Dorji K, Lakey L, Chophel S, Dorji SD, Tamang B (2016). Adoption of improved citrus orchard management practices: A micro study from Drujegang growers, Dagana, Bhutan. *Agriculture & Food Security, 5*, 1–8.

FAOSTAT (2020). UN Food and Agriculture Organization, Corporate Statistical Database. https://www.fao.org/faostat/en/#data/QCL/visualize (access date: 02.05.2024).

Gebre GG, Isoda H, Amekawa Y, Nomura H (2019). Gender differences in the adoption of agricultural technology: The case of improved maize varieties in southern Ethiopia. In *Women's studies international forum* (Vol. 76, p. 102264). Pergamon.

Hussen S, Yimer Z (2013). Assessment of production potentials and constraints of mango (Mangifera indica) at Bati, Oromia zone, Ethiopia. *International Journal of Sciences: Basic and Applied Research*, *11*(1), 1–9.

Jahanban-Esfahlan A, Ostadrahimi A, Tabibiazar M, Amarowicz R (2019). A comprehensive review on the chemical constituents and functional uses of walnut (Juglans spp.) husk. *International Journal of Molecular Sciences, 20*(16), 3920.

Kattel RR, Regmi PP, Sharma MD, Thapa YB (2020). Factors influencing adoption of major post-harvest handling practices of large cardamom in Nepal. *Cogent Food & Agriculture, 6*(1), 1796201.

Lavison R (2013). Factors influencing the adoption of organic fertilizers in vegetable production in Accra. Master's Thesis, Accra (Unpublished), Ghana.

Levy Y (1998). Citrus irrigation. *Nouveaux Acquis de La Recherche En Agrumiculture. Inst. Agronomique Veterinaire Hassan II, Agadir, Morocco*, 113–126.

Loevinsohn M, Sumberg J, Diagne A, Whitfield S (2013). Under what circumstances and conditions does adoption of technology result in increased agricultural productivity? A Systematic Review. Institute of Development Studies, Brighton, UK. p. 31.

Melesse B (2018). A review on factors affecting adoption of agricultural new technologies in Ethiopia. *Journal of Agricultural Science and Food Research*, *9*(3), 1–4.

Mesfin A (2005). Analysis of factors influencing adoption of triticale and its Impact. PhD Thesis, Alemaya University (Unpublished), Ethiopia.

Mignouna DB, Manyong VM, Rusike J, Mutabazi KDS, Senkondo EM (2011). Determinants of adopting imazapyr-resistant maize technologies and its impact on household income in Western Kenya. AgBioForum, 14(3), 158-163.

Mir M, Kumar A (2011). Effect of different methods, time and environmental conditions on grafting in walnut. *International Journal of Farm Sciences*, *1*(2), 17–22.

MoALD (2021). Statistical information on Nepalese agriculture. Ministry of Agriculture, Livestock and Development. https://moald.gov.np/publication-types/agriculture-statistics/ (access date: 02.05.2024)

MOALD (2022). *Agriculture Statistics | Publication Category | Ministry of Agriculture and Livestock Development*. https://moald.gov.np/publication-types/agriculture-statistics/ (access date: 02.05.2024)

Mwangi M, Kariuki S (2015). Factors determining adoption of new agricultural technology by smallholder farmers in developing countries. *Journal of Economics and Sustainable Development*, *6*(5), 208-216.

Naing NN (2003). Determination of sample size. *The Malaysian Journal of Medical Sciences: MJMS*, *10*(2), 84.

Namara RE, Weligamage P, Barker R (2003). Prospects for adopting system of rice intensification in Sri Lanka: A socioeconomic assessment (Vol. 75). IWMI.

NCFD (2020). Publications | National Centre for Fruit Development (NCFD) | Kirtipur, Kathmandu. https://ncfd.gov.np/en-publications (access date: 02.05.2024)

Omonona BT, Oni OA, Uwagboe AO (2006). Adoption of improved cassava varieties and its welfare impact on rural farming households in Edo State, Nigeria. *Journal of Agricultural & Food Information*, *7*(1), 39–55.

Poudel A, Sapkota S, Pandey N, Oli D, Regmi R (2022). Causes of citrus decline and its management practices adopted in Myagdi district, Nepal. *Heliyon*, *8*(7), e09906.

Radovic Markovic M (2021). Gender and technology adoption among farmers in Bangladesh. *International Review*, 3-4, 12-28.

Sharma RM, Pandey MK, Shankar U (2012). Pest management in walnut: An overview. *Ecologically Based Integrated Pest Management*, New India Publishing Agency, New Delhi, India. pp. 765-785.

Subedi S, Ghimire YN, Adhikari SP, Devkota D, Poudel HK, Sapkota BK (2019). Adoption of improved wheat varieties in eastern and western Terai of Nepal. *Journal of Agriculture and Natural Resources*, *2*(1), 85–94.

Thakur RB (2003). *A compendium of tree species of Nepal*. Mr. & Mrs. RB Thakur. p.194.

Thapa MB, Dhimal S (2017). Horticulture development in Nepal: Prospects, challenges and strategies. *Universal Journal of Agricultural Research*, *5*(3), 177–189.

Thapa R, Thapa P, Ahamad K, Vahdati K (2021). Effect of grafting methods and dates on the graft take rate of Persian Walnut in open field condition of Rukum (East), Nepal. *Methods*, *544*, 503–509.

Uaiene RN (2011). Determinants of agricultural technology adoption in Mozambique. *10th African Crop Science Conference Proceedings, Maputo, Mozambique, 10-13 October 2011*.

Ulrich BN, Roth AJ, Bobich EG (2008). PS 30-152: Influences of microclimate on water relations and phenology of southern California black walnut (*Juglans californica*). *The 93rd ESA Annual Meeting*.

Zewdie T (2021). Review on factors affecting adoption of improved maize seed technology in Ethiopia. *International Journal of Agriculture Extension and Social Development*, *1*(2), 62-68. https://www.researchgate.net/publication/352933562

Zhang YY, Ni ZJ, Elam E, Zhang F, Thakur K, Wang S, Zhang JG, Wei ZJ (2021). Juglone, a novel activator of ferroptosis, induces cell death in endometrial carcinoma Ishikawa cells. *Food & Function*, *12*(11), 4947–4959.

APPENDICES

Appendix 1. Number of years of experience of commercial walnut orchard growers in Rukum-East

Years of experience of walnut growing farmers	Frequency
1 year	30(25)
2 years	42(35)
3 years	40(33.3)
More than 4 years	8(6.7)
Total	120(100)

Figure in the parentheses indicates the percentage

Appendix 2. Different age groups of the respondents on the study site

Age groups (Years)	Frequency
Below 30 (15-29 years old)	27(22.5)
Between 30-60 (30-60 years old)	75(62.5)
Above 60 (61-75 years old)	18(15)
Total	120(100)

Figure in the parentheses indicates the percentage

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Potential Use of Aloe Vera (*Aloe barbadensis* Miller) Leaf Gel in the Development of Functional Ice Cream: Physicochemical, Bioactive, Thermal, Rheological, and Sensory Properties

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HIGHLIGHTS

- Aloe vera leaf gel was successfully incorporated into the ice cream formulation.
- Ice cream samples containing aloe vera leaf gel had lower viscosity compared to control sample.
- The addition of aloe vera leaf gel into the ice cream formulation, with or without an emulsifier, resulted in an increase in both dry matter and total phenolic content.
- Aloe vera leaf gel can be incorporated into ice cream formulations without compromising their sensory characteristics.

Abstract

Aloe vera leaf contains a transparent mucilaginous gel possessing notable chemical, pharmacological, biological, and therapeutic activities. The unique structure and colloidal composition of ice cream make it a highly suitable medium for the efficient delivery of bioactive compounds, as well as its ability to be stored at low temperatures. This study investigated the effects of different proportions of aloe vera leaf gel (AVLG, 1 g/100 g and 2 g/100 g) on the physicochemical, rheological, total phenolic content (TPC), thermal, and sensory characteristics of ice creams, formulated with and without an emulsifier, in comparison to reference samples. The findings revealed that the pH values of the samples were nearly neutral, ranging from 6.55 to 6.61. The incorporation of AVLG, with or without an emulsifier, increased both dry matter content from 33.09% (control) to 40.57% (A2E2), and TPC values, from 0.073 (control) to 0.099 mg GAE/g (A1E2). Additionally, the AVLG supplementation played a significant role in altering the color features of the enriched samples compared to the reference sample, with *L** values ranging from 82.37 to 86.01, *a** values from -2.25 to -2.86, *b** values from 1.96 to 4.97, and *ΔE** values from 3.19 to 4.73. The thermal analysis indicated that enriching the samples with AVLG, with or without an

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emulsifier, reduced the ΔH_m from 160.80 to 53.60 J g^{-1}. The steady-state shear data of ice cream mixtures, which exhibited pseudoplastic flow characteristics, were accurately described by the Ostwald de Waele model (R^2>0.9974), with ice cream consistency coefficient values of 0.25-0.68 Pa.s, flow behavior index values of 0.50-0.80, and apparent viscosity values of 0.0980-0.1540 Pa.s. The sensory evaluation results showed that there were no statistically notable differences in sensory perception among the evaluated samples. Overall, the findings highlighted that AVLG can serve as a functional ingredient and natural colorant in ice cream production, offering various advantages.

Keywords: Functional ice cream; Aloe vera leaf gel; Physicochemical properties; Thermal properties; Rheological properties

1. Introduction

Consumers seek a well-rounded diet that not only satisfies their hunger but also offers disease-preventive benefits (Kutlu and Erol 2025). In order to meet health-conscious consumers expectations, food producers seek for incorporation of functional ingredients into their food products (Goraya and Bajwa, 2015). Ice cream and frozen milk-based desserts are widely regarded as delightful and popular treats, appreciated for their sweetness, creamy texture, rich flavors, and visual appeal, offering a combination of indulgence and nutritional value ice cream (Khider et al. 2021). Due to the growing interest in foods that improve human nutrition, market proportion and consumer satisfaction, academia and the food industry feel the need for production of functional and novel ice creams (Genovese et al. 2022).

Aloe vera (also called Gheegwar or Ghritkumari) scientifically belongs to a number of species including *Aloe vera* L., *Aloe rubescens, Aloe elongate, Aloe barbadensis* in *Aloe officinalis* in *Aloeaceae* family (Ahlawat et al. 2011; Sonawane et al., 2021). The term "aloe" comes from the Arabic word "alloeh" meaning "bitter". It is popular for its therapeutic and functional properties and is often referred to as a miracle plant (Sonawane et al. 2021). This miraculous plant can withstand extremely harsh conditions, even in very hot and dry climates, where most other vegetation is destroyed due to the low water-holding capacity of its leaves. Removing the green epidermis of a leaf reveals a transparent and mucilaginous substance known as a gel (Olaleye et al. 2015). Although the majority of the content of aloe vera is water, it has been determined that there are more than 200 chemicals in its dry matter. Moreover, the dry matter of aloe vera is rich in carbohydrate (~60%), followed by protein (6-8%) and lipids (2-5%), of which proportions can vary depending on the leaf parts (Rodríguez et al. 2010). This unique leaf gel is a low molecular weight glucomannan consisting of mainly mannose and glucose (Reynolds and Dweck 1999; Comas-Serra et al. 2023). The composition, chemical and biological activities of AVLG can vary depending on several factors including gel extraction techniques, geographical location, and sample preparation methods (Hamman 2008).

Globally, the AVLG market expands quickly, reaching a value of 649.41 million USD in 2020. The food (15%), cosmetics (24%), and pharmaceutical industry (61%), are the main industries propelling this rise. AVLG is used in the food business to produce functional foods, as a natural preservative, and as a component of coatings and edible films (Maan et al. 2021). In earlier papers, aloe vera was incorporated into the various food formulations such as ice cream (Srisukh et al. 2008; Manoharan et al. 2012; Manoharan and Ramasamy 2013a,b,c), juices (Alemdar and Agaoglu 2009), candies (Shibinshad 2023), chewing gum (Aslani et al. 2015), as well as edible coating (Misir et al. 2014). Corresponding to the ice cream studies, different formulations were developed using AVLG. For instance, Srisukh et al. (2008) manufactured ice cream with AVLG and specified their organoleptic properties. As well, Manoharan et al. (2012) studied the effects of the level of aloe vera pulp addition on sensory characteristics. Furthermore, Manoharan and Ramasamy (2013a), Manoharan and Ramasamy (2013b), and Manoharan and Ramasamy (2013c) prepared aloe vera (pulp) ice cream formulated with both artificial sweeteners (with low-calorie) and natural colorant and evaluated their physicochemical, microbial and sensory properties. Moreover, Verma et al. (2018) prepared different ice cream samples containing different proportions of mint extract and aloe vera juice and investigated their physicochemical, microbiological and sensory characteristics. In addition, Chathurangi et al. (2018) supplemented ice cream with aloe vera-gel and analyzed their physical, sensory, bioactive, and anti-inflammatory properties. Additionally, Mule et al. (2020) produced probiotic (*L. acidophilus*) ice cream samples formulated with different ratios of ginger and aloe vera juices and determined the cost calculation as well as physicochemical and sensory characteristics. However, to the best of our knowledge, no study has been conducted to evaluate the color, thermal and rheological properties of aloe vera gel-incorporated ice creams. Additionally, we explore the interaction between AVLG and emulsifiers, which has not been extensively addressed in previous literature studies. Overall, considering the growing demand for highly consumed natural dairy products with natural ingredients and health-promoting properties, the goals of current study were (i) to develop different functional ice cream formulations using different combinations of aloe vera leaf gel and emulsifier (ii) and to specify and characterize their physicochemical, total phenolic content, rheological, thermal, and sensory properties. By examining these factors collectively, our study offers a broader and more integrated understanding of AVLG's role in ice cream formulation, highlighting its potential as a functional ingredient for enhancing the product quality and health benefits.

2. Materials and Methods

2.1. Materials

Sugar (Ismen Food Company, Istanbul, Türkiye), pasteurized milk cream (35% milkfat; Mis, Ak Gida Company, Türkiye), and pasteurized cow's milk (3.1% milkfat; Mis, Ak Gida Company, Türkiye) were supplied from the local markets in Istanbul, Türkiye. Pure salep was purchased from a local seller of medicinal herbs (Aktar Diyari, Istanbul, Türkiye). Moreover, plant-based ice cream emulsifier (glycerol monostearate, Tito BUZ610, Izmir, Türkiye), Folin–Ciocalteu phenol reagent (Sigma-Aldrich, Steinheim, Germany), sodium carbonate (Na_2CO_3) (Sigma-Aldrich, Germany), gallic acid (Sigma-Aldrich, Germany), were used. Fresh aloe vera leaf (*Aloe barbadensis* Miller) were collected in September in Istanbul, Türkiye (Longitude: 28° 51' 13.878'' E, Latitude: 41° 4' 45.8868'' N).

2.2. Preparation of Aloe Vera Leaf Gel (AVLG)

AVLG preparation was conducted based on the former protocol of Munoz et al. (2015) with minor modifications. For this purpose, leaf samples were washed and cleaned with distilled water. Then, the inner part of the aloe vera leaf was successfully separated from its outer leaf pulp and green rind of cuticle with the aid of a sharp metal knife. The gel (AVLG) was separated and ground by a kitchen blender, then stored in falcon tubes at -18 ºC until use (Figure 1).

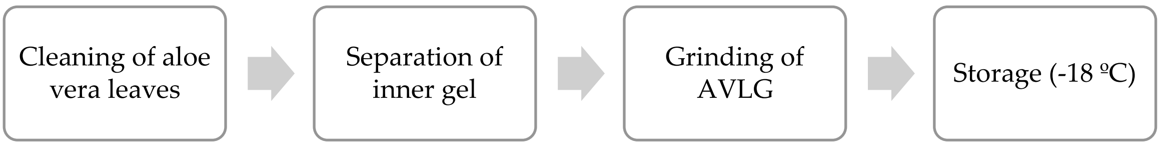

Figure 1. Flowchart for the preparation of aloe vera leaf gel (AVLG).

2.3. Ice Cream Preparation Process

In this study, four different ice cream mixes were made using the procedure flow chart shown in Figure 2. After cooling to 40 °C, each prepared sample was put in the refrigerator and kept for 24 h at 4 °C. The samples were coded as follows: A1E2 (ice cream mix with 1 g/100 g AVLG and 2 g emulsifier), A2E2 (ice cream mix with 2 g/100 g AVLG and 2 g emulsifier), A2 (ice cream mix with 2 g/100 g AVLG), and the control sample (containing 0 g/100 g AVLG and 2 g emulsifier). After that, the ice cream samples were produced in a Delonghi, II Gelataio, ICK5000, China ice cream machine for 25 min at a steady rotation speed.

2.4. Physicochemical Analysis

pH measurement was performed by immersing the electrode of a pH meter (Mettler Toledo™ S220 SevenCompact pH/Ion Benchtop Meter) standardized with buffer solutions into the melted ice cream samples (Karaman et al. 2014). Determination of dry matter content was performed by drying of a ~ 3 g ice cream sample in a hot air oven (Memmert UF-110, Germany) at 105 °C for 4 h, by following the AOAC 2000 standard method. L^* (0, white and 100, black), a^* ($-a^*$, green coordinate and $+a^*$, red coordinate), and b^* values ($-b^*$, blue coordinate and +*b** yellow coordinate) of melted ice creams were measured using a colorimeter (CR-400, Minolta Camera Co., Osaka, Japan) and then the *ΔE** (visual color differences) were calculated following the instructions outlined by Atlar et al. (2023).

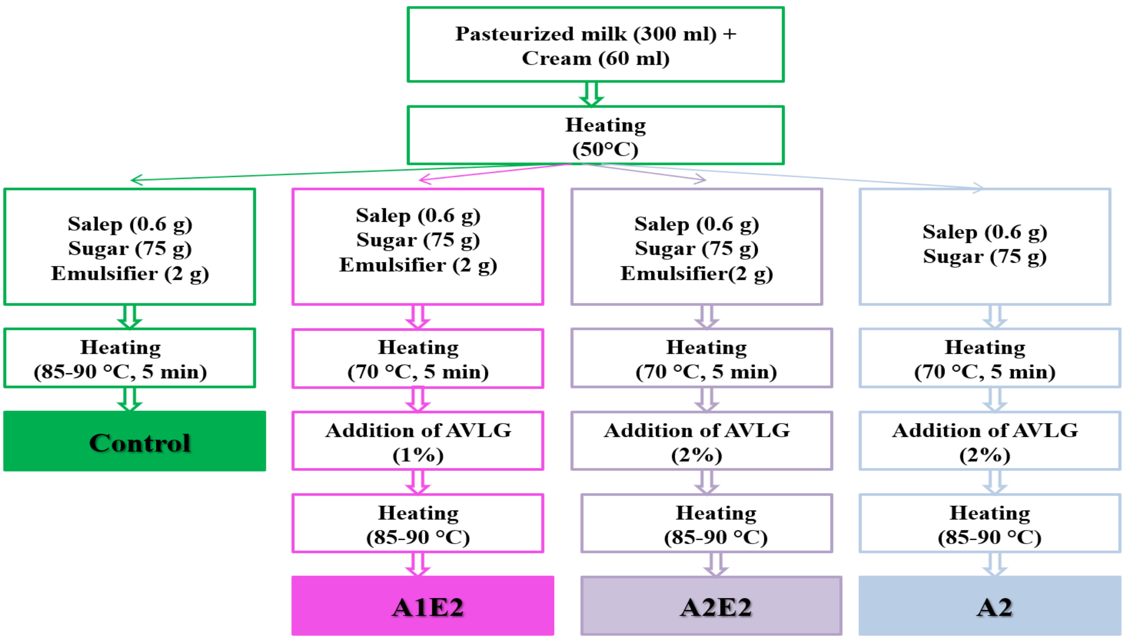

Figure 2. Process flowchart for the production of ice cream mixes coded with control, A1E2, A2E2 and A2.

* AVLG: Aleo vera leaf gel.

2.5. Bioactive Properties

2.5.1. Preparation of ice cream extract for TPC analysis

The ice cream extract for TPC analysis was prepared according to the protocol previously reported by Karaman and Kayacier (2012) with some modifications. Firstly, each sample (10 g) was combined with 5 mL of hexane and 50 mL of 80% methanol in a 100 mL glass bottle and then vigorously mixed. Afterwards, each sample was kept in dark at ambient conditions for 24 h and then centrifuged at 9000 rpm for 10 min at 4 °C. Subsequently, the oil layer was collected with the aid of a plastic syringe (Steriject, with a 0.80x38 mm diameter needle) and the centrifugation was performed under the same conditions. This procedure repeats the respective steps (the collection of oil layer and centrifugation) until complete separation of the oil was achieved and subsequently, it was filtered by using coarse filter paper.

2.5.2. Total phenolic content (TPC)

Each sample (0.5 mL) was taken into the glass test tubes, and then 2.5 mL of 10% Folin-Ciocalteau solution was added at 10-sec intervals, followed by a 3-min incubation period. Subsequently, 2 mL of 7.5% Na_2CO_3 solution was pipetted into this mixture. Then, the mixture was allowed to stand for 30 min at room temperature in dark. Then, a UV-visible spectrophotometer (Shimadzu, UV-1800, Japan) was used to measure the absorbance values of the extracts at a wavelength of 760 nm. The resulting values were expressed as milligram gallic acid equivalent per gram (mg GAE g^{-1}) (Erol et al. 2023; Kutlu 2024; Demirkan et al. 2024).

2.6. Thermal Properties by DSC

The thermal characteristics of the ice cream samples were analyzed using a differential scanning calorimeter (DSC, Q100, TA Instruments Inc., New Castle, DE, USA) from -20 °C to 25 °C at a heating rate of 10 °C min^{-1}, following the procedure described by Akman et al. (2023). For this purpose, 10 mg of the ice cream mix or AVLG was carefully placed in hermetically sealed aluminum pans and loaded into the instrument. The

DSC thermograms were utilized to determine the onset, midpoint, offset temperatures, and enthalpy values of the ice cream specimens.

2.7. Steady-Shear Analysis

The rheological characteristics of ice cream samples were analyzed using a stress or strain-controlled rheometer (Anton Paar, model MCR 302, Austria) outfitted with a parallel plate probe (50 mm diameter) set to a gap distance of 1 mm, temperature of 4 °C, and shear rate of 0.1-100 s^{-1}. The results were subjected to statistical analysis by Statistica version 7 (Stat Soft Inc., Tulsa, OK, USA) computer software program for modeling the parameters. Using the parameters obtained from this software, η_{50} was determined. η_{50} represents the apparent viscosity (Pa·s) measured at a shear rate of 50 s^{-1}, which provides a quantitative evaluation of the fluid's resistance to flow under the applied shear (Demir et al. 2017; Kutlu et al. 2024b). Moreover, the Oswalt-de-Waele model was utilized to assess the determination coefficient (R^2), flow behavior index (n), and consistency coefficient (K) of the collected data (Kutlu et al. 2020).

$$\sigma = K(\dot{\gamma})^n \quad (1)$$

(σ: shear stress, K: consistency coefficient, γ: shear rate and n: flow behavior index)

2.8. Sensorial Attributes

Ten individuals, including undergraduate and graduate students as well as faculty members from the Department of Food Engineering at Yıldız Technical University, were chosen as semi-trained panelists. The ice cream samples were randomly coded with three-digit numbers and presented to the panelists under controlled conditions in a well-lit room at 22 ± 2 °C. Each panelist was provided with water and unsalted crackers to cleanse their palate between the samples. Five quality parameters (color, consistency, taste and flavor, odor, overall acceptability) of the samples were assessed using an evaluation form. Panelists provided evaluations within a range of 1-5 points (1: very poor; 5: excellent) (Karaman and Kayacier 2012).

2.9. Statistical Evaluation

The analyses were conducted with two replicates and at least three parallels. One-way analysis of variance (ANOVA) was employed for data analysis, and Duncan's test ($p<0.05$) was utilized for comparing the means. The statistical evaluations were performed using JMP software package program (version 6.0; SAS Institute Inc., Cary, North Carolina, USA) (Kutlu et al. 2024a).

Results

3.1. Physico-Chemical Properties

3.1.1. pH

The pH level directly affects how the taste and aroma of dairy products are perceived (Murtaza et al. 2004). As seen in Table 1, the pH values of tested samples were close to neutral and ranged between 6.55 and 6.61. Although the incorporation of AVLG into the ice creams slightly decreased the pH value compared to the control ice creams, there was no statistically significant decline ($p>0.05$). Similarly, Güzeler et al. (2012) also reported that the stabilizers (salep or guar gum) and emulsifiers (glycerol monostearate and/or polysorbate 80) used in the preparation of ice cream had no significant effect on pH values. As well, Baer et al. (1997) also reported that emulsifier type (mono-diglycerides, polysorbate 80, α-monoglyceride and lecithin) and level did not significantly alter the pH of ice cream. However, the use of different stabilizers/emulsifier blends (guar gum-xanthan gum) in the preparation of ice creams significantly affected the pH values ($p<0.05$) (Murtaza et al. 2004). The pH findings were in line with the findings published by Baer et al. (1997) (6.48-6.50), Güzeler et al. (2012) (6.43-6.54), and Karaca et al. (2009) (6.27-6.52).

Table 1. Some physicochemical and bioactive properties of ice cream samples.

Samples	pH	Dry matter (%)	L^*	a^*	b^*	ΔE^*	TPC (mg GAE/g)
Control	6.61 ± 0.02^{a}	33.09 ± 0.08^{c}	82.37 ± 1.27^{b}	-2.25 ± 0.10^{a}	1.96 ± 0.49^{b}	0.00	0.073 ± 0.01^{b}
A1E2	6.60 ± 0.03^{a}	37.30 ± 0.02^{b}	86.01 ± 0.51^{a}	-2.56 ± 0.37^{ba}	4.97 ± 0.68^{a}	4.73	0.099 ± 0.01^{a}
A2E2	6.56 ± 0.03^{a}	40.57 ± 0.05^{a}	85.03 ± 1.53^{ba}	-2.75 ± 0.04^{ba}	4.32 ± 1.23^{a}	3.59	0.081 ± 0.01^{ba}
A2	6.55 ± 0.01^{a}	33.99 ± 0.06^{c}	84.39 ± 0.65^{ba}	-2.86 ± 0.03^{b}	4.35 ± 0.60^{a}	3.19	0.090 ± 0.01^{ba}

L^*: Whiteness/darkness, a^*: Redness/greenness, b^*: Yellowness/blueness, ΔE^*: total colour change, TPC: Total phenolic content, GAE: Gallic acid equevalents, AVLG: Aleo vera leaf gel, Control: The ice cream mix formulations with 2 g emulgator and 0 g/100 g AVLG, A1E2: The ice cream mix formulations with 2 g emulgator and 1 g/100 g AVLG, A2E2: The ice cream mix formulations with 2 g emulgator and 2 g/100 g AVLG, A2: The ice cream mix formulations with 2 g/100 g AVLG.

3.1.2. Dry matter content

The samples exhibited dry matter contents ranging from 33.09% to 40.57% (Table 1). The A2E2 sample and the control ice cream had the highest and lowest dry matter contents, respectively. These results were as expected due to the particular combination and amounts of AVLG used, both with and without the emulsifier. The rise in total solids directly correlated with the increase in AVLG, regardless of the presence of the emulsifier, when compared to the control sample. The aloe vera whole leaf primarily comprises water, with the remaining dry matter content consisting of carbohydrates, dietary fibers, ashes, proteins, and lipids, accounting for approximately 2.51% (Palaniyappan et al. 2023). The statistical analysis showed that there was no significant difference between control and A2 ($p>0.05$); however, the differences between other ice cream samples were statistically significant ($p<0.05$).

3.1.3. Color properties

The enriched samples resulted in noticeably higher brightness values (L^*, 84.39-86.01) compared to the control ice cream samples (82.37), as indicated in Table 1. The addition of AVLG, with/without an emulsifier, had a significant effect in increasing the brightness values of the samples. Furthermore, all ice cream samples possessed negative a^* values with the range of -2.25 to -2.86 (Table 1). The incorporation of AVLG increased the negativeness of a^* value and the highest greenness was determined in A2. The incorporation of AVLG resulted in a reduction in the greenness values $(-a^*)$ of the samples, indicating that the ice cream samples enriched with AVLG had a greener color compared to the control ice cream sample. Moreover, b^* values of the samples were between 1.96 and 4.97 (Table 1), indicating that the highest yellowness was found in A1E2. The incorporation of AVLG had a substantial impact on elevating the yellowness values of the ice cream samples ($p < 0.05$). Among the coded samples, the highest positive b^* values were observed in A1E2, indicating a greater degree of yellowness with increasing AVLG content. However, it is noteworthy that there were no significant differences in b^* values of A1E2, A2E2 and A2 ($p<0.05$). Similar trend for L^*, a^* and b^* values was also noted with the incorporation of grape wine lees (Hwang et al. 2009).

ΔE^* is a widely used colorimetric parameter for assessing color variations resulting from various processing conditions. It combines the L^*, a^*, and b^* values to quantify the extent of color variation (Sagdic et al. 2012; Bagdat et al., 2024; Süren et al., 2024). As seen in Table 1, the ΔE^* values ranged from 0.00 to 4.73, indicating that the observed differences were noticeable to the human eye, as ΔE^* values exceeding 3 were considered perceptible (Yavuz et al. 2022). The findings showed that the combination of AVLG along with/without glycerol monostearate had a significant role in the variation of color properties for the ice creams. All the evidence gathered in these findings indicated that the utilization of AVLG along with/without emulsifier in the formulation led to an increase in the brightness values of the samples, while simultaneously increasing the levels of greenness and yellowness.

3.2. TPC

Phenolics are important components found in plants and possess antioxidant properties that can be attributable to their redox properties. Phenolic compounds are the predominant secondary metabolites in plants and play crucial roles in pigmentation, growth, and defense against pathogens. These compounds display a diverse range of chemical and biological characteristics (Erol and Kutlu, 2025; Kutlu, 2024; Yasar et al. 2022). In this context, the incorporation of AVLG in ice cream may be beneficial as it provides a substantial quantity of phenolic antioxidants, owing to its rich phenolic content. Table 1 presents the TPC values of the ice cream samples. The control sample exhibited the lowest TPC (0.073 ± 0.01 mg GAE/g) compared to the AVLG-incorporated samples. Specifically, the TPC values were 0.099 ± 0.01 mg GAE/g for A1E2, 0.081 ± 0.01 mg GAE/g for A2E2, and 0.090 ± 0.01 mg GAE/g for A2. The addition of AVLG resulted in a significant diffusion of phenolic compounds into the ice cream ($p<0.05$), leading to an increase in the TPC values of the tested samples. According to the findings of O'Connell and Fox (2001), TPC in milk may originate from several sources such as amino acid degradation, animal feed, or environmental contamination. Furthermore, the pasteurization process of ice cream mixtures may lead to the formation of phenolic compounds through the Maillard reaction, which could be the main source of phenolics in the control ice cream. In a related study, Chathurangi and Gunathilake (2018) reported TPC values for the ice creams enriched with lyophilized aloe vera gel powder as 69.67 ± 2.02 µmol GAE g^{-1} in dry weight. The aqueous and methanol extract of aloe vera were reported 8 and 20 mg GAE g^{-1} extract, respectively (Manye et al. 2023). Also, Gorsi et al. (2019) examined the TPC of aloe vera gel powder using different solvents such as ethanol (28.44 mg GAE g^{-1}), methanol (27.15 mg GAE g^{-1}), and acetone (11.48 mg GAE g^{-1}). The exact content of phenolic compounds can vary depending on the type of aloe vera plant, the geographical region where it grows and the growing conditions (Aida et al. 2022; Sánchez-Machado et al., 2017). Aloe vera is composed of approximately 110 bioactive compounds, which can be grouped into six categories: flavonoids; phenylpropanoids and coumarins; phytosterols and other components; chromones and their glycoside derivatives; anthraquinones and their glycoside derivatives; and phenylpyrone along with phenolic compounds (Kahramanoğlu et al. 2019).

3.3. DSC

The melting resistance of ice cream is a critical indicator of its ability to endure elevated temperatures without significant melting, directly influencing the functionality of its components (Pintor-Jardines et al. 2018). Within the increasing temperature during the DSC heating process, an endothermic peak was formed. The onset temperature for the control sample (-0.90 °C) was higher than that of the AVLG-containing samples, where onset temperatures were measured as -6.71 °C (A1E2), -6.95 °C (A2E2), and -7.48 °C (A2). These results showed that the inclusion of AVLG shifted the onset of ice crystal formation to lower temperatures, thereby slowing down the freezing process. Similarly, midpoint crystallization temperatures decreased with increasing AVLG content, with A2 showing the lowest value at -2.35 °C. Offset temperatures were also lower in AVLG-containing samples, measured at 3.09 °C (A1E2), 2.24 °C (A2E2), and 1.45 °C (A2), compared to the control sample's offset temperature of 9.41 °C. The ΔHm value, indicating the enthalpy change during melting, was highest in the control sample (160.80 J g^{-1}), reflecting a higher energy requirement for ice melting due to greater ice crystal formation (Brown and Brown 2000). Conversely, the ΔHm values decreased in AVLG-containing samples to 76.63 J g^{-1} (A1E2), 68.76 J g^{-1} (A2E2), and 53.60 J g^{-1} (A2). Notably, the lowest ΔHm value was observed in the A2 sample (2 g/100 g AVLG), emphasizing AVLG's efficacy in modifying the thermal behavior of the ice cream matrix (Scholten, 2013). This suggests that the presence of AVLG contributed to lower energy consumption for ice melting, which correlated with an altered structural composition of the ice cream mix (Scholten, 2013). These findings underlined that the reduced melting enthalpy was associated with higher water content, increased ice formation, and a decrease in protein content, which otherwise binds water and stabilizes the structure. In model systems with lower solids content and the absence of proteins, ice formation was more pronounced, leading to higher melting enthalpies (Soukoulis et al. 2009; Süren et al. 2024).

The DSC findings were consistent with the rheological studies. Due to the decrease in viscosities, reductions in onset, midpoint, and offset temperatures were observed in comparison to the control samples (Soukoulis et al. 2009). Moreover, the decrease in midpoint temperatures indicated a reduction in the thermodynamic stability of the ice creams (Süren et al. 2024). These observations highlighted the critical role of stabilizers and emulsifiers in determining the thermal properties of ice cream. Similarly, as noted by Kavaz-Yüksel (2015), the thermal conductivity of ice cream is influenced by various factors, including density, air fraction, temperature, and other related parameters. Overall, the incorporation of AVLG, with or without the addition of an emulsifier, significantly affected the thermal characteristics of the ice cream by reducing the amount of ice and free water while increasing the proportion of bound water.

Table 2. Thermal and steady state rheological properties of ice cream samples.

Samples	Onset (°C)	Midpoint (°C)	Offset (°C)	ΔHm (J g^{-1})	R^2	*K* (Pa.s)	*n*	η_{50} (Pa.s)
Control	-0.90	2.65	9.41	160.80	0.9998	0.68 ± 0.01^a	0.50 ± 0.01^d	0.1540
A1E2	-6.71	-1.64	3.09	76.63	0.9998	0.44 ± 0.00^b	0.73 ± 0.00^c	0.1505
A2E2	-6.95	-2.48	2.24	68.76	0.9995	0.36 ± 0.00^c	0.77 ± 0.00^b	0.1140
A2	-7.48	-2.35	1.45	53.60	0.9993	0.25 ± 0.00^d	0.80 ± 0.00^a	0.0980

K: consistency coefficient, *n:* flow behavior index, AVLG: aleo vera leaf gel, AVLG: Aleo vera leaf gel, η_{50}: aparent viscosity, Control: The ice cream mix formulations with 2 g emulgator and 0 g/100 g AVLG, A1E2: The ice cream mix formulations with 2 g emulgator and 1 g/100 g AVLG, A2E2: The ice cream mix formulations with 2 g emulgator and 2 g/100 g AVLG, A2: The ice cream mix formulations with 2 g/100 g AVLG.

3.4. Steady shear flow properties

The alteration of viscosity in relation to changes in shear rate plays a crucial role in the ice cream production, as it provides insights into the aggregation of fat globules and their susceptibility to fragility under specific shear conditions. These factors are significant in the transition of clumps into a cohesive fat network. Figure 3 shows typical plots illustrating the relationship between shear stress (Pa) and shear rate (s^{-1}) for non-Newtonian fluids in ice cream samples. As can be seen clearly from the figure that the shear stress of ice creams increased with increasing shear rate as expected. Typically, ice cream mixes exhibit colloidal properties and consist of fat droplets that are enveloped by a protein-emulsifier layer, acting as the dispersed phase (Aime et al. 2001; Karaca et al. 2009). As a result, the flow properties of the majority of ice cream mixes are often characterized as pseudoplastic. The relationship between shear stress and shear rate of all ice cream samples was suitably characterized by the Oswalt-de-Waele equation, with determination coefficients of 0.9993 or higher (Table 2). Likewise, Kavaz-Yüksel (2015) reported that rise in viscosity values in ice cream was attributed to protein aggregation and the clustering of fat globules at lower temperatures, as well as the formation of small air cells during storage. In the present study, the addition of AVLG decreased the viscosity by altering the water-binding and structural properties of the ice cream mix, potentially influencing the overall textural and melting properties of the final product. Overall, the combined use of AVLG and emulsifier significantly influenced the rheological properties of the ice cream mixes.

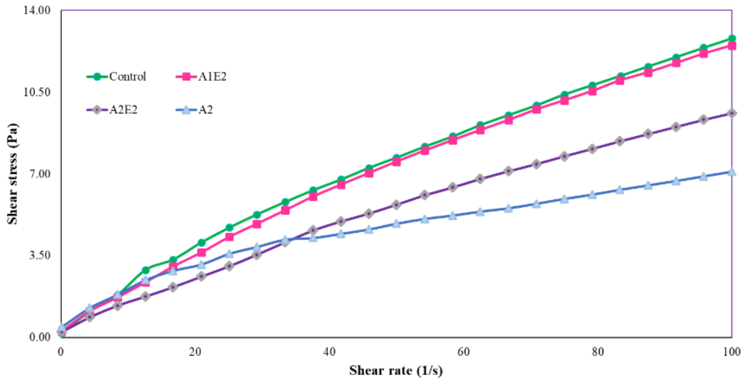

Figure 3. Shear rate versus shear stress plots of ice cream mixes.

AVLG: Aleo vera leaf gel, Control: The ice cream mix formulations with 2 g emulgator and 0 g/100 g AVLG, A1E2: The ice cream mix formulations with 2 g emulgator and 1 g/100 g AVLG, A2E2: The ice cream mix formulations with 2 g emulgator and 2 g/100 g AVLG, A2: The ice cream mix formulations with 2 g/100 g AVLG.

3.5. Sensory evaluation

The ice creams enriched with AVLG received generally acceptable sensory scores from the panelists. The sensory assessment scores of the ice cream samples were presented in Table 3. The control sample achieved the highest sensory score in overall acceptability (4.20 ± 0.79). Among the enriched formulations, A1E2 exhibited similar sensory performance to the control, with slightly lower scores across most attributes but no statistically significant differences ($p > 0.05$). The A2E2 sample showed comparable scores to A1E2, particularly in color (4.60 ± 0.52) and consistency (4.10 ± 0.88). However, a slight decline was noted in taste and aroma (3.70 ± 1.34), odor (3.90 ± 0.10) and overall acceptability (4.00 ± 1.10), indicating that a higher concentration of AVLG might slightly influence flavor perception. In contrast, the A2 sample recorded the lowest sensory scores across most parameters, particularly in consistency (3.40 ± 0.84), taste & aroma (3.60 ± 0.51) and overall acceptability (3.70 ± 0.83). This suggested that the absence of emulsifiers negatively impacted the sensory properties, highlighting their role in improving texture and overall sensory quality parameters. Notably, the color attribute was consistently rated high across all formulations, with no significant differences observed ($p > 0.05$), indicating that the incorporation of AVLG did not negatively affect the visual appeal of the samples. Similarly, the odor scores remained stable, with minor variations among the formulations. These findings demonstrated that the addition of AVLG at moderate concentrations, particularly in conjunction with an emulsifier, could maintain the sensory quality of ice cream formulations. However, the absence of emulsifiers or higher concentrations of AVLG may slightly diminish sensory acceptance, particularly in terms of consistency and overall flavor perception. The findings obtained in our study in terms of taste and flavor also supported the accuracy of the results obtained from pH measurements. Additionally, according to Baer et al. (1997), the presence of emulsifier did not have a notable impact on the flavor scores of low-fat ice creams. However, according to a study conducted by Chathurangi and Gunathilake (2018), the ice cream formulations incorporated with aloe vera gel cubes were identified as the most preferred choice among consumers.

Table 3. Sensory properties of ice cream samples.

Samples	Color	Consistency	Taste and aroma	Odor	Overall acceptability
Control	4.50 ± 0.52^a	4.00 ± 0.82^a	4.00 ± 1.15^a	4.00 ± 0.67^a	4.20 ± 0.79^a
A1E2	4.60 ± 0.52^a	3.90 ± 0.88^a	3.90 ± 0.74^a	4.10 ± 0.57^a	4.10 ± 0.57^a
A2E2	4.60 ± 0.52^a	4.10 ± 0.88^a	3.70 ± 1.34^a	3.90 ± 0.10^a	4.00 ± 1.10^a
A2	4.40 ± 0.70^a	3.40 ± 0.84^a	3.60 ± 0.51^a	3.80 ± 0.64^a	3.70 ± 0.83^a

Control: The ice cream mix formulations with 2 g emulgator and 0 g/100 g AVLG, A1E2: The ice cream mix formulations with 2 g emulgator and 1 g/100 g AVLG, A2E2: The ice cream mix formulations with 2 g emulgator and 2 g/100 g AVLG, A2: The ice cream mix formulations with 2 g/100 g AVLG.

The slight reduction in pH observed in the AVLG-enriched samples corresponds to the taste & aroma and overall acceptability trends. Lower pH values may subtly influence flavor perception, particularly in formulations with higher AVLG concentrations. For instance, the A2E2 and A2 samples, which had the lowest pH values (6.56 ± 0.03 and 6.55 ± 0.01, respectively), also received slightly lower scores in taste and aroma (3.70 ± 1.34 and 3.60 ± 0.51, respectively). This suggests that even minor changes in acidity could interact with the flavor profile of the ice cream. Despite the slight decline in pH, the sensory scores for most attributes, including overall acceptability, remained relatively high, particularly for the control and A1E2 samples. The A1E2 sample showed a pH of 6.60 ± 0.03 and maintained a sensory profile comparable to the control. This indicates that moderate levels of AVLG, when paired with an emulsifier, do not negatively impact the sensory qualities of ice cream. On the other hand, the A2 sample, which lacked an emulsifier, not only had the lowest pH but also recorded the lowest sensory scores across consistency, color, taste & aroma, odor and overall acceptability (3.70 ± 0.83). This suggests that the absence of an emulsifier, combined with the slight reduction in pH, may have contributed to less favorable sensory properties, particularly in texture and flavor. These findings demonstrated that the incorporation of AVLG at moderate levels, particularly in conjunction with an emulsifier, could maintain both pH stability and sensory quality in ice cream formulations. However, higher AVLG concentrations or the absence of emulsifiers may slightly alter acidity and negatively impact sensory acceptance; although this difference is not statistically significant, it is noteworthy.

4. Conclusions

Effect of AVLG incorporation on the physico-chemical, TPC, rheological, thermal, and sensory characteristics of ice cream samples were investigated in this study. The results showed that the pH values of all samples were not significantly affected by the addition of AVLG, while the dry matter and TPC values of the tested samples increased with the addition of AVLG. As well, the supplementation of AVLG along with/without emulsifier had a significant role in the variation of color properties of ice creams. Corresponding to the thermal analysis findings, enrichment of ice creams with AVLG provided lower energy consumption for ice melting. The Ostwald de Waele model was efficiently used to accurately characterize the steady shear data of ice cream mixes showing pseudoplastic flow tendencies. All the ice cream samples had acceptable sensory evaluation scores irrespective of AVLG addition. In conclusion, the findings indicated that AVLG could serve as a multifunctional ingredient for the production of ice cream with better physicochemical, bioactive, rheological, thermal, and sensory properties.

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References

Ahlawat KS, Khatkar BS (2011). Processing, food applications and safety of aloe vera products: a review. *Journal of Food Science and Technology,* 48: 525-533.

Aida PUIA, Chedea VS, Levai AM, Bocsan IC, Buzoianu AD (2022). Pot Aloe vera gel–a natural source of antioxidants. *Notulae Botanicae Horti Agrobotanici Cluj-Napoca,* 50(2): 12732-12732.

Aime DB, Arntfield SD, Malcolmson LJ, Ryland D (2001). Textural analysis of fat reduced vanilla ice cream products. *Food Research International,* 34(2-3): 237-246.

Akman PK, Kutlu G, Tornuk F (2023). Development and characterization of a novel sodium alginate based active film supplemented with *Lactiplantibacillus plantarum* postbiotic. *International Journal of Biological Macromolecules,* 125240.

Alemdar S, Agaoglu S (2009). Investigation of *in vitro* antimicrobial activity of aloe vera juice. *Journal of Animal and Veterinary Advances,* 8(1): 99-102.

Aslani A, Ghannadi A, Raddanipour R (2015). Design, formulation and evaluation of Aloe vera chewing gum. *Advanced Biomedical Research*, 4:175.

Atlar GC, Kutlu G, Tornuk F (2023). Design and characterization of chitosan-based films incorporated with summer savory (*Satureja hortensis* L.) essential oil for active packaging. *International Journal of Biological Macromolecules,* 127732.

AOAC International (2000). AOAC International Official Methods of Analysis (17th), AOAC International, Arlington, VA (2000).

Bagdat ES, Kutlu G, Tornuk F (2024). The effect of free and encapsulated probiotic bacteria on some physicochemical, microbiological, and textural properties of apricot leather (pestil) during storage. *Journal of Food Science,* 1-16.

Baer RJ, Wolkow MD, Kasperson KM (1997). Effect of emulsifiers on the body and texture of low fat ice cream. *Journal of Dairy Science,* 80(12): 3123-3132.

BahramParvar M, Razavi SM, Khodaparast MH (2010). Rheological characterization and sensory evaluation of a typical soft ice cream made with selected food hydrocolloids. *Food Science and Technology International,* 16(1): 79-88.

Brown RJC, Brown RFC (2000). Melting point and molecular symmetry. *Journal of Chemical Education*, *77*(6): 724.

Chathurangi, BMV, Gunathilake KDPP (2018). Development of Aloe barbadensis incorporated ice cream and evaluation of antioxidant and anti-inflammatory properties. *Research Symposium on Pure and Applied Sciences*, Faculty of Science, University of Kelaniya, Sri Lanka. P57.

Comas-Serra F, Estrada P, Minjares-Fuentes R, Femenia A (2023). Evaluation of Acemannan in different commercial beverages containing Aloe vera (*Aloe barbadensis* Miller) gel. *Gels,* 9(7): 552.

Demir MK, Kutlu G, Yilmaz MT (2017). Steady, dynamic and structural deformation (three interval thixotropy test) characteristics of gluten-free Tarhana soup prepared with different concentrations of quinoa flour. *Journal of Texture Studies*, 48(2): 95-102.

Demirkan EB, Akyurek SN, Bayraktar D, Kutlu G, Tornuk F (2024). Potential use of hazelnut (*Corylus avellana* L.) shell in muffin production by substitution of wheat flour: Color, bioactive, textural, and sensory properties. *European Food Science and Engineering,* 5(1): 1-7.

Erol KF, Kutlu G (2025). Assessment of antimicrobial, anti-diabetic, anti-Alzheimer and anti-cancer properties, along with bioaccessibility and bioactive potential of *Polygonum cognatum* Meissn extracts from various cities in Türkiye. *Food Bioscience, 63*: 105669.

Erol KF, Kutlu G, Tornuk F, Guzel M, Donmez IE (2023). Determination of antioxidant, anticancer, antidiabetic and antimicrobial activities of Turkish red pine (*Pinus brutia* Ten.) bark ultrasound-assisted extract as a functional food additive. *Acta Alimentaria, 52*(1): 102-112.

Genovese A, Balivo A, Salvati A, Sacchi R (2022). Functional ice cream health benefits and sensory implications. *Food Research International,* 111858.

Goraya RK, Bajwa U (2015). Enhancing the functional properties and nutritional quality of ice cream with processed amla (*Indian gooseberry*). *Journal of Food Science and Technology,* 52: 7861-7871.

Güzeler N, Kaçar A, Keçeli T, Dilek SAY (2012). Effect of different stabilizers, emulsifiers and storage time on some properties of ice cream. *Akademik Gıda, 10*(2): 26-30.

Hamman JH (2008). Composition and applications of Aloe vera leaf gel. *Molecules,* 13(8): 1599-1616.

Hwang JY, Shyu YS, Hsu CK (2009). Grape wine lees improves the rheological and adds antioxidant properties to ice cream. *LWT-Food Science and Technology,* 42(1): 312-318.

Kahramanoğlu İ, Chen C, Chen J, Wan C (2019). Chemical constituents, antimicrobial activity, and food preservative characteristics of Aloe vera gel. *Agronomy,* 9(12): 831.

Karaca OB, Güven M, Yasar K, Kaya S, Kahyaoglu T (2009). The functional, rheological and sensory characteristics of ice creams with various fat replacers. *International Journal of Dairy Technology,* 62(1): 93-99.

Karaman S, Kayacier A (2012). Rheology of ice cream mix flavored with black tea or herbal teas and effect of flavoring on the sensory properties of ice cream. *Food and Bioprocess Technology,* 5: 3159-3169.

Karaman S, Toker ÖS, Yüksel F, Çam M, Kayacier A, Dogan M (2014). Physicochemical, bioactive, and sensory properties of persimmon-based ice cream: Technique for order preference by similarity to ideal solution to determine optimum concentration. *Journal of Dairy Science,* 97(1): 97-110.

Kavaz-Yuksel A (2015). The Effects of blackthorn (*Prunus Spinosa* L.) addition on certain quality characteristics of ice cream. *Journal of Food Quality,* 38(6): 413–421.

Khider M, Ahmed N, Metry WA (2021). Functional ice cream with coffee-related flavor. *Food and Nutrition Sciences,* 12(8): 826-847.

Kutlu G (2024). Valorization of various nut residues grown in Turkiye: Antioxidant, anticholinesterase, antidiabetic and cytotoxic activities. *Food Science & Nutrition,* 12:4362–4371.

Kutlu G, Erol KF (2025). An in vitro assessment of the bioactive, cytotoxic, and antidiabetic potential of different parts of tumbleweed (*Gundelia tournefortii* L.). *The Journal of Food,* 50(1): 28-41.

Kutlu G, Bozkurt F, Tornuk F (2020). Extraction of a novel water-soluble gum from nettle (Urtica dioica) seeds: Optimization and characterization. *International Journal of Biological Macromolecules,* 162: 480-489.

Kutlu G, Yılmaz S, Karabulut AE (2024a). Development of a new vegan muffin formulation: Assessing its quality and sensory characteristics. *European Food Science and Engineering,* 5(1): 26-34.

Kutlu G, Baslar M, Yilmaz MT, Karaman S, Vardar US, Poyraz-Yildirim E, Sagdic O (2024b). Manufacturing of emulsion type garlic paste: Characterization of rheological properties as affected by lecithin, guar gum, and olive oil using steady, dynamic, and three interval thixotropy tests. *Journal of Food Process Engineering,* 47(1): e14522.

Maan AA, Ahmed ZFR, Khan MKI, Riaz A, Nazir A (2021). Aloe vera gel, an excellent base material for edible films and coatings. *Trends in Food Science & Technology,* 116: 329-341.

Manoharan A, Ramasamy D, Kumar CN, Dhanalashmi B, Balakrishnan V (2012). Organoleptic evaluation of herbal ice creams prepared with different inclusion levels of Aloe vera pulp. *Health,* 1(2):25-28.

Manoharan AP, Ramasamy D (2013a). Physico-chemical, microbial and sensory analysis of Aloe vera (pulp) ice cream with natural colour in different artificial sweeteners. *Indian Journal of Fundamental and Applied Life Sciences,* 3(2): 114-121.

Manoharan AP, Ramasamy D (2013b). Physico-chemical, microbial and sensory analysis of Aloe vera (pulp) ice cream with natural colour curcumin in different artificial sweeteners. *Indian Journal of Fundamental and Applied Life Sciences,* 3(2): 122-130.

Manoharan AP, Ramasamy D (2013c). Physico-chemical, microbial and sensory analysis of Aloe vera (pulp) ice cream with natural identical vanilla flavour in different artificial sweeteners. *Indian Journal of Fundamental and Applied Life Sciences,* 3(2): 106-113.

Misir J, Brishti FH, Hoque MM (2014). Aloe vera gel as a novel edible coating for fresh fruits: A review. *American Journal of Food Science and Technology,* 2(3): 93-97.

Mule SM, Kadam SS, Dandekar VS, Ramod SS, Desai B, Narkhede SL (2020). Manufacturing technology and production cost of ginger (*Zingiber officinale* L.) and Aloe vera (*Aloe barbadensis*) juice enriched probiotic (*L. acidophilus*) ice cream. *International Journal of Chemical Studies,* 8(2): 185-188.

Munoz OM, Leal X, Quitral V, Cardemil LJMAP (2015). Extraction, characterization and properties of the gel of Aloe vera (*Aloe barbadensis* Miller) Cultivated in Chile. *Medicinal and Aromatic Plants,* 4(3): 2-7.

Murtaza MA, Mueenuddin G, Huma N, Shabbir MA, Mahmood S (2004). Quality evaluation of ice cream prepared with different stabilizers/emulsifier blends. *International Journal of Agriculture and Biology,* 6(1): 65-67.

O'Connell JE, Fox PF (2001). Significance and applications of phenolic compounds in the production and quality of milk and dairy products: a review. *International Dairy Journal,* 11: 113– 120.

Olaleye MT, Bello-Michael CO (2005). Comparative antimicrobial activities of Aloe vera gel and leaf. *African Journal of Biotechnology,* 4(12): 1413-1414.

Pintor-Jardines A, Arjona-Román JL, Totosaus-Sánchez A, Severiano-Pérez P, González-González LR, Escalona-Buendia HB (2018). The influence of agave fructans on thermal properties of low-fat, and low-fat and sugar ice cream. *LWT,* 93: 679-685.

Reynolds T, Dweck AC (1999). Aloe vera leaf gel: a review update. *Journal of Ethnopharmacology* 68(1-3): 3-37.

Rodríguez ER, Martín JD, Romero CD (2010). Aloe vera as a Functional Ingredient in Foods. *Critical Reviews in Food Science and Nutrition,* 50(4): 305–326.

Sagdic O, Ozturk I, Cankurt H, Tornuk F (2012). Interaction between some phenolic compounds and probiotic bacterium in functional ice cream production. *Food and Bioprocess Technology,* 5: 2964–2971.

Sánchez-Machado DI, López-Cervantes J, Sendón R, Sanches-Silva A (2017). Aloe vera: Ancient knowledge with new frontiers. *Trends in Food Science & Technology,* 61: 94-102.

Scholten, E (2013). Ice cream. In Particulate products: Tailoring properties for optimal performance. Cham: Springer International Publishing, pp. 273-294.

Shibinshad K (2023). Development of Aloe vera candy (Doctoral dissertation).

Sonawane SK, Gokhale JS, Mulla MZ, Kandu VR, Patil S (2021). A comprehensive overview of functional and rheological properties of aloe vera and its application in foods. *Journal of Food Science and Technology*, 58: 1217-1226.

Soukoulis C, Lebesi D, Tzia C (2009). Enrichment of ice cream with dietary fibre: Effects on rheological properties, ice crystallisation and glass transition phenomena. *Food Chemistry*, 115(2): 665-671.

Srisukh V, Tungrugsasut W, Bunyapraphatsara N, Khuwattanasil S, Wongpoomchai S (2008). Aloe ice cream and sherbet. *Thai Journal of Phytopharmacy,* 13(2): 2549-2551.

Süren BN, Salman S, Kaya E, Büyükkal Y, Kutlu G, Törnük F (2024). Symbiotic ice cream production using Lactiplantibacillus plantarum and oleaster (*Elaeagnus angustifolia* L.) flour. *Harran Tarım ve Gıda Bilimleri Dergisi*, *28*(3): 444-458.

Verma A, Ansari R, Broadway AA (2018). Preparation of herbal ice cream by using Aloe vera with mint flavour. *Journal of Pharmacognosy and Phytochemistry,* 7(3): 391-394.

Yasar B, Kutlu G, Tornuk F (2022). Edible flowers as sources of bioactive compounds: Determination of phenolic extraction conditions. *International Journal of Gastronomy and Food Science,* 30: 100618.

Yavuz Z, Kutlu G, Tornuk F (2022). Incorporation of oleaster (*Elaeagnus angustifolia* L.) flour into white bread as a source of dietary fibers. *Journal of Food Processing and Preservation,* 46(11): e17050

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Research Article

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Classifying *Apis mellifera* Breeds Using Data Mining Techniques Based on Morphological Traits

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HIGHLIGHTS

- The classification of bee breeds is crucial for genetic diversity and efficiency.
- The morphological traits of honeybee breeds are different.
- The random forest model achieved 99.8% success, proving highly effective for classification.
- Naïve Bayes consistently performed the worst across all evaluation metrics.

Abstract

The classification of bee breeds is significant for breeding, maintaining genetic diversity, increasing productivity and protecting the health of the bee colonies. Therefore, this study aims to classify different honeybee breeds based on their morphological traits using data mining techniques, which are cost-effective and straightforward. It were used a total of 35 colonies from a private bee farm for morphometric analysis in the study, which included seven different bee breeds and 404 bee samples. A range of data mining techniques (Support Vector Machines (SVM), Random Forest (RF), Artificial Neural Networks (ANN), Naive Bayes (NB) and k-Nearest Neighbors (k-NN)), and model fit criteria were used for the classification of bee breeds. Overall, the study shows significant differences in the morphological traits of different bee breeds, highlighting the diversity and different traits of each bee breed. In addition, the study shows that the RF model is superior in all criteria and therefore the most effective for classifying honeybee breeds. In contrast, the NB model consistently performs the worst, as evidenced by the consistently minimum values of all metrics. In conclusion, RF model exhibiting a 99.8% success rate, stands out as highly effective in the classification of bee breeds based on the morphological traits, supporting its applicability in future classification research.

Keywords: bee breeds, classification, data mining, morphological traits, performance metrics

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1. Introduction

Bees are essential pollinators that contribute significantly to global food production and biodiversity (Potts et al. 2010). For sustainability in these; the classification of bee breeds is significant, which has a crucial role in understanding their behavior, ecological impacts, and potential for various applications in agriculture and environmental conservation. However, traditional methods of classification have relied on manual observation and subjective judgment, often leading to inconsistencies, misclassification (Berlocher 1984) and high costs (Tapkan et al. 2016). In addition, accurately classification of bee breeds only based on their morphological traits (MTs) could be difficult due to their few variations. Accordingly, the application of data mining methods has emerged as a promising approach to overcome these challenges in recent years. Data mining methods can be defined as the process of extracting previously unknown information from large-scale databases and utilizing it effectively in the decision-making process using computational techniques, statistical analysis, and machine learning algorithms (Tapkan et al. 2016; Han et al. 2022). Because of these features, it offers a more objective and systematic approach to classification of bee breeds. The primary data mining methods Support Vector Machines (SVM), Random Forest (RF), Artificial Neural Networks (ANN), Naive Bayes (NB) and k-Nearest Neighbors (k-NN) were investigated. SVM is a powerful supervised learning technique that constructs a hyperplane (Vapnik 1999) to classify different bee breeds in a high-dimensional feature space. RF is an ensemble learning method that combines multiple decision trees to improve classification accuracy (Breiman 2001). ANN, inspired by the biological neural network, utilizes interconnected layers of artificial neurons to learn (Yegnanarayana 1999) and classify bee breeds based on their MTs. NB is based on Bayes' theorem, which assumes that the presence of a particular feature is independent of the presence of any other trait in an insect class (Antony and Pratheepa 2018). k-NN classifies based on their proximity to the nearest neighbors in the feature space (Cover and Hart 1967) and the k-NN algorithm achieves a better identification rate for species (Li and Xiong 2018). By harnessing the power of these data mining methods, researchers and beekeepers could improve the efficiency and accuracy of breed classification processes. In the classification studies, the MTs of bees include length of the wing, width of the wing, length of the tongue and other distinguishing traits (Buco et al. 1987; Rinderer et al. 1993; Crewe et al. 1994; Ftayeh et al. 1994; Diniz-Filho and Malaspina 1995; Szymula et al. 2010). However, the MTs of bees can be generally categorized into three main groups: length measurements, color measurements, and wing venation traits (Abou-Shaara et al. 2013) and the third were investigated in detail by Abou-Shaara (2013). The classification studies in terms of the MTs efficiently compile and organize a vast amount of data regarding variations (Alpatov 1929; Guler and Bek 2002). Additionally, they assess the significance of these variations for honeybee populations (Estoup et al. 1995), facilitate visual comparisons in pure breed breeding through morphometric data (Gençer 2004), and distinguish between geographically adjacent types (Ruttner et al. 1978). For example, Frunze et al. (2022) studied to regularize by six best traits (width of the abdomen, length of tergites 3 and 4, width of the head, length of the antenna, and length of the forewing). Accurate breed classification could enable more targeted management practices, selective breeding programs, informed decision-making in areas such as pollination services, disease management, and ecological conservation.

The aim of this study is to determine whether bee breeds can be effectively classified using various data mining techniques such as SVM, RF, ANN, NB and k-NN, based on the analysis of certain MTs (e.g., scutellum color (SC), second abdominal tergit color (TC-2), fourth abdominal tergit color (TC-4), length of the tibia (LTI), length of the wing (LW), width of the wing (WW), cubital index (CI), width of the vein-a (WV-a), width of the vein-b (WV-b), wing vein angles (J10, B4, A4, E9), length of the tongue (LTO), length of the glossa (LG), length of the prementum (LPR) and length of the postmentum (LPM)) obtained from a diverse set of bee breeds including Pure Anatolia, Anatolia Hybrid, Carniolan Hybrid, Carpathian Hybrid, Caucasian hybrid, Italian Hybrid, and Kerkuk bees. The findings of this study may have practical implications for beekeepers, researchers, interested in improving bee breed identification as well as understanding the ecological roles and behaviors of different bee breeds.

2. Materials and Methods

This study was conducted in *Apis mellifera L.* (*Hymenoptera*: *Apidae*) honeybees raised Bozkır district (Latitude: 37.171860 and Longitude: 32.216846), Konya province, Türkiye. In the study, some MTs of seven different honeybee breeds in thirty-five colonies were used for breed classification. For the Carniolan hybrid: 1 colony and 27 bee samples; for the Kerkuk: 1 colony and 10 bee samples; for the Anatolian hybrid: 2 colony and 21 bee samples; for the Italian hybrid: 2 colony and 20 bee samples; for the Pure Anatolian: 1 colony and 23 bee samples; for the Carpathian hybrid: 3 colonies and 32 bee samples; and finally, for the Caucasian hybrid: 25 colonies and 271 bee samples were used. Totally, 35 colonies, 7 different breeds and 404 bee samples were used for the study. No morphometric analyzes and/or any practices related to the MTs were performed prior to the study to determine the breeds of used in the study; The colonies and bee samples were randomly selected. The collection of bee samples and testing techniques followed the animal welfare guidelines outlined in Article 9 of the "Veterinary Services, Plant Health, Food, and Feed Law" of Türkiye. The colonies used in the study were raised in ten-frame Langstroth hives in Konya during the summer months (May-September) and Antalya province during the winter months (October-April). The colonies were fed with sugar syrup and bee cake, except in January and February when temperatures are not suitable for feeding. In the current apiary, pure queens naturally mate with different drones and form hybrid colonies. For example, by mating pure Caucasian queens with different drones, Caucasian hybrids were formed and new queens were bred from these hybrid colonies. Colonies with pure queens were formed through purchases. Also, Kerkuk bees were used in the study to strengthen morphological distinction.

Bee samples from the colonies were taken in the summer of 2020 and were placed in alcohol and stored at +4 °C until morphological analysis. The specimens were initially preserved in 70% ethanol until the dissection. Subsequently, the forewings, proboscis and legs of honeybees were precisely severed and sequentially immersed in 60%, 70%, 80%, 90% and 100% ethanol solutions for gradual hydration. For all parts measurements, after immersing in alcohol series, were maintained in clove oil at least for 24 hours to facilitate tissue softening and enhance resolution. Following the dissection, the forewings, proboscis, and legs were carefully positioned in the microscope slides with hoyer medium (Distilled water, Gum Arabic, Chloral hydrate, and Glycerin) developed by Anderson (1954). The MTs scutellum color (SC); second abdominal tergit color (TC-2); fourth abdominal tergit color (TC-4); length of the tibia (LTI); length of the wing (LW); width of the wing (WW); cubital index (CI); width of the vein-a (WV-a); width of the vein-b (WV-b); wing vein angles (J10, B4, A4 and E9); length of the tongue (LTO); length of the glossa (LG); length of the prementum (LPR); and length of the postmentum (LPM) of honeybees were used. CI was calculated following equation reported by Özbakır (2011).

$$CI = (\frac{WV-a}{WV-b}) \quad (1)$$

In the equation, WV-a is the width of the vein-a and WV-b is the width of the vein-b. While SC was determined using a 5-point color scale from 0 to 4, TC-2 and TC-4 were determined using a 10-point color scale from 0 to 9 (Dodoloğlu 2000). Each bee sample was scored by two assessors and the average value was used for the evaluation. Optical microscopic observations and photography were conducted using an integrated 16-megapixel digital camera (AmScope MU1603, Irvine, CA, USA) under 20X magnification. All comparative morphological observations and the preparation of slides for colony morphology were carried out with the assistance of a Nikon SMZ 745T stereo microscope, specifically the stereomicroscope equipped with a G-AL-2X objective. This microscope model offered essential magnification capabilities and stereo vision, facilitating in-depth examination, including color detection, and precise documentation of MTs in the specimens under investigation. To conduct measurements, the 3-point circle measurement, arc measurement, and line measurement tools available in AmScope software version x64, 4.8 were utilized. These tools provided a comprehensive means of quantifying and analyzing various aspects of the captured images (Figure 1).

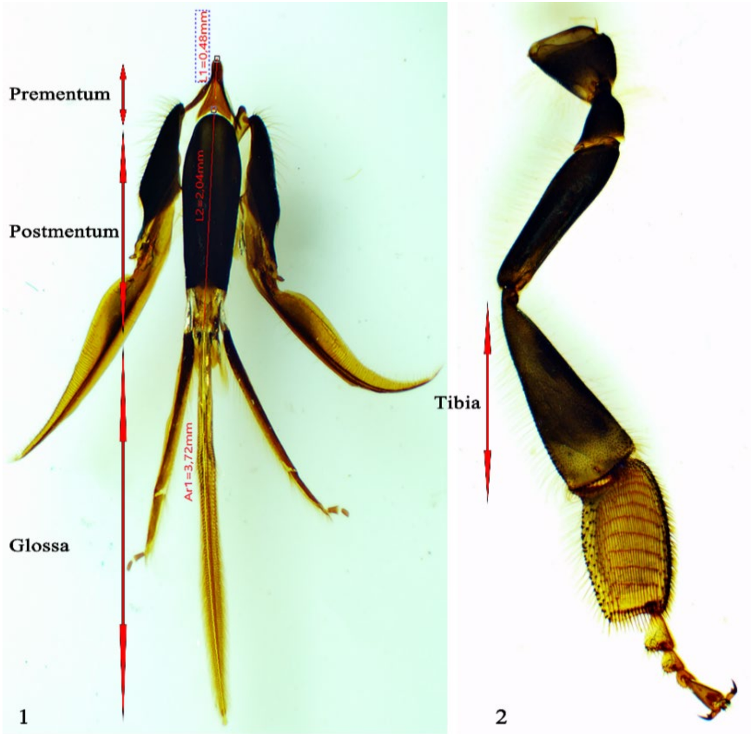

Figure 1. The analyzed some morphological traits. The honeybee tongue parts: (1) the prementum, the postmentum, the glossa. The honeybee leg part: (2) the tibia

Statistical analyzes were made using the ORANGE program version 3.3.1. (Demsar et al. 2013). Five different data mining algorithms SVM, RF, ANN, NB and k-NN models were used for classification based on the MTs of seven different bee breeds. A 70% training – 30% test ratio was used in ANN. Classification methods allow test data to be accurately assigned to specific classes by learning a model from the training data set (Bhavsar and Ganatra 2012). The data mining classification methods (SVM, RF, ANN, NB and k-NN) and model fit criteria (AUC, CA, F1, Precision, Recall and Specificity) used are explained below. SVM belongs to the family of generalized linear classifiers and is a supervised learning method for classification and regression. In other words, SVM is a classification and regression prediction method that leverages machine learning theory to maximize predictive power (Khan et al. 2023). Breiman (2001) developed the RF method in which different classification assignment algorithms are used together. RF combines multiple decision trees, and the majority vote based on each tree's predictions together determines the final prediction (Breiman 2001). Additionally, it is quite user-friendly in that it has only two parameters, the number of variables in the random subset at each node and the number of trees in the forest, and is generally not very sensitive to their values (Liaw and Wiener 2002). ANN, an information processing system, simulates the thinking system of the human brain. The fact that ANN is non-linear and designed to perform its tasks in a similar manner to the human brain makes ANN unique, and the sigmoid (logistic) activation function was used in this study. Therefore, it is a suitable method for processing the signals sent by various sensors and communication devices (Pan et al. 2023). NB is a well-known probabilistic classification algorithm that is simple but efficient and has a wide range of real-world applications including product recommendations, medical diagnostics, and autonomous vehicle control (Wickramasinghe and Kalutarage 2021). The nearest neighbor rule assigns an unclassified sample point the classification of the closest point from a set of previously classified points (Cover and Hart 1967). Given an unlabeled example, k-NN classifier searches for the k objects in the sample space that are closest to it and assigns the class based on the most common class label among them (Bhavsar and Ganatra 2012). If the value of k is too small, the k-NN classifier may be prone to overfitting due to noise in the training data set. On the other hand, if k is too large, the nearest neighbor classifier might misclassify the test example because the nearest neighbor list may contain data points that are far from the neighborhood. A value of k=1 is called nearest neighbor classification (Jadhav and Channe 2016). Model comparison was performed using the six following criteria. First, Area Under the Curve (AUC) is a criterion for measuring the performance of classification models. It represents the area under the Receiver Operating Characteristic (ROC) curve. AUC takes a value between 0 and 1 and indicates how well the classifier is performing. A value close to 1 indicates high accuracy, while a value close to 0.5 corresponds to random classification and indicates low model performance. Second, Classification Accuracy (CA) is a commonly used criterion for measuring the accuracy of classification models and represents the model's ability to classify correctly. It is calculated by dividing the total number of correctly classified examples by the total number of examples, ranges from 0% to 100%. A CA of 0% means that the model has not made any correct classifications, while a CA of 100% indicates that the model has classified all instances correctly. In general, a CA value above 80% is considered sufficient. Third, F1 score is the harmonic mean of a classification model's precision and recall measurements. Precision measures the ratio of true positive predictions to the overall prediction, and recall measures the rate of true positive predictions detected. The F1 score effectively measures the accuracy of the model by calculating these two criteria together. The F1 score takes a value between 0 and 1. The higher the value, the better the performance of the model. Fourth, Precision measures the ratio of truly positive predictions to the overall prediction. This evaluates how accurately the classifier identifies positive examples. Precision takes a value between 0 and 1, and a higher value indicates how accurately the model predicts positive classes. Fifth, Recall is a criterion for measuring the performance of a classification model and indicates how accurately the model detects true positives. Recall takes a value between 0 and 1, and a higher value indicates how accurately the model detects true positives. Finally, specificity typically is defined as the ability of a screening test to detect a true negative, based on the true negative rate, and to correctly exclusion of irrelevant MTs (Trevethan 2017).

3. Results

Morphological breed discrimination in honeybees is important for improving productivity, disease resistance, climate adaptation, and behavioral traits in beekeeping. Also, it is vital in determining genetic diversity and maintaining biological diversity worldwide. The means and standard deviations for seventeen different MTs of seven bee breeds used in the study were presented (Table 1). Table 1 shows an extensive comparison of different traits in different bee breeds. The fact that the scutellum color (3.60±0.52 pt) and TC-2 (9.0±0.0 pt) are higher in the Kerkuk breed than in other breeds indicates a distinctive trait of this breed. Conversely, TC-4 was lower in the Caucasian hybrid (1.19±0.89 pt) and Pure Anatolian (1.2±0.78 pt) breeds. It was determined that Italian hybrid bees had higher values in terms of LW (8.65±0.17 mm) and WW (2.97±0.08 mm). These various values may indicate specific traits or deficiencies of these breeds compared to others. Overall, the data shows significant differences in the traits of different bee breeds, highlighting the diversity and different traits of each bee breed.

In the present study, it can be seen that there are differences between the breeds in terms of the mean values of the MTs. It is noteworthy that SC and TC-2 are higher in the Kerkuk breed compared to other breeds. The observed differences in LTI, wing dimensions and CI suggest differences in flight abilities and foraging behavior between the breeds. Additionally, WV and wing vein angles are important traits that indicate the structural integrity and aerodynamics of the wings and are critical for efficient flight and maneuverability. The measurements of LTO and LG as well as LPR and LPM provide insights into the nutritional mechanisms and possible flowering preferences of the breeds. Morphological distinctions between bee breeds utilized in the current population could be regarded as an important gene pool for pollination efficiency, nectar collection, maintenance of ecological balance and sustainability of biodiversity. Gençer and Günbey (2020) reported that there were statistically significant differences between bees in the Caucasian, Yığılca and Korgan groups in terms of LTO, WW, LW and CI (P<0.001). Same researchers stated that the Caucasian breed is higher in LTO, but lower in other traits than other groups. Kambur and Kekeçoğlu (2018) found that the average of J10, B4, A4, and E9 vein angles were 53.86±0.09, 101.75±0.11, 33.69±0.05 and 19.22±0.03, respectively.

Table 1. The means and standard deviations of morphological traits of breeds

Traits	Breeds							Mean
	Carniolan hybrid	Pure Anatolian	Caucasian hybrid	Italian hybrid	Anatolian hybrid	Carpathian hybrid	Kerkuk	
SC (pt)	1.58±1.31	1.33±1.18	1.53±1.28	2.35±1.54	1.056±1.06	1.27±1.34	3.60±0.52	1.58±1.33
TC-2 (pt)	6.92±2.39	2.67±2.19	4.8±3.35	6.82±3.21	4.17±3.26	4.83±3.70	9.0±0.0	4.99±3.39
TC-4 (pt)	1.75±1.06	1.2±0.78	1.19±0.89	1.94±1.44	1.5±0.99	2.03±1.07	1.6±0.70	1.37±0.98
LTI (mm)	3.08±0.06	3.07±0.06	3.06±0.09	3.1±0.10	3.05±0.08	3.09±0.08	2.9±0.08	3.06±0.09
LW (mm)	8.64±0.10	8.56±0.12	8.52±0.20	8.65±0.17	8.47±0.24	8.43±0.14	8.49±0.21	8.52±0.19
WW (mm)	2.98±0.09	2.97±0.05	2.93±0.10	2.97±0.08	2.92±0.07	2.9±0.06	3.01±0.08	2.94±0.09
CI (mm)	2.51±0.36	2.26±0.39	2.25±0.41	2.36±0.53	2.2±0.30	2.36±0.37	2.74±0.61	2.29±0.42
WV-a (mm)	0.53±0.05	0.52±0.05	0.52±0.05	0.52±0.06	0.51±0.03	0.53±0.04	0.55±0.03	0.52±0.05
WV-b (mm)	0.21±0.02	0.23±0.03	0.23±0.03	0.23±0.03	0.24±0.03	0.23±0.02	0.21±0.04	0.23±0.03
J10 (°)	54.69±5.16	51.47±3.38	52.6±3.99	52.09±4.29	50.55±2.97	50.63±3.64	52.25±1.78	52.28±3.96
B4 (°)	105.94±5.31	97.75±5.58	102.44±6.65	102.99±5.11	99.56±5.38	105.19±6.97	106.65±4.07	102.62±6.58
A4 (°)	31.23±1.77	33.81±2.44	31.37±3.10	31.14±2.54	32.5±2.09	31.51±5.77	30.95±1.43	31.53±3.31
E9 (°)	20.96±1.25	17.9±1.29	19.42±1.53	19.84±1.33	19.72±1.23	20.47±1.46	22.08±1.38	19.63±1.62
LTO (mm)	6.01±0.56	5.67±0.65	5.9±0.60	6.01±0.53	6.03±0.58	6.1±0.45	5.7±0.55	5.92±0.58
LG (mm)	3.47±0.49	3.25±0.52	3.41±0.50	3.48±0.41	3.56±0.48	3.55±0.38	3.32±0.44	3.43±0.48
LPR (mm)	0.48±0.06	0.53±0.04	0.48±0.13	0.5±0.05	0.51±0.04	0.51±0.05	0.41±0.09	0.49±0.11
LPM (mm)	2.06±0.10	1.89±0.16	2.01±0.19	2.04±0.14	1.97±0.15	2.04±0.12	1.98±0.11	2.01±0.17

SC: Scutellum color; TC-2: Second abdominal tergit color; TC-4: Fourth abdominal tergit color; LTI: Length of the tibia; LW: Length of the wing; WW: Width of the wing; CI: Cubital index; WV-a: Width of the vein-a; WV-b: Width of the vein-b; J10, B4, A4 and E9: Wing vein angles; LTO: Length of the tongue; LG: Length of the glossa; LPR: Length of the prementum; LPM: Length of the postmentum; pt: Point; mm: Millimeter; °: Angle degree

Table 2. Classes assigned to bee breeds by different classification models

Models		Breed	Predicted 1	2	3	4	5	6	7	Total
SVM	Actual	1	2	0	25	0	0	0	0	27
		2	0	0	23	0	0	0	0	23
		3	0	0	271	0	0	0	0	271
		4	0	0	20	0	0	0	0	20
		5	0	0	21	0	0	0	0	21
		6	0	0	29	0	0	3	0	32
		7	0	0	4	0	0	0	6	10
	Total		2	0	393	0	0	3	6	404
RF	Actual	1	22	0	5	0	0	0	0	27
		2	0	22	1	0	0	0	0	23
		3	0	0	271	0	0	0	0	271
		4	0	0	5	15	0	0	0	20
		5	0	0	3	0	18	0	0	21
		6	0	0	6	0	0	26	0	32
		7	0	0	1	0	0	0	9	10
	Total		22	22	292	15	18	26	9	404
ANN	Actual	1	12	0	15	0	0	0	0	27
		2	1	7	15	0	0	0	0	23
		3	2	2	264	0	2	0	1	271
		4	0	0	18	2	0	0	0	20
		5	0	0	16	0	4	1	0	21
		6	0	0	23	0	2	7	0	32
		7	0	0	2	0	0	0	8	10
	Total		15	9	353	2	8	8	9	404
NB	Actual	1	20	1	2	0	0	0	4	27
		2	0	18	3	1	0	1	0	23
		3	15	29	135	26	26	13	27	271
		4	0	3	1	10	1	2	3	20
		5	0	2	2	1	15	1	0	21
		6	1	2	3	4	5	13	4	32
		7	0	0	0	0	0	0	10	10
	Total		36	55	146	42	47	30	48	404
k-NN	Actual	1	17	0	9	0	0	0	1	27
		2	1	9	13	0	0	0	0	23
		3	7	7	256	0	0	1	0	271
		4	2	1	13	3	0	0	1	20
		5	3	2	14	0	2	0	0	21
		6	1	1	25	0	0	5	0	32
		7	1	0	3	1	0	0	5	10
	Total		32	20	333	4	2	6	7	404

Kekeçoğlu et al. (2023) found that the average J10, B4, A4, and E9 vein angles were 53.63±0.80, 99.63±1.16, 33.78±0.49 and 19.87±0.33, respectively. Cariveau et al. (2018) found that the average LG and LPR were 2.05 mm and 1.57 mm. The results of the current study were found to be generally similar to Gençer and Günbey (2020) and different from Kambur and Kekeçoğlu (2018). However, the current study was found to be higher than Kekeçoğlu et al. (2023) and Cariveau et al. (2018) in terms of B4 vein angle and LG, respectively, and lower than Cariveau et al. (2018) in terms of LPR. Furthermore, Souza et al. (2002) reported that there were statistically significant correlations between tibia width (0.46), LTI (0.55), and LG (0.41) with honey production. Zemskova et al. (2020) stated that the necessity for thorough monitoring of honeybee MTs arises from the declining bee populations and the reduced marketability of apiaries. When the results of the current study are evaluated together with the literature, it becomes evident that identifying and classifying the MTs of bees is important for the sustainability of the bee industry and genetic resources, as well as for increasing productivity per colony.

In addition, the classification of bee breeds was carried out and presented using some data mining algorithms (Table 2). A matrix showed how each model classified the different bee breeds (numbered 1 to 7). In these matrices, the actual and model-predicted breed class is in the rows and columns, respectively. The values in each cell of the matrix represent the frequency with which the model predicted each actual bee breed class as a certain class. For instance, in the case of the SVM model, out of the bees that truly belonged to breed class "1", the model predicted "1" for two bees and "3" for twenty-five bees. Comparing the number of samples correctly assigned to each class shows the classification success of the models. This table can be considered as an accuracy matrix that shows how accurately different data mining algorithms classify bee breeds. This analysis can be used to determine which model is more effective in automatically classifying bee breeds.

After the classification analysis, model fit criteria were used to evaluate the success of the models. The evaluation metrics for comparison of the models are presented in Table 3. Table 3 compares the performance metrics of various machine learning models SVM, RF, ANN, NB, and k-NN by their AUC, CA, F1, Precision, Recall and Specificity values.

In addition, Figure 2 is provided to facilitate a deeper comprehension of the classification performance of the models. Figure 2 indicate the models' performances and predictions. RF model is found to be most efficient, with the highest AUC values of 0.998, indicating superior ability in distinguishing between breeds. Additionally, it has the CA of 0.948 and an impressive F1 score of 0.946, highlighting its balanced precision and recall. The excellence of this model indicates its robustness in tackling complex classification tasks in beekeeping research. Conversely, NB model is identified as the least effective model, as evidenced by the lowest CA of 0.547 and a modest AUC of 0.853. Despite its simplicity and ease of implementation, the performance of the NB model highlights limitations in addressing the intricacies of bee classification, particularly in scenarios that require fine-grained differentiation between similar species. The SVM and k-NN models exhibit commendable performance with AUC values of 0.950 and 0.919, respectively, positioning them as viable alternatives for certain applications. However, they do not exceed the overall effectiveness of the RF model. Although the ANN model shows potential with an AUC of 0.905, it falls short of expectations compared to RF in terms of CA and F1 score, suggesting that further optimization is required to improve classification accuracy.

Table 3. The evaluation metrics used for model comparison

Model	AUC	CA	F1	Precision	Recall	Specificity
SVM	0.950	0.698	0.589	0.633	0.698	0.385
RF	0.998	0.948	0.946	0.952	0.948	0.894
ANN	0.905	0.752	0.703	0.766	0.752	0.549
NB	0.853	0.547	0.584	0.744	0.547	0.922
k-NN	0.919	0.735	0.688	0.750	0.735	0.607

AUC: Criterion for measuring the performance of classification models; CA: Criterion for measuring the accuracy of classification models; F1: Harmonic mean of a classification model's precision and recall measurements; Precision: Measures the ratio of truly positive predictions to the overall prediction; Recall: Criterion indicates how accurately the model detects true positives; Specificity: Criterion indicates how accurately the model detects true negatives

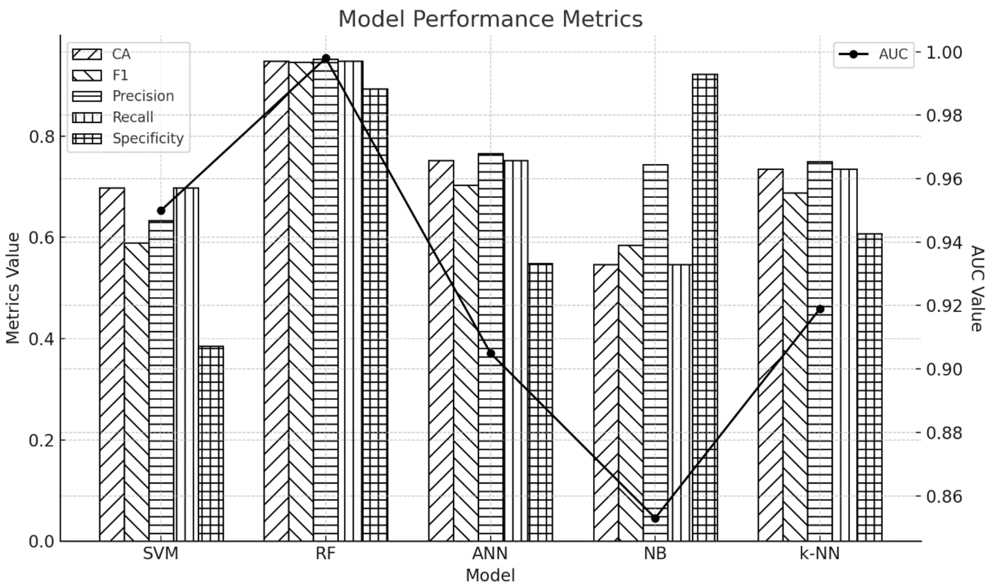

Figure 2. Evaluation of machine learning models in bee classification

AUC represents the area under the ROC curve, indicating the balance between sensitivity and specificity of a classification model. A higher AUC value, closer to 1, signifies a better balance and, consequently, a more accurate classification. Among these models, RF exhibited the highest AUC value (0.998), indicating its superior performance in classifying bee breeds based on their MTs. Conversely, NB demonstrated the lowest AUC value (0.853), suggesting a comparatively lower accuracy in classifying bee breeds. These results highlight that the RF was the most effective model for classifying bee breeds. CA calculates the ratio of correct predictions to the total number of samples and could be misleading for imbalanced class data. Since CA considers total correct predictions rather than distinguishing between classes, it can lead to erroneous interpretations. These values indicate that, in classifying bee breeds based on their MTs, RF demonstrated superior accuracy, outperforming other models, whereas NB exhibited comparatively lower accuracy. In this study, despite the CA determined by the SVM model being lower than the ANN model, the AUC value was found to be higher for the ANN model. This situation underscores the importance of not relying solely on CA as a singular criterion for evaluation. Although the SVM model made fewer correct predictions, it better explained the differences between bee breeds. This observation highlights that the performance of the model is not solely contingent on the number of correct predictions. The F1 score is a balanced measure that takes precision and recall into account and is calculated using the harmonic mean of these two metrics. A high F1 score plays a crucial role in distinguishing true positives (correct identification of the MTs under consideration) from true negatives (correct exclusion of irrelevant MTs). When false positives (misclassifying non-relevant traits as relevant) and false negatives (misclassifying relevant traits as not relevant) are of equal importance, the F1 score becomes a significant metric. In the context of beekeeping, the F1 score is of great importance, especially in applications such as selection processes that influence the efficiency of bee breed. According to the F1 score, RF exhibited the highest classification accuracy, while NB demonstrated the lowest performance among the models. Therefore, when it is essential to precisely differentiate between the MTs of bee breeds, choosing the model with the highest F1 score is crucial. The precision metric signifies the proportion of samples predicted as positive (possessing a specific feature) by the model that are actually positive. This measure holds particular importance in scenarios where false positive predictions (predicting a feature that is not actually present as present) carry significant implications. In the context of apiculture, when selecting colonies based on the MTs of disease-resistant bee breeds, misclassifying non-disease-resistant bees as disease-resistant (resulting in false positive predictions) can markedly diminish the efficacy of the selection process. Consequently, RF exhibited the highest accuracy in identifying colonies with the desired MTs, whereas SVM demonstrated the lowest performance in this aspect. The recall metric indicates the ratio of true positives (correctly identified traits) to all actual positives. If identifying the presence of a specific trait (false negative prediction) holds significance, the recall value becomes crucial. For instance, in apiculture, when selecting hygienic colonies based on MTs, including non-hygienic colonies in the selection can increase the colony's susceptibility to diseases, making false negative predictions important in this context. Therefore, it is essential to consider a model with a high recall value. Consequently, the RF model, with the highest recall value, will be more effective in selecting hygienic colonies. The NB model, which yielded the lowest recall value, should not be considered due to the high likelihood of misclassifying non-hygienic colonies as hygienic ones. Significant differences have been found in the data mining algorithms used to classify breeds of bees based on their MTs. When all criteria used in the classification were evaluated together, it was found that the RF model achieved the highest values for all criteria and performed the best classification. The RF model is followed by ANN. On the other hand, the lowest ranking was generally achieved in the NB model, which consistently had the lowest values. The RF model has demonstrated superior performance in key metrics such as AUC, CA, F1 score, precision, and recall, establishing itself as an effective method for classifying bee breeds. However, other algorithms such as SVM, ANN, NB, and k-NN have shown lower and varying degrees of success in classification compared to the RF model. Rodrigues et al. (2022) reported that the SVM algorithm classified the forewings with an average accuracy of 86.6±6.9% across the 26 subspecies represented in the test data set, although the number of images used for classification was small and many of them were low quality. The same researchers found classification accuracy of 98.9%, 97.7%, 91.1%, 96.4%, and 95% for the Carniolan, Caucasian, Iberiensis (Spanish bee), Italian bee, and Mellifera subspecies, respectively. In the current study, the RF model showed the highest accuracy in classifying bees and was different from Rodrigues et al. (2022).

4. Conclusions

As a result, RF model is the most effective tool for classifying bee breeds within the evaluated dataset and offers a promising approach for researchers seeking to improve the sustainability and productivity of beekeeping through genetic and morphological analyses. While NB model is valuable for its computational efficiency, it requires careful application in this context due to its comparatively low performance. In addition, the present study demonstrates the possible applications and limitations of each algorithm in the context of classifying bee breeds based on their MTs.

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References

Abou-Shaara HF (2013). Wing venation characteristics of honey bees. *Journal of Apicultural* 28: 79-86.

Abou-Shaara HF, Al-Ghamdi AA, Mohamed AA (2013). Body morphological characteristics of honey bees. *Agricultura,* 10: 45-49.

Alpatov WW (1929). Biometrical studies on variation and races of the honey bee (*Apis mellifera L.*). *The Quarterly Review of Biology,* 4: 1-58. https://www.jstor.org/stable/2808231

Anderson LE (1954). Hoyer's solution as a rapid permanent mounting medium for bryophytes. *The Bryologist,* 57: 242-244. https://doi.org/10.2307/3240091

Antony JC, Pratheepa M (2018). A Bayesian classification approach for predicting Gesonia gemma Swinhoe population on soybean crop in relation to abiotic factors based on economic threshold level. *Journal of Biological Chemistry,* 32: 68-73. https://doi.org/10.18311/jbc/2018/16309

Berlocher SH (1984). Insect molecular systematics. *Annual Review of Entomology,* 29: 403-433. https://doi.org/10.1146/annurev.en.29.010184.002155

Bhavsar H, Ganatra A (2012). A comparative study of training algorithms for supervised machine learning. *International Journal of Soft Computing and Engineering,* 2: 74-81.

Breiman L (2001). Random forests. *Journal of Machine Learning Research,* 45: 5-32. https://doi.org/10.1023/A:1010933404324

Buco SM, Rinderer TE, Sylvester HA, Collins AM, Lancaster VA, Crewe RM (1987). Morphometric differences between South American Africanized and South African (*Apis mellifera scutellata*) honey bees. *Apidologie,* 18: 217-222. https://doi.org/10.1051/apido:19870301

Cariveau DP, Nayak GK, Bartomeus I, Zientek J, Ascher JS, Gibbsand J, Winfree R (2018). The allometry of bee proboscis length and its uses in ecology. *PloS One,* 13: Article e0207900. https://doi.org/10.1371/journal.pone.0207900

Cover T, Hart P (1967). Nearest neighbor pattern classification. *IEEE Transactions on Information Theory,* 13: 21-27. https://doi.org/10.1109/TIT.1967.1053964

Crewe RM, Hepburn HR, Moritz RFA (1994). Morphometric analysis of 2 southern African races of honey bee. *Apidologie,* 25: 61-70. https://doi.org/10.1051/apido:19940107

Demsar J, Curk T, Erjavec A, Gorup C, Hocevar T, Milutinovic M, Mozina M, Polajnar M, Toplak M, Staric A, Stajdohar M, Umek L, Zagar L, Zbontar J, Zitnik M, Zupan B (2013). Orange: Data Mining Toolbox in Python. *Journal of Machine Learning Research,* 14: 2349–2353.

Diniz-Filho JAF, Malaspina O (1995). Evolution and population structure of Africanized honey bees in Brazil: Evidence from Spital analysis of morphometric data. *Evolution,* 49: 1172-1179. https://doi.org/10.1111/j.1558-5646.1995.tb04444.x

Dodoloğlu A (2000). Kafkas ve Anadolu balarısı (*Apis smellifera L.*) ırkları ile karşılıklı melezlerinin morfolojik, fizyolojik ve davranış özellikleri (Publication No. 96437). PhD Thesis, Atatürk University (Unpublished), Türkiye.

Estoup A, Garnery L, Solignac M, Cornuet JM (1995). Microsatellite variation in honey bee (*Apis mellifera L.*) populations: hierarchical genetic structure and test of the infinite allele and stepwise mutation models. *Genetics,* 140: 679-695. https://doi.org/10.1093/genetics/140.2.679

Frunze O, Kim DW, Kang EJ, Kim K, Park BS, Choi YS (2022). The accuracy of morphometric characteristic analysis depends on the type of the assessed traits of honey bees (*Apis cerana F.* and *Apis mellifera L.*). *Journal of Asia-Pacific Entomology,* 25: Article 101991. https://doi.org/10.1016/j.aspen.2022.101991

Ftayeh A, Meixner M, Fuchs S (1994). Morphometrical investigation in Syrian honey bees. *Apidologie,* 25: 396-401. https://doi.org/10.1051/apido:19940406

Gençer HV (2004). The graphic evaluation of morphological characters in honey bees (*Apis mellifera L.*) by Chernoff Faces. *Journal of Agricultural Sciences,* 10: 245-249. https://doi.org/10.1501/Tarimbil_0000000901

Gençer HV, Günbey B (2020). The morphological characteristics of distinctive honey bee (*Apis mellifera L.*) genotypes in Black Sea Region. *Journal of Animal Science and Products,* 3: 40-53.

Guler A, Bek Y (2002). Forewing angles of honey bee (*Apis mellifera*) samples from different regions of Turkey. *Journal of Apicultural Research,* 41: 43-49. https://doi.org/10.1080/00218839.2002.11101067

Han J, Pei J, Tong H (2022). Data mining: concepts and techniques. Morgan kaufmann. Amsterdam, Elsevier, p.735.

Jadhav SD, Channe HP (2016). Comparative study of K-NN, naive Bayes and decision tree classification techniques. *International Journal of Science and Research,* 5: 1842-1845. https://www.ijsr.net/getabstract.php?paperid=NOV153131

Kambur M, Kekeçoğlu M (2018). The current situation of Turkey Honey Bee (*Apis mellifera L.*) biodiversity and conservations studies. *Biological Diversity and Conservation,* 11: 105-119. https://doi.org/10.13140/RG.2.2.12203.54568

Kekeçoğlu M, Bir S, Acar MK (2023). Determination of Anatolian honeybee biodiversity by wing characters. *Sociobiology,* 70: Article e8333. https://doi.org/10.13102/sociobiology.v70i3.8333

Khan SN, Khan SU, Aznaoui H, Şahin CB, Dinler ÖB (2023). Generalization of linear and non-linear support vector machine in multiple fields: a review. *Computer Science and Information Technology,* 4: 226-239. https://doi.org/10.11591/csit.v4i3.p226-239

Li F, Xiong Y (2018). Automatic identification of butterfly species based on HoMSC and GLCMoIB. *The Visual Computer,* 34: 1525-1533. https://doi.org/10.1007/s00371-017-1426-1

Liaw A, Wiener M (2002). Classification and regression by random Forest. *R News,* 2: 18-22. http://CRAN.R-project.org/doc/Rnews/

Özbakır GÖ (2011). Morphological characterization of honey bee populations (*Apis mellifera L.*) along the southeast border of Turkey (Publication No. 299725). PhD Thesis, Ankara University(Unpublished), Türkiye.

Pan X, Wang Y, Qi Y (2023). Artificial neural network model and its application in signal processing. *Asian Journal of Advanced Research and Reports,* 17: 1-8. https://doi.org/10.9734/AJARR/2023/v17i1459

Potts SG, Biesmeijer JC, Kremen C, Neumann P, Schweiger O, Kunin WE (2010). Global pollinator declines: trends, impacts and drivers. *Trends in Ecology and Evolution,* 25: 345-353. https://doi.org/10.1016/j.tree.2010.01.007

Rinderer TE, Buco SM, Rubink WL, Daly HV, Stelszer JA, Rigio RM, Baptista C (1993). Morphometric identification of Africanized and European honey bees using large reference populations. *Apidologie,* 24: 569-585. https://doi.org/10.1051/apido:19930605

Rodrigues PJ, Gomes W, Pinto MA (2022). DeepWings©: automatic wing geometric morphometrics classification of honey bee (*Apis mellifera*) subspecies using deep learning for detecting landmarks. *Big Data and Cognitive Computing,* 6: 70. https://doi.org/10.3390/bdcc6030070

Ruttner F, Tassencourt L, Louveaux J (1978). Biometrical-statistical analysis of the geographic variability of *Apis mellifera L.* I. Material and methods. *Apidologie,* 9: 363-381. https://doi.org/10.1051/apido:19780408

Souza D, Cruz CD, Campos L, Regazzi AJ (2002). Correlation between honey production and some morphological traits in africanized honey bees (*Apis melifera*). *Ciência Rural,* 32: 869-872. https://doi.org/10.1590/S0103-84782002000500020

Szymula J, Skowronek W, Bienkowska M (2010). Use of various morphological traits measured by microscope or by computer methods in the honeybee taxonomy. *Journal of Apicultural Science,* 54: 91-97.

Tapkan P, Özbakır L, Kulluk S, Baykasoğlu A (2016). A cost-sensitive classification algorithm: BEE-Miner. *Knowledge-Based Systems,* 95: 99-113. https://doi.org/10.1016/j.knosys.2015.12.010

Trevethan R (2017). Sensitivity, specificity, and predictive values: foundations, pliabilities, and pitfalls in research and practice. *Frontiers in Public Health,* 5: 307. https://doi.org/10.3389/fpubh.2017.00307

Vapnik V (1999). The nature of statistical learning theory. *Springer Science and Business Media*, pp.314. https://doi.org/10.1007/978-1-4757-3264-1

Wickramasinghe I, Kalutarage H (2021). Naive Bayes: applications, variations and vulnerabilities: a review of literature with code snippets for implementation. *Soft Computing,* 25: 2277-2293. https://doi.org/10.1007/s00500-020-05297-6

Yegnanarayana B (1999). Artificial neural networks. PHI Learning Pvt. Ltd.: New Delhi, India, pp. 476.

Zemskova NE, Sattarov VN, Skvortsov AI, Semenov VG (2020). Morphological characteristics of honey bees of the Volga region. BIO Web Conf 17, 35. EDP Sciences. https://doi.org/10.1051/bioconf/20201700035

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Comparative Analysis of Empirical and AI-Supported Models in Global Solar Radiation Prediction for İzmir Province

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HIGHLIGHTS

- Model 15 achieved the highest accuracy with RMSE: 0.1451 and R^2: 0.9995.
- AI-supported models outperformed 14 traditional empirical models.
- The study emphasizes İzmir's solar energy potential of 1611.5 kWh·m^{-2}·year^{-1}.
- Hybrid models adapt better to İzmir's unique microclimatic features.

Abstract

In this study, the performances of different models that can be used to predict global solar radiation for İzmir province were analyzed comparatively. Using ATATEK-Solar software, 14 empirical models commonly used in the literature and a newly developed AI-supported model were tested. Each model was analyzed using three different optimization algorithms (Nelder-Mead Simplex, Pattern Search, Simulated Annealing). Long-term average meteorological data obtained from Turkish State Meteorological Service were used. According to the analysis results, Model 15 performed the most successful predictions with RMSE:0.1451 and R^2:0.9995 values. This was followed by Model 5 with RMSE:0.2016 and R^2:0.9990 values and Model 6 with RMSE:0.2017 and R^2:0.9990 values. When model performances were examined on a monthly basis, it was observed that the lowest prediction errors occurred in spring and summer months. As a result of the study, it is recommended to use Model 15 in evaluating the solar energy potential of İzmir province and system design.

Keywords: Solar energy; Global solar radiation; Empirical models; Artificial intelligence; İzmir

1. Introduction

The continuous increase in global energy demand and efforts to combat climate change have heightened interest in renewable energy sources. Solar energy stands out as a sustainable energy solution with low carbon emissions and high potential (Külcü and Ersan 2021). Owing to its geographical location, Turkey possesses significant solar energy potential, with a long-term annual average of 2741 hours of sunshine and a mean total global solar radiation value of 1527.46 kWh·m^{-2}·year^{-1} (Türkiye Enerji Bakanlığı 2024).

Accurate prediction of global solar radiation is critically important for designing and evaluating the performance of solar energy systems. Due to atmospheric conditions, geographical characteristics, and

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climatic factors, regional variations in radiation predictions are observed. Therefore, it is essential to determine suitable prediction models for each region (Almorox et al. 2013). Considering the installation and operational costs of solar observation equipment, the regional assessment of prediction models is gaining increasing importance (Süslü and Külcü 2024).

Located at 38.43° N latitude and 27.17° E longitude, İzmir is an Aegean city with an annual average sunshine duration of 7.92 hours. The region's microclimatic features, coastal proximity, and topographic structure influence the distribution of solar radiation. While these measurements represent average values from the central meteorological station, the solar energy potential shows significant variation across the province. According to a report by İzmir Development Agency (2021), the solar energy potential in İzmir reaches 1750 $kWh{\cdot}m^{-2}{\cdot}year^{-1}$ in southern districts, while it hovers around 1500 $kWh{\cdot}m^{-2}{\cdot}year^{-1}$ in northern districts. These variations are attributed to local geographical and climatic characteristics.

Various empirical models have been developed in the literature for predicting global solar radiation. These models generally rely on parameters such as sunshine duration, temperature differences, and geographical data. In recent years, alongside traditional empirical models, new models supported by artificial intelligence (AI) techniques have been proposed. These approaches aim to enhance prediction accuracy by combining traditional methods with modern optimization techniques (Ertürk et al. 2023).

In recent studies, Süslü (2024) compared different empirical models for global solar radiation prediction in Turkey's Lakes Region using the ATATEK-Solar software. In the study, 15 different models were tested with three different optimization algorithms, and Model 13 was determined to provide the most suitable results for regional predictions (Süslü, 2024). This finding aligns with the results of the current study conducted for İzmir and supports the notion that AI-supported models offer higher accuracy than traditional empirical models.

In this study, 15 different models for predicting global solar radiation in İzmir province were comparatively analyzed. The analyses were performed using ATATEK-Solar software, where each model was solved using three different optimization algorithms: Nelder-Mead Simplex, Pattern Search, and Simulated Annealing. The study's objective is to identify the most suitable prediction model by considering İzmir's unique climatic and geographical features. The results obtained will provide a reliable foundation for the design and performance evaluation of solar energy systems in the region.

2. Materials and Methods

2.1. Study Area and Dataset

This study was conducted for İzmir province, located in western Turkey (38.43° N, 27.17° E). Situated at an average elevation of 32 meters above sea level, İzmir encompasses a geography characterized by diverse microclimatic conditions (Figure 1). The region is dominated by a Mediterranean climate, with hot and dry summers and mild, rainy winters. İzmir has a total area of 12,012 km², consisting of 11 central districts and 19 peripheral districts. The annual average temperature of the city is 18.2°C, with an annual total precipitation of 695.9 mm.

Figure 1. The location of İzmir on the map of Turkey.

The meteorological data used in this study were obtained from the Turkish State Meteorological Service. The dataset includes the following parameters:

- Monthly average sunshine duration (hours)
- Monthly average temperature (°C)
- Maximum and minimum temperature difference (°C)
- Monthly total global solar radiation ($MJ \cdot m^{-2} \cdot day^{-1}$)
- Theoretical sunshine duration (hours)
- Extraterrestrial radiation values ($MJ \cdot m^{-2} \cdot day^{-1}$)

The climatic characteristics of İzmir are summarized in Table 1. The annual average sunshine duration was determined as 7.92 $hours \cdot day^{-1}$, the average temperature as 18.2°C, and the average global solar radiation as 16.05 $MJ \cdot m^{-2} \cdot day^{-1}$.

Table 1. Long-term average climatic data for İzmir province

Month	*Sunshine Duration (hours)*	*ΔT (°C)*	*Global Radiation ($MJ \cdot m^{-2} \cdot day^{-1}$)*
January	4.34	6.6	7.62
February	5.04	7.5	10.40
March	6.49	8.6	14.71
April	7.52	9.7	18.36
May	9.74	10.6	22.27
June	11.74	10.8	25.08
July	12.19	10.8	24.69
August	11.71	10.6	22.30
September	10.08	10.5	18.73
October	7.49	9.4	13.31
November	5.43	7.9	8.74
December	4.02	6.5	6.35

The long-term monthly average variation of İzmir's sunshine duration and global radiation values is shown in Figure 2. The highest average sunshine duration is observed in July (12.19 $hours \cdot day^{-1}$), while the lowest is in December (4.02 $hours \cdot day^{-1}$). Average global radiation values reach a maximum in June (25.08 $MJ \cdot m^{-2} \cdot day^{-1}$) and a minimum in December (6.35 $MJ \cdot m^{-2} \cdot day^{-1}$).

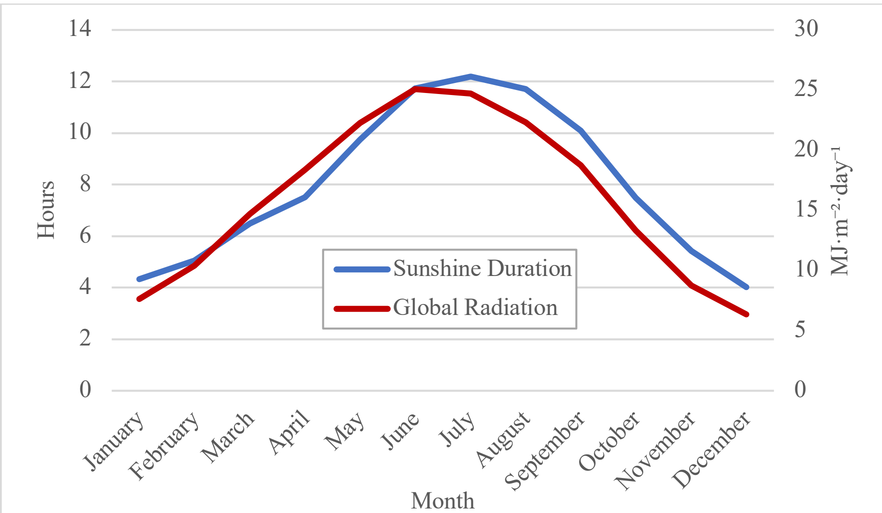

Figure 2. Monthly variation of sunshine duration and global radiation values in İzmir province

Based on the values calculated using meteorological observation data, the annual total global solar radiation in İzmir was determined to be 5801.4 $MJ \cdot m^{-2} year^{-1}$. This value is above the national average of Turkey (5498.8 $MJ \cdot m^{-2} year^{-1}$), indicating the high solar energy potential of the region.

2.2. Global Solar Radiation Prediction Models

In this study, 15 different models were analyzed. As shown in Table 2, these models are categorized into three main groups based on their mathematical structure and the parameters they utilize:

- **Models Based on Sunshine Duration (Models 1–9)**

These models estimate global solar radiation primarily using sunshine duration data. They are widely used due to their simplicity and reliance on easily accessible meteorological data.

- **Advantages:**

Require minimal input data (only sunshine duration and extraterrestrial radiation).

Suitable for locations where temperature and humidity records are not available.

Computationally efficient and easy to implement.

- **Disadvantages:**

Accuracy is highly dependent on sunshine duration records, which may not always be reliable.

Performance decreases in regions with frequent cloud cover or sudden weather changes.

- **Models Based on Temperature Data (Models 10–12)**

These models use temperature-based parameters, such as maximum-minimum temperature differences, to estimate solar radiation.

- **Advantages:**

Useful in regions where sunshine duration data is unavailable.

Can capture seasonal variations in solar radiation better than sunshine duration-based models.

- **Disadvantages:**

Less accurate in regions where temperature variations are not strongly correlated with solar radiation.

Performance may be affected by microclimatic conditions and elevation differences.

- **Hybrid Models (Models 13–15)**

Hybrid models integrate multiple meteorological variables, such as sunshine duration, temperature, and atmospheric parameters, often using advanced computational techniques like AI.

- **Advantages:**

Provide higher accuracy by considering multiple influencing factors.

More adaptable to varying climatic and geographical conditions.

AI-supported models can improve prediction performance over time with additional data.

- **Disadvantages:**

Require more complex calculations and computational power.

Depend on the availability and quality of multiple meteorological inputs.

Table 2. Models used in the study and their mathematical expressions

No	Model Expression	References
1	$\frac{H}{H_0} = c_1 + c_2\left(\frac{S}{S_0}\right)$	Angstrom (1924); Prescott (1940)
2	$\frac{H}{H_0} = c_1 + c_2\left(\frac{S}{S_0}\right)^{c_3}$	Elagib and Mansell (2000)
3	$\frac{H}{H_0} = c_1^{\left(\frac{1}{S}\right)}$	El-Metwally (2005)
4	$\frac{H}{H_0} = \left[\frac{c_1\left(\frac{S}{S_0}\right)}{c_2 w_s}\right] + c_3 w_s$	Külcü (2015)
5	$\frac{H}{H_0} = c_1 + c_2\left(\frac{S}{S_0}\right) + c_3\left(\frac{S}{S_0}\right)^2 + c_4\left(\frac{S}{S_0}\right)^3$	Bahel et al. (1987)
6	$\frac{H}{H_0} = c_1 + c_2\left(\frac{S}{S_0}\right) + c_3 log\left(\frac{S}{S_0}\right)$	Ampratwum and Dorvlo (1999)
7	$\frac{H}{H_0} = c_1 + c_2 exp\left(\frac{S}{S_0}\right)$	Almorox and Hontoria (2004)
8	$\frac{H}{H_0} = c_1 + \left[c_2\left(\frac{S}{S_0}\right) + c_3\right]\varphi + c_3\left(\frac{S}{S_0}\right)$	Dogniaux and Lemoine (1983)
9	$\frac{H}{H_0} = c_1 + c_2 log\left(\frac{\frac{S}{S_0}}{w_s}\right) + c_3\left(\frac{S}{S_0}\right)$	Külcü (2019)
10	$\frac{H}{H_0} = c_1(\Delta T)^{0.5} + c_2$	Hargreaves et al. (1985)
11	$\frac{H}{H_0} = c_1 ln(\Delta T) + c_2$	Coppolino (1994)
12	$\frac{H}{H_0} = c_1[1 - exp - c_2(\Delta T)^{c_3}]$	Bristow and Campbell (1984)
13	$\frac{H}{H_0} = c_1 log\left[\left(c_2\frac{S}{S_0}\right) + (c_3\Delta T)\right] + c_4$	Ersan and Külcü (2024)
14	$\frac{H}{H_0} = c_1 log[(c_2 w_s) + (c_3\Delta T)] + c_4$	Ersan and Külcü (2024)
15	$\frac{H}{H_0} = c_1\left(\frac{S}{S_0}w_s\right)^{c_2} + c_3 log_{10}(1 + \Delta T) + c_4 sin(\varphi) cos\left(\frac{2\pi n}{365}\right) + c_5$	Süslü and Külcü (2024)

Where;

H : Daily global solar radiation reaching the Earth's surface (MJ·m^{-2}·day^{-1})

H_0 : Extraterrestrial radiation (MJ·m^{-2}·day^{-1})

S : Daily sunshine duration (hours)

S_0 : Theoretical sunshine duration (hours)

ΔT : Daily maximum and minimum temperature difference (°C)

w_s : Sunset hour angle

φ : Latitude angle

n : Day of the year (1–365)

c_1, c_2, c_3, c_4, c_5 : Model coefficients

The daily extraterrestrial solar radiation (H_0) is calculated using the following equation (Duffie and Beckman 2006):

$$H_0 = \frac{24x3600xG_{sc}}{\pi}\left[1 + 0.033cos\left(\frac{360n}{365}\right)\right]\left[cos\varphi cos\delta sinw_s + \frac{\pi}{180}w_s sin\varphi sin\delta\right] \quad (1)$$

Where;

G_{sc} : Solar constant (1367 W·m⁻²)

δ : Declination angle

w_s : Sunset hour angle

Declination Angle (δ)

$$\delta = 23.45sin\left[360\left(\frac{284 + n}{365}\right)\right] \quad (2)$$

Sunset Hour Angle (w_s)

$$w_s = arccos[-tan(\varphi)tan(\delta)] \quad (3)$$

2.3. Optimization Methods

Three different optimization algorithms provided by the ATATEK-Solar software were used to determine the model coefficients:

- **Nelder-Mead Simplex Algorithm:** Developed by Nelder and Mead (1965), this method is widely used for solving nonlinear optimization problems.
- **Pattern Search Algorithm:** Proposed by Hooke and Jeeves (1961), this is a fundamental approach among direct search methods that do not require derivatives.
- **Simulated Annealing Algorithm:** Introduced by Kirkpatrick et al. (1983), this stochastic optimization method is inspired by the annealing process in metallurgy.

Coefficient optimization for each model was performed separately using these three methods, and the results with the lowest RMSE value were selected.

2.5. Statistical Analysis

The performance of the models was evaluated using the following statistical parameters:

Coefficient of Determination (R^2):

$$R^2 = \frac{\sum_{i=1}^{N}(H_{ip} - H_{ipa})(H_{io} - H_{ioa})}{\sqrt{\left[\sum_{i=1}^{N}(H_{ip} - H_{ipa})^2\right]\left[\sum_{i=1}^{N}(H_{io} - H_{ioa})^2\right]}} \quad (4)$$

Root Mean Square Error ($RMSE$):

$$RMSE = \sqrt{\frac{1}{N}\sum_{i=1}^{N}(H_{ip} - H_{io})^2} \quad (5)$$

Mean Percentage Error (MPE):

$$MPE = \frac{1}{N}\sum_{i=1}^{N}\frac{H_{ip} - H_{io}}{H_{io}} \times 100 \quad (6)$$

Mean Absolute Error (MAE):

$$MAE = \frac{1}{N}\sum_{i=1}^{N}\left|H_{ip} - H_{io}\right| \tag{7}$$

Where:

H_{ip}: Predicted value

H_{io}: Observed value

H_{ipa}: Mean of predicted values

H_{ioa}: Mean of observed values

N: Number of data points

When evaluating model performances, the $RMSE$ value was primarily considered. For models with equal $RMSE$ values, the R^2 value was used for comparison.

3. Results and Discussion

The performance of each model was evaluated based on multiple statistical metrics, including RMSE, R^2, and MPE, to ensure a comprehensive assessment of their predictive capabilities. The analysis was conducted across different time scales, considering both annual and seasonal variations in solar radiation.

To better understand the impact of optimization algorithms on model accuracy, the performance of each model was compared under three different optimization approaches: Nelder-Mead Simplex, Pattern Search, and Simulated Annealing. The results indicated that while the Nelder-Mead Simplex algorithm yielded optimal solutions for most models, the Pattern Search algorithm was particularly effective for Model 15, which demonstrated the highest predictive accuracy.

Furthermore, the monthly and seasonal performance of the top three models (Models 15, 5, and 6) was examined in detail. The analysis revealed that prediction errors were lower during spring and summer months, while higher deviations were observed in winter. This can be attributed to increased cloud cover and atmospheric variations in colder months, which introduce additional uncertainties into radiation modeling.

Another critical aspect of the evaluation was the comparison between empirical and AI-supported models. The findings demonstrated that AI-enhanced models, particularly Model 15, outperformed traditional empirical models by incorporating multiple climatic and geographical parameters into their predictive framework. This suggests that hybrid modeling approaches, integrating empirical equations with advanced computational techniques, can significantly improve the accuracy of solar radiation predictions, especially in regions with complex microclimatic conditions like İzmir.

In Süslü (2024)'s study, the results of 15 models tested with different optimization methods were compared. While Model 15 was identified as the most successful model in the study conducted for İzmir, Model 13 was determined to be the best model for the Lakes Region. This difference highlights the significant impact of regional microclimate and geographical factors on global solar radiation prediction.

3.1. Analysis of Model Performance

The performance metrics of all models analyzed for İzmir province are summarized in Table 3.

According to the analysis results, Model 15 achieved the most accurate predictions with RMSE: 0.1451, R^2: 0.9995, and MPE: 0.08. This was followed by Model 5 with RMSE: 0.2016, R^2: 0.9990, and MPE: 0.15, and Model 6 with RMSE: 0.2017, R^2: 0.9990, and MPE: 0.10. The lowest performance was observed in Model 3, with RMSE: 1.8284, R^2: 0.9208, and MPE: -8.34.

Table 3. Performance metrics and optimization methods of the models

Model No	RMSE	R^2	MPE	MAE	Best Method
15	0.1451	0.9995	0.08	0.1182	Pattern Search
5	0.2016	0.9990	0.15	0.1756	Nelder-Mead Simplex
6	0.2017	0.9990	0.10	0.1842	Nelder-Mead Simplex
13	0.2165	0.9989	0.12	0.1980	Nelder-Mead Simplex
2	0.2232	0.9988	0.37	0.1861	Nelder-Mead Simplex
1	0.2824	0.9981	0.71	0.2297	Nelder-Mead Simplex
8	0.2824	0.9981	0.71	0.2297	Pattern Search
9	0.3070	0.9978	1.02	0.2546	Nelder-Mead Simplex
7	0.3369	0.9973	0.98	0.2891	Nelder-Mead Simplex
4	0.4428	0.9954	-1.72	0.3939	Nelder-Mead Simplex
10	0.6243	0.9908	-0.47	0.4784	Nelder-Mead Simplex
12	0.6311	0.9906	-0.51	0.4822	Nelder-Mead Simplex
11	0.6362	0.9904	-0.50	0.4854	Nelder-Mead Simplex
14	0.6364	0.9904	-0.50	0.4856	Nelder-Mead Simplex
3	1.8284	0.9208	-8.34	1.6692	Nelder-Mead Simplex

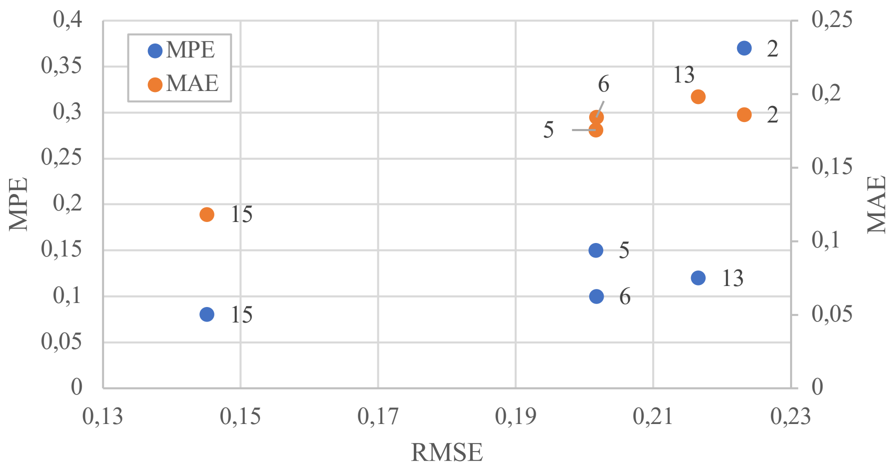

Figure 4. Comparison of the top 5 models' performances based on RMSE, MAE, and MPE values.

From the perspective of optimization algorithms, the Nelder-Mead Simplex algorithm provided the best results for 13 models, while the Pattern Search algorithm performed better for Models 15 and 8. This can be attributed to the more complex structure of Model 15 and the Pattern Search algorithm's ability to avoid local minima.

3.2. Monthly Performance Evaluation

The monthly prediction performances of the top three models are compared in Table 4.

The monthly performance analysis of the top three models shows that all models performed better in spring and summer months. The mean absolute error (MAE) of the predicted and observed values was calculated as follows:

- Model 15: 0.1182 $MJ \cdot m^{-2} \cdot day^{-1}$
- Model 5: 0.1756 $MJ \cdot m^{-2} \cdot day^{-1}$
- Model 6: 0.1842 $MJ \cdot m^{-2} \cdot day^{-1}$

Table 4. Monthly relative error values (%) for the top three models

Month	Model 15	Model 5	Model 6
January	-1.87	-1.50	-1.82
February	-0.17	-1.68	-1.69
March	-0.39	-1.66	-1.40
April	0.09	0.20	0.43
May	0.59	1.46	1.32
June	-0.89	-0.51	-0.68
July	-0.31	0.05	0.09
August	1.41	0.85	1.05
September	-0.29	-1.54	-1.63
October	-0.96	-0.84	-0.95
November	0.90	1.83	2.10
December	2.80	5.09	4.35

Table 5. Seasonal Performance Summary:

	MAE			
	Spring (Mar-May)	**Summer (Jun-Aug)**	**Autumn (Sep-Nov)**	**Winter (Dec-Feb)**
Model 15	0.0688	0.2044	0.0871	0.1125
Model 5	0.2021	0.1100	0.1862	0.2042
Model 6	0.1927	0.1425	0.2048	0.1968

In conclusion, Model 15 demonstrates the highest reliability across all seasons due to its consistently lower error rates, while Model 5 performs better during the summer months but exhibits lower overall accuracy. This highlights the superiority of Model 15, which benefits from comprehensive data integration and advanced optimization techniques.

3.3. Evaluation of Models Specific to İzmir Province

Seasonal differences in model performance are closely related to the unique climatic characteristics of İzmir. Particularly, the microclimatic conditions created by the coastal influence and topographic structure significantly affect prediction accuracy. The following key findings were obtained:

- Models Based on Sunshine Duration (Models 1–9):
    - Provide more consistent results during summer months.
    - Perform better in coastal regions where the marine influence is stronger.
    - Accuracy improves during periods with low cloud cover.
- Models Based on Temperature Data (Models 10–12):
    - Perform better during transitional seasons when temperature differences are more pronounced.
    - Are influenced by the temperature gradient, which increases from coastal to inland areas.
    - Experience reduced accuracy during periods with high humidity.
- Hybrid Models (Models 13–15):
    - Better adapt to seasonal variations by combining different parameters.
    - Model 15, in particular, captures İzmir's microclimatic features more effectively.
    - Provide more stable results during transitional seasons compared to other model groups.

The superior performance of Model 15 stems from its ability to incorporate multiple influential parameters and sophisticated mathematical techniques. By simultaneously utilizing sunshine duration and hour angle, the model effectively captures key solar radiation dynamics. Additionally, it accounts for the logarithmic effects of temperature variation, enhancing its ability to adapt to different climatic conditions. The model further leverages trigonometric functions to represent the influence of latitude and day length, enabling it to accurately reflect seasonal and geographic variations.

These advanced features make Model 15 particularly well-suited for predicting changes driven by İzmir's Mediterranean climate and coastal influences. Similarly, the strong performance of Models 5 and 6 can be attributed to their use of polynomial and logarithmic functions, which provide a detailed representation of sunshine duration and contribute to their accuracy in capturing solar radiation patterns.

3.4. Model Selection for Practical Applications

The selection of an appropriate model for predicting global solar radiation in İzmir province depends on the purpose of the application and the required level of accuracy. For high-precision applications, such as concentrated solar energy systems, photovoltaic plants, and detailed feasibility studies, Models 15, 5, and 6 are recommended due to their superior performance, with RMSE values of 0.1451, 0.2016, and 0.2017, respectively.

For medium-precision needs, which include small-scale solar energy systems, preliminary feasibility studies, and general planning, Models 1, 8, and 9 provide sufficient accuracy with RMSE values ranging from 0.2824 to 0.3070. These models strike a balance between simplicity and reliability, making them suitable for less critical applications.

In scenarios where basic evaluations and approximate calculations are sufficient, models with RMSE values exceeding 0.35 can be utilized. These models, while not as precise, are still useful for general assessments and regional potential analysis, particularly in cases where detailed accuracy is not a priority.

3.5. Limitations and Recommendations

The results obtained in this study should be interpreted considering several limitations. From the perspective of the dataset, the use of long-term average data without accounting for hourly variations affects the precision of the analysis. Additionally, the reliance on a single meteorological station to represent the entire İzmir province restricts the evaluation of regional differences.

There are also limitations in the modeling approach. The exclusion of key parameters such as cloud cover types and atmospheric transparency can influence the accuracy of predictions. Furthermore, the absence of direct representation of İzmir's distinctive features, such as the coastal influence and topographic variations, limits the ability of the models to fully capture the region's microclimatic effects.

To overcome these limitations in future studies, several recommendations can be made. Utilizing data from multiple meteorological stations across the province and conducting analyses at an hourly resolution can enhance prediction accuracy. Additionally, integrating coastal influence and topographic factors into the models would allow for better representation of regional characteristics. Lastly, a more extensive application of machine learning techniques could improve the modeling of complex atmospheric interactions, further enhancing prediction reliability.

4. Conclusion

In this study, 15 different models for predicting global solar radiation in İzmir province were comparatively analyzed. Using the ATATEK-Solar software, each model was evaluated with three different optimization algorithms. According to the analysis results, the most accurate predictions were achieved by Model 15, with RMSE: 0.1451, R^2: 0.9995, and MPE: 0.08. This was followed by Model 5 (RMSE: 0.2016, R^2: 0.9990, MPE: 0.15) and Model 6 (RMSE: 0.2017, R^2: 0.9990, MPE: 0.10).

An evaluation of seasonal performance revealed that the models performed more consistently during spring and summer months. Notably, Model 15 demonstrated stable performance across all seasons, effectively reflecting İzmir's microclimatic characteristics. From the perspective of optimization algorithms, the Nelder-Mead Simplex algorithm yielded the best results for most models, while the Pattern Search algorithm performed better for Model 15.

The coastal influence and the microclimatic variations from coastal to inland areas in İzmir significantly affect model performance. While Model 15 is recommended for high-precision projects, Model 5 or Model 6 provides sufficient accuracy for preliminary feasibility studies. Future studies could improve prediction accuracy by incorporating data from multiple meteorological stations, performing analyses at an hourly resolution, and integrating region-specific geographical factors into the models.

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References

Almorox J, Hontoria C (2004). Global solar radiation estimation using sunshine duration in Spain. *Energy Conversion and Management*, 45(9–10): 1529–1535. https://doi.org/10.1016/j.enconman.2003.08.022

Almorox J, Bocco M, Willington E (2013). Estimation of daily global solar radiation from measured temperatures at Cañada de Luque, Córdoba, Argentina. *Renewable Energy*, 60: 382–387.

Ampratwum DB, Dorvlo ASS (1999). Estimation of solar radiation from the number of sunshine hours. *Applied Energy*, 63: 161–167.

Angstrom A (1924). Solar and terrestrial radiation. Report to the international commission for solar research on actinometric investigations of solar and atmospheric radiation. *Quarterly Journal of the Royal Meteorological Society*, 50(210): 121–126.

Bristow KL, Campbell G (1984). On the relationship between incoming solar radiation and daily maximum and minimum temperature. *Agricultural and Forest Meteorology*, 31(2): 159–166.

Coppolino S (1994). A new correlation between clearness index and relative sunshine. *Renewable Energy*, 4(4): 417–423.

Dogniaux R, Lemoine M (1983). Classification of radiation sites in terms of different indices of atmospheric transparency. Solar Energy Research and Development in the European Community, Series F, Vol. 2. Dordrecht, Holland: Reidel.

Duffie JA, Beckman WA (2006). Solar engineering of thermal processes (3rd ed.). New York: John Wiley & Sons.

Elagib N, Mansell M (2000). New approaches for estimating global solar radiation across Sudan. *Energy Conversion and Management*, 41(5): 419–434.

El-Metwally M (2005). Sunshine and global solar radiation estimation at different sites in Egypt. *Journal of Atmospheric and Solar-Terrestrial Physics*, 67(14): 1331–1342.

Ersan R, Külcü R (2024). Development of new models using empirical modeling of global solar radiation and its application in Usak city, Turkey. *Tekirdağ Ziraat Fakültesi Dergisi*, 21(5): 1235-1251. https://doi.org/10.33462/jotaf.1512442

Ertürk S, Kara H, Akkuş C, Genç G (2023). Türkiye'de farklı iklim kuşakları için yapay sinir ağları kullanılarak güneş ışınımının tahmini. *Gazi University Journal of Science Part C: Design and Technology*, 11(3): 885–892. https://doi.org/10.29109/gujsc.1331788

Hargreaves G, Riley J (1985). Irrigation water requirements for Senegal River basin. *Journal of Irrigation and Drainage Engineering*, 111(3): 265–275.

Hooke R, Jeeves TA (1961). "Direct Search" solution of numerical and statistical problems. *Journal of the ACM*, 8(2): 212–229. https://doi.org/10.1145/321062.321069

İzmir Kalkınma Ajansı (2021). İzmir'in güneş enerjisi potansiyeli ve yatırım fırsatları. Kalkınma Sözlüğü. https://kalkinmasozlugu.izka.org.tr/gunes-enerjisi/ (Accessed November 21, 2024).

Kirkpatrick S, Gelatt CD, Vecchi MP (1983). Optimization by simulated annealing. *Science*, 220(4598): 671–680. https://doi.org/10.1126/science.220.4598.671

Külcü R (2015). Modelling of solar radiation reaching the earth to Isparta province. *Süleyman Demirel University, Journal of the Faculty of Agriculture*, 10(1): 19–26.

Külcü R (2019). Development of a new model using empirical modeling of global solar radiation and its application in Çankırı city. *Süleyman Demirel University, Yekarum e-Dergi*, 4(2): 1–8.

Külcü R, Ersan R (2021). Empirical modelling of global solar radiation in Hatay (Turkey) province. *Tekirdağ Ziraat Fakültesi Dergisi*, 18(3): 446–456. https://doi.org/10.33462/jotaf.828187

NelderJA, Mead R (1965). A simplex method for function minimization. *The Computer Journal*, 7(4): 308–313. https://doi.org/10.1093/comjnl/7.4.308

Prescott J (1940). Evaporation from a water surface in relation to solar radiation. *Transactions of the Royal Society of South Australia*, 64(1): 114–118.

Süslü A (2024). Determining the most suitable empirical model for global solar radiation prediction in the lakes region. *International Journal of Agriculture, Environment and Food Sciences*, 8(4): 904-912. https://doi.org/10.31015/jaefs.2024.4.20

Süslü A, Külcü R (2024). Global güneş ışınımı tahmin modelleri için ATATEK-Solar yazılımının geliştirilmesi. *Akademia Doğa ve İnsan Bilimleri Dergisi,* 10(1): 62–73.

Türkiye Enerji Bakanlığı. (2024). Güneş enerjisi. T.C. Enerji ve Tabii Kaynaklar Bakanlığı. https://enerji.gov.tr/bilgi-merkezi-enerji-gunes (Accessed November 17, 2024).

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Prediction of Time-series Friction Data using ANFIS

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HIGHLIGHTS

- ANFIS model optimised friction data for peanut grading machines.
- Simulation using DEM took 63 days for 60 seconds of real-time data.
- Peanut classification model achieved a correlation of 0.798854.
- ANFIS eliminates data pre-processing and enhances performance.

Abstract

Modelling is frequently used in science and industry. Friction, wear, and corrosion issues are the main design criteria in peanut kernel grading machines. In this study, the time-series of friction force data is modelled with adaptive neuro-fuzzy inference system (ANFIS). Machine learning focuses on developing models for prediction and classification without explicit programming. The data on the friction force is obtained from a simulation based on the discrete element method. The simulation takes 63 days, 18 hours and 27 minutes to calculate the real time of 60 seconds. A Takagi-Sugeno type ANFIS network is constructed. The network is clustered using grid partitioning method. ANFIS helps to optimise machine performance by modelling friction data. In the obtained peanut kernel classification model, the correlation value is 0.799 and the root of the mean square error is 0.514 N. The percentage of the mean absolute error is found to be 1.666%. 100 iterations are run. Calculations take 20.7 seconds. The model has a high linear relationship. It is also observed that the ANFIS network eliminates the need for any pre-processing of the data. Background of the network used, its hyper-parameters, and the prediction performance are presented in the study.

Keywords: Peanut kernel classification; discrete element method; adaptive neuro-fuzzy inference system; modelling; prediction

1. Introduction

Modelling is a well-established and effective tool widely used in science and industry. It is based on the functions in mathematics. Words such as ‘prediction’, ‘estimation’, ‘insight’, ‘foresight’, or ‘intuition’ can be used to explain the task of a model. Curve fitting, simulation, and artificial intelligence are frequently used for

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modelling. In this study, the words ‘network’ and ‘model’ are used in the same mean because there are function(s) behind a network, as well.

Machine Learning (ML) is a rapidly developing technique that focuses on the development and implementation of algorithms and codes that allow computers to learn from data and make predictions or decisions without explicit programming. It has attracted great interest in recent years due to its ability to predict, classify, and solve complex problems in almost all fields. ML techniques can be generally categorized as supervised, unsupervised, or reinforced learning approaches (Brockwell and Davis 2002). Supervised learning requires training before predicting or classifying. Unsupervised learning needn't to be trained initially. Its training takes place in the realm of working condition. Reinforcement learning focuses on training before usage (Sutton RS 2018). Advances in ML algorithms have accelerated due to the availability of large datasets, increased computing power, and improvements in algorithmic frameworks. Deep learning techniques such as Convolutional Neural Networks (CNNs) and Recurrent Neural Networks (RNNs) have emerged as powerful models for image recognition, Natural Language Processing (NLP), speech recognition, and other complex tasks (LeCun et al. 2015). Transfer learning has also gained importance as it allows fine-tuning of pre-trained models on large datasets to specific applications with limited data (Pan and Yang 2009). In addition, interpretations that provide information on how models make predictions or judgements are also increasing interest in ML methods. These efforts aim to address concerns about model reliability and accuracy (Caruana et al. 2015). Time-series analysis is a fundamental technique used in a variety of disciplines to understand and analyze data collected over time (Murphy 2022). Time-series data can be found in fields as diverse as finance, economics, environmental science and engineering. In the analysis of time-series, many more sophisticated modelling techniques are often used, such as Autoregressive Integrated Moving Average (ARIMA), exponential smoothing methods, state space models, and RNN. In recent years, there have been significant developments in time-series analysis methods. ML algorithms have been included in traditional time-series models to improve forecasting performance. Researchers have investigated the use of deep learning techniques such as Long Short-Term Memory (LSTM) networks to model complex temporal patterns (Lipton 2015). Furthermore, attention mechanisms have been used to improve the interpretability of the model by highlighting important features (Chen and Shi 2021). Another emerging trend is to hybridize single models to obtain more accurate forecasts. Adaptive Network Based Fuzzy Inference System (ANFIS) is one of the popular hybrid methods. It combines the fuzzy logic and neural networks to create the hybrid model so that it can effectively handle uncertainty and non-linearity in data. This approach has attracted great interest in various fields, including control systems, pattern recognition, and decision-making processes (Jang and Sun 1995). While an ANFIS model uses the concept of fuzzy clusters and fuzzy rules to represent linguistic variables and the relationships between them, its neural network part helps to adapt the parameters of these fuzzy rules to the input-output data pairs. The adaptive nature of ANFIS allows it to learn iteratively from the data and thus perform accurate modelling and prediction. Recent research has focused on improving the capabilities of ANFIS models through various techniques such as optimization algorithms (Khalaf et al. 2024; Tien Bui et al. 2018). For example, evolutionary algorithms such as genetic algorithms have been used to optimize the learning process by searching for optimal hyper-parameters that minimize the error between predicted and actual outputs (Chou et al. 2020). Furthermore, hybrid approaches combining ANFIS with other ML techniques such as support vector machines or deep learning architectures have been proposed to improve model performance (Irshaid and Abu-Eisheh 2023). These developments aim to improve the accuracy, interpretability and efficiency of ANFIS models to address complex real problems.

Discrete Element Method (DEM) is a simulation technique used to simulate the interaction between solid particles and their environment. It has attracted great interest in recent years thanks to its ability to calculate complex interactions between individual particles. DEM is widely applied in various fields such as geotechnical engineering, energy, chemical, mining, pharmaceuticals, agriculture and food processing (Zhang et al. 2024; Zhou et al. 2024). The method involves in analysing every single particle by calculating its motion based on contact forces and other multi-physics, taking into account both macroscopic behaviour and microscopic interactions. Advances in computing power have enabled more efficient simulations using DEM techniques (Asylbekov et al. 2024; Reineking et al. 2024; Siegmann et al. 2021). Researchers have introduced improved algorithms for faster calculations and increased accuracy in capturing complex phenomena such as breakage, segregation, and flow patterns within the drum (Ge and Zheng 2024; Ramirez et al. 2024; Zhang et al. 2024). Furthermore, the coupling of DEM with other numerical methods such as Computational Fluid Dynamics (CFD) provides a more comprehensive understanding of particle-fluid interactions (Adhav et al. 2024; Strö̈m et al. 2024; Yao et al. 2020; Zhao et al. 2024). In addition, advances in visualisation techniques have provided more realistic particle motion and system behaviour (Mahboob et al. 2023). In this way, it provides suitable tools for the study of issues such as friction, breakage, and wear. Ansys Fluent DEM©, Ansys Rocky DEM©, Altair DEM© etc. are more widely used software.

In the agricultural industry, peanut grading machines are mostly used to sort by size and ensure consistent quality. However, their performance can be direly affected by friction and wear, leading to reduced productivity, increased maintenance and product damage. To overcome these challenges, researchers frequently use DEM-simulations (Cui et al. 2024), which offer a valuable tool for understanding the complex dynamics in the peanut grading machine. DEM offers various advantages when simulating friction or wear in peanut grading machines: a) Particle level resolution: DEM provides a detailed understanding of particle interactions, allowing friction and wear mechanisms to be accurately characterised. b) Realistic boundary conditions: DEM simulations are able to simulate the real operating environment of peanut grading machines by considering realistic contact and boundary conditions. c) Parametric analysis: DEM allows the study of various operating conditions such as particle size, shape, and material properties, providing information on their influence on friction and wear. Although researchers commonly argue that ML methods should only be applied on experimental data, it is seen that ML can also be performed on simulated data, as well. It may provide shorter prediction time than that of simulations (Bui et al. 2019; Kibriya et al. 2023; Wu et al. 2024; Zhang et al. 2024). Peanut grading machines play a critical role in the agricultural industry by ensuring the quality and efficiency of peanut sizing processes (Akcali et al. 2014). Since wear and friction significantly affect the performance and lifetime of such machines, accurate simulations are necessary to optimize their design and operation. Accuracy also depends on the multi-physics models and assumptions, as well.

Despite significant advancements in time-series prediction techniques, the application of ANFIS for forecasting friction data in peanut kernel drying within rotary drum dryers remains an underexplored area. Existing studies have primarily focused on modeling the drying kinetics, heat and mass transfer mechanisms, and optimization of drying parameters, while limited attention has been given to the dynamic behavior of frictional forces acting on peanut kernels. Friction data is crucial for understanding the mechanical interactions within the drum, which directly influence energy efficiency, product quality, and equipment wear. However, conventional modeling approaches, such as regression-based or purely data-driven machine learning techniques, may lack the interpretability and adaptability required for complex nonlinear systems like rotary drum drying. ANFIS, with its hybrid structure combining neural networks and fuzzy logic, offers a promising alternative, yet its potential for accurate friction prediction in this specific application has not been sufficiently validated. Addressing this research gap could enhance process control strategies, optimize dryer performance, and contribute to the broader goal of automating industrial drying systems through intelligent modeling approaches.

This study aims to simulate using Ansys Rocky DEM© most essentially to the actual conditions and to model the friction data using the ANFIS method, which is a hybrid model. Thus, the effect of wear, which causes significant costs and operational difficulties in the industry, will be better determined. The friction force data from the simulation in a cylindrical sieve are employed. The performance of the ANFIS network is evaluated.

2. Materials and Methods

2.1. The Net Force Generated in Peanut Kernel Grading

In this study, a peanut grading machine located in Çukurova University Faculty of Agriculture as shown in Figure 1 is simulated. Normal force and coefficient of friction between grain and sieve wall results in friction force (tangential contact force). How to calculate the net force in the tangential direction according to the domain is explained in this section. Torque can be calculated after the force calculation. Power can be calculated using the torque. Energy is calculated using the power.

Figure 1. The peanut grading (sorting, sieving, classification) machine used in the experiments.

To calculate the net force that will be the basis for friction force, equilibrium equations are written for the circular and spiral path as shown in Equation (1) and Equation (2) (Ugurluay and Akcali 2021).

$$F(\beta) = g[\beta(n - cos\beta) - (n-1)sin\beta] - g\mu_d[\beta sin\beta + (n-1)(1 - cos\beta)] - \frac{1}{2}r\mu_d\omega^2\beta^2(1+n) = 0 \quad (1)$$

$$F(\beta) = \frac{\rho re}{\beta}\left[(1 + n(\beta - 1))\left(g(cos\varphi - \mu_h sin\varphi)(sin\beta - \beta cos\beta) + (g - \frac{1}{2}r\omega^2\beta^2 - gcos\beta - g\beta cos\beta\right)\mu_d\right] = 0 \quad (2)$$

where $\omega, r, \mu_d, \alpha,$ and g are rotational velocity of the drum, drum's radius, friction coefficient of granular material, rotation angle of the drum, and gravitational acceleration, respectively. β is the angle of the arc sweept from bottom of the cylinder. e is the minimum height of the layer on the drum surfaces. $n = \frac{h_0}{e}$ and h_0 is the maximum thickness of the layer on the drum surfaces.

2.2. Discrete Element Method

The basis of this method is to use equilibrium equations to predict the trajectory of particles. These equations are based on d'Alembert's principle, angular momentum, and Newton's second law of motion. The equilibrium equations are given in Equation (3) and Equation (4). Note that both are ordinary differential equations.

$$m_i\frac{dv_i}{dt} = \sum_j\left(F_{n,ij} + F_{t,ij}\right) + m_i g + F_{f\to p} \quad (3)$$

$$I_i\frac{d\omega_i}{dt} = \sum_j\left(M_{t,ij} + M_{r,ij}\right) + M_{f\to p} \quad (4)$$

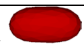
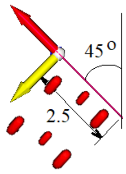
where *i, j* are the particle number, *m* is the mass, *I* is the moment of inertia of the particle. *n* and *t* are the direction vectors in the normal and tangential directions, respectively. *v* is the linear velocity vector, *ω* is the rotational velocity vector of the particle. $F_{f\to p}$ and $M_{f\to p}$ are the force and moment, respectively. Both arises if there is a particle-fluid interaction in the case of CFD-DEM coupling. Since there is no coupling in this study, the expressions for $F_{f\to p}$ and $M_{f\to p}$ are zero. F is the force, $M_{t,ij}$ is the net tangential torque produced by all tangential forces (such as gravity or drag as well as the tangential force component) that cause the particle to rotate. $M_{r,ij}$ is the rolling resistance torque acting on particle *i* by particle *j* or the wall. The direction of $M_{r,ij}$ is the same as that of the rotational velocity, but the sense is opposite. The normal force does not contribute to particle rotation.

Contact occurs as a result of any collision. A contact model is used to calculate the contact forces. Contacts are calculated according to soft or hard sphere techniques. In the hard sphere method, the particles are rigid and there is no deformation in contact. Instead of contact forces, restitution coefficients and shock laws are used to calculate the motion and energy loss during the contact. However, multiple contacts are not allowed. In any collision, forces occur such as Van der Waals forces, liquid bridge forces, and electrostatic forces. In the soft sphere method, although the particles are rigid, the deformation of the particles in contact is calculated using a method called ′′overlap′′. In contact, forces that occur without physical contact, such as Van der Waals forces, liquid bridge forces, and electrostatic forces, can also be included in the calculations.

2.3. Simulation and Data Collection

Ansys Rocky DEM© software package is used in this study (Rocky 2021). It offers a user-friendly interface and many advanced features. These features are particle shape libraries, different contact detection algorithms, and parallel computing capabilities. The parameters used in the DEM simulation are given in Table 1. The calculation is performed in 60 steps. Each step has a time period of 1 second. In addition, each time period is divided into sub-steps having intervals of 0.1 seconds. 60 seconds of real-time calculation is performed. As a result, a time-dependent friction force dataset consisting of 600 data is obtained.

Table 1. Parameters of DEM simulation

Injection properties	**Value**	
Particle shape	216 polyhedra with triangular faces, 	
Particle type, material	Single component (Peanut kernels)	
Particle material behaviour	Hard sphere	
Particle equivalent diameter distributions, *dp* (mm)	22 (100%)+7.6 (22%)	
Particle volume, (m^3)	5.575e-06	
Particle mass flow rate (kgh^{-1})	0.169	
Velocity vector of the particles at the injection point		3 m s^{-1} (normal to the input surface)
Time step size (s)	0.01	
Real time duration (s)	60	
Simulation physics	**Value**	
Normal force	Linear spring-damper	
Tangential force	Coulomb friction	
Rolling resistance torque	Costant, $\mathbf{M}_{\mathbf{r,ij}} = -\mu_r |\mathbf{r}| \times \mathbf{F}_{\mathrm{cn,ij}} \frac{\boldsymbol{\omega}_{\mathbf{p}}}{|\boldsymbol{\omega}_{\mathbf{p}}|}$	
Numerical softening factor	1	
Gravity (m s^{-2})	9.81	
Material properties	**Particle**	**Wall (steel)**
Young's modulus, E	10.11 MPa	200 GPa
Poisson ratio, ν	0.201	0.3
Solid density, ρ_s (kgm^{-3})	416	7850
Bulk density, ρ_b (kgm^{-3})	250	--
Drum's rotational speed	--	10 rev min^{-1}
Material interaction properties	**Particle-particle**	**Particle-wall**
Coefficient of friction, $\mu(\dot{s}_t)_{static}$, $\mu(\dot{s}_t)_{dynamic}$	0.408, 0.318	0.326, 0326
Rolling resistance coefficient, μ_r	0.2	0.2
Coefficient of restitution, ε	0.224	0.224

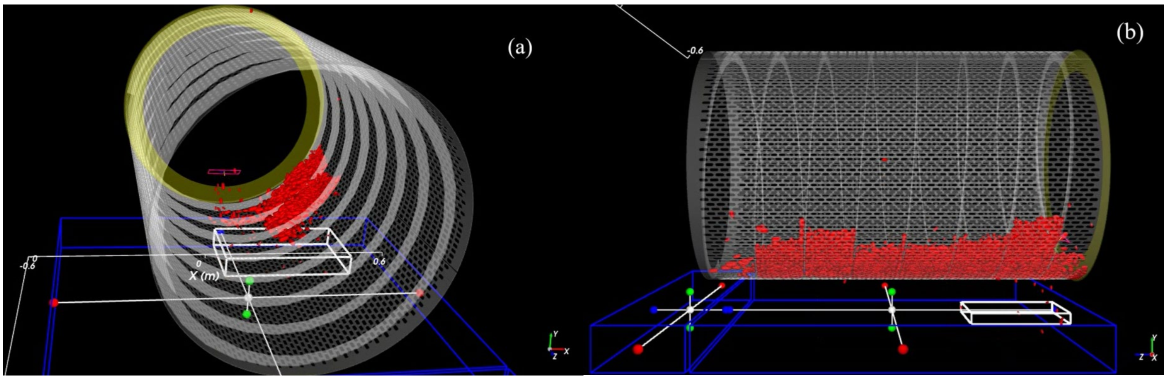


Figure 2. DEM based simulation of peanut grading machine and its grading boxes (a) peanut distribution at *t* = 10 s and (b) *t* = 35 s

Figure 2 shows the geometry of the screen used in the simulation and screenshots taken at certain time steps. In the images, the flow of the peanut kernels into the domain, the rectangular opening at the inlet, the two accumulation boxes under the screen and the helical structure inside the drum are clearly seen.

Figure 3 shows the time-series data of the calculated friction force. The horizontal axis is the time step. The data are time-dependent friction forces. The elapsed calculation time for a 60-second real-time simulation is 63 days, 18 hours and 27 minutes. The calculations are performed using a computer with 8 GB RAM and a 2.8 GHz quad-core CPU.

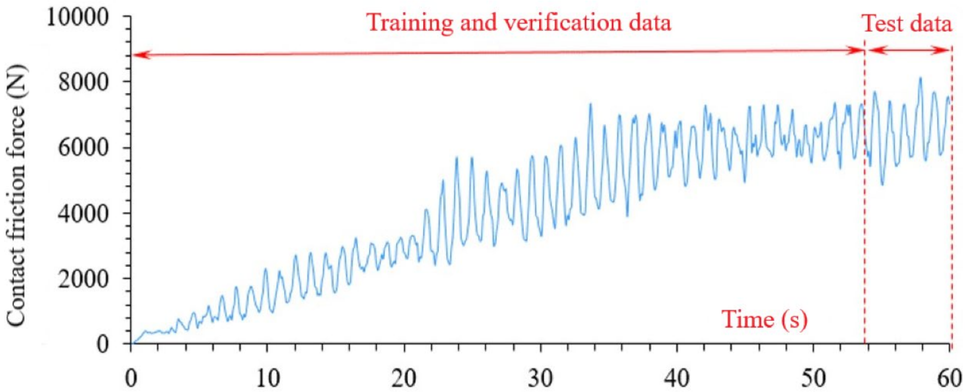


Figure 3. Time-series data of the contact friction force and its training/test portions

2.4. Understanding Insight of the Data

Prior to modelling, a critical step is to understand the nature of the time-series data. It helps the selection of hyper-parameters that are good for the data. In time-series analyses, Auto-Correlation (ACF) and Partial Auto-Correlation (PACF) plots are usually examined together to identify time-lags. Also, hypothesis tests can be performed.

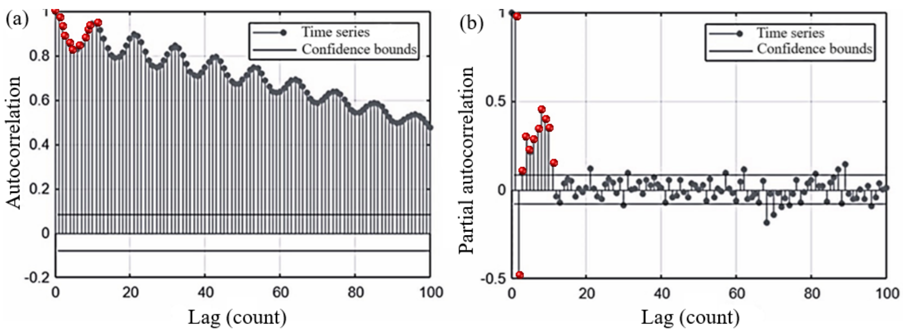


Figure 4. For the friction data; (a) ACF, (b) PACF

In Figure 4, ACF and PACF correlograms are given. Both are unitless. Delays are expressed in units of 'time steps'. Each peak in the graph represents one lag. 100 lags are evaluated. A confidence limit of 2 standard deviations is taken as its boundaries. Both graphs show that the data in the series are not randomly distributed, but rather correlated with their lags. It is also observed that the ACF values decrease over time, while the PACF drops suddenly below the confidence limit after the 11^{th} lag (white noise). White noise mean the values that have no effect on the lags. Both the gradual decrease in ACF and the sudden drop in PACF indicate that this series has an AutoRegressive (AR) structure with 11 lags, shown AR(11) (Korkmaz and Kacar 2024).

In this study, time-series data representing the variation of friction force over time was utilized. Accordingly, a 11-lag of output was fed as input.

2.5. ANFIS

Figure 5 shows the topology of the ANFIS network used.

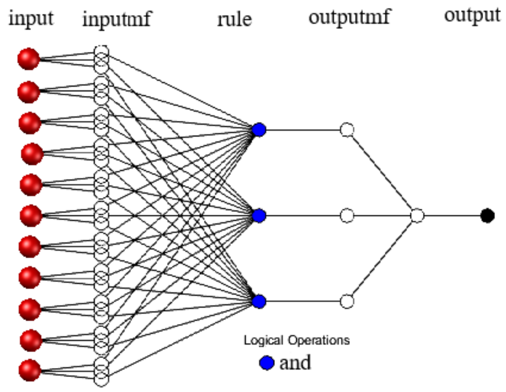


Figure 5. ANFIS network topology

In order to create an ANFIS network, the first stage is to select the hyper-parameters of the network, appropriately. The hyper-parameters are as follows:

- Data partition: Training 90 % (54 s), test 10 % (6 s)
- Number of lags in the data : 11
- Number of inputs :11
- Number of outputs : 1
- Clustering method: Grid Partitioning (Number of clusters: 3, Membership function: Gaussian, output function: Linear (Takagi-Sugeno))
- Maximum number of iterations: 100
- Initial step: 0.01
- Initial acceleration: 0.9
- Initial deceleration: 1.1
- The data were normalised using the formula

$$o(t)^{normalize} = \frac{o(t)-\overline{o(t)}}{\sigma}. \quad (5)$$

  where, $o(t)$ is the time-series data at time step t and $\overline{o(t)}$ is the mean data and σ is the standard deviation, calculated by

$$\sigma = \sqrt{\frac{\sum_{t=1}^{NN}\left[\left(p(t)-\overline{o(t)}\right)\right]^2}{NN}}. \quad (6)$$

  NN is the number of total data. $p(t)$ is the value estimated by the network at time step t. t is the time step.
- Error criterion : Mean Square Error,

$$MSE = \frac{1}{NN}\sum_{t=1}^{NN}\left(p(t)-o(t)\right)^2. \quad (7)$$

- $RMSE$ is Root of MSE. The unit of MSE is the square of the unit of the data. Since the friction force data is in unit of Newton (N), MSE will be N^2. Similarly, $RMSE$ will be in units of (N).

MAE is Mean Absolute Error. Its unit is the data unit. $MAPE$ is Percentage of MAE. MSE ($unit^2$), $RMSE$ ($unit$), MAE ($unit$) and $MAPE$ (%) are error metrics used to measure model success. The closer these criteria to zero means the smaller error. Similarly, the closer the R (or R^2) values to 1 (or -1) means the more successful model.

- Correlation coefficient,

$$R = \frac{NN \sum_{t=1}^{NN} p(t)o(t) - (\sum_{t=1}^{NN} p(t))(\sum_{t=1}^{NN} o(t))}{\left(\sqrt{NN \sum_{t=1}^{NN} p(t)^2 - (\sum_{t=1}^{NN} p(t))^2}\sqrt{NN \sum_{t=1}^{NN} o(t)^2 - (\sum_{t=1}^{NN} o(t))^2}\right)} \quad (8)$$

- and the determinant coefficient is R^2.

Although the 80/20 training/testing ratio is commonly used in the literature, it is no strict limitation. However, while an insufficient number of samples in the training-set may lead to underfitting, a larger dataset generally has a positive impact on learning performance. In this study, a 90/10 ratio was adopted to ensure better training and to make predictions for the next 6 seconds. This choice is justified by the fact that a total of 60 seconds of friction data was collected, and a 10% data partition is sufficient for a 6-second prediction.

3. Results and Discussions

The network's prediction performances are presented in this section. Statistical methods are used in the evaluation of the models.

3.1. Prediction Curve and Verification

Figure 6 (a) shows the prediction curve of the ANFIS network on the training data. Although it is seen that the prediction curve given in (a) perfectly passes over the target data, it is seen that there is some error when looking at the error distribution given in (b). RMSE of this difference is given on the figure. The so-called 'target data' is the friction force data obtained from the simulation. (c) shows error histogram where 'average error' indicates the mean error in the histogram, while "S.D. Error" indicates the Standard Deviation of the error. The average error is 3.6378 x10^{-6} N, which is very small. The histogram shows a normal distribution. It is desirable to have a bell curve shape in the histograms, which is called 'normal distribution'. The narrowness of this curve indicates that the errors are small. The horizontal axis is the error value. The vertical axis in the histogram is the 'incidence'. Looking at the histogram, it is seen that the highest lines are centred around 'zero error'. It means that small errors occur frequently. In other words, the model predicted well on the training set. This is an expected case because the network is familiar with the data since it is trained with this data. The whole training data had already been used during training, so, the prediction performance was high.

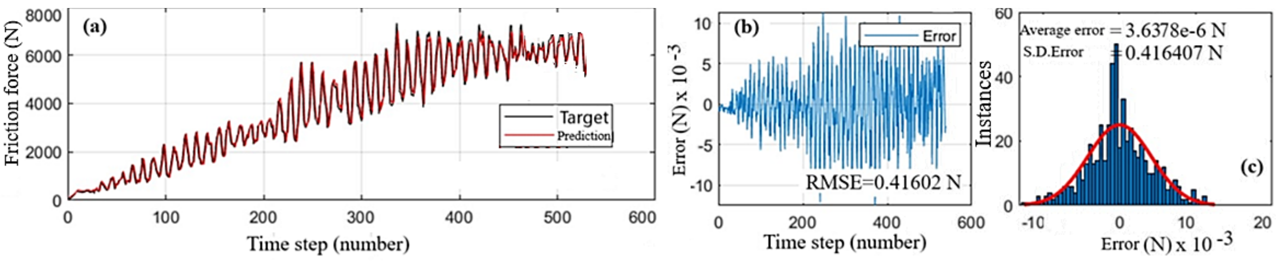


Figure 6. (a) Prediction curve on the training data, (b) error distribution and (c) error histogram of the network on the training data

The response curve for the network's prediction on the test data is given in Figure 7 (a), error distribution in (b) and error histogram in (c). This graph makes it possible to analyse only the performance on the test data in more detail. It is noticeable that this model has a high ability to represent peaks and dips, although a constant shift is also present. The performance is lower than that of training performance. It is an expected case because the network has never seen the forecasting data before. Therefore, it has a greater error in prediction. Looking at the histogram, it can be seen that the error frequently goes outside the bell curve. This is an undesirable situation indicating that the error is greater.

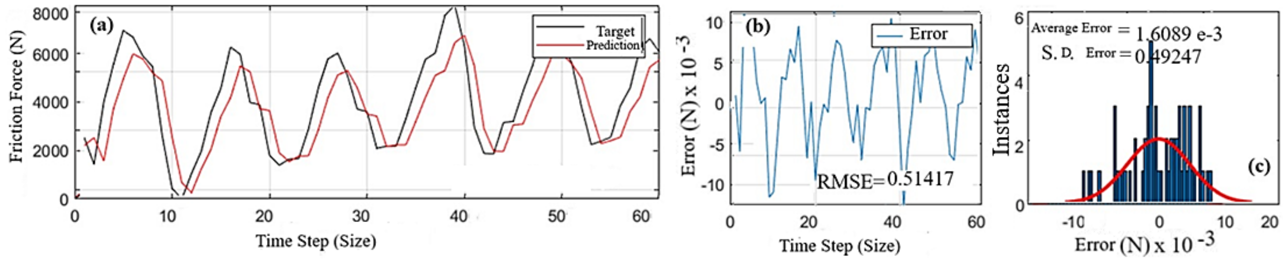


Figure 7. (a) Prediction curve on the forecasting data, (b) error distribution and (c) error histogram of the network on the test data

The response curve for the prediction of the network on the full data is given in Figure 8 (a), the difference (error) distribution is given in (b) and the difference (error) histogram is given in (c).

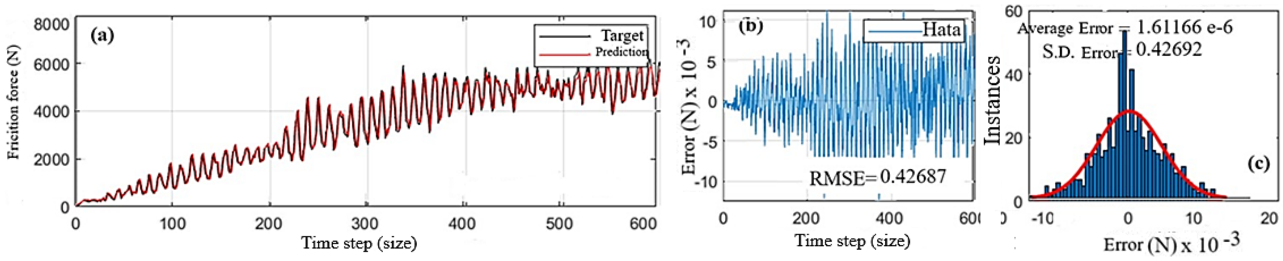


Figure 8. (a) Prediction curve on all data set (b) difference (error) distribution and (c) difference (error) histogram of the network on all data

Another frequently-used model performance metric is analysis of variance. One of the basic assumptions of modelling is that the residual has constant variance at all levels. If the model has constant variance, it means that its generalization ability is high. The most common way to determine whether a model has constant variance is to plot the points of model prediction and corresponding residual. If all points are randomly distributed between two parallel lines, then the variance is constant. Otherwise, if the distribution increases or decreases systematically, then the variance is variable. When Figure 9 is analyzed, it can be seen that the variance is homoscedastic (Korkmaz and Kacar 2024). It shows the variance of the model on the test data. The variances in training and validation could also be determined.

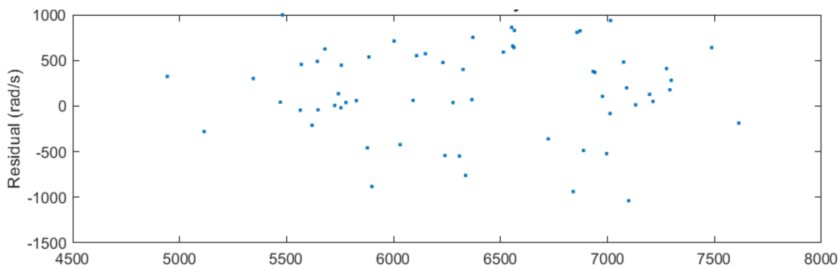


Figure 9. Variance analysis

Regression analysis is another technique frequently used in the evaluation of model performance. It gives linear correlation between the prediction of the model and the values in the data set. The correlation coefficient is denoted by R, while R^2 is called the determinant coefficient. Both are unitless. For R, values between 0.01-0.29 mean a low level of correlation, 0.3-0.7 means a medium level of correlation, and 0.71-0.99 means a high level of correlation. Zero means no relationship, while negative values mean an inverse relationship. The increase in absolute R indicates that the relationship becomes more apparent. Figure 10 shows the results of the regression analysis. The horizontal axis is the target while the vertical axis is the predicted values. As expected, the network has the highest correlation on the training data (Figure 10 *a*). This is because it is very familiar with this data during training. It has recognized this data already. Figure 10-*b* shows that the network also has a high correlation on the test data. Figure 10-*c* shows the correlation of the network's prediction on all data. Since the number of data in the training set is much higher, the data points on the graph is also much more. Correlation on the training set is greater than that of test.

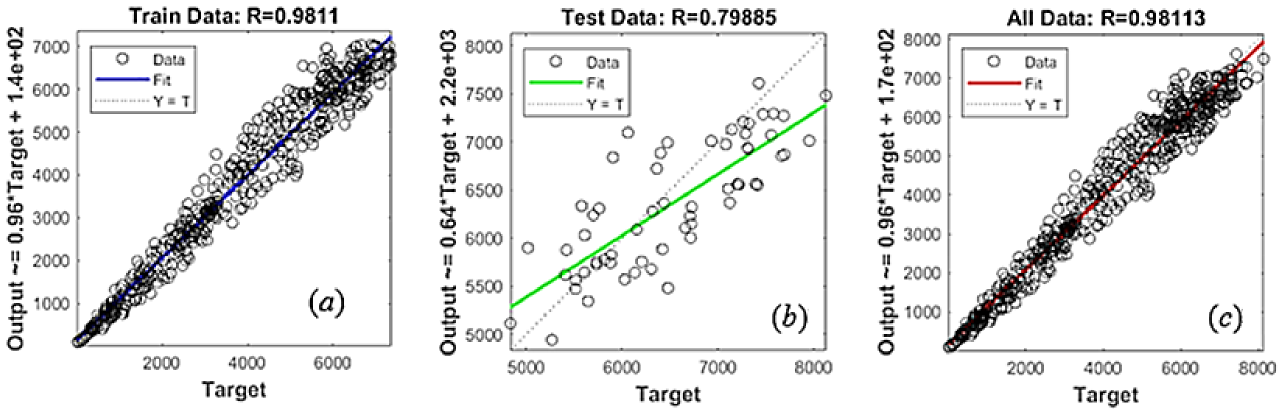


Figure 10. Correlation between the model prediction and (a) training, (b) test, (c) all data

The correlation is 0.799 and RMSE is 0.514 N. MAE is found to be 1.6658%. 100 iterations are performed. The calculations take 20.7 s. There is a high linear relationship between the model outputs and the data set. It is also observed that the ANFIS network eliminates the need for any preprocessing of the data.

Table 2. Indicators recorded during the training of the network

Performance metrics	Values
MAE (unit)*	4.219
MAPE(%)	1.666
Mean Error (unit)	1.61E-03
MSE (unit^2)	0.264
RMSE (unit)	0.514
Elapsed calculation duration (s)	20.7
R_{test}	0.799
R_{training}	0.981
Iteration number	100

3.2. Discussion

ML is rarely used for modelling process parameters in peanut classification whereas curve fitting techniques are more common (Cunha et al. 2024). However, preprocess applied to the data before the curve fitting process disturbs the naturalness of the data to some extent. The generalisation of ANFIS network on the data obtained from Ansys Rocky DEM© simulation is an important motivation for future studies on peanut classification. The most important limitation of DEM simulations is the computational cost. The calculation time increases exponentially as the number of particles in the region increases over time. ML techniques offer reducing these costs. The simulation took 63 days, 18 hours and 27 minutes to calculate the real time of 60 seconds. In the ANFIS method, the total computation time for the 6-second prediction process is 20.7 seconds. When compared, the difference is remarkable. Moreover, no pore-process was applied. The present model has a correlation of 0.799 and an RMSE of 0.514 N. The variance is constant. In addition to general curve shape, peaks-dips could be predicted accurately, as well. Although the terms R or R^2 are expected to be close to ±1, the use of R^2 provides a more conservative perspective. While the R value (0.799) indicates a strong correlation, the lower R^2 value (0.638) suggests that the explanatory power of the model is somewhat limited. In this context, despite the low R^2 value, the model can still be considered to have achieved a certain level of success. However, improving the model’s performance may require training with a larger dataset and optimizing model parameters. Obtaining more accurate and precise friction and wear data can provide a significant advantage in predicting the lifetime and predictive-maintenance on the grading machines. Recent research shows that DEM-based simulations are an effective method to study these factors in peanut grading machines (Kacar 2023; Korkmaz 2023; Qiao et al. 2024). DEM software provides valuable information on machine design and performance by modelling particle interactions and boundary conditions realistically. With the help of coupled CFD-DEM analysis, the APE (Absolute percentage error) of the ML-based model for the transitional flow of sand particles was determined as minimum 7.36% and maximum 29.99% (Hu et al. 2024). Hybrid CNN-LSTM hybrid model are applied onto the data obtained from the CFD-DEM simulation of liquid-solid particle mixing and separation in a bi-dispersed fluidised bed. The prediction results are compared with the CFD-DEM results. In terms of computation cost, LSTM takes 2.1-2.5 h and CNN-LSTM takes 6.2-6.5 h while CFD-DEM takes 72-165 h. The best accuracy is obtained from CNN-LSTM in reference with velocity prediction with R^2 = 0.86-0.92, MAE = 0.0006-0.0046 m s^{-1}, RMSE = 0.0030-0.0138 m s^{-1} (Xie et al. 2022).

Future work should aim to increase the efficiency, durability and reliability of these machines by improving DEM techniques.

4. Conclusions

This study highlights the potential of using ANFIS for predicting time-series friction data in the peanut grading process. By integrating data obtained from DEM-based simulations, the study demonstrates the feasibility of employing machine learning techniques in agricultural and food processing applications. The findings indicate that ANFIS provides an effective framework for capturing the complex frictional interactions within a rotary drum dryer, offering an efficient alternative to traditional simulation-based approaches.

The significance of this research lies in its contribution to process optimization and automation in industrial drying systems. The results suggest that ANFIS can effectively model and predict friction forces without requiring extensive data preprocessing, thereby reducing computational costs and improving efficiency. This is particularly relevant given the substantial time investment required for high-fidelity DEM simulations, underscoring the necessity of alternative predictive methods that balance accuracy and computational efficiency. The DEM simulation had taken 63 days, 18 hours, and 27 minutes to compute 60 seconds of real-time data. 6 seconds ahead is predicted. The ANFIS method completed both training and forecasting within a total computation time of 20.7 seconds. The remarkable difference between these results is noteworthy.

Future research should focus on enhancing the predictive accuracy and generalizability of the proposed model. Comparative analyses with alternative machine learning techniques, such as deep learning or hybrid approaches, could provide valuable insights into performance improvements. Additionally, incorporating real-world experimental data alongside simulation results could further validate the model's reliability. Investigating the effects of different peanut varieties, processing conditions, and time-series delays may also refine the applicability of the proposed framework. Moreover, integrating external factors such as humidity, temperature, and kernel size distribution into the predictive model could enhance its robustness and practical relevance in industrial applications.

Ultimately, this study serves as a foundation for future advancements in the intelligent modeling of friction forces in food processing systems. By leveraging data-driven methodologies, future research can contribute to the development of more adaptive, efficient, and scalable solutions for agricultural automation and process optimization.

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References

Adhav P, Besseron X, Estupinan AA, Peters B (2024). Development and validation of CFD-DEM coupling interface for heat & mass transfer using partitioned coupling approach. *International Communications in Heat and Mass Transfer,* 157: 107801.

Akcali İD, Mutlu H, Ercan U (2014). Mathematical Model of a Sorting Machine. *Journal of Agricultural Machinery Science,* 10(3): 229-234.

Asylbekov E, Poggemann L, Dittler A, Nirschl H (2024). Discrete Element Method Simulation of Particulate Material Fracture Behavior on a Stretchable Single Filter Fiber with Additional Gas Flow. *Powders,* 3(3): 367-391.

Brockwell PJ, Davis RA (2002). *Introduction to time-series and forecasting*: Springer. New York , USA, p.449

Bui VH, Bui MD, Rutschmann P (2019). Combination of discrete element method and artificial neural network for predicting porosity of gravel-bed river. *Water,* 11(7): 1461.

Caruana R, Lou Y, Gehrke J, Koch P, Sturm M, Elhadad N (2015). *Intelligible models for healthcare: Predicting pneumonia risk and hospital 30-day readmission.* Paper presented at the Proceedings of the 21th ACM SIGKDD international conference on knowledge discovery and data mining.

Chen W, Shi K (2021). Multi-scale attention convolutional neural network for time-series classification. *Neural Networks,* 136: 126-140.

Chou K-Y, Yeh Y-W, Chen Y-T, Cheng Y-M, Chen Y-P (2020). *Adaptive neuro fuzzy inference system based MPPT algorithm applied to photovoltaic systems under partial shading conditions.* Paper presented at the 2020 International Automatic Control Conference (CACS).

Cui X, Li X, Du Y, Bao Z, Zhang X, Hao J, Hu Y (2024). Macro-micro numerical analysis of granular materials considering principal stress rotation based on DEM simulation of dynamic hollow cylinder test. *Construction and Building Materials,* 412: 134818.

Cunha N, da Silva LHM, da Cruz Rodrigues AM (2024). Drying of Curcuma longa L. slices by refractance window: Effect of temperature on thermodynamic properties and mass transfer parameters. *Heat and Mass Transfer,* 60(4): 617-626.

Ge M, Zheng G (2024). Fluid–Solid Mixing Transfer Mechanism and Flow Patterns of the Double-Layered Impeller Stirring Tank by the CFD-DEM Method. *Energies,* 17(7): 1513.

Hu G, Zhou B, Zheng W, Li C, Wang H (2024). A ML-based drag model for sand particles in transition flow aided by spherical harmonic analysis and resolved CFD-DEM. *Acta Geotechnica*, 20(1):461-474.

Irshaid M, Abu-Eisheh S (2023). Application of adaptive neuro-fuzzy inference system in modelling home-based trip generation. *Ain Shams Engineering Journal,* 14(11): 102523.

Jang J-S, Sun C-T (1995). Neuro-fuzzy modeling and control. *Proceedings of the IEEE,* 83(3): 378-406.

Kacar İ (2023). *Scientific Principles of Mechanical Design and Analysis* (Vol. 381): Academician bookshop.

Khalaf AH, Lin B, Abdalla AN, Han Z, Xiao Y, Tang J (2024). Enhanced prediction of corrosion rates of pipeline steels using simulated annealing-optimized ANFIS models. *Results in Engineering,* 24: 102853.

Kibriya G, Orosz Á, Botzheim J, Bagi K (2023). Calibration of micromechanical parameters for the discrete element simulation of a masonry arch using artificial intelligence. *Infrastructures,* 8(4): 64.

Korkmaz C (2023). The place of oganic and organomineral fertilizer production in sustainable agriculture. In A Bayat (Ed.), *Sustainable Agriculture Technologies-II*), Ankara: İKSAD. Vol. 244, pp. 183-205

Korkmaz C, Kacar İ (2024). Explaining data preprocessing methods for modeling and forecasting with the example of product drying. *Journal of Tekirdag Agricultural Faculty,* 21(2): 482-500.

LeCun Y, Bengio Y, Hinton G (2015). Deep learning. *Nature,* 521(7553): 436-444.

Lipton ZC (2015). A Critical Review of Recurrent Neural Networks for Sequence Learning. *arXiv Preprint, CoRR, abs/1506.00019*.

Mahboob A, Hassanshahi O, Tabrizi AS (2023). Three-dimensional simulation of granular materials by discrete element method (DEM) by considering the fracture effect of particles. *Journal of Civil Engineering Researchers,* 5(2): 14-28.

Murphy KP (2022). *Probabilistic ML: an introduction*: MIT press, London, England, p:864.

Pan SJ, Yang Q (2009). A survey on transfer learning. *IEEE Transactions on knowledge and data engineering,* 22(10): 1345-1359.

Qiao J, Hu K, Yang J, Wang Y, Liu J, Zhou E, . . . Duan C (2024). Research on enhancement of screening performance of a novel drum screen based on the Discrete Element Method simulation. *Powder Technology,* 437: 119567.

Ramirez CE, Sermet Y, Demir I (2024). HydroCompute: An open-source web-based computational library for hydrology and environmental sciences. *Environmental Modelling & Software,* 175: 106005.

Reineking L, Fischer J, Mjalled A, Illana E, Wirtz S, Scherer V, Mönnigmann M (2024). Convective drying of wood chips: Accelerating coupled DEM-CFD simulations with parametrized reduced single particle models. *Particuology,* 84: 158-167.

Rocky DEM© Ansys (2021). DEM technical manual 4.4. 3.

Siegmann E, Enzinger S, Toson P, Doshi P, Khinast J, Jajcevic D (2021). Massively speeding up DEM simulations of continuous processes using a DEM extrapolation. *Powder Technology,* 390: 442-455.

Strö̈m H, Luo H, Xiong Q (2024). Perspectives on Particle–Fluid Coupling at Varying Resolution in CFD-DEM Simulations of Thermochemical Biomass Conversion. *Energy & Fuels,* 38(18): 17179-17190.

Sutton RS (2018). Reinforcement learning: An introduction. *A Bradford Book.* The MIT Press, London, England, p. 548.

Tien Bui D, Khosravi K, Li S, Shahabi H, Panahi M, Singh VP, . . . Chen W (2018). New hybrids of anfis with several optimization algorithms for flood susceptibility modeling. *Water,* 10(9): 1210.

Ugurluay S, Akcali ID (2021). Development of a vibrationless sorting system. *Spanish journal of agricultural research,* 19(1): 204.

Wu W, Chen K, Tsotsas E (2024). Prediction of rod-like particle mixing in rotary drums by three ML methods based on DEM simulation data. *Powder Technology,* 448: 120307.

Xie Z, Gu X, Shen Y (2022). A ML study of predicting mixing and segregation behaviors in a bidisperse solid–liquid fluidized bed. *Industrial & Engineering Chemistry Research,* 61(24): 8551-8565.

Yao L, Xiao Z, Liu J, Zhang Q, Wang M (2020). An optimized CFD-DEM method for fluid-particle coupling dynamics analysis. *International Journal of Mechanical Sciences,* 174: 105503.

Zhang C, Chen Y, Wang Y, Bai Q (2024). Discrete element method simulation of granular materials considering particle breakage in geotechnical and mining engineering: A short review. *Green and Smart Mining Engineering*, 1(2): 190-207.

Zhang T, Li S, Yang H, Zhang F (2024). Prediction of constrained modulus for granular soil using 3D discrete element method and convolutional neural networks. *Journal of Rock Mechanics and Geotechnical Engineering,* 16(11): 4769-4781.

Zhang Y, Cao Z, Liu C, Huang H (2024). Fluid-solid coupling numerical simulation of micro-disturbance grouting treatment for excessive deformation of shield tunnel. *Underground Space,* 19:87-100.

Zhao Z, Zhou L, Bai L, Wang B, Agarwal R (2024). Recent advances and perspectives of CFD–DEM simulation in fluidized bed. *Archives of Computational Methods in Engineering,* 31(2): 871-918.

Zhou L, Wang B, Cao Y, Zhao Z, Agarwal R (2024). Fluidisation of spherocylindrical particles: computational fluid dynamics–Discrete element method simulation and experimental investigation. *Engineering Applications of Computational Fluid Mechanics,* 18(1): 2297537.

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Research Article

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Determination of The Effects of Different Applications on Some Forage Quality Characteristics in Alfalfa Harvesting with Disc Mowers

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HIGHLIGHTS

- Harvesting alfalfa with three different harvesting systems
- Determination of drying rates and dry matter losses of alfalfa with different harvesting systems
- Some factors affecting forage quality

Abstract

In this study, the effects of different harvesting systems on the drying process, dry matter loss and some forage quality characteristics of alfalfa were investigated. In the research, alfalfa, a forage plant, was harvested with three different harvesting systems: disc mower (UI), disc mower + rubber roller conditioning unit (UII), and disc mower + finger type conditioning unit (UIII) and baling has been done. The trials were conducted according to the randomized block trials design. In the research, moisture content, drying rate, dry matter loss, forage quality values and relative forage values were determined depending on the drying time of the harvesting systems. At the end of the third day of harvesting applications, moisture losses were determined as 34.94%, 39.13% and 31.51%, drying ratios as 3.05, 2.95 and 3.37, and total dry matter losses as 3.72%, 2.26% and 7.80% in UI, UII and UIII applications, respectively. CP values, which express forage quality after baling, are on average 17.63%, 21.33% and 16.87% in UI, UII and UIII applications has been determined respectively, ADF values are 26.93%, 28.27% and 30.76%, NDF values if 31.97%, 30.93% and 34.77%. The relative forage value was determined as 205.18%, 201.21% and 173.75%, respectively. As a result of the research, it was seen that the UII application was the best application.

Keywords: Alfalfa; disc mower; dry matter losses; drying rate; forage quality

1. Introduction

Living things need nutrition to survive. Agriculture and animal husbandry activities form the basis of human nutrition. For people to have a healthy and balanced diet, the proteins contained in meat and milk of animal origin and the products obtained from them are very important.

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According to TurkStat 2021 data, it is understood that there are 17.7 million head of livestock, 1.2 million tons of red meat and 23 million tons of milk production in our country as of 2019. According to the values of the last 10 years, there has been an increase of 55.6% in the number of cattle, 53.9% in red meat production and 69.5% in milk production. 91% of milk production and 89.5% of red meat production are obtained from cattle (TurkStat 2021).

Feeds used in animal nutrition are divided into two main groups: concentrated feed and roughage. The high cost of concentrated feed also increases the feeding cost of animals. The biggest cost in animal production is feed input (Mavruk 2017). Feed input is of even greater importance for small and medium-sized agricultural enterprises in our country. For this reason, the ability of agricultural enterprises to carry out animal husbandry activities with the highest profitability depends on their ability to obtain quality roughage at the lowest cost (Toruk 1997).

In our country, roughages used in animal nutrition are generally supplied from meadow-pasture areas, forage crops grown in agricultural lands and dry grass and silage obtained from crop production residues.

According to TurkStat's 2001 agricultural census, meadow-pasture areas in our country are 14.6 million hectares (12.95 million hectares of land have been determined as of 2020), corresponding to approximately 5% of Turkey's surface area and 38.8% of the total agricultural areas (Anonymous 2020). Since the early 2000s, meadow-pasture areas have become unable to meet the required roughage due to reasons such as heavy and overgrazing. Since the meadow-pasture areas are insufficient for grazing, the roughage needs of the animals are tried to be met with forage plants such as alfalfa, silage corn and vetch.

With the government support given to livestock farming in recent years, forage crop cultivation areas have increased by approximately 50%, reaching 2.3 million hectares from 1.5 million hectares (TurkStat 2021). As a result of this increase, the roughage need in livestock enterprises has begun to be met to a large extent by forage plants such as corn silage, dry alfalfa and legume straw.

According to TurkStat 2021 data in our country, when the last 10-year production values of alfalfa, silage corn, vetch and sainfoin plants are examined, it is understood that there is an increase of 59.7% in alfalfa, 2.3% in vetch, 104.5% in silage corn and 23.1% in sainfoin (TurkStat 2021).

In the same period, it was observed that there was an increase of 90.2% in alfalfa production, 88.9% in vetch and 320% in silage corn in Konya province, and a 36.6% decrease in sainfoin.

In our country, alfalfa which is produced in the largest area among all forage crops, is consumed with pleasure by animals. Demiroğlu et al. (2008) stated that alfalfa is the queen of forage crops due it has a high quality forage value.

Feed quality can be defined as whether the feed is liked by the animal and can be converted into animal products at most. Feed quality is closely related to the nutritional and energy value, the proportion of crude protein and vitamins, the amount of mineral substances, in addition to fiber and digestibility rates, it is also closely related to the productivity of animals.

Budak and Budak (2014) stated that the quality of forage plants is determined by milk yield and live weight gain; They also stated that the palatability of the forage, the amount eaten by the animals, its digestibility, nutritional element content, toxic content and the performance of the animals are indicators. They emphasized that if the crude protein content of the forage plant is 12% or less, it is low quality forage, if it is 15% or so, it is medium, and if it is 18% or more, it is high quality forage.

In determining forage quality, crude protein (CP), acid detergent fiber (ADF), neutral detergent fiber (NDF) analyses, forage consumption and forage digestibility level are also used as important quality criteria (Vurarak 2016).

If the forage plants are harvested in accordance with the technique, the high quality forages needed by the animals can be obtained. Harvesting consists of sequential processes that influence each other. These operations to contain mowing or mowing + conditioning, raking, baling and transporting. During harvest collecting and storing the green and very moist product from the field in a short time without losing its nutritional value is only possible with mechanization practices. Forages plants harvested green are stored either by making silage or by drying.

The main purpose of drying is to reduce the moisture content from 70-80% at the time of harvest to 20-25% moisture content where it can be safely stored (Dursun and Güner 1999; Sessiz at al., 2020). There are three ways of drying to a safe moisture level in practice. These are drying completely in the field with meteorological factors, drying in the field up to a certain level and then artificial drying in closed places and artificially removing all the water rapidly (Evcim 1979).

Today, as in the past, natural drying continues to play an important role in forage production. Freshly harvested forages may require several days to reduce to the ideal moisture desired for baling. Although this period is affected to some extent by the type of forage plant, the harvesting method and the equipment used, it is mainly determined by the weather conditions prevailing during the drying period. During this time, the forage crop can be exposed to various factors leading to quality loss.

Losses that start immediately after harvesting are due to two main causes, biological and mechanical (Rotz 1995). In some cases, artificial drying methods are applied to reduce this possible quality loss, the energy cost required makes forage production more expensive. Under these circumstances, the traditional drying method continues to be a widely accepted practice. It is important to introduce practices that will minimize the drying time without damaging the forage quality.

Harvesting of forage crops is usually done with rotary or double blade mowers and raking is done with grass rakes. Baling is done with balers that make cylindrical and prismatic bales. Mavruk (2017) emphasized that the production of roughage at the lowest cost and the best quality depends on the most effective use of these machines.

In this study, it was aimed to determine the effects of different harvesting practices on field drying rate, dry matter losses and forage quality in the harvest of alfalfa a roughage crop.

2. Materials and Methods

2.1. Material

2.1.1. Trial area

The trials were carried out in the alfalfa cultivated field numbered 29 987, parcel 102 in Yukarıpınarbaşı neighborhood of Selçuklu district of Konya province. Geographic location of the trial plot: (Latitude: 38˚06'22"N, Longitude: 32˚63'33 "E).

The trial study has been made out with the 3rd mowing (harvest) on 20.08.2021 in an area of 2.5 da (20 m x 125 m) on the south-western edge of the field.

2.1.2. Trial material

Bilensoy-80 alfalfa plant, the first synthetic variety registered in our country in 1984 by the Central Research Institute of Field Crops Directorate, was used as trial material.

2.1.3. Tools and machines used in the trial

New Holland brand TD 110D model tractor was used in the trials. The technical specifications of the disc mowers used in the trials are given in Table 1.

2.2. Method

2.2.1. Mowing and baling time

The trials has been made on 20-21-22 August 2021. The mowing process has been made on 20.08.2021out as the 3rd mowing when there was approximately 10-15% flowering in clover (average plant height 61 cm) (Figure 1).

Table 1. Technical specifications of the harvesting machines used in the trials

Harvesting method	Working width (cm)	Number of discs (pieces)	Conditioning system	Swath width (cm)
Disc mower (UI)	245	6	None	160
Disc mower + rubber roller conditioning (UII)	315	8	Zigzag patterned rubber roller (2 pieces)	80
Disc mower + finger type conditioning (UIII)	245	6	Plastic fingers (52 pieces)	140

(**a**) (**b**)

Figure 1. (**a**) Disc mower + rubber roller conditioning (UII); (**b**) Disc mower + finger conditioning (UIII).

On the first day, mowing was started at 9 am and finished at 10 am. Samples were taken between 11-12 and 17-18 hours on the same day. Samples were taken at the same times on the second and third days. At the end of the second day, swath joining was done with a weed harrow and at the end of the third day, baling was done with a baling machine. The created bale dimensions are 36 x 46 x 90 cm, and the average bale weight is 25 kg.

2.2.2. Meteorological data

The averages of temperature, relative humidity and wind speed values measured during the trials are given in Table 2.

Table 2. Values measured in the trial area.

	1.day		2.day		3.day	
	1st Sample	2nd Sample	1st Sample	2nd Sample	1st Sample	2nd Sample
Temperature (°C)	28.5	33.3	33.2	29.4	34.9	31.2
Relative humidity (%)	50.18	27.35	33.80	28.90	30.05	27.43
Wind speed (m/s)	2.85	1.97	0.50	2.16	1.33	2.01

2.2.3. Finding field fresh alfalfa yield

In order to find the field fresh herbage yield, 5 samples of 0.25 m^2 (50x50 cm) randomly selected from the trial plots were mowed at the normal mowing height of the mower (8-10 cm) and weighed with a precision balance immediately after mowing. The field fresh alfalfa yield obtained from the weighed samples was determined by the following formula (Dursun and Güner 1999; Gökkaya 2019).

$$Tw = \frac{Ww}{0.25} x1000 \tag{1}$$

Tw : Field wet alfalfa yield (kg da^{-1})

Ww : Average amount of alfalfa sampled (kg)

After determining the field fresh grass yield, the samples taken were dried in an oven at 70 ^{o}C for 48 hours, then the samples that reached a constant weight were weighed and the field dry grass yield was found in kg da^{-1}.

2.2.4. Taking samples and determining moisture content

To determine the moisture content and drying rate of the material after mowing, 5 samples were taken from each parcel in each block, twice a day for three days. They were weighed on a scale with a precision of 0.5 g, disconnected from air, brought to the laboratory, and weighed again after being dried in an oven at 70 ^{o}C for 48 hours (Ayaz 2010; Mutlu 2019). The moisture content was calculated on a wet weight basis with the following formula (Toruk 1997). Additionally, the dry matter ratio was determined.

$$Mw = \frac{Ww - Wd}{Ww} x100 \tag{2}$$

Mw : Moisture content of the product (w.b.) (%)

Ww : Initial weight of the samples (g)

Wd : Weight of the samples after drying (g)

2.2.5. Determination of drying rate

The drying rate was calculated with the following formula, based on the % moisture lost per drying hour depending on the drying time (Evcim 1979; Toruk 1997; Dursun and Güner 1999).

$$DR = \frac{Mwa - Mwi}{\sum_a^i Dt} x100 \tag{3}$$

DR : Drying rate (%h^{-1})

Σ Dt : Time from start to time i (h)

Mwa: Initial moisture content (w.b.) %

Mwi: Moisture content at time i (w.b.) %

2.2.6. Calculation of dry matter losses

To determine the dry matter loss of different harvesting systems, the material falling on the soil surface was collected along the harvest width in five replicates from the trial plots after harrowing and after bale making.

In order to determine the dry matter loss of different harvesting systems, the materials remaining on the soil surface (spilled) were collected along the harvest width in five replicates from the trial plots after raking and after baling. Dry matter losses of harvesting systems were calculated as a percentage (Rotz and Sprott 1984; Rotz et al. 1987).

$$DML = \frac{\sum m\, xDMY}{BxBs} x100 \quad (4)$$

DML: Dry matter loss (%)

m: Mass (g)

DMY: Dry matter yield (w.b.)

B : Working width (m)

Bs : Sampling width (m)

2.2.7. Determination of nutritional value

From samples taken in triplicate after baling, CP (crude protein) ratio (%), ADF (acid detergent fiber) ratio (%), NDF (neutral detergent fiber) ratio (%), CA (crude ash) ratio (%) and CO (crude oil) ratio (%) was determined in the NIR device (Bulgurlu and Ergül 1978; Gökkaya 2019).

2.2.8. Determination of relative forage value

ADF and NDF values in the alfalfa content are used to determine the relative forage value. Relative forage value was calculated with the following formula (Schroeder, 1994; Van Dyke and Anderson, 1994; Budak and Budak 2014).

$$RFV = DRMxDMIx0.775 \quad (5)$$

$$DRM = 88.9 - (0.779xADF) \quad (6)$$

$$DMI = \frac{120}{NDF} \quad (7)$$

RFV: Relative forage value (%)

DRM: Digestible dry matter ratio (%)

DMI: Dry matter intake percentage (%)

ADF: Acid detergent fiber ratio (%)

NDF: Neutral detergent fiber ratio (%)

2.2.9. Organizing and conducting trials

The trials were carried out with three replications according to the randomized block trial design, and the parcel dimensions were formed by the working width of the machines and a length of 125 m. The machines used are symbolized in the text as follows;

UI : Disc mower

UII : Disc mower + rubber roller conditioning unit

UIII : Disc mower + finger type conditioning unit

For humidity measurement values, six measurements were made over three days and these measurements were; T1 (between 11.00-12.00 hours on the 1st day), T2 (between 17.00-18.00 hours on the 1st day), T3 (between 11.00-12.00 hours on the 2nd day), T4 (between 17.00-18.00 hours on the 2nd day), T5 (between 11.00-12.00 hours on the 3rd day) and T6 (between 17.00-18.00 hours on the 3rd day) it is shown as. Windrow joining has been done between T4 and T5 with a hay rake. The time intervals of these six measurements were used for the drying rate and A1 (T1-T2 interval), A2 (T2-T3 interval), A3 (T3-T4 interval), A4 (T4-T5 interval) and A5 (T5-T6 interval) it is shown as.

The working widths of the machines used in the trials are: 245 cm in UI, 315 cm in UII and 245 cm in UIII. The swath widths resulting from the harvest were measured as 160 cm (65% of the mowed area), 80 cm (25.4% of the mowed area) and 140 cm (57% of the mowed area), respectively. At the end of the second day, the windrows were combined with a hay rake to create new windrows with a width of 90 cm. At the end of the third day, rectangular (36 cm x 46 cm x 90 cm) bales were made using a baler.

2.2.10. Statistical analysis

On the data obtained from the trials, variance analysis was performed with the Minitab 16 package program and MSTAT-C, and the LSD test was applied to the significant results.

3. Results and Discussion

3.1. Wet and Dry Grass Yield

The average field wet grass yield was found to be 1 831 kg da^{-1}, and the dry grass yield was 421 kg da^{-1}, by calculation from the samples taken before harvest.

3.2. Result on Moisture Content

The moisture content values obtained as a result of the harvest of alfalfa using different harvesting systems, depending on the drying times, are given in Table 3.

By examining Table 3, we can state that moisture loss is close to each other in all three harvest systems on the first day of harvest. It is seen that the alfalfa loses moisture faster in UIII starting from the second day of harvest and the moisture content of the material is 15.9% at the end of the day of windrow assembly.

Table 3. Moisture content changes (% w.b.) in alfalfa harvested with different harvesting systems

Applications	1.day		2.day		3.day		Average of applications
	(T1)	(T2)	(T3)	(T4)	(T5)	(T6)	
UI	75.06±2.25a	48.57±0.76b	35.56±2.75d	19.63±1.29f	18.57±0.57fg	12.23±1.21h	34.94b
UII	77.57±3.21a	49.80±1.35b	42.70±1.42c	26.23±1.06e	18.63±2.16fg	19.87±1.46f	39.13a
UIII	76.47±0.67a	50.77±1.11b	25.87±1.12e	15.93±0.85f	11.50±0.80hı	8.50±0.66ı	31.51c
	LSD=3.440						LSD=1.986

From the evaluation of Table 3, it was determined that the lowest moisture value before baling occurred in UIII, followed by UI and UII applications. We can explain the high humidity value in UII as the high density of the windrow (working width of 315 cm and the created windrow width of 80 cm) delaying moisture loss.

As a result of the variance analysis applied to the moisture loss values, the harvesting systems, time and harvesting systems x time interaction were found to be statistically significant ($p<0.01$). Moisture loss values decreased depending on the time periods in which the samples were taken and the moisture loss between the time intervals was found to be statistically significant (Table 4).

Table 4. Results of variance analysis applied to the drying time index values of alfalfa for different harvesting practices

Source of variation	DF	SS	MS	F	
Application	2	525.4	262.7	108.83**	Time
Time	5	26530.6	5306.1	2198.33**	76.367a
ApplicationxTime	10	385.2	38.5	15.96**	49.711b
Error	36	869	2.4		34.711c
Total	53	27528.1			20.600d
					16.233e
					13.533f
					LSD=1.986

(**) sign indicates that the difference is significant according to the 1% probability limit.

When the binary interaction was evaluated, it was determined that the lowest moisture value was reached in the UIIIT6 interaction and there was no statistical difference between it and the UIIIT5 interaction.

3.3. Results Regarding Drying Rate

Depending on the drying time, the drying rate data calculated based on the % moisture lost per drying hour is given in Table 5.

Table 5. Effect of different harvesting systems on the drying rate of alfalfa (%)

Applications	A1	A2	A3	A4	A5	Average of applications
UI	4.41±0.50a	3.31±0.18bc	3.07±0.17bcd	2.36±0.08ghı	2.09±0.12hı	3.05b
UII	4.63±0.50a	2.91b±0.19cde	2.85±0.14cdef	2.44±0.11efgh	1.93±0.13ı	2.95b
UIII	4.29±0.17a	4.22±0.15a	3.37±0.05b	2.71±0.01defg	2.25±0.04ghı	3.37a
			LSD=0.4930			LSD=0.286

Examining Table 5, it is seen that on the first day of harvest (A1), the drying rate of the alfalfa harvested in UIII is low (dries slowly), while the drying rate is high in UII. At the end of the second day, in the samples taken from the windrows created as a result of the raking process in all parcels, the drying rates of the harvest systems were determined as 2.36 %, 2.44 and 2.71 and 2.09, 1.93 and 2.25, respectively, in the A4 and A5 time periods.

As a result of the variance analysis applied to the drying rate values, the harvesting systems, measurement interval (time period) and the harvesting systems x measurement interval interaction were found to be statistically significant ($p<0.01$) (Table 6).

When the binary interaction was examined, it was found that the highest drying rate value was reached in the UIA1, UIIA1, UIIIA1 and UIIIA2 interactions and there was no statistical difference between them (Table 5).

The average drying rate among the harvesting systems was 3.37 % in UIII, 3.05 % in UI and 2.95 % in UII. This difference resulted from the differences in swath widths during harvest (80 cm in UII, 140 cm in UI, 160 cm in UIII) and the function of the conditioning units. The values found, Shinners et al. (1991) reported that drying in a wide swath increased the drying rate by 34% compared to drying in a narrow swath, Rotz and Sprott (1984) reported that alfalfa dried 20-30% slower in a narrow swath than in a wide swath, and Greenlees et al. (2000) is consistent with the findings that the forage obtained from mowers with finger conditioning dries faster than from mowers with roller conditioning.

Table 6. Results of variance analysis applied to the drying rate index values of alfalfa in different harvesting practices

Source of variation	DF	SS	MS	F	
Application	2	1.4283	0.7141	14.8**	Time
Time	4	29.8794	7.4699	154.84**	4.4433a
ApplicationxTime	8	2.2298	0.2787	5.78**	3.4789b
Error	30	1.4473	0.0482		3.0956c
Total	44	34.9848			2.5011d
					2.0922e
					LSD=0.286

(**) sign indicates that the difference is significant according to the 1% probability limit.

It is noteworthy that the drying rate value is high in the second time interval of UIII. It can be said that the finger conditioner is effective in breaking down the material.

3.4. Results Regarding Dry Matter Losses

The average dry matter losses of the samples taken before and after baling during the trials are given in Table 7.

Table 7. Dry matter loss (%)

Applications	Before baling (mowing + raking)	After baling	Total dry matter loss
UI	1.49±0.031e	2.23±0.036c	3.72b
UII	1.86±0.031d	0.40±0.025f	2.26c
UIII	3.01±0.121b	4.79±0.193a	7.80a
	LSD=0.2444		LSD=0.558

Examining Table 7, it can be seen that after the mowing and raking process, the lowest dry matter loss was in UI (1.49%), followed by UII (1.86%) and UIII (3.01), respectively. After baling, it was determined that the highest dry matter loss was in UIII with a value of 4.79%, followed by UI with 2.23% and UII with 0.40%. When the total dry matter loss of the systems was evaluated, it was determined that the lowest dry matter loss was obtained in the UII application with a rate of 2.26%, and the highest was obtained in the UIII application with 7.80%. It was determined that dry matter loss occurred 1.65 times more in UI and 3.45 times more in UIII compared to UII. When Table 8 is examined, this situation was found to be statistically significant ($p<0.01$).

Table 8. Results of variance analysis applied to total dry matter loss index values of alfalfa for different harvesting practices

Source of variation	DF	SS	MS	F
Application	2	49.487	27.744	719.75**
Error	6	0.206	0.034	
Total	8	49.693		

(**) sign indicates that the difference is significant according to the 1% probability limit.

It is thought that the moisture content of the windrows before baling is effective in dry matter losses. Before bale making, the moisture content of the windrows was found to be 12% in UI, 20% in UII and 9% in UIII. In addition, it is considered that the finger conditioner has an effect on the high dry matter loss in UIII. Shinners et al. (1991) that increasing the violence of the conditioner in the mower increased leaf loss, and Greenlees et al. (2000) reported that finger conditioner caused more leaf loss. Rotz and Muck (1994) reported in their study that increasing swath density caused the loss rate to decrease, while Savoie (1988) reported that leaf loss was greatly affected by moisture content and that leaf breakage increased exponentially when the humidity dropped below 30%.

3.5. Forage Quality Values

As a result of the research, the results obtained by analyzing the samples on the NIR device are shown in Table 9.

Table 9. Forage values of alfalfa (%)

Applications	CP	ADF	NDF
UI	17.63b	26.93b	31.97b
UII	21.33a	28.27b	30.93b
UIII	16.87b	30.76a	34.77a
	LSD=2.210	LSD=1.508	LSD=3.319

Crude protein (CP) values of dry alfalfa turned into bales after harvest with different systems were obtained as 17.63% in UI, 21.33% in UII and 16.87% in UIII. Budak and Budak (2014) stated that if the CP ratio is 12% and below, the forage is low quality, if it is 15%, it is medium quality, and if it is 18% and above, it is high quality forage. As a result of the variance analysis applied to the crude protein values, the crude protein value obtained in the UII application was found to be statistically significant ($p<0.01$) compared to the other two applications (Table 10). According to this classification, we can state that the roughage obtained as a result of applications numbered UI and UIII is of medium quality, and the roughage obtained as a result of application numbered UII is of high quality.

Table 10. Results of variance analysis applied to crude protein values

Source of variation	DF	SS	MS	F
Application	2	34.229	17.114	32.09**
Error	6	3.20	0.533	
Total	8	37.429		

(**) sign indicates that the difference is significant according to the 1% probability limit.

ADF value is a forage criterion used to determine the digestibility of roughage by the animal. For a good hay quality, ADF values are required to be as low as possible. In applications, the ADF values of the forage were determined as 26.93%, 28.27% and 30.76%, respectively. The highest ADF value was obtained in the UIII application. While there was a statistically significant difference ($p<0.05$) between the other two applications, no statistically significant difference was found between the UI and UII applications (Table 11).

Table 11. Results of variance analysis applied to ADF values

Source of variation	DF	SS	MS	F
Application	2	22.722	11.361	9.82*
Error	6	6.940	1.157	
Total	8	29.662		

(*) sign indicates that the difference is significant according to the 5% probability limit.

NDF value is a forage criterion used to determine the ability of roughage to be taken by animals. Since the high NDF value in the forage slows down digestion, it causes the animal to feel physically full and the amount of forage the animal consumes decreases. In the trial, the lowest NDF value was obtained in UII (30.93%), which has a high protein content. However, no statistically significant ($p<0.05$) relationship was found between the NDF value obtained in UI (31.97%) (Table 12).

Russell and Johnson (1993) reported that the higher the NDF value of the forage, the lower its consumption by animals and that forages containing less than 31% ADF and less than 40% NDF are good quality forage. In the studies conducted, Ayaz (2010) reports that ADF and NDF values vary between respectively 23.22% - 26.67% and 33.67% - 38.74%, respectively, and Gökkaya (2019) varies between 25.23% - 31.87% and 40.26% - 46.84%.

Table 12. Results of variance analysis applied to NDF values

Source of variation	DF	SS	MS	F
Application	2	29.136	14.568	5.27*
Error	6	16.600	2.767	
Total	8	45.736		

(*) sign indicates that the difference is significant according to the 5% probability limit.

3.6. Relative Forage Values

RFV values, which are the index of forage intake and digestibility by animals, were calculated as 205.18 for UI, 201.21 for UII, and 173.75 for UIII, and the results are shown in Figure 2.

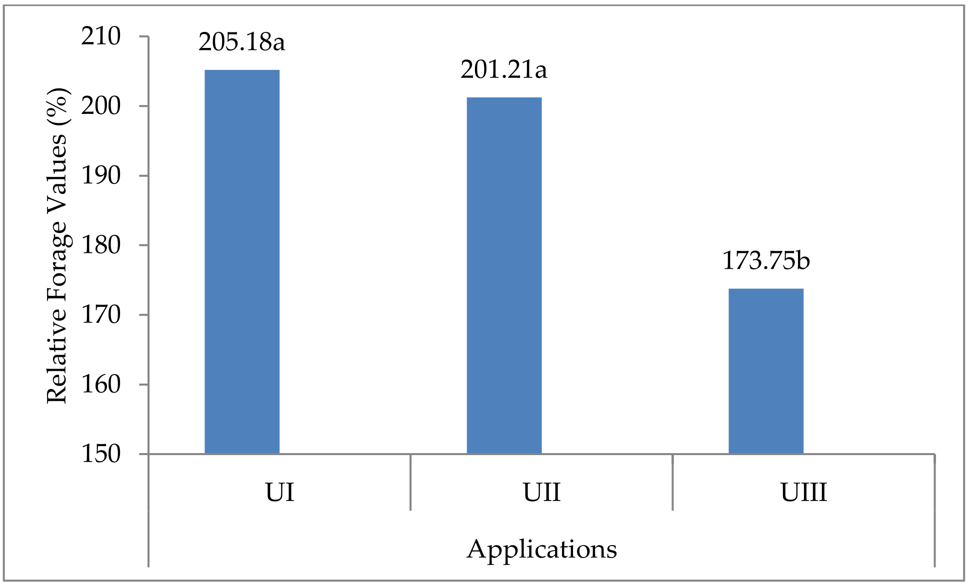

Figure 2. Relative forage values of alfalfa obtained as a result of different applications (%)

By examining Figure 2, RFV values were determined as 205.18%, 201.21% and 173.75%, respectively, according to the applications. As a result of variance analysis, the difference between the applications is significant ($p<0.05$), and the source of the difference is the UIII application. Buckmaster (1993) stated that RFV in animal nutrition should be between 54 and 230. The results of the variance analysis applied to the RFV values are given in Table 13.

Table 13. Results of variance analysis applied to RFV values

Source of variation	DF	SS	MS	F
Application	2	1756.9	878.4	5.14*
Error	6	1028.8	171.5	
Total	8	2785.6		

(*) sign indicates that the difference is significant according to the 5% probability limit.

4. Conclusions

The average fresh grass yield of the trial material alfalfa was 1 831 kg da^{-1} and the dry grass yield was found to be 421 kg da^{-1} on average.

Alfalfa was harvested with three different harvesting systems: disc mower (UI), disc mower + rubber roller conditioning unit (UII) and disc mower + finger type conditioning unit (UIII).

The working widths of these machines measured as 245 cm, 315 cm and 245 cm, respectively. As a result of the harvest, swath widths were 160 cm in UI (65% of the mowed area), 80 cm in UII (25.4% of the mowed area) and 140 cm in UIII (57% of the mowed area). At the end of the second day, the swaths were combined with a hay rake to create new windrows with a width of 90 cm. At the end of the third day, rectangular (36 cm x 46 cm x 90 cm) bales were made using a baler.

It was determined that moisture loss was close to each other in all three harvest systems on the first day of harvest, and that alfalfa lost moisture faster in UIII from the second day of harvest. While alfalfa harvested with UIII reached 20% moisture content after approximately 25 hours, alfalfa harvested with UI reached 20% moisture content after 31 hours and alfalfa harvested with UII after 49 hours. At the end of the day of windrow combining, the moisture content of the material was found to be at its lowest value at 15.9% at UIII.

The average drying rates among the harvesting systems were 3.37, 3.05 and 2.95 in UIII, UI and UII, respectively. This difference resulted from the differences in swath widths during harvest and the function of the conditioning units.

When total dry matter losses are evaluated; It was detected in the UII application with a rate of 2.26%, in the UI application with a rate of 3.72% and in the UIII application with a value of 7.80%. The dry matter loss was caused by differences in the moisture content of the windrows before baling.

Crude protein values (CP) of the trial material after baling were determined as 17.6% in UI, 21.3% in UII and 16.9% in UIII. ADF values of the forage obtained after different applications were determined as 26.9%, 28.3% and 30.8% in UI, UII and UIII applications, respectively, and NDF values were determined as 32.0%, 30.9% and 34.8%, respectively. Relative forage values (RFV) were determined as 205.18%, 201.21% and 173.75% according to the applications, respectively, and the highest value was obtained in the UI application.

As a result of this trial, it was determined that the disc mower (UIII) with a finger type conditioning unit would not be appropriate to use in its current form in the harvest of materials such as alfalfa, as it caused a high rate of dry matter loss. However, it is thought that it would be appropriate to conduct research as a result of improvements that can be made on the machine (change in the speed of the conditioner unit, preventing fingers from breaking the material, etc.).

The formation of narrow swath heaps as a result of harvesting with a disc mower (UII) equipped with a rubber roller conditioning unit caused a longer drying time. In order to shorten the drying time, it would be beneficial to increase the swath width at the time of harvest.

Research should be conducted on different forage crops in terms of forage quality values using disc mowers with rubber rollers and finger type conditioners.

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References

Anonymous (2020). Çayır mera ve yem bitkileri. https://www.tarimorman.gov.tr/Konular/Bitkisel-Uretim/Cayir-Mera-ve-Yem-Bitkileri: (access date: 15.11.2020).

Ayaz H (2010). Elçi yoncasında farklı biçim zamanlarında tarımsal ve fizyolojik özelliklerin gün içerisindeki değişimi. Master Thesis, Kahramanmaraş Sütçü İmam University (Unpublished), Turkey.

Buckmaster D (1993). Alfalfa raking losses as measured on artificial stubble. *Transactions of the ASAE*, 36 (3): 645-651.

Budak F, Budak F (2014). Yem bitkilerinde kalite ve yem bitkileri kalitesini etkileyen faktörler. *Türk Bilimsel Derlemeler Dergisi*, 7 (1): 01-06.

Bulgurlu Ş, Ergül M (1978). Yemlerin fiziksel, kimyasal ve biyolojik analiz metotları. *Ege Üniversitesi Ziraat Fakültesi Yayınları*, İzmir, Türkiye, pp. 58-76.

Demiroğlu G, Geren H, Avcıoğlu R (2008). Farklı yonca (*Medicago sativa* L.) genotiplerinin Ege bölgesi koşullarına adaptasyonu. *Ege Üniversitesi Ziraat Fakültesi Dergisi,* 45(1):1-10.

Dursun E, Güner M (1999). Biçme-ezme makinasının yoncanın tarlada kuruma hızına etkisi. *Tarım Bilimleri Dergisi*, 5 (2): 23-29.

Evcim Ü (1979). Tarlada kurutma amacıyla yonca hasadında değişik biçim ve biçim sonrası işlemlerin performans irdelemesi ve kuruma olgusunu kestirmede optimizasyon. *E.Ü. Ziraat Fakültesi Yayınları*, İzmir, Türkiye, p. 413.

Gökkaya G (2019). Önemli bazı yonca çeşitlerinde (*Medicago sativa* L.) biçim zamanının verim ve kalite özellikleri üzerine etkilerinin saptanması. Master Thesis, Tekirdağ Namık Kemal University (Unpublished), Turkey.

Greenlees WJ, Hanna HM, Shinners KJ, Marley S, Bailey T (2000). A comparison of four mower conditioners on drying rate and leaf loss in alfalfa and grass. *Applied Engineering in Agriculture*, 16 (1): 1.

Mavruk A (2017). Kaba yem hasadında farklı tip çayır biçme makinelerinin iş başarılarının belirlenmesi. Master Thesis, Çukurova University (Unpublished), Turkey.

Mutlu Z (2019). Bazı yonca çeşitlerinde farklı biçim zamanı uygulamalarının ot verimi ve kalitesi üzerine etkileri. PhD Thesis, Ankara University (Unpublished), Turkey.

Rotz C, Muck R (1994). Changes in feed quality during harvest and storage. In: Feed quality, evaluation, and utilization, Eds: George C, Fahey Jr. American Society of Agronomy, Madison, USA, pp. 828-868.

Rotz C, Sprott D (1984). Drying rates, losses and fuel requirements for mowing and conditioning alfalfa, *Transactions of the ASAE*, 27 (3): 715-0720.

Rotz C, Abrams S, Davis R (1987). Alfalfa drying, loss and quality as influenced by mechanical and chemical conditioning, *Transactions of the ASAE*, 30 (3): 630-0635.

Rotz C (1995). Loss models for feed harvest, *Transactions of the ASAE*, 38 (6): 1621-1631.

Russell M, Johnson K (1993). Selecting quality hay for horses, *ID-Cooperative Extension Service, Purdue University (USA).*

Savoie P (1988). Hay tedding losses. *Canadian Agricultural Engineering*, 30 (1): 39-42.

Schroeder J (1994). Interpreting feed analysis. North Dakota State Univ. Coop. Ext. Serv. Pub. AS-1080.

Sessiz A, Eliçin AK, Turgut MM, Pekitkan FG (2020). Tarım Makinaları Esasları. *Nobel Akademik Yayıncılık*. Yayın No:3056. ISBN: 978-625-406-658-0.

Shinners KJ, Koegel RG, Straub RJ (1991). Leaf loss and drying rate of alfalfa as affected by conditioning roll type, *Applied Engineering in Agriculture*, 7 (1): 46-49.

Toruk F (1997). Yonca hasadında farklı hasat sistemlerinin otun kuruma olgusu ve kalitesi üzerine etkilerinin saptanması. PhD Thesis, Trakya University (Unpublished), Turkey.

TurkStat (2021). Turkish Statistical Institute. https://data.tuik.gov.tr/Kategori/GetKategori?p=tarim-111&dil=1 (access date: 13.05.2021).

Van Dyke N, Anderson P (1994). Interpreting a feed analysis, *Circular ANR,* USA, p. 890.

Vurarak Y (2016). Bazı yem bitkileri karışımlarında farklı hasat ve muhafaza sistemlerinin makina işletmecilik değerleri ve yem kalitesi üzerine olan etkilerinin belirlenmesi. PhD Thesis, Çukurova University (Unpublished), Turkey.

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Some Germination and Emergence Characteristics of Cocklebur (*Xanthium strumarium* L.) Seeds

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HIGHLIGHTS

- The study determined that there was no dormancy in the large seeds of cocklebur, but there was a dormancy state in the small seeds.
- In germination studies conducted with seeds of cocklebur from different provinces; It was found that seeds had a wide germination temperature range between 10 and 40°C.
- The research demonstrated that the application of gibberellic acid (GA_3) had no effect on the germination rates of large seeds. However, it was observed that GA_3 increased the germination rates of small seeds.

Abstract

Cocklebur (*Xanthium strumarium* L.) is an annual broadleaf weed that causes significant yield losses in many crops and reproduces by seed. This study was conducted in 2020-2022 to contribute to alternative control by determining some biological characteristics of cocklebur seeds. For this purpose, in seed studies, the lower (large) and upper (small) seeds in the cocklebur fruit were placed in the incubator at 27±1°C and the germination rates were determined. Furthermore, the minimum, optimum and maximum germination temperatures of cocklebur seeds collected in 23 provinces of Türkiye were also determined. For this purpose, the studies were carried out at temperatures of 5, 10, 15, 20, 25, 30, 35, 40 and 45°C. In the studies to determine the optimum emergence depth of cocklebur; 25 cocklebur fruits were planted in pots at soil depths of 2, 5, 10, 15, 20 and 25 cm. The study found that the maximum germination rate of cocklebur was 93.1% for large seeds, while this rate was 21.2% for small seeds. The study revealed that the highest germination rate of cocklebur in the soil is at a depth of 2-10 cm. In the studies on seed germination temperatures, it was determined that the minimum temperature was 5°C, the optimum temperature was 15-35°C and the maximum temperature was 40°C, although it varied according to the province where seeds were collected. These findings indicate that this weed exhibits a substantial temperature requirement and will spread more in agricultural areas in different ecologies.

Keywords: Germination biology; germination temperature; depth; *Xanthium strumarium*

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1. Introduction

Weeds are defined as plants that are undesirable and grow spontaneously in their habitat. Weeds compete with cultivated plants for mineral nutrients, water, light and space, and due to this competition, they cause yield, quality and quantity losses in products (Güncan and Karaca 2014).

Knowledge of the biology of weeds is important for the development of economical and environmentally friendly weed control methods, which are the basic principles of integrated control. Weed biology refers to plant characteristics such as plant morphology, seed dormancy and germination, growth physiology, competitive ability and reproductive biology. The biological concepts of populations such as the dynamics of seed banks and root reserves in annual plants and the longevity of vegetative reproduction in perennial plants can be used to better the prediction of weed invasion and the evaluation of sustainable management strategies (Bhowmik 1997). Weeds differ in species and genetic diversity depending on geographic region, crop, season and planting date (Pala et. al. 2018). Therefore, the seeds of plants growing in different ecologies have different germination characteristics (Wang et al. 2023). Accurate information on seed germination biology is required for the successful control of any weed species

Cocklebur (*Xanthium strumarium* L.) is an annual, broad-leaved weed that belongs to the Asteraceae family and reproduces by seed (Venodha 2016; Eymirli and Torun 2015; Cesur et al. 2017; Özaslan 2021). The plant is capable of growing to between 20 and 150 centimeters in height (Weaver and Lechowicz 1983) and one plant can produce an average of 70 to 600 fruits (Eymirli and Torun 2015). Each fruit usually contains two seeds, one of which is usually larger than the other, and they differ in their germination requirements (Barton 1962; Hicks 1971).

Cocklebur is an extremely invasive and spreads rapidly. It spreads to agricultural land, pastures, national parks, rivers, dams, roadsides and urban green spaces, causing major problems for biodiversity and significant economic losses (Eymirli and Torun 2015; Tadesse et al. 2017; Chikuruwo et al. 2017; Machado et al. 2021; Özaslan 2021). It has been reported to cause significant yield losses in row crops such as soybean, cotton, corn and peanut in many parts of the world. (Amini and Izadkhah 2013; Tepe 2014; Özaslan 2021). In Turkey, cocklebur is a plant that can grow in almost all regions and under all types of climate and soil conditions (TBL 2024). It has been reported that cocklebur is currently distributed in the Mediterranean, Black Sea and Southeastern Anatolia regions and has the highest density in the Southeastern Anatolia region (Özaslan 2021). Additionally, cocklebur can increase invasion success with its high reproductive capacity, long-term seed survival and ability to adapt to different ecological conditions, as well as rapid spread and dispersal.

Cocklebur not only causes significant yield losses in agricultural areas, but is also a highly toxic plant (Tepe 2014). The toxicity of cocklebur is due to the presence of xanthostrumarin and carboxyatractyloside glycosides in the seeds and seedlings of the plant. These glycosides are particularly harmful to cattle, horses, sheep and pigs. Ingestion of the cotyledons by grazing animals can result in a range of adverse effects, including vomiting, muscle cramps, liver damage and even death (Eymirli and Torun 2015).

Knowing some of the biological characteristics of cocklebur seeds such as germination temperature and emergence depth will help to use alternative control options. In the context of weed control, the emergence depth of weeds is a crucial factor in determining the appropriate tillage depth. This knowledge is also instrumental in determining the tillage depth of herbicides applied prior to sowing. (Güncan ve Karaca 2014). Although there are many studies on the ecological development characteristics of this weed, studies on germination temperatures and soil vigor under different ecological conditions are very limited. For this reason, this study was conducted to determine some germination and emergence characteristics of cocklebur seeds from different geographical regions of Türkiye.

2. Materials and Methods

2.1. Site and Plant Material

The main material of the research consisted of cocklebur fruits collected from 23 provinces in different regions of Turkey. (Table 1). The cocklebur fruits intended for the germination studies were stored in liquid nitrogen for a duration of 1-2 minutes, followed by a cracking process on the wood surface to remove the lower (large) and upper (small) seeds within the fruit.

The seeds were stored at +4°C until they were used in the germination experiments. The cocklebur seeds used to determine seed germination rates and emergence depth were obtained from the Diyarbakır province. The experiments were repeated twice, with four replications, according to the randomized plot design.

Table 1. Regions, provinces and altitudes where cocklebur seeds are obtained.

Regions	Provinces	Climatic properties	Altitudes (m)
Mediterranean	Antalya	Mediterranean	15m
	Adana	Mediterranean	2 m
Eastern Anatolia Region	Bitlis	Terrestrial	1793m
	Erzincan	Terrestrial	1290m
	Elazığ	Terrestrial	1089m
	Malatya	Terrestrial	815m
	Muş	Terrestrial	1959m
	Iğdır	Terrestrial	1455m
	Van	Terrestrial	1770 m
Aegean	Aydın	Mediterranean	22m
	İzmir	Mediterranean	5m
	Afyonkarahisar	Terrestrial	1066m
Southeastern Anatolia	Diyarbakır	Terrestrial	636m
	Batman	Terrestrial	569m
	Mardin	Terrestrial	602m
	Şanlıurfa	Terrestrial	445 m
	Siirt	Terrestrial	615m
Central Anatolia	Kayseri	Terrestrial	1180m
	Konya	Terrestrial	1184m
	Niğde	Terrestrial	1367m
Black Sea	Tokat	Transitionclimate of Black Seaand Central Anatolian	1272m
	Ordu	Temperate	521m
Marmara	Tekirdağ	Mediterranean	176m

2.2. Germination Experiments

The germination rates and germination speeds of the lower and upper seeds of cocklebur were determined according to their position within the fruit. In the experiments, the upper and lower seeds of the weed were placed in 9 cm Petri dishes containing Whatman filter paper No. 1 (moistened with 6 ml pure water). After 20 seeds had been placed in each Petri dish, they were placed in germination cabinets set to a temperature of 27±1°C and a 16-hour light/8-hour dark period. Pure water was added to the Petri dishes as required throughout the experiment. Observations were made every day after sowing. The plants whose radicle length reached 0.5 cm on day 14 were considered germinated and removed from the Petri dish.

Application of gibberellic acid (GA_3); to determine the effect of gibberellic acid application on seed germination, cocklebur seeds were subjected to two different applications of GA_3.

In the first application, the upper and lower seeds were immersed in pure water (control) and different concentrations of GA_3 (500, 1000, 1500, 2000 and 2500 ppm) for a duration of 24 hours (Cesur et al. 2017). After application, the cocklebur seeds were planted in 9 cm petri dishes. Following this, 6 ml of pure water was added to the Petri dishes and placed in germination cabinets.

In the second application, the upper and lower seeds were planted in 9 cm Petri dishes. After planting the seeds, 6 ml of the solution prepared from pure water (control) and different concentrations of GA_3 (500, 1000, 1500, 2000 and 2500 ppm) were added to the Petri dishes (Ateş and Üremiş 2018). After 20 seeds were placed in each petri dish, they were placed in germination cabinets with a temperature of 27±1°C and a 16-hour light / 8-hour dark period. In the control application, pure water was added, while the solution prepared from different GA_3 concentrations was added as required in the other applications.

Since the difference between the two repetitions was insignificant, statistical analyses were performed by calculating the average of the two repetitions of the data obtained from the applications. As a result of the analyses, germination rate (G_{max}) and germination times (T_{50} and T_{90}) were calculated.

$$G_{max} = (G/T) \times 100 \quad (1)$$

G: Number of germinating seeds (number)
T: Total number of seeds used in the experiment (pieces)
T_{50} = Time taken for 50% of the germinated seeds to germinate (days)
T_{90} = Time taken for 90% of the germinated seeds to germinate (days)

2.2.1. Determination of germination temperatures of cocklebur seeds from different provinces

To determine the minimum, optimum and maximum germination temperatures of cocklebur seeds, seeds from 23 provinces were collected in order to represent each region. Large seeds of the cocklebur without dormancy were used for the germination temperature study. After surface sterilization with 1% (NaClO), large seeds were placed in 9 cm Petri dishes containing Whatman filter paper No. 1 (moistened with 6 ml pure water). Each Petri dish placed 10 seeds of similar size. The Petri dishes were then placed in germination cabinets, which were set to constant temperatures of 5, 10, 15, 20, 25, 30, 35, 40 and 45°C. Pure water was added to the Petri dishes as needed during the experiment. Observations were made every day from planting. Those whose radicle length reached of 0.5 cm by the 21st day were considered germinated and removed from the Petri dish (Üremiş and Uygur 1999).

2.2.2. Determination of the emergence depth of cocklebur seeds

In the studies conducted to determine the optimum emergence depth of cocklebur, 25 cocklebur fruits were planted in pots at soil depths of 2, 5, 10, 15, 20 and 25 cm. The soil used for the experiment was sterilized by placing it in an oven at 120°C for a duration of 30 minutes. The mixture of soil, peat and sand (1:1:1) was then added to the pots (Özkil 2021). After sowing was completed, observations were made at one-week intervals for 60 days and the germinated seeds were counted when they had cotyledons or two leaves and were removed from the pot (Erciş et al. 1993). In the evaluations, the emergence percentages were calculated.

2.2.3. Data analysis

The data obtained from the studies was analysed using a one-way Generalised Linear Model (GLM) with Analysis of Variance (ANOVA) in IBM SPSS 25 statistical software package. The difference between the applications was determined according to the significance level $P \leq 0.05$ using the Duncan multiple comparison test.

3. Results

3.1. Seed Germination Trials

In the seed germination experiments, the difference between the germination rates of large and small seeds was found to be statistically significant at the $P \leq 0.05$ level. However, no statistical difference was found in the germination times of the seeds.

In the control application, the maximum germination rate (G_{max}) of the large seeds of cocklebur was found to be 93.1% when pure water was applied, while the maximum germination rate (G_{max}) of the small seeds was 21.2%. When the seeds were evaluated for germination times, it was found that the time taken for 50% of both the large and small seeds to germinate T_{50} was 1.2 days and the time taken for 90% of the germinated seeds to germinate T_{90} was 2.5 days (Table 2).

Table 2. Effect of pure water application on seed germination rates (%) and duration

Pure water application	G_{max}	T_{50}	T_{90}
Large seeds	93.1±1.8a	1.2±0.2a	2.5±0.8a
Small seeds	21.2±1.2b	1.2±0.3a	2.5±0.6a

Each column was evaluated on its own.

When cocklebur seeds were kept in different GA_3 concentrations for a duration of 24 hours, it was found that large seeds showed 100% germination rate with the 24-hour application of 1500 ppm GA_3 and the 24-hour application of pure water (control). In the study, it was found that the application of GA_3 had no effect on the germination rates of large seeds of cocklebur. However, an increase in the concentration of gibberellic acid was observed to result in an increase in the germination rates of small seeds. Specifically, while the lowest germination rate of 45.0% was achieved in small seeds with a 24-hour application of pure water, the highest germination rate of 81.2% was achieved with a 24-hour application of 2000 ppm GA_3. When the effects of the different GA_3 applications on seed germination times were evaluated, the highest T_{50} (germination times of 50% of germinating seeds) and T_{90} (germination times of 90%) times were obtained for large seeds by the 24-hour application of 500 ppm GA_3. Conversely, for small seeds, the statistical difference in the T_{50} and T_{90} times of the applications was not significant (Table 3).

Table 3. Effect of different GA_3 applications on germination rates (%) and duration (days) of cocklebur seeds

	Large seeds			Small seeds		
Treatments	G_{max}	T_{50}	T_{90}	G_{max}	T_{50}	T_{90}
24 hours pure water	100.0±0.0^{a}	1.2±0.2^{b}	2.0±0.4^{b}	45.0±3.5^{c}	1.1±0.9^{a}	1.4±0.2^{a}
24 hours 500 ppm GA_3	87.5±4.7^{b}	4.5±0.9^{a}	4.7±1.1^{a}	62.5±5.9^{b}	1.3±0.0^{a}	3.0±1.0^{a}
24 hours 1000 ppm GA_3	97.5±2.5^{a}	1.2±0.2^{b}	1.7±0.4^{b}	72.5±2.5^{ab}	1.2±0.8^{a}	3.5±0.8^{a}
24 hours 1500 ppm GA_3	100.0±0.0^{a}	2.0±1.0^{b}	3.5±0.8^{ab}	63.0±7.2^{b}	1.3±0.0^{a}	1.8±0.3^{a}
24 hours 2000 ppm GA_3	95.0±2.8^{ab}	1.2±0.2^{b}	2.0±0.0^{b}	81.2±1.2^{a}	1.2±0.8^{a}	3.2±0.8^{a}
24 hours 2500 ppm GA_3	97.0±2.5^{a}	2.0±0.40^{b}	2.7±0.7^{ab}	75.0±4.1^{ab}	1.4±0.8^{a}	3.4±0.7^{a}

Each column was evaluated on its own. Values marked with the same letter are not statistically different. ($P>0.05$).

Table 4. Effect of different GA_3 applications on germination rates (%) and duration (days) of cocklebur seeds

	Large seeds			Small seeds		
Treatments	G_{max}	T_{50}	T_{90}	G_{max}	T_{50}	T_{90}
Pure water (control)	98.7±1.2^{a}	2.0±0.0^{a}	2.0±0.0^{a}	18.7±2.3^{c}	1.7±0.1^{a}	2.5±0.7^{a}
500 ppm GA_3	97.5±1.4^{a}	1.7±0.2^{a}	2.2±0.2^{a}	40.0±4.0^{a}	1.8±0.4^{a}	3.0±0.8^{a}
1000 ppm GA_3	100.0±0.0^{a}	1.7±0.2^{a}	2.0±0.0^{a}	25.0±7.9^{bc}	1.7±0.1^{a}	3.7±0.9^{a}
1500 ppm GA_3	98.7±1.25^{a}	1.5±0.3^{a}	2.0±0.0^{a}	33.7±2.3^{ab}	2.2±0.8^{a}	3.6±0.7^{a}
2000 ppm GA_3	100.0±0.0^{a}	1.7±0.2^{a}	2.0±0.0^{a}	42.5±2.5^{a}	2.1±0.6^{a}	3.7±0.8^{a}
2500 ppm GA_3	100.0±0.0^{a}	1.7±0.2^{a}	2.0±0.0^{a}	36.7±3.7^{ab}	1.8±0.2^{a}	5.5±0.5^{a}

Each column was evaluated on its own.

When cocklebur seeds of different GA_3 concentrations were added to 6 ml of Petri medium, it was found that the difference between the treatments was not statistically significant in terms of germination rates of the large seeds at the $P \leq 0.05$ level. In the study, it was determined that different concentrations of GA_3 had no effect on the germination rates of large seeds. When the germination rates of small seeds were evaluated, difference between the treatments was found to be statistically significant ($P \leq 0.05$). For small seeds, the lowest germination rate was obtained with the application of pure water (control) at 18.7%, while the highest germination rate was obtained with the application of 2000 ppm GA_3 at 42.5%. When the effect of applications on germination times was evaluated, it was found that the difference between the T_{50} and T_{90} times of large seeds was not statistically significant. When evaluating the effects of small seeds on germination times, it was found that the difference between the T_{50} germination times of the treatments was not statistically significant, while the effects on the T_{90} germination times were significant. The highest T90 germination time for small seeds was achieved with the application of 2500 ppm GA3 at 5.5 days (Table 4).

3.2. Germination Temperatures of Cocklebur Seeds from Different Provinces

In germination studies conducted with large seeds of cocklebur from different provinces; it was found that the lowest germination rates were at temperatures of 5 °C and 40°C. When the germination rates of cocklebur seeds at 5°C were examined, it was found that there was no statistical difference between the provinces. Notably, no germination was observed in 14 provinces (Bitlis, Diyarbakır, Elazığ, Iğdır, İzmir, Kayseri, Konya, Malatya, Mardin, Muş, Siirt, Şanlıurfa, Tokat, Van), while a very low germination rate of 2.5% was recorded in 9 provinces (Adana, Afyon, Antalya, Aydın, Batman, Erzincan, Niğde, Ordu, Tekirdağ). In the experiments, the most suitable temperature for seed germination was determined as 15-35°C, although it varies by province. (Table 5).

Table 5. Minimum, optimum and maximum germination temperatures of cocklebur from different provinces

Provinces	5°C	10°C	15°C	20°C	25°C	30°C	35°C	40°C	45 °C
Adana	2.5 ±1.4$_{a}^{D}$	65.0±6.1$_{a}^{B}$	96.0±2.6$_{a}^{A}$	97.5±2.5$_{a}^{A}$	100.0±0$_{a}^{A}$	100.0±0a^{A}	98.7±1.2$_{a}^{A}$	35.0±3.5$_{ab}^{C}$	0.0±0.0$_{a}^{D}$
Afyon	2.5±1.4$_{a}^{D}$	30.0±7.9$_{d}^{B}$	67.5±3.2$_{bc}^{A}$	81.2±2.3$_{def}^{A}$	81.2±1.2$_{f}^{A}$	81.2±5.5$_{f}^{A}$	81.2±3.7$_{cde}^{A}$	16.2±7.4$_{bcde}^{C}$	0.0±0.0$_{a}^{D}$
Antalya	2.5±2.5$_{a}^{E}$	61.2±3.7$_{ab}^{C}$	73.7±3.1$_{b}^{B}$	100.0±0$_{a}^{A}$	98.7±1.2$_{ab}^{A}$	100.0±0$_{a}^{A}$	95.0±3.5$_{ab}^{A}$	11.2±5.1$_{cde}^{D}$	0.0±0.0$_{a}^{E}$
Aydın	2.5±1.4$_{a}^{D}$	7.5±1.4$_{e}^{D}$	43.7±5.5$_{cd}^{C}$	78.7±5.1$_{def}^{B}$	98.7±1.2$_{ab}^{A}$	92.5±3.2$_{abcd}^{A}$	93.7±1.2$_{ab}^{A}$	6.2±3.1$_{e}^{D}$	0.0±0.0$_{a}^{D}$
Batman	2.5±1.4$_{a}^{C}$	41.2±3.1$_{bc}^{B}$	70.0±14.8$_{b}^{A}$	84.7±4.6$_{bcde}^{A}$	85.0±2.8$_{ef}^{A}$	86.2±3.1$_{def}^{A}$	87.5±4.7$_{abcde}^{A}$	17.5±5.2$_{bcde}^{C}$	0.0±0.0$_{a}^{C}$
Bitlis	0.0±0.0$_{a}^{F}$	8.7±1.2$_{e}^{E}$	47.5±4.7$_{bcd}^{C}$	88.7±3.1$_{abcd}^{AB}$	92.5±1.4$_{bcd}^{A}$	91.2±2.3$_{abcde}^{AB}$	83.7±3.7$_{bcde}^{B}$	17.5±1.4$_{bcde}^{D}$	0.0±0.0$_{a}^{F}$
Diyarbakır	0.0±0.0$_{a}^{D}$	5.0±3.5$_{e}^{D}$	72.7±8.1$_{b}^{B}$	95.0±2.0$_{abc}^{A}$	95.0±0$_{abcd}^{A}$	92.5±2.5$_{abcd}^{A}$	100.0±0.0$_{a}^{A}$	20.0±7.0$_{bcde}^{C}$	0.0±0.0$_{a}^{D}$
Elazığ	0.0±0.0$_{a}^{D}$	2.5±1.4$_{e}^{D}$	26.7±4.1$_{d}^{C}$	71.7±3.8$_{fg}^{B}$	98.7±1.2$_{ab}^{A}$	93.7±1.2$_{abcd}^{A}$	76.2±6.5$_{e}^{B}$	21.2±5.1$_{bcde}^{C}$	0.0±0.0$_{a}^{D}$
Erzincan	2.5±2.5$_{a}^{EF}$	13.7±4.2$_{e}^{DE}$	52.5±2.5$_{c}^{C}$	77.5±10.3$_{def}^{B}$	93.7±1.2$_{abcd}^{A}$	83.7±3.1$_{ef}^{AB}$	80.7±1.4$_{cde}^{B}$	17.5±5.9$_{bcde}^{D}$	0.0±0.0$_{a}^{F}$
Iğdır	0.0 ±0.0$_{a}^{D}$	6.2±4.7$_{e}^{D}$	77.5±7.5$_{b}^{B}$	97.5±2.5$_{a}^{A}$	100.0±0$_{a}^{A}$	100.0±0$_{a}^{A}$	98.7±1.2$_{a}^{A}$	50.2±1.6$_{a}^{C}$	0.0±0.0$_{a}^{D}$
İzmir	0.0 ±0.0$_{a}^{D}$	3.7±2.3$_{e}^{D}$	18.7±3.7$_{d}^{C}$	74.7±5.2$_{efg}^{B}$	73.7±3.7$_{g}^{B}$	95.0±2.8$_{abcd}^{A}$	100.0±0$_{a}^{A}$	12.5±6.2$_{cde}^{CD}$	0.0±0.0$_{a}^{D}$
Kayseri	0.0 ±0.0$_{a}^{D}$	38.0±9.3$_{cd}^{C}$	75.0±2.0$_{b}^{B}$	95.0±0$_{abc}^{A}$	93.7±1.2$_{abcd}^{A}$	95.0±2.0$_{abcd}^{A}$	76.2±5.5$_{e}^{B}$	7.5±4.7$_{de}^{D}$	0.0±0.0$_{a}^{D}$
Konya	0.0 ±0.0$_{a}^{C}$	1.2±1.2$_{e}^{C}$	81.2±5.1$_{ab}^{B}$	93.7±3.7$_{abc}^{A}$	95.0±2.0$_{abcd}^{A}$	92.5±4.3$_{abcd}^{A}$	93.7±2.3$_{ab}^{A}$	7.5±3.2$_{de}^{C}$	0.0±0.0$_{a}^{C}$
Malatya	0.0±0.0$_{a}^{D}$	7.5±3.2$_{e}^{CD}$	62.5±6.6$_{bc}^{B}$	90.0±2.0$_{abcd}^{A}$	100.0±0$_{a}^{A}$	97.5±1.4$_{abc}^{A}$	98.7±1.2$_{a}^{A}$	16.2±6.2$_{bcde}^{C}$	0.0±0.0$_{a}^{D}$
Mardin	0.0 ±0.0$_{a}^{F}$	2.5±2.5$_{e}^{F}$	22.5±2.5$_{d}^{E}$	78.7±2.3$_{def}^{C}$	88.7±1.2$_{de}^{B}$	92.5±1.4$_{abcd}^{B}$	98.7±1.2$_{a}^{A}$	28.7±3.7$_{bc}^{D}$	0.0±0.0$_{a}^{F}$
Muş	0.0±0.0$_{a}^{F}$	12.5±4.7$_{e}^{E}$	26.2±3.7$_{ed}^{D}$	66.7±4.8$_{g}^{C}$	91.2±1.2$_{cde}^{A}$	96.2±1.2$_{abc}^{A}$	80.0±3.5$_{cde}^{B}$	10.0±2.0$_{cde}^{E}$	0.0±0.0$_{a}^{F}$
Niğde	2.5±1.4$_{a}^{D}$	10±4.5$_{e}^{D}$	30.0±3.5$_{d}^{C}$	82.5±4.7$_{cdef}^{B}$	92.5±3.2$_{bcd}^{AB}$	96.2±1.2$_{abc}^{A}$	91.2±1.2$_{abc}^{AB}$	27.5±7.5$_{bcd}^{D}$	0.0±0.0$_{a}^{D}$
Ordu	2.5 ±1.4$_{a}^{D}$	50.0±3.5$_{bc}^{C}$	78.7±7.7$_{ab}^{B}$	96.2±1.2$_{ab}^{AB}$	97.5±1.4$_{abc}^{AB}$	100.0±0$_{a}^{A}$	95.0±0.0$_{ab}^{AB}$	36.2±15.9$_{ab}^{C}$	0.0±0.0$_{a}^{D}$
Siirt	0.0±0.0$_{a}^{D}$	5.0±2.0$_{e}^{D}$	49.0±2.4 $_{bc}^{C}$	90.0±2.8$_{abcd}^{A}$	95.0±2.0$_{abcd}^{A}$	90.0±2.0$_{bcde}^{A}$	78.7±4.2$_{de}^{B}$	6.2±3.7$_{e}^{D}$	0.0±0.0$_{a}^{D}$
Şanlıurfa	0.0±0.0$_{a}^{F}$	8.7±2.3$_{e}^{EF}$	16.2±3.1$_{d}^{E}$	63.7±6.5$_{g}^{C}$	81.2±2.3$_{f}^{B}$	93.7±2.3$_{abcd}^{A}$	97.5±1.4$_{a}^{A}$	34.0±5.5$_{ab}^{D}$	0.0±0.0$_{a}^{F}$
Tekirdağ	2.5±2.5$_{a}^{E}$	48.7±3.1$_{c}^{C}$	65.0±7.3$_{bc}^{B}$	90.0±0$_{abcd}^{A}$	95.0±3.5$_{abcd}^{A}$	98.7±1.2$_{a}^{A}$	88.7±3.7$_{abcd}^{A}$	36.2±6.2$_{ab}^{D}$	0.0±0.0$_{a}^{E}$
Tokat	0.0±0.0$_{a}^{D}$	7.5±1.4$_{e}^{D}$	51.2±4.2$_{c}^{B}$	83.7±1.2$_{bcdef}^{A}$	93.7±1.2$_{abcd}^{A}$	90.0±4.0$_{bcde}^{A}$	85.0±6.7$_{bcde}^{A}$	18.7±6.2$_{bcde}^{C}$	0.0±0.0$_{a}^{D}$
Van	0.0±0.0$_{a}^{D}$	10.0±2.8$_{e}^{D}$	41.2±3.1$_{bcd}^{C}$	96.2±1.2$_{ab}^{A}$	85.0±4.5$_{ef}^{A}$	88.7±3.7$_{cdef}^{A}$	62.5±8.5$_{f}^{B}$	6.2±4.7$_{e}^{D}$	0.0±0.0$_{a}^{D}$

While uppercase letters represent groups between temperatures, lowercase letters represent groups between provinces ($P \leq 0.05$).

3.3. Determination of the Emergence Depth of Cocklebur Seeds

The studies show that the emergence rates of cocklebur at soil depths of 2, 5 and 10 cm are statistically in the same group and have the highest emergence rate. The highest emergence rates of cocklebur seeds at soil depths were observed to be 90.6% at a depth of 2 cm, 99.3% at a depth of 5 cm and 91.8% at a depth of 10 cm. The lowest emergence rate of cocklebur fruits was observed at a soil depth of 25 cm with 0.6% (Table 6).

Table 6. Emergence rates of cocklebur seeds according to different soil depths (%)

Soil depths (cm)	Emergence rate (%)
2	90.6a±4.8
5	99.3a±0.3
10	91.8a±4.0
15	50.0b±5.1
20	36.2c±4.8
25	0.6d±0.3

Values marked with the same letter are statistically the same ($P>0.05$).

4. Discussion

Studies have shown that dormancy is not to be found in the large seeds of the cocklebur, but in the small seeds. Similar to the results of our study, various researchers reported in their studies on dormancy that there are two seeds in the cocklebur, that the germination of these seeds is different, that there is no dormancy in the large seeds, that the small seeds do not germinate under normal growing conditions, and that they germinate about one year after the germination of the large seeds (Barton 1962; Esashi et al. 1983).

In the study to determine the highest germination rate of seeds as a result of different applications of GA_3, the highest germination rate of large seeds, 100%, was obtained by applying 1500 ppm GA_3 for 24 hours. Cesur et al. (2017) reported that the highest germination rate of 100% in different GA_3 applications in the germination of cocklebur seeds was obtained by applying 1500 ppm GA_3 for 24 hours, and they provided the same results as our study. Karimmojeni et al. (2010) reported in dormancy-breaking studies that the combination of cold (5±2 °C for 2 months) and GA_3 led to a slight increase in germination in cocklebur seeds. In the results of the study, it was found that the different applications of GA_3 had no effect on the germination of large seeds, but there was a slight increase in small seeds. In the study, it was found that different applications of GA_3 had no effect on the T_{50} and T_{90} germination times of cocklebur seeds. Similar to our study, Cesur et al. (2017) reported that different applications of GA_3 for 24 hours was had no effect on the germination time of seeds.

The study revealed that the optimal temperature range for cocklebur seeds in the Mediterranean region is 15-35°C, with a minimum of 5°C and a maximum of 40°C. Similar to our study results, in the study conducted by Kadıoğlu (1997) in the Mediterranean region, it was reported that the optimum germination temperature of cocklebur seeds was 25-30°C, with a maximum of 40°C. In the temperature studies conducted in the Aegean region, it was determined that the cocklebur seeds germinated under a minimum of 10°C, while the maximum germinated temperature was recorded as 35°C. (Kaya and Nemli 2004). In the present studies, the minimum temperature in the Aegean region was found to be 5°C, the optimum temperature was determined to be 25-35°C, and the maximum temperature was found to be 40°C. These results are consistent with the studies conducted in the Aegean region. When the germination temperature of cocklebur seeds in the Southeastern Anatolia region was examined, it was found that the highest seed germination rate was obtained at 35°C. This finding aligns with the results of other researchers, who have also reported that the optimal germination temperature for cocklebur seeds ranges from 25 to 40 °C (Bükün 1997; Norsworthy and Oliveira 2007; Saric-Krsmanovic et al. 2012; Goharian et al. 2019). In studies conducted in different countries, Saed et al. (2020) found that germination of cocklebur seeds in Pakistan occurred at temperatures between 10 and 45 °C, and Ahmedi et al. (2024) found that seeds germinated at temperatures between 10 and 40 °C in Iran. The study shows that there are differences in the germination temperatures of seeds from different provinces. In the study conducted by Karaman and Tursun (2021), it was reported that the germination temperatures of seeds grown in different ecological regions differ.

In the study, it was found that the highest emergence rate of cocklebur seeds in soil was at a depth of 2 to 10 cm, and the lowest emergence rate was at a depth of 25 cm. Similar our study, Kadıoğlu (1997) found that the highest emergence rate of cocklebur was 2 to 10 cm at different soil depths and the lowest emergence was 25 cm. In a separate study, Tao et al. (2021) reported that the majority of cocklebur fruits emerged between 0 and 3 cm, while Toledo et al. (1993) stated that the highest emergence rate of hogweed seeds occurred at a soil depth of 0 to 8 cm James et al. (2016) have stated that there is no emergence of cocklebur seeds at a depth of 10 cm. However, the our study found that emergence occurred even at a depth of 25 cm.

5. Conclusions

In the study, it should be taken into consideration that there are differences in the germination temperatures of seeds from different provinces and therefore the cocklebur control period may vary from province to province. A further finding of the study is that the seeds have a wide germination temperature range between 10 and 40 °C, which increases the invasive character of this weed species and causes considerable damage to the ecosystem and agricultural land. One of the most important practices in weed control is tillage. Tillage can influence on the distribution and germination of weed seeds in the soil. This study will help determine the depth of tillage by determining the depth of cocklebur emergence from the soil. Thus, it will help reduce the population of this weed species in agricultural areas by indicating to growers the depth at which tillage should be done. The results of this research will form the basis for future scientific studies to control the damage that cocklebur may cause in agricultural areas.

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References

Amini R, Izadkhah S (2013). Effect of seed position on parental plant on seed germination and seedling growth of common cocklebur (*Xanthium strumarium* L.). *Journal of Biodiversity and Environmental Sciences,* 3(10): 123-129.

Ahmadi A, Zaefarian F, Rezvani M, Mansouri I, Sindel BM (2024). Response of spiny cocklebur (*Xanthium spinosum* L.) and common cocklebur (*X. strumarium* L.) seed germination to environment and soil. Factors. *Australian Journal of Botany*, 72(4): 1-9. http://dx.doi.org/10.2139/ssrn.4374173

Ateş E, Üremiş İ (2018). Determination of germination temperatures of *Sinapis arvensis* L. and *Avena sterilis* L. *International Journal of Agricultural and Natural Science,s* 1(2): 154-159

Barton LV (1962). The germination of weed seeds. *Weeds,* 10(3):174-182. https://doi.org/10.2307/4040776

Bhowmik PC (1997). Weed biology: importance to weed management. *Weed Science,* 45: 349-356. https://doi.org/10.1017/S0043174500092973

Bükün B (1997). The Weeds found in cotton growing areas of Harran plain and determination of the most appropriate time of their control. M.Sc. Thesis, Harran University (Unpublished), Turkey.

Cesur C, Coşge Ş B, Uskutoğlu T, Yaman C, Yurteri T (2017). A study on the determination of optimum germination methods for cocklebur seeds (*Xanthium strumarium* L.). *Turkish Journal of Agricultural Research,* 4(2): 124-130. https://doi.org/10.19159/tutad.270676

Chikuruwo C, Masocha M, Murwira A, Ndaimani H (2017). Predicting the suitable habitat of the invasive *Xanthium strumarium* L. in Southeastern Zimbabwe. *Applied Ecology and Environmental Research,* 15(1): 17-31. https://doi.org/10.15666/aeer/1501_017032

Erciş A, Taştan B, Yıldırım A (1993). Yabani hardal (*Sinapis arvensis*)'ın bazı biyolojik özellikleri üzerinde araştırmalar. *Türkiye I. Herboloji Kongresi*, 3-5 Şubat 1993; Adana, Türkiye, pp. 55-61.

Esashi Y, Ishihara N, Kuraishi R, Kodama H (1983). Light actions in the germination of cocklebur seeds: I. dıfferences in the light responses of the upper and lower seeds. *Journal of Experimental Botany,* 34(7): 903-914. https://doi.org/10.1093/jxb/34.7.903

Eymirli S, Torun H (2015). *Xanthium strumarium*. Türkiye istilacı bitkiler kataloğu. T.C. Gıda, Tarım ve Hayvancılık Bakanlığı, Tarımsal Araştırmalar ve Politikalar Genel Müdürlüğü, Bitki Sağlığı Araştırmaları Daire Başkanlığı, Ankara, p.521-533.

Goharian A, Asilan KS, Mansourifar S (2019). Effect of environmental factors on some seed germination aspects of two populations of common cocklebur (*Xanthium strumarium* L.). *Iranian Journal of Seed Science and Research,* 6(3): 411-425.

Güncan A, Karaca M (2014). Yabancı ot mücadelesi (3. Baskı). Selçuk Üniversitesi Basımevi, Konya, p.334.

Hicks AJ (1971). Systematic studies of *Xanthium* (Compositae: Ambrosieae); The cockleburs of Tazewell County, Illinois. Ph.D Thesis, University of Illinois, Champaign-Urbana.

James TK, Rahman A, Trolove MR, Dowsett CA (2016). Seed germination characteristics and control options for noogoora bur (*Xanthium strumarium*) in commercial maize production. *New Zealand Plant Protection,* 69: 270-277. https://doi.org/10.30843/nzpp.2016.69.5947

Kadıoğlu İ (1997). Akdeniz Bölgesi pamuk ekim alanlarında görülen bazı yabancı ot tohumlarının çimlenme biyolojileri ve çıkış derinlikleri üzerine araştırmalar. *Türkiye II. Herboloji Kongresi*, 1-4 September 1997; İzmir, Türkiye, pp. 205-217.

Karaman Y, Tursun N (2021). Germination biology of field bindweed seeds collected from different provinces. *Bulgarian Journal of Agricultural Science,* 27 (6): 1168–1177.

Karimmojeni H, Zare A, Keshtkar E, Mashhadi HR, Alizadeh H (2010). Dormancy breaking of cocklebur (*Xanthium strumarium* L.) seeds. *Iranian Journal of Field Crop Science,* 41(3): 503-511.

Kaya İ, Nemli Y (2004). Determination of minimum and maximum germination temperature of certain weed species in cotton fields in Nazilli and Menemen. *Türkiye Herboloji Dergisi,* 7(1): 13-19.

Machado M, Queiroz CRR, Wilson TM, Sousa DER, Castro MB, Saravia A, Lee ST, Armién AG, Barros SS, Riet-Correa F (2021). Endemic *Xanthium strumarium* poisoning in cattle in flooded areas of the Araguari river, Minas Gerais, Brazil. *Toxicon,* 200: 23-29. https://doi.org/10.1016/j.toxicon.2021.06.019

Norsworthy K, Oliveira MJ (2007). Light and temperature requirements for common cocklebur (*Xanthium strumarium*) germination during after-ripening under field conditions. *Weed Science,* (3): 227-234. https://doi.org/10.1614/WS-06-184

Özaslan C (2021). Current distribution map of common cocklebur (*Xanthium strumarium* L.) in Türkiye. *Fresenius Environmental Bulletin,* 30: 3649-3656.

Özkil M (2021). Determination of distribution, density, biology and control strategies of species of ivyleaf morningglory (*Ipomoea* spp.) and field bindweed (*Convolvulus* spp.) in agricultural areas of the Mediterranean region. Ph.D Thesis, Hatay Mustafa Kemal University, Turkey.

Pala F, Mennan H, Demir A (2018). Determination of the weed species, frequency and density in lentil fields in Diyarbakır province. *Turkish Journal of Weed Science,* 21(1): 33-42.

Saeed A, Hussain A, Khan MI, Arif M, Maqbool MM, Mehmood H, Iqbal M, Alkahtani J, Elshikh MS (2020). The influence of environmental factors on seed germination of *Xanthium strumarium* L.: Implications for management. *PLoS ONE*, 15(10): 1-16. https://doi.org/10.1371/journal.pone.0241601

Saric-Krsmanovic M, Bozic D, Pavlovic D, Elezović I, Vrbnicanin S (2012). Temperature effects on common cocklebur (*Xanthium strumarium* L.) seed germination. *Romanian Agricultural Research,* 29: 389-393.

Tadesse NS, Assefa AS, Motbaynor MM, Betsiha EM, Hailu AA, Beyene GF, Hordofa TB (2017). Invasion and impacts of *Xanthium strumarium* in Borena zone of Oromia region, Ethiopia. *Journal of Coastal Life Medicine,* 5(8): 350-355. https://doi.org/10.12980/jclm.5.2017J7-26

Tao Y, Sang T, Yan J, Hu Y, Zhao Y, Liu Y (2021). Effects of sand burial depth on *Xanthium spinosum* seed germination and seedling growth. *Research Square,* 1-17. https://doi.org/10.21203/rs.3.rs-871392/v1

Tepe I (2014). Yabancı otlarla mücadele. Sidas Yayıncılık, Turkiye, p.290.

TBL (2024). Türkiye bitkileri listesi. https://bizimbitkiler.org.tr/yeni/demos/technical/ (access date: 04/02/2024).

Toledo REB, Kuva MA, Alves PLCA (1993). Factors affecting germination and emergence of *Xanthium strumarium* L.: Dormancy, light quality and sowing depth. *Planta Daninha,* 11(1-2): 15-20. https://doi.org/10.1590/S0100-83581993000100003

Üremiş İ, Uygur FN (1999). Minimum, optimum and maxsimum germination temperatures of some important weed species in the Çukurova region of Turkey. *The Journal of Weed Science,* 2(2): 1-12.

Venodha PM (2016). Landscape degradation and restoration—a planning approach. *International Journal of Environmental Science and Development,* 7(3): 229-233. https://doi.org/10.7763/IJESD.2016.V7.774

Wang Q, Xu D, Yin B, Zheng Y, Guo X, Li Y, Sun X, Wang L, Wu N (2023). Differences in seed characteristics, germination and seedling growth of Suaeda salsa grown in intertidal zone and on saline inland. *Frontiers in Plant Science,* 14. https://doi.org/10.3389/fpls.2023.1175812

Weaver SE, Lechowicz MJ (1983). The biology of Canadian weeds. 56. *Xanthium strumarium* L. *Canadian Journal of Plant Science,* 63(1): 211-225. https://doi.org/10.4141/cjps83-021

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Research Article

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Quality and Mineral Elements Contents of *Trifolium* spp. Ecotypes Collected from Eastern Anatolia (Türkiye)

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HIGHLIGHTS

- In this study, plant identification and quality of pastures were measured from pastures in different provinces of the Southeast and comparisons were made.

Abstract

The flora of Turkey is rich plant species but there is less known about some species. Hence, there is need to determine species properties and their forage potentials for animal feeding. The objective of this study was determine to 14 different *Trifolium* species (*T. ambiguum, T. bullatum, T. campestre, T. cherleri, T. dasyurum, T. echinatum, T. grandiflorum, T. hirtum, T. nigrescens, T. pauciflorum, T. resupinatum, T. spumosum, T. stellatum* and *T. tomentosum*) that taken from natural habitats during the flowering period of the plants by cutting from the root collar of the plants from 12 different locations. The differences between *Trifolium* genotypes were found to be statistically very significant for all the examined traits. Crude protein (CP), Dry matter (DM), acid detergent insoluble fiber (ADF), neutral detergent insoluble fiber (NDF), acid detergent insoluble protein (ADP), digestible dry matter (DDM), dry matter intake (DMI), calcium (Ca), magnesium (Mg), phosphorus (P) and potassium (K) ratios and relative feed values (RFV), Ca/P and K/(Ca+Mg) values varied between 12.02-25.54%, 90.25-96.56%, 11.61-35.26%, 27.27-47.82%, 0.47-1.24%, 61.43-79.87%, 2.51-4.40%, 1.41-2.19%, 0.26-0.43%, 0.31-0.50%, 1.19-3.65%, 119.66-268.62, 3.24-6.09 and 0.56-1.97, respectively. Among the *Trifolium* species collected from different locations examined in the research, genotype number 25 (*Trifolium tomentosum*) stood out with high CP, DM, DDM, DMI and RFV values and low ADF and NDF ratios. Genotype number 17 (*Trifolium nigrescens* subsp. *petrisavii*) in terms of the same characteristics followed this genotype. While genotypes number 1 (*Trifolium ambiguum*), 16 (*Trifolium nigrescens* subsp. *petrisavii*) and 21 (*Trifolium resupinatum* var. *resupinatum*) stood out with high K values, genotypes number 25 (*Trifolium tomentosum*), 9 (*Trifolium cherleri*) and 21 (*Trifolium resupinatum* var. *resupinatum*) stood out with high Ca, Mg and P values, respectively. Among the *Trifolium* species examined, the best genotypes in terms of yield and quality were genotypes number 17 (*Trifolium nigrescens* subsp. *petrisavii*) and 25 (*Trifolium tomentosum*).

Keywords: *Trifolium* spp.; ecotype; herbage quality; mineral element content

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1. Introduction

One of the most significant obstacles to the development of grazing livestock is generally acknowledged to be undernutrition. Low levels of protein and energy are frequently the reason of cattle productivity that is not at its best. However, ruminants sometimes deteriorate in spite of an abundant feed supply (McDowell and Valle 2000). Reduced protein efficiency is typically the result of rapid and widespread ruminal breakdown of proteins in grass and legume forages. The rumen microbial community consumes amino acids for energy and releases ammonia through deamination when energy is limited but there is an oversupply of peptides and amino acids derived from plants. Therefore, forages with high water-soluble carbohydrates (WSC) may enhance the synchronization and balance of carbon (C) and nitrogen (N), boost rumen microbial protein production, and raise animal productivity. Cattle diets' protein degradability may be impacted by the species of forage chosen (Da Silva et al. 2014).

According to recent studies, dairy calves are frequently overfed protein; yet, milk and component yields can be maintained, and occasionally even enhanced, with lower protein consumption (Broderick 2018). Fiber often makes up over 25% of the total mixed rations and is a key component of diets offered to dairy ruminants. Fiber is essential for ruminant energy and rumen health maintenance. CP concentration has a negative connection with NDF, ADF, and lignin, which is consistent with multiple research (Du et al. 2016).

Low productivity and reproductive issues in grazing ruminants have long been attributed to mineral excesses or deficiencies in soils and forages. Worldwide, mineral deficiencies are frequently indicated by hair loss, skin conditions, non-infectious abortion, diarrhea, anemia, appetite loss, abnormalities of the bones, tetany, and low fertility. Calcium (Ca), chlorine (Cl), phosphorus (P), magnesium (Mg), potassium (K), sodium (Na), and sulfur (S) are the seven main minerals that forages provide. With the exception of Cl, each of these minerals has been shown to be inadequate for grazing animals under particular circumstances (McDowell and Valle 2000).

Lack of P is the most common mineral-element deficit in grazing animals worldwide. In tropical grazing zones, P insufficiency is more common than in temperate ones. Reproductive failure is the most detrimental economic effect of P shortage; P treatment significantly raises fertility levels in grazing cattle from throughout the globe. With the exception of cows that produce considerable amounts of milk or those that graze on acidic, sandy, or organic soils in humid regions where the herbage is primarily composed of fast-growing grasses devoid of legume species, Ca shortage is uncommon in grazing cattle. Ca deficiency is rarely observed in grazing beef cattle, especially during lactation, despite the fact that it can easily occur in young, growing animals and breastfeeding dairy cows fed local forages supplemented with concentrates (Fadlalla 2022).

The connection between magnesium and the dangerous metabolic disease grass tetany (hypomagnesaemia) makes it practically significant. Forage species and mineral composition, soil characteristics, fertilizer techniques, season, temperature, animal species, breed, and age all have an impact on grass tetany, a complex metabolic disease that affects ruminants. More severe cases may cause the affected cows to lose their appetite, walk stiffly, and reject the other cows in the herd. When older cattle graze grass or small-grain forages in chilly weather, such as in the early spring or an exceptionally rainy fall, grass tetany typically occurs. Non-specific symptoms of ruminant K insufficiency include sluggish development, decreased intake of feed and water, decreased feed efficiency, muscular weakness, and neurological problems. Very few cases of K insufficiency in ruminants that only graze forages have been confirmed. Other forage nutrient shortages are likely the primary cause of the lack of widespread K insufficiency, even when forages contain less than the necessary amount (McDowell and Valle 2000). Requirements of ruminants for Ca is 1.8-8.2 g kg^{-1} DM; for Mg is 1-2 g kg^{-1} DM; for P is 1.8-4.8 g kg^{-1} DM; for K is 6-8 g kg^{-1} DM (McDowell 1997).

Numerous factors, such as soil, plant species, maturation stages, yield, pasture management, and climate, interact to determine the concentration of mineral elements in plants from different parts of the world. For instance, the following factors influence the mineral content of forage: (i) plant maturity; (ii) species and species-specific variation; (iii) management practices like crop removal or grazing systems; (iv) fertilization, especially with K and N; and (v) soil and environmental conditions. Forage mineral concentrations are also influenced by interactions with other elements. The majority of naturally occurring mineral deficiencies in livestock are regionally specific and strongly correlate with the mineral content and properties of the soil. Young and alkaline geological formations are more abundant in most elements than are the older, more acid, coarse, sandy formations. However, availability factors, including soil pH, texture, moisture content and organic matter, are probably more often the limiting factors rather than soil mineral content. Animals' mineral intake is more influenced by the kind of plant and its amount of consumption than by the parent rock that produced the soil and supported the plants. The mineral content of several plant species growing in the same soil varies greatly. Legumes are often thought to be richer than grasses in a variety of mineral elements. The mineral value of most forages decreased as they became older, thus grazing cattle may not get enough K and P elements. P, K, Mg, Na, and Cl typically decrease as the plant ages (McDowell and Valle 2000).

In this study, 14 different *Trifolium* species (*T. ambiguum, T. bullatum, T. campestre, T. cherleri, T. dasyurum, T. echinatum, T. grandiflorum, T. hirtum, T. nigrescens, T. pauciflorum, T. resupinatum, T. spumosum, T. stellatum* and *T. tomentosum*) were taken from natural habitats during the flowering period of the plants by cutting from the root collar of the plants from 12 different locations in Batman, Diyarbakır, Şanlıurfa, Mardin, Siirt provinces of Türkiye, and determined to some properties such as crude protein (CP), acid detergent insoluble fiber (ADF), neutral detergent insoluble fiber (NDF), Ca, K, Mg and P ratio were measured and ADF, NDF, digestible dry matter (DDM), dry matter intake (DMI) ratio and relative forage values (RFV).

2. Materials and Methods

The materials of the research consist of 25 plant samples of 14 species of the genus *Trifolium* collected from different locations in the Southeastern Anatolia Region at 2023 (Table 1) (Figure 1).

Table 1. Locations, collection dates, and geographic coordinates of the plant samples

Location	Altitude (m)	Date
Batman-I	572	07.05.2023
Diyarbakır-I	887	10.05.2023
Diyarbakır-II	763	10.05.2023
Diyarbakır-III	920	10.05.2023
Diyarbakır-IV	1113	10.05.2023
Diyarbakır-V	885	10.05.2023
Diyarbakır-VI	667	13.05.2023
Diyarbakır-VII	652	15.05.2023
Şanlıurfa-I	1469	21.05.2023
Şanlıurfa-II	1329	21.05.2023
Mardin-I	1036	07.05.2023
Siirt-I	846	07.05.2023

2.1. Climatic Data of Locations

Mean temperature and total precipitation for all locations were in Tables 2. In Batman, the lowest temperature was recorded in January with 2.9 °C; the highest temperature was recorded in August with 31.6°C. The highest humidity was observed in December (85.6%), and the lowest humidity was observed in August (25.3%). Total precipitation was heavy in winter and spring (106.7 mm in November) and zero in summer. In Diyarbakır, the lowest temperature was recorded in February with 4.0°C; the highest temperature was recorded in August with 32.9°C. The highest humidity was recorded in December (84.4%), and the lowest humidity was recorded in July and August (23.3 and 23.6%, respectively). The highest rainfall was recorded in March with 131 mm, and there was almost no rainfall in the summer months (June and August). In Siirt, the lowest temperature was recorded in February with 3.6°C and the highest in August with 33.1°C. The highest humidity was recorded in December with 74.9% and the lowest in August with 18.7%. The highest rainfall was in November (115.6 mm) and almost none in the summer months. In Sanliurfa, the lowest temperature was recorded in February with 6.7 °C, and the highest temperature was recorded in August with 34.1 °C. The highest humidity was observed in December (71.3%), and the lowest humidity was observed in July (22.7%). The highest amount of rainfall was recorded in March with 255 mm (Table 2).

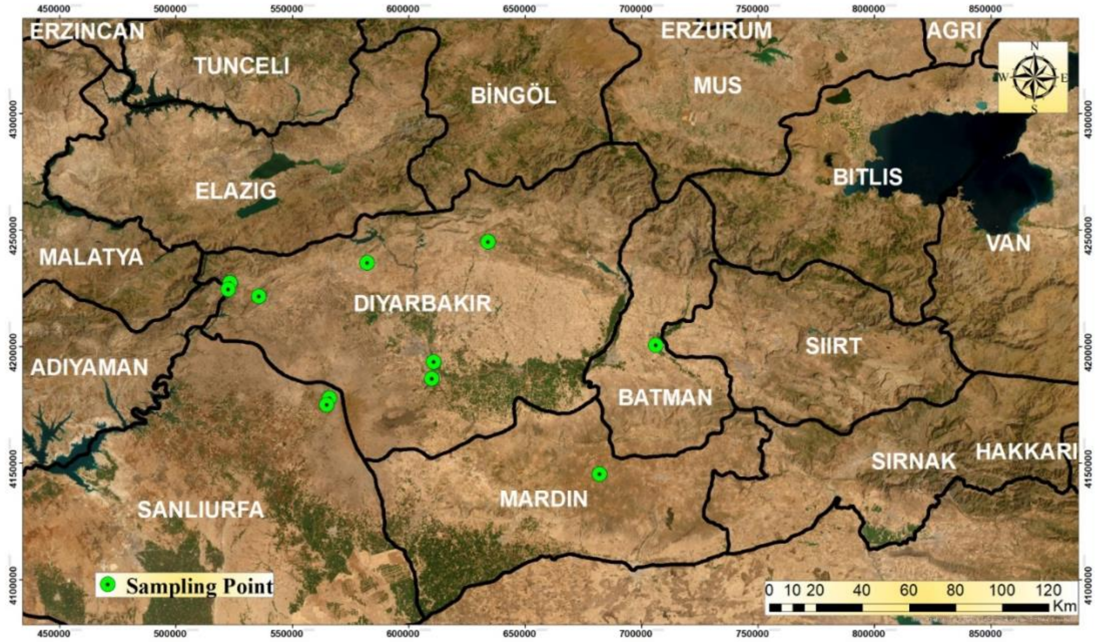

Figure 1. Location of work area

Table 2. Climatic data of Batman, Diyarbakır, Siirt, Şanlıurfa province

Months	Average Temperature (°C)				Total Precipitation (mm)			
	Batman	Diyarbakır	Siirt	Şanlıurfa	Batman	Diyarbakır	Siirt	Şanlıurfa
January	2.9	5.7	5.1	7.8	18.4	87.5	18.6	21.6
February	3.5	4.0	3.6	6.7	55.4	57.6	70.6	78.3
March	12.5	12.0	12.2	13.7	57.2	131.0	61.4	255.0
April	14.7	14.7	14.5	16.4	36	79.4	88.2	41.8
May	19.9	20.1	20.0	22.6	24.7	16.0	37.2	5.7
June	26.9	27.6	27.2	28.7	0.0	0.0	0.0	0.0
July	30.9	32.3	32.3	33.3	0.0	1.9	0.8	0.0
August	31.6	32.9	33.1	34.1	0.0	0.0	2.6	0.1
September	26.3	27.2	27.7	29.1	0.0	0.5	9.6	0.0
October	18.6	18.8	18.9	22.5	25.3	23.4	51.2	2.3
November	12.5	12.0	12.7	15.3	106.7	125.9	115.6	46.7
December	6.7	6.9	7.8	10.8	23.4	60.4	79.0	41.8

2.2. Measurements

Trifolium species were taken during the flowering period of the plants. Approximately 200 g plant samples from each species were cut and dried in a drying cabinet (Memmert ULM 800) at 70 °C and until a constant weight was achieved. The samples were ground in a laboratory-type mill (IKA, A11). It was sieved with a 1 mm diameter sample sieve (Retsch DIN-ISO 3310/2) and made ready for analysis.

The quality analyses of the species were analyzed at the Dicle University Science and Technology Application and Research Center laboratory with NIRS (Near Infrared Spectroscopy-Foss Model 6500) analyzer. In the analysis, crude protein (CP), acid detergent insoluble fiber (ADF), neutral detergent insoluble fiber (NDF), Ca, K, Mg and P values were measured. In addition, with the help of the determined ADF and NDF, digestible dry matter (DDM), dry matter intake (DMI) and relative forage values (RFV) were calculated and found. The following equations were used for calculations to validate the results of FT-NIR spectroscopy, randomly selected 20 samples were analyzed using the Dumas method for CP content, and using Ankom Fiber Analyzer for NDF and ADF contents. Dumas and Ankom Fiber Analyzer results had a significant correlation ($r \geq 0.9$, $P \leq 0.01$) with FT- NIR results. Therefore, FT-NIR results were used in the statistical analyses (Morrison 2003).

$$\text{DDM (\%)} = 88.9 - (0.779 \times \text{ADF}) \tag{1}$$

$$\text{DMI (\%)} = 120 / \text{NDF} \tag{2}$$

$$\text{RFV} = (\text{DDM} \times \text{DMI}) / 1.29 \tag{3}$$

2.3. Data analysis

Data analysis was realized using the Jump-Pro13 statistical package program using randomized completed parcel. Differences between means were compared using the Tukey HSD test. Principal component analysis (biplot) using the scatter plot model was performed using the Genstat 12th (Copyright 2011 VSN International Ltd) statistical package program (Genstat 2009).

3. Results and Discussion

The results showed that significant differences were found among the genotypes for all examined parameters ($P \leq 0.01$). The mean of the examined parameters were presented in Tables 6, 7 and 8.

The lowest CP ratio were measured in Siirt I (12.02%) and Diyarbakır V (12.27%) locations from genotypes number 9 and 10, respectively, and the highest CP ratio (between 23.11% and 25.54%) in Diyarbakır IV, Diyarbakır IV, Şanlıurfa I, Diyarbakır VII, Şanlıurfa I and Şanlıurfa II locations from genotypes number 17, 19, 20, 21, 22 and 25. DM ratio were lowest (between 90.89% and 91.17%) in Diyarbakır III, Batman I and Mardin II locations, from genotypes number 5, 7 and 14, and the highest DM rate (96.56%) in Şanlıurfa II location, from genotype number 25. The lowest ADF ratio (between 13.1% and 16.71%) were measured in Batman I, Diyarbakır I, Diyarbakır IV, Diyarbakır VII and Şanlıurfa I locations, from genotypes number 7, 16, 17, 21 and 22 and the highest ADF ratio (35.26% and 35.16%) were measured in Siirt I and Diyarbakır V locations, from genotypes number 9 and 10, respectively. *Trifolium cherleri, Trifolium angustifolium, Trifolium hybridum, Trifolium nigrescens, Trifolium lappaceum, Trifolium pilulare, Trifolium scabrum, Trifolium resupinatum, Trifolium spumosum* and *Trifolium tomentosum* were collected from ten different points in Hatay in a pasture by Ertekin (2021) to determine the chemical composition of collected clover species, and values of CA, CP, NDF, ADF, ADL, of species varied between 5.36-9.85%, 18.5-22.1%, 30.3-49.8%, 21.3-34.3%, 3.25-5.04%, respectively. It was determined that the values of DMD, DMI ratio, and RFV calculated for the nutritive values of these plants varied between 62.2-72.3%, 2.41-3.97%, and 116.2-222.2, respectively. On the other hand, *Trifolium alexandrinum*, *Trifolium spumosum*, *Trifolium resupinatum*, *Trifolium lappaceum*, *Trifolium campastre*, *Trifolium angustifolium* and *Trifolium purpureum* were collected from ten different points in Adana in a pasture by Kökten et al. (2011) to determine the chemical composition of collected clover species, and values of CP of species obtained 35.00%, 23.57%, 31.93%, 28.13%, 33.63%, 36.60%, 36.17%, respectively. While CP, ADF, NDF, DDM, DMI rates, and RFV in *Trifolium resupinatum* were obtained as 12.99%, 35.0%, 47.4%, 61.64%, 2.54%, 121.44, respectively, in Elazığ (Karadeniz and Kökten 2022), CP, ADF, NDF rates and RFV in *Trifolium repens* were obtained as 20.0%, 22.9%, 31.1%, 215, respectively, in Bingol (Cacan et al. 2024).

NDF ratio were lowest (between 27.27% and 30.95%) in Diyarbakır I, Diyarbakır IV and Şanlıurfa II locations, from genotypes number 16, 17 and 25, and the highest NDF ratio (43.98% and 47.82%) were measured in Diyarbakır VII and Diyarbakır V locations, from genotypes number 6 and 10. Pereira-Crespo et al. (2012) examined 316 samples of several clover species and concluded that the NDF concentrations ranged from 14.5 to 51.6%. The study's NDF contents fell within the ranges that Pereira-Crespo et al. (2012) reported.

The ADF results from our investigation are supported by Pereira-Crespo et al. (2012), who discovered that the ADF levels of several clover species ranged from 11.55 to 44.35%.

Table 6. Mean values of CP, DM, ADF and NDF

Location	Genotypes	CP	DM	ADF	NDF
Diyarbakır II	1. *Trifolium ambiguum*	17.49ıjk	91.75f	25.97cd	42.76bc
Sanlıurfa I	2-*Trifolium ambiguum*	22.76de	91.58fgh	19.24gh	39.17ef
Sanlıurfa I	3. *Trifolium bullatum*	20.21f	91.60fg	21.88efg	38.97efg
Sanlıurfa I	4. *Trifolium campestre*	18.36ghı	92.17de	23.15def	41.76cd
Diyarbakır III	5. *Trifolium campestre* subsp. *campestre* var. *campestre*	15.17l	91.17j	20.93fg	40.03de
Diyarbakır VII	6. *Trifolium campestre* subsp. *campestre* var. *campestre*	17.86hıj	92.37c	24.44de	43.98b
Batman I	7. *Trifolium campestre* subsp. *campestre* var. *campestre*	19.08fgh	90.89k	16.21ıj	37.08ghı
Sanlıurfa I	8. *Trifolium campestre* subsp. *campestre* var. *campestre*	22.29e	92.30cd	17.52hı	38.24e-h
Siirt I	9. *Trifolium cherleri*	12.02m	91.28ıj	35.26a	46.65a
Diyarbakır V	10. *Trifolium dasyurum*	12.27m	92.73b	35.16a	47.82a
Diyarbakır VII	11. *Trifolium echinatum*	22.72e	92.05e	22.14efg	38.77efg
Mardin I	12. *Trifolium grandiflorum*	18.67ghı	90.25l	21.65efg	37.23fgh
Diyarbakır III	13. *Trifolium hirtum*	16.45kl	92.05e	27.55bc	39.47e
Mardin II	14. *Trifolium hirtum*	18.53ghı	91.15j	23.53def	35.04ıj
Sanlıurfa I	15. *Trifolium nigrescens* subsp. *petrisavii*	19.34fg	91.41hı	19.54gh	35.15ıj
Diyarbakır I	16. *Trifolium nigrescens* subsp. *petrisavii*	22.50e	91.62fg	15.85ıjk	30.95l
Diyarbakır IV	17. *Trifolium nigrescens* subsp. *petrisavii*	24.11bc	91.64fg	13.10kl	27.27m
Diyarbakır VII	18. *Trifolium nigrescens* subsp. *petrisavii*	22.90cde	92.10e	21.41fg	37.00ghı
Diyarbakır IV	19. *Trifolium pauciflorum*	23.59b-e	92.40c	20.97fg	36.65hı
Sanlıurfa I	20. *Trifolium pauciflorum*	23.11cde	92.39c	21.79efg	39.57e
Diyarbakır VII	21. *Trifolium resupinatum* var. *resupinatum*	25.54a	92.46c	16.71hıj	33.20jk
Sanlıurfa I	22. *Trifolium resupinatum* var. *resupinatum*	24.07bcd	92.03e	14.49jkl	31.48kl
Diyarbakır VII	23. *Trifolium spumosum*	17.37ıjk	92.38c	29.10b	42.72bc
Diyarbakır IV	24. *Trifolium stellatum* var. *stellatum*	16.75jk	91.49gh	21.50fg	43.84b
Sanlıurfa II	25. *Trifolium tomentosum*	24.50ab	96.56a	11.60l	27.96m
	Mean	19.90	91.99	21.62	38.11
	CV(%)	**4.02**	**0.12**	**8.28**	**3.25**

Levels not connected by the same letter are significantly different.

Table 7. Mean values of ADP, DDM, DMI, RFV and K

Location	Genotypes	ADP	DDM	DMI	RFV	K
Diyarbakır II	1. *Trifolium ambiguum*	0.69hı	68.67ıj	2.81ı	149.41j	3.63a
Sanlıurfa I	2-*Trifolium ambiguum*	0.94c	73.91ef	3.09fg	177.71dfg	2.45ı
Sanlıurfa I	3. *Trifolium bullatum*	0.79fg	71.86fgh	3.08fg	171.59fgh	2.22j
Sanlıurfa I	4. *Trifolium campestre*	0.84ef	70.87ghı	2.88hı	158.60hıj	1.88l
Diyarbakır III	5. *Trifolium campestre* subsp. *campestre* var. *campestre*	0.66ı	72.59fg	3.02gh	170.49fgh	1.53o
Diyarbakır VII	6. *Trifolium campestre* subsp. *campestre* var. *campestre*	0.84ef	69.86hı	2.74ıj	148.43j	1.61n
Batman I	7. *Trifolium campestre* subsp. *campestre* var. *campestre*	0.56k	76.28cd	3.24ef	191.48de	1.80m
Sanlıurfa I	8. *Trifolium campestre* subsp. *campestre* var. *campestre*	0.94c	75.25de	3.14efg	183.29d-g	1.42p
Siirt I	9. *Trifolium cherleri*	0.68hı	61.43l	2.58jk	122.85k	1.19r
Diyarbakır V	10. *Trifolium dasyurum*	0.81efg	61.51l	2.51k	119.66k	1.34q
Diyarbakır VII	11. *Trifolium echinatum*	1.05b	71.65fgh	3.10fg	171.96fgh	2.68g
Mardin I	12. *Trifolium grandiflorum*	0.65ıj	72.03fgh	3.23ef	180.33d-g	2.16k
Diyarbakır III	13. *Trifolium hirtum*	0.73h	67.44jk	3.04gh	159.04hıj	2.80f
Mardin II	14. *Trifolium hirtum*	0.60jk	70.57ghı	3.44d	188.67de	2.55h
Sanlıurfa I	15. *Trifolium nigrescens* subsp. *petrisavii*	0.60jk	73.68ef	3.42d	195.21d	2.25j
Diyarbakır I	16. *Trifolium nigrescens* subsp. *petrisavii*	0.59k	76.55bcd	3.88b	230.34b	3.65a
Diyarbakır IV	17. *Trifolium nigrescens* subsp. *petrisavii*	0.47l	78.70ab	4.40a	268.62a	3.25c
Diyarbakır VII	18. *Trifolium nigrescens* subsp. *petrisavii*	0.86de	72.21fg	3.28de	184.17def	2.99e
Diyarbakır IV	19. *Trifolium pauciflorum*	0.90cd	72.56fg	3.28de	184.39def	2.77f
Sanlıurfa I	20. *Trifolium pauciflorum*	1.06b	71.93fgh	3.03gh	169.16ghı	2.59h
Diyarbakır VII	21. *Trifolium resupinatum* var. *resupinatum*	0.83ef	75.88cde	3.62c	212.76c	3.60a
Sanlıurfa I	22. *Trifolium resupinatum* var. *resupinatum*	0.81efg	77.61abc	3.82b	229.71b	3.43b
Diyarbakır VII	23. *Trifolium spumosum*	0.93c	66.23k	2.81ı	144.55j	3.11d
Diyarbakır IV	24. *Trifolium stellatum* var. *stellatum*	0.78g	72.15fg	2.78ı	154.89ıj	2.48ı
Sanlıurfa II	25. *Trifolium tomentosum*	1.24a	79.87a	4.29a	265.74a	2.68g
	Mean	0.79	72.05	3.22	181.32	2.48
	CV (%)	**3.79**	**1.92**	**3.11**	**5.00**	**1.20**

Levels not connected by same letter are significantly different.

The lowest ADP ratio (0.47% and 0.56%) were measured in Batman I and Diyarbakır IV locations, from genotypes number 7 and 17, and the highest ADP rate (1.24%) was measured in Şanlıurfa II location, from genotype number 25. DDM ratio were lowest (61.43% and 61.51%) in Siirt I and Diyarbakır V locations, from genotypes number 9 and 10, and the highest DDM ratio (between 75.88% and 79.87%) were measured in Batman I, Diyarbakır I, Diyarbakır IV, Diyarbakır VII and Şanlıurfa II locations, from genotypes number 7, 16, 17, 21 and 25. The lowest DMI ratio (between 2.51% and 2.81%) were measured in Diyarbakır II, Diyarbakır VII, Siirt I, Diyarbakır V and Diyarbakır VII locations, from genotypes number 1, 6, 9, 10, 23 and 24, and the highest DMI ratio (4.4% and 4.29%) were measured in Diyarbakır IV and Şanlıurfa II locations, from genotypes number 17 and 25, respectively. This study's DMD, DMI, and RFV values were comparable to those of Gürsoy and Macit's (2017) investigation of similar results.

RFV values were lowest (between 119.66 and 144.55) in Siirt I, Diyarbakır V and Diyarbakır VII locations, from genotypes number 9, 10 and 23, and the highest RFV values (268.62 and 265.74) were measured in Diyarbakır IV and Şanlıurfa II locations, from genotypes number 17 and 25. The lowest K rate (1.19%) was measured in Siirt I location, from genotype number 9, and the highest K ratio (between 3.6% and 3.65%) were measured in Diyarbakır II, Diyarbakır I and Diyarbakır VII locations, from genotypes number 1, 16 and 21. Around the world, forage crops constitute a vital source of animal feed and are integral to livestock husbandry and animal production systems. According to Lüscher et al. (2014), perennial forage legumes used as base feed make ideal fodder for a variety of livestock classes, such as dry dairy cows, dairy heifers, dairy beef, or beef cows. According to cultivar availability and seed production and marketing volumes, clover (*Trifolium* spp.) is the second most significant farmed perennial fodder legume in Europe, behind alfalfa (Hoekstra et al. 2018). This perennial plant has many benefits, including a high protein content and soil-improving qualities that improve livestock feed intake and lessen the need for synthetic nitrogen fertilizers (Tucak et al. 2023).

Table 8. Mean values of Ca, Mg, P, Ca/P and K/(Ca+Mg)

Location	Genotypes	Ca	Mg	P	Ca/P	K/(Ca+Mg)
Diyarbakır II	1. *Trifolium ambiguum*	1.58fgh	0.26k	0.44e	3.62ıj	1.97a
Sanlıurfa I	2-*Trifolium ambiguum*	1.78cd	0.36c	0.42f	4.20g	1.15j
Sanlıurfa I	3. *Trifolium bullatum*	1.70de	0.32e	0.40h	4.27fg	1.10jkl
Sanlıurfa I	4. *Trifolium campestre*	1.41k	0.28j	0.39ı	3.64ı	1.12jk
Diyarbakır III	5. *Trifolium campestre* subsp. *campestre* var. *campestre*	1.71de	0.24l	0.33m	5.19c	0.78no
Diyarbakır VII	6. *Trifolium campestre* subsp. *campestre* var. *campestre*	1.68e	0.29hı	0.34l	4.95d	0.82n
Batman I	7. *Trifolium campestre* subsp. *campestre* var. *campestre*	1.96b	0.30h	0.32n	6.09a	0.80n
Sanlıurfa I	8. *Trifolium campestre* subsp. *campestre* var. *campestre*	1.67ef	0.30h	0.37j	4.46ef	0.72o
Siirt I	9. *Trifolium cherleri*	1.70de	0.43a	0.31o	5.56b	0.56p
Diyarbakır V	10. *Trifolium dasyurum*	1.48jk	0.29h	0.32n	4.61e	0.75no
Diyarbakır VII	11. *Trifolium echinatum*	1.84c	0.31fg	0.41g	4.50e	1.25ı
Mardin I	12. *Trifolium grandiflorum*	1.84c	0.32ef	0.36k	5.04cd	1.00m
Diyarbakır III	13. *Trifolium hirtum*	1.70de	0.28j	0.38ıj	4.49e	1.41gh
Mardin II	14. *Trifolium hirtum*	1.65efg	0.32de	0.40h	4.16g	1.30ı
Sanlıurfa I	15. *Trifolium nigrescens* subsp. *petrisavii*	1.83c	0.27j	0.40h	4.59e	1.07kl
Diyarbakır I	16. *Trifolium nigrescens* subsp. *petrisavii*	1.64efg	0.28ıj	0.49b	3.36klm	1.89b
Diyarbakır IV	17. *Trifolium nigrescens* subsp. *petrisavii*	1.67e	0.28j	0.48b	3.46ı-l	1.67e
Diyarbakır VII	18. *Trifolium nigrescens* subsp. *petrisavii*	1.69e	0.33d	0.47c	3.57ıjk	1.48f
Diyarbakır IV	19. *Trifolium pauciflorum*	1.55hıj	0.35c	0.47c	3.33lm	1.46fg
Sanlıurfa I	20. *Trifolium pauciflorum*	1.55hıj	0.33d	0.45d	3.48ı-l	1.38h
Diyarbakır VII	21. *Trifolium resupinatum* var. *resupinatum*	1.63e-h	0.33d	0.50a	3.24m	1.83c
Sanlıurfa I	22. *Trifolium resupinatum* var. *resupinatum*	1.64efg	0.30gh	0.48b	3.41j-m	1.76d
Diyarbakır VII	23. *Trifolium spumosum*	1.49ıjk	0.33d	0.41g	3.59ıj	1.72de
Diyarbakır IV	24. *Trifolium stellatum* var. *stellatum*	1.57ghı	0.16m	0.40h	3.93h	1.43fgh
Sanlıurfa II	25. *Trifolium tomentosum*	2.19a	0.38b	0.45d	4.90d	1.04lm
	Mean	1.69	0.31	0.40	4.23	1.25
	CV (%)	**2.97**	**2.00**	**1.00**	**3.08**	**2.40**

Levels not connected by same letter are significantly different.

Kura clover (*Trifolium ambiguum*) has better CP and in vitro digestibility than alfalfa, but lower amounts of NDF, acid detergent fiber (ADF), and acid detergent lignin (ADL) (Seguin et al. 2002). Interest in using Kura clover in permanent pastures has increased as a result of these qualities. It is not advised to store Kura clover as hay because to its high leaf content (>85%), as field losses may be substantial (Peterson et al. 1994). Seguin and Mustafa (2003) determined quality of *Trifolium ambiguum* for two cultivars which had similar NDF (average of 355 g/kg of DM), ADF (281 g/kg of DM), EE (24 g/kg of DM), total carbohydrate (672 g/kg of DM), non-structural carbohydrate (354 g/kg of DM), TDN (650 g/kg of DM), and NEl (6.17 MJ/kg DM) contents.

The lowest Ca ratio (between 1.41% and 1.64%) were measured from genotypes number 4, 10, 19, 20, 21, 22, 23 and 24, and the highest Ca rate (2.19%) was measured from genotype number 25. For Mg ratio, the lowest value (0.16%) was measured in Diyarbakır IV location, from genotype number 24, and the highest value (0.43%) was measured in Siirt I location, from genotype number 9. The lowest P rate (0.31%) was measured in Siirt I location, from genotype number 9, and the highest P rate (0.5%) was measured in Diyarbakır VII location, from genotype number 21. Lee (2018) have reported that plant mineral matter contents vary significantly among forage species. K, Ca, Mg and P rates in *Trifolium repens* were obtained as 2.05%, 1.93%, 0.41%, 0.32%, respectively, in Bingol (Cacan et al. 2024).

Ca/P values were lowest (between 3.24% and 3.64%) at genotypes number 1, 4, 16, 17, 18, 19, 20, 21, 22 and 23, and the highest Ca/P value (6.09%) was measured from genotype number 7. The lowest K/(Ca+Mg) value (0.56%) was measured in Siirt I location, from genotype number 9, and the highest K/(Ca+Mg) value (1.97%) was measured in Diyarbakır II location, from genotypes number 1.

3.1. Biplot Analysis

The equality lines divided the biplot into sectors and the winning genotype for each sector was the one placed on the vertex.

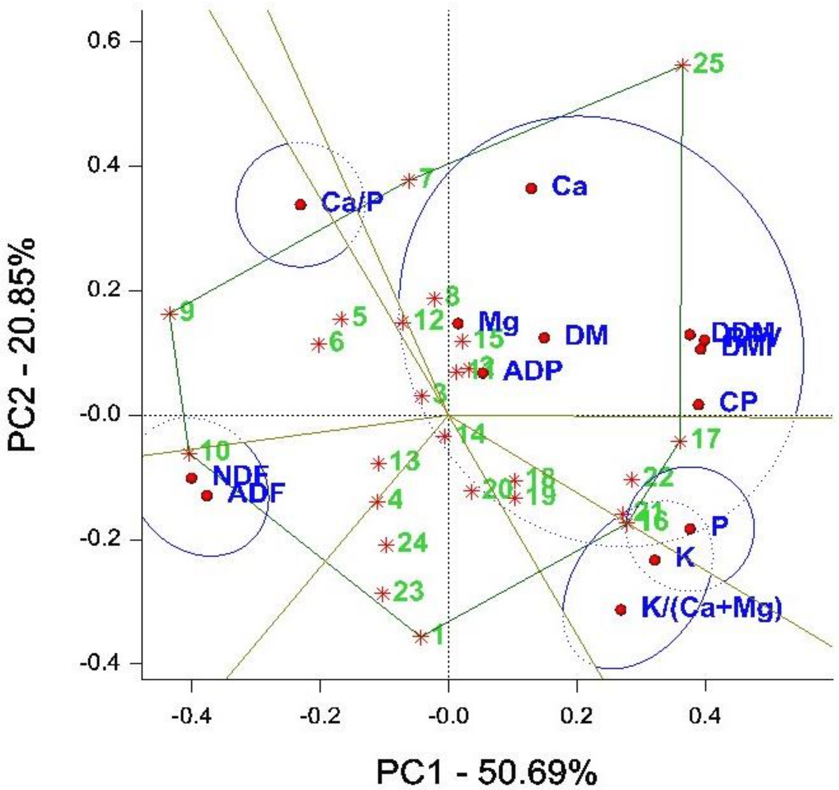

Figure 1. Polygons show suitable genotypes in each environment and show which genotype won where. Codes; **1.** *Trifolium ambiguum*, **2.** *Trifolium ambiguum*, **3.** *Trifolium bullatum*, **4.** *Trifolium campestre*, **5.** *Trifolium campestre* subsp. *campestre* var. *campestre*, **6.** *Trifolium campestre* subsp. *campestre* var. *campestre*, **7.** *Trifolium campestre* subsp. *campestre* var. *campestre*, **8.** *Trifolium campestre* subsp. *campestre* var. *campestre*, **9.** *Trifolium cherleri*, **10.** *Trifolium dasyurum*, **11.** *Trifolium echinatum*, **12.** *Trifolium grandiflorum*, **13.** *Trifolium hirtum*, **14.** *Trifolium hirtum*, **15.** *Trifolium nigrescens* subsp. *petrisavii*, **16.** *Trifolium nigrescens* subsp. *petrisavii*, **17.** *Trifolium nigrescens* subsp. *petrisavii*, **18.** *Trifolium nigrescens* subsp. *petrisavii*, **19.** *Trifolium paucifloru*, **20.** *Trifolium pauciflorum*, **21.** *Trifolium resupinatum* var. *resupinatum*, **22.** *Trifolium resupinatum* var. *resupinatum*, **23.** *Trifolium spumosu*, **24.** *Trifolium stellatum* var. *stellatum*, **25.** *Trifolium tomentosum*. Abbreviations: ADF; Acid detergent fiber, NDF; Nötral detergent fiber; ADP; Neutral detergent insoluble protein, CPY; Crude protein yield, DDM; Digestible dry matter, RFV; Relative feed value, DMI; Dry matter intake, K; Potassium, Mg; Magnesium, P; Phosphorus, Ca; Calcium, K; Potassium.

The studied environments were divided into seven sectors. The graph describes the 50.69 and 20.85% variation for PC1 and PC2, respectively. The total variation was 71.54%. The genotypes placed on the vertices of the polygon had the best or the poorest in examined parameters. Accordingly, genotypes number 16 and 21 had the best performance for P, K and K/Ca+Mg values. Genotype number 25 was the best for Ca, Mg, ADP, DM, DDM, DMI and RFV values. Genotype numbered 9 was the best for Ca/P ratio. Genotype number 10 was the best for ADF and NDF values. The parameters located on the left of the coordinate plane were negatively correlated with those located on the right, and the genotypes located in these areas are far from each other in terms of the traits in question.

4. Conclusions

Information about 14 different *Trifolium* species collected in the 12 different locations in Batman, Diyarbakır, Şanlıurfa, Mardin, Siirt provinces of Türkiye was determined. This study depicted that (i) genotypes number 17 and 25 were the most favorable genotypes according to CP, DM, DDM, DMI, ADF and NDF ratio and RFV values, (ii) genotypes number 1, 16 and 21 were the best genotypes for the K rate, (iii) genotypes number 25, 9 and 21 were the most stable genotypes according to Ca, Mg and P ratio.

When the obtained results are evaluated as a whole, it is suggested that more studies should be conducted on the genotypes number 17 and 25 from the examined *Trifolium* species and that studies should be conducted for their use in the improvement of meadows and pastures.

Author Contributions: The authors declare that they have contributed equally to the article. All authors declare that they have seen/read and approved the final version of the article ready for publication.

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References

Broderick GA (2018). Optimizing ruminant conversion of feed protein to human food protein. *Animal*, 12(8): 1722-1734.

Cacan E, Ozdemir S, Kokten K, Ucar R, Mokhtarzadeh S, Ekmekci M, Kutlu MA (2024). Change of forage yield and quality characteristics of white clover (*Trifolium repens* L.) at different harvest time. *Journal of Tekirdag Agricultural Faculty*, 21(5): 1099-1111.

Da Silva MS, Tremblay GF, Bélanger G, Lajeunesse J, Papadopoulos YA, Fillmore SA, Jobim CC (2014). Forage energy to protein ratio of several legume–grass complex mixtures. *Animal Feed Science and Technology*, 188: 17-27.

Du S, Xu M, Yao J (2016). Relationship between fibre degradation kinetics and chemical composition of forages and by-products in ruminants. *Journal of Applied Animal Research*, 44(1): 189-193.

Ertekin İ (2021). Comparison of chemical composition and nutritive values of some clover species. *International Journal of Chemistry and Technology*, 5(2): 162-166.

Fadlalla IM (2022). The interactions of some minerals elements in health and reproductive performance of dairy cows. IntechOpen, https://doi.org/10.5772/intechopen.101626

Genstat (2009). Genstat for Windows (12th edition) Introduction. Vsn International, Hemel Hempstead.

Gürsoy E, Macit M (2017). Determination of relative feed values of different legume forages grown as naturally in pastures of Erzurum Province. *Anadolu Journal of Agricultural Sciences*, 32(3): 407-412.

Hoekstra NJ, De Deyn GB, Xu Y, Prinsen R, Van Eekeren, N (2018). Red clover varieties of Mattenklee type have higher production, protein yield and persistence than Ackerklee types in grass-clover mixtures. *Grass and Forage Science*, 73(2): 297-308.

Karadeniz M, Kökten K (2022). The effects on yield and quality of berseem clover and Italian ryegrass mixture ratios in Elazig conditions. *Journal of the Institute of Science and Technology*, 12(1): 509-517.

Kökten K, Koçak A, Kaplan M, Akçura M, Bakoğlu A, Bağcı, E (2011). Tannin, protein contents and fatty acid composition of the seeds of some *Trifolium* L. species from Turkey. *Asian Journal of Animal and Veterinary Advances*, 6(1): 88-95.

Lee MA (2018). A global comparison of the nutritive values of forage plants grown in contrasting environments. *Journal of Plant Research*, 131(4): 641-654.

Lüscher A, Mueller-Harvey I, Soussana JF, Rees RM, Peyraud JL. (2014). Potential of legume-based grassland-livestock systems in Europe: a review. *Grass and Forage Science*, 69(2): 206-228.

McDowell LR, Valle G (2000). Major minerals in forages. In forage evaluation in ruminant nutrition. CABI Publishing. ISBN 0-85199-344-3.

McDowell LR (1997) Minerals for grazing ruminants in tropical regions, 3rd edn. University of Florida, Gainesville, Florida, USA, 81 pp

Morrison J.A. (2003). Hay and pasture management, Chapter 8. Extension Educator, Crop Systems Rockford Extension Center. http://iah.aces.uiuc.edu/pdf/Agronomy_HB/ 08chapter.pdf. (Access date: 30.06.2017).

Pereira-Crespo S, Valladares J, Flores G, Fernández-Lorenzo B, Resch C, Piñeiro J, Rodríguez-Díz, X (2012). Prediction of the nutritive value on annual forage clovers and serradella by near infrared spectroscopy (NIRS). In Options Méditerranéennes-Series A: Mediterranean Seminars, 102: 241-244.

Peterson PR, Sheaffer CC, Jordan RM, Christians CJ (1994). Response of Kura clover to sheep grazing and clipping. II. Below-ground morphology, persistence, and total nonstructural carbohydrates. *Agronony of Journal*, 86: 660-667.

Seguin AF, Mustafa P (2003). Chemical composition and ruminal nutrient degradabilities of fresh and ensiled Kura clover (*Trifolium ambiguum* MB). *Canadian Journal of Animal Science*, 83(3): 577-582.

Seguin P, Mustafa AF, Sheaffer CC. (2002). Effects of soil moisture deficit on forage quality, digestibility, and protein fractionation of Kura clover. *Journal of Agronomy and Crop Science*, 188: 260-266.

Tucak M, Čupić T, Horvat D, Ravlić M, Krizmanić G, Maćešić D, Meglič V (2023). Changes in agronomic and forage nutritive values of red clover in response to different development stage. *Romanian Agricultural Research*, 40: 215-224.

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Research Article

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The Exploration of Scientific Studies on Vegetable Seed Coating from The Past to The Future

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HIGHLIGHTS

- This study consists of data from a total of 631 scientific publications in scientific studies on vegetable seed coating between 1980 and 2024.
- More than 50% of the scientific studies in the field of vegetable seed coating have been carried out in India, USA and China respectively.
- The most commonly used keywords in scientific studies in the field of vegetable seed coating were; vegetable-oils, seed oil, performance, identification, vegetables, seed coat, coating, antioxidant activity and expression.

Abstract

Seed coating is a very hands-on technology that involves applications of external materials to seeds; this is done with great goals in mind, hence improved handling, protection to an extent, and, to certain small extents, germination and growth establishment. Despite its wide acceptability, the implementation of seed coating procedures has still got many challenges nowadays. This study, therefore, applies bibliometric analyses to deduce the trends and gaps between the seed coating research produced from 1980 up to 2024, particularly under the Web of Science database outlet. This work will integrate quantitative and qualitative methodologies regarding the analysis of publications, citations, affiliations, patents, grants, and data on other relevant indicators. The results showed that in the final years, there was an incredible rise in citations about this area, which, put differently, means continuously improving publishing and citation rates throughout the period. Publication data showed that India, the United States, and China represented important contributors to seed coating-related publications, as witnessed through both high publication outputs by its authors and numerous affiliation establishments. The present study comprehensively outlines the trends of research so that the researcher can identify the knowledge gap and assess developments to shape the future of research in seed coating. The results also provide insights for strategic guidance of future research and product development in the area.

Keywords: Bibliometric analysis; germination; seed; seed coating; vegetable

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1. Introduction

Seeds are the fundamental components of agricultural production, with germination and emergence being pivotal and delicate phases in a crop's life cycle (Villalobos et al. 2016). Low-quality seeds and inadequate planting circumstances can directly or indirectly impede plants' growth, health, and yield, beginning at the seedling development stage. Besides, environmental stress factors that include biotic stresses related to pests and diseases and abiotic stresses such as drought, salinity, extreme temperatures have been discouraging the process of germination and seedling development. In fact, such environmental stresses may be the cause for drastic yield reductions in crops (Lamichhane et al. 2018).

High-quality seeds are crucial for the establishment of stands in both transplant production and direct seeding. Seed performance is optimized via modifications to provide maximum emergence in poor settings (Taylor 2020). In order to overcome these limitations, seed coating methods were developed to seed performance, as well as protect them against biotic and abiotic stressors. Seed coating is defined as the application of biological, chemical, or physical agents to the surface of seeds with the principal intention of increasing the rate of germination of the seeds, while accelerating the rate of emergence of the seedling and supporting the early growth of the plant. Coating technologies applied to the seeds can be used to improve accurate seed placement at sowing and as a delivery system for compounds and agents which protect and enhance both seed and plant performance (Afzal et al. 2020).

The technology of coating seeds has immensely enhanced over the years with different techniques being developed to meet specific agricultural needs. Major processes of seed coating techniques include pelleting, encrusting, film coating, and seed dressing (Taylor 2003). Pelleting is a process where materials like clays or polymers are used to create a smooth, rounded coating on the seed. This technique enhances handling and sowing of seeds while concurrently achieving accuracy in mechanical planting (Pedrini et al. 2020). Encrusting is a sort of pelleting, the original shape of the seed can be preserved with an added outer protective layer. This just slightly increases the size and weight of the seeds, whereby the seeds become suitable for higher sowing systems without compromising germination (Pedrini et al. 2018; Taylor 2020). Film coating is the new approach in which thin, smooth, and coloured film is applied to seeds. Film coating is little modification of the size and shape of the seed surface. The application of this technique helps the incorporation of pesticides, fungicides, and nutrients without harming the seed so that it may be well protected at its initial stage of development (Taylor and Harman 1990; Taylor et. al. 2001). Further, seed dressing is the simpler and effective way of dressing the seeds with the protecting agent from pests and diseases in use for many decades because of the availability of effective fungicides, its relatively low cost, and easiness of the treatment (Gullino et al. 2014).

Among seed coating technologies, multi-seed coating is one of the great recent innovations. It is a technology that involves coating several seeds in one pellet. Specific developments for small-seeded plants have been done with the intention of assuring regular dispersion during sowing and attaining better uniformity in seedling emergence. The multi-seed coating thus confers advantages under low germination rate conditions and in intercropping systems. In consideration of the foregoing, the concurrent improvement of seed performance and handling along with the early development of a plant makes those techniques vital for further advancement in modern agricultural practices. At the same time, despite such developments, there are a few bottlenecks regarding proper documentation and dissemination of knowledge on different seed coating methods.

Even though different researchers try to define and explain about seed coating, there were problems regarding to ways of publication time, which reference and organization used, number of citation and affiliation collected from different data base was not appropriately documented. Inclusive all information and available data to know performance rate knowledge about seed coating process. The information gathered from data base very important to show right direction and pattern about seed coating to researcher and reviewers. Gathering and collecting research out puts in seed coating and related fields are crucial to get more and detail information about number publication, citation and, affiliation.

Numerous systematic reviews in seed coating examine scientific literature to investigate research directions and trends. Bibliometric analysis is a method that quantitatively assesses scientific research using their bibliographies (Kaplan and Altay 2023). This method objectively evaluates scientific production, the efficacy of scientific investigations, collaboration networks, commonality in research domains, and the performance of authors and scientific institutions, among other factors (Çelik Ş 2020; Altay and Kaplan 2023). This quantitative methodology offers a substantial benefit in the objective assessment of scientific research, monitoring trends within the discipline, and facilitating strategic decision-making.

The negative features of bibliometric analysis, on the other hand, are insufficient scientific publications related to the area under investigation, the lack of any kind of data entry control within databases, and biased citation practices of scientific studies. Despite all such disadvantages, bibliometric analysis allows assessing historical progress of scientific studies, developing policies about their field, reinforcing interdisciplinary networks, and optimizing the use of resources for conducting scientific research (Onder and Tirink 2022; Kaplan and Altay 2023).

The main aim of the study is to indicate, by applying the method of bibliometric analysis-a quantitative method being increasingly applied in the field of science studies-historical development trends faced within scientific publications regarding the topic of seed coating. For that, this research critically presents studies on seed coating within a time frame from 1980 to 2024. The result of the presented study is bound to enable researchers to trace the past and ongoing research outputs and, thereafter, make full and informed decisions concerning seed coating based on the facts so identified. In this perspective, the ultimate goal is to have seed coating technologies advanced, including their potentials for sustainable agriculture.

2. Materials and Methods

2.1. Material

The material of this study consists of bibliometric data of 631 scientific studies obtained in a search with the help of the keywords "Seed Coating" and “Vegetable” conducted in the Web of Science database, which covers the periods between 1980 and 2024. The publication document content type distribution of the scientific studies on seed coating is presented as follows: Articles: 456; article book chapter: 15; article early access: 4; article proceedings paper: 5; Editorial material: 2; meeting abstracts: 3; Proceedings paper: 56; review: 88; and review book chapter: 2, totalling 631 publications. Among these, articles are the most frequent form of scientific publication. Reviews and proceedings papers are ranked next in the number of publications. In contrast, early access articles, book chapters, article proceedings papers, meeting abstracts, editorial materials, and review book chapters are much fewer compared to other types of publications.

2.2. Method

Great care was taken in selecting appropriate keywords and relevance of scientific publications to identify relevant studies on seed coating. After this step, data concerning the types of publications were extracted from these studies' bibliographies through the Web of Science database. The data were downloaded from the Web of Science database in plaintext format and processed using the “convert2pdf” function in R software (R Core Team 2024). After organizing the data, the dataset underwent comprehensive bibliometric analysis with the "bibliometrix" package in R software (Aria and Cuccurullo 2017).

3. Results

The curve in the following chart indicates that the number of publications each year, from 1980 to 2024, continuously shows an obvious uptrend in annual scientific research related to vegetable seed coating. The quantity has remained at a low level before 2000, with an average annual output below 10 papers; after 2001, the publication rate gradually went up and had an accelerated growth after 2011. The most productive period was from 2021 to 2024, where more than 60 articles were published in a year. Though slight ups and downs are present, the data indicate continuous growth in research outputs concerning seed coating technologies and, hence, increased interest in this topic by the scientific community, as presented in Figure 1.The curve in the following chart indicates that the number of publications each year, from 1980 to 2024, continuously shows an obvious uptrend in annual scientific research related to vegetable seed coating. The quantity has remained at a low level before 2000, with an average annual output below 10 papers; after 2001, the publication rate gradually went up and had an accelerated growth after 2011.

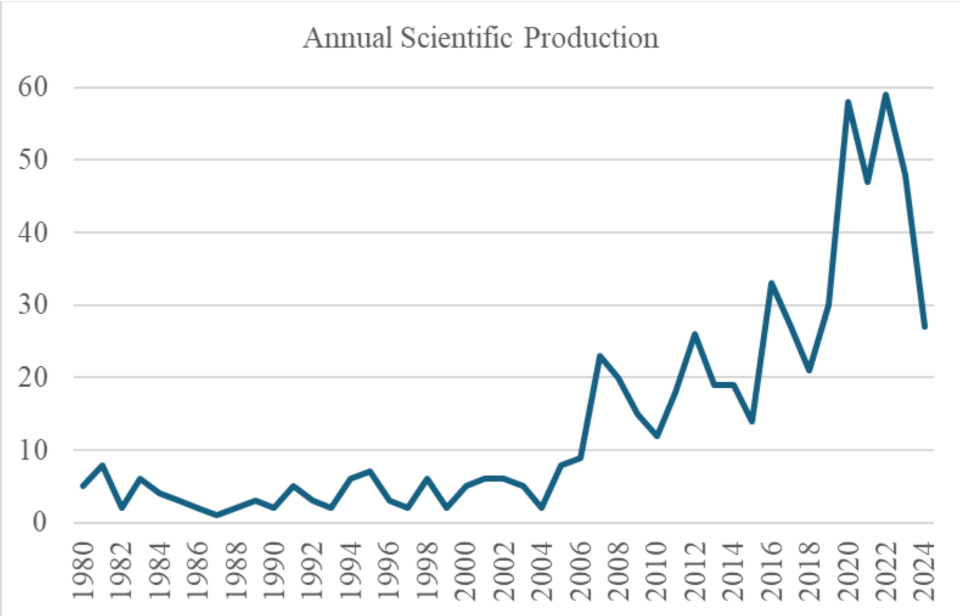

Figure 1. Trends in annual scientific publications on seed coating (1980-2024)

In addition to the growing number of publications, citation analysis represents an interesting indicator about the impact of scientific published works. The annual average citation counts from 1980 through 2024 deserves consideration with regard to the impact of research output on the scientific community. Figure 2 shows that citations increased rapidly, especially for works published after 2010. The annual average citation count is more than 10 for the period between 2014 and 2018, indicating that the works published within this period were highly cited. Therefore, from 2019 onward, the annual average cite counts decreased due to the relative freshness of the publications and hence fewer citations it would get. Another hypothesis could be that such studies beyond 2020 might have drawn on consequences of changed research dynamics in light of the COVID-19 pandemic.

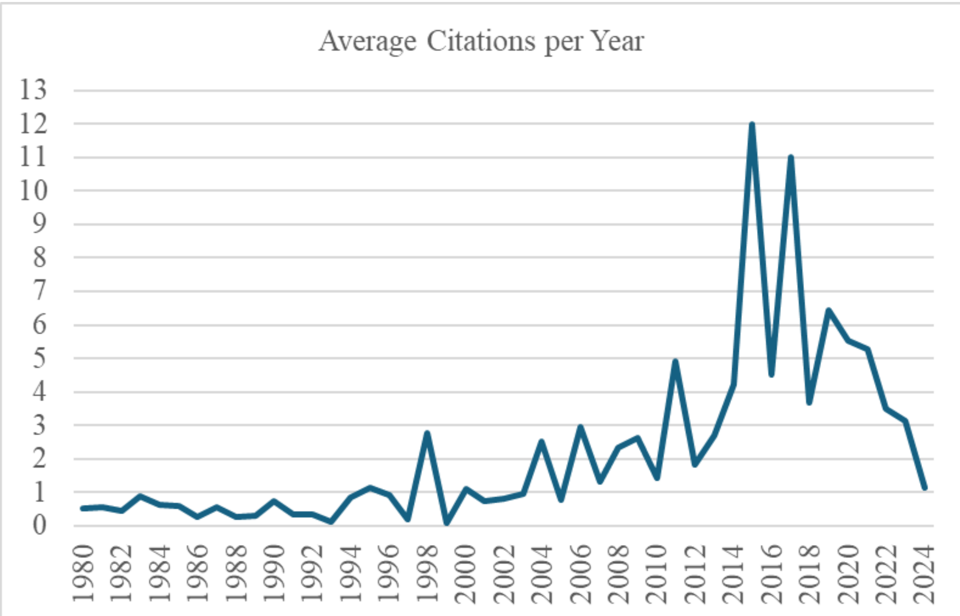

Figure 2. Yearly average citations for scientific publications on seed coating

The top 10 journals with the highest number of publications in the field of vegetable seed coating are shown in Figure 3.

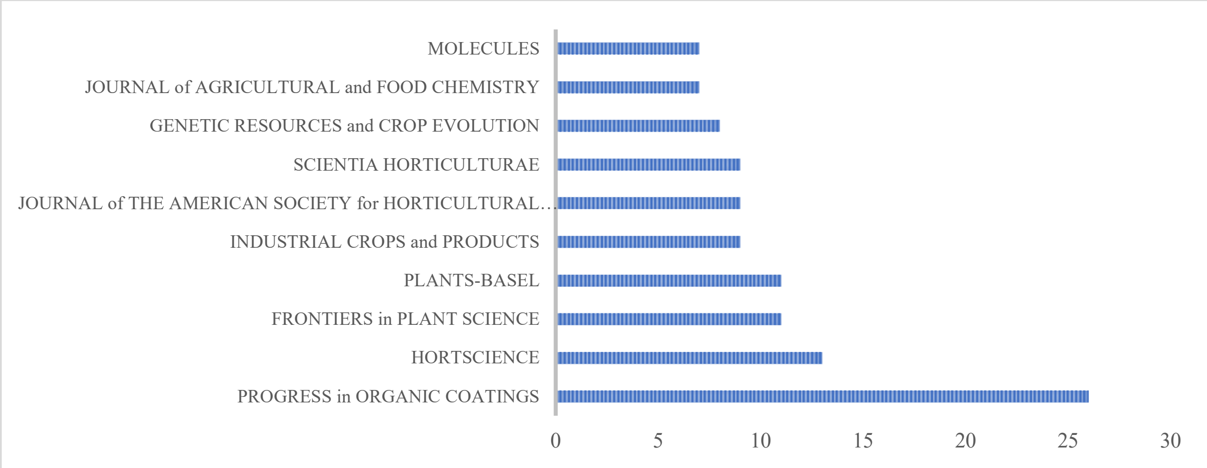

Figure 3. Journals with the highest number of scientific publications in the field of vegetable seed coating.

With a total of 26, the journal publishing most articles on vegetable seed coating is Progress in Organic Coatings, followed by Hortscience, with 13 articles; at 11 each are the journals of Frontiers in Plant Science and Plants-Basel, which come immediately after Progress in Organic Coating as the most publishing. Among the top 10 journals, the least published journals are *Journal of Agricultural and Food Chemistry* and *Molecules*, each with 7 articles. The 10 authors who have conducted the most scientific studies on vegetable seed coating are presented in Figure 4.

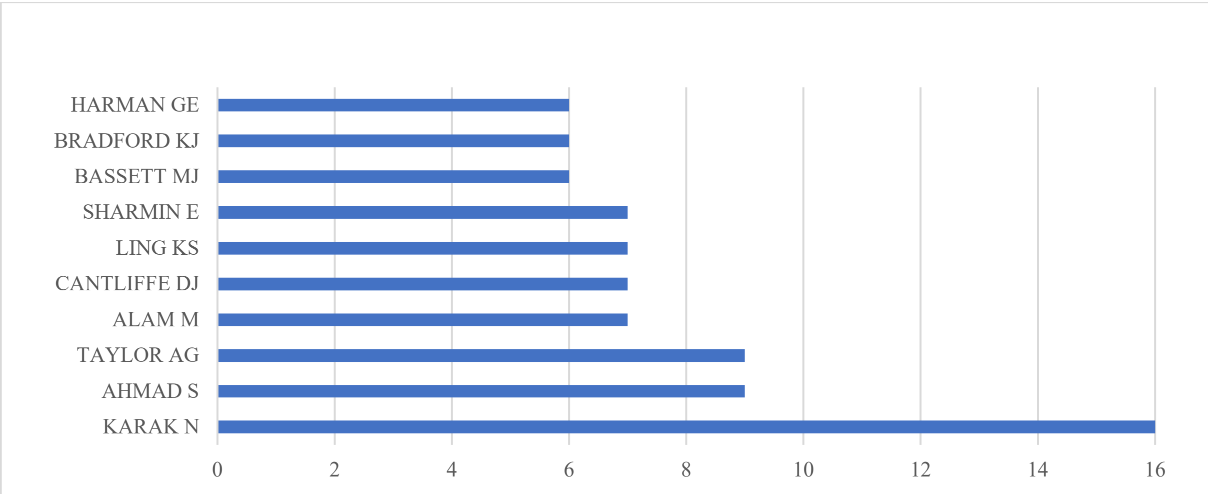

Figure 4. Journals with the highest number of scientific publications in the field of vegetable seed coating.

In 1980, Harman GE is recognized as the first author to publish a scientific paper on vegetable seed coating. Upon examining Figure 3, Karak N emerges as the leading author in the field, with 16 publications. Ahmad S and Taylor AG, with 9 publications each, are also significant contributors to the field. Among the top 10 authors with the most scientific publications, Harman GE, who conducted the first study on the topic, ranks last. The top 10 authors with the most citations for their scientific works are shown in Figure 5.

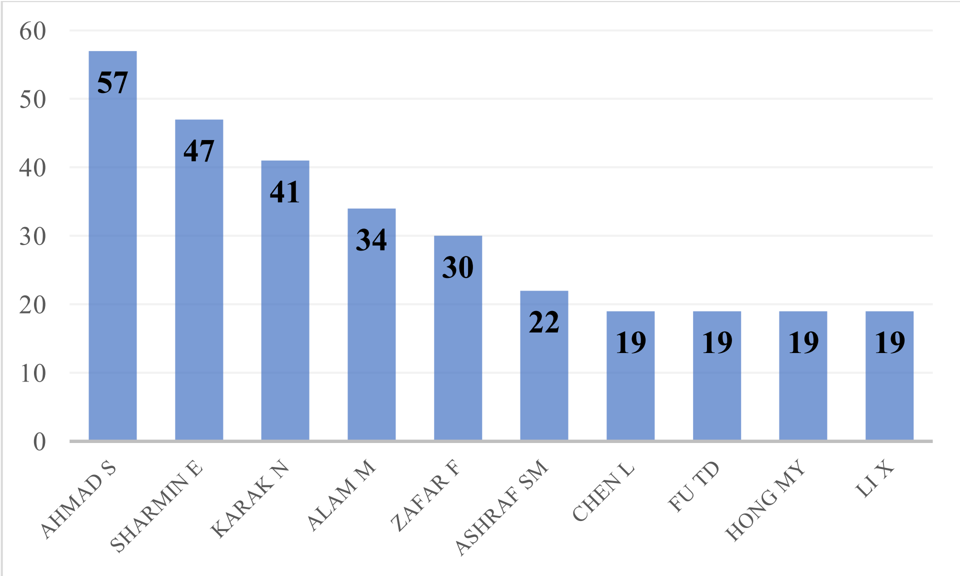

Figure 5. Authors with the most citations in the field of vegetable seed coating.

Upon examining Figure 4, Ahmad S stands out as the most cited author with 57 citations. Karak N, the author with the highest number of publications in the field of vegetable seed coating, ranks third with 41 citations. Among the top 10 authors with the most publications in the field, Sharmin E, with 7 articles, ranks seventh but stands out as the second most cited author with 47 citations. Harman GE, who conducted the first study on the topic, does not appear among the top 10 most cited authors. The distribution of corresponding authors' countries and the collaboration patterns between these countries in the field of vegetable seed coating are presented in Figure 6.

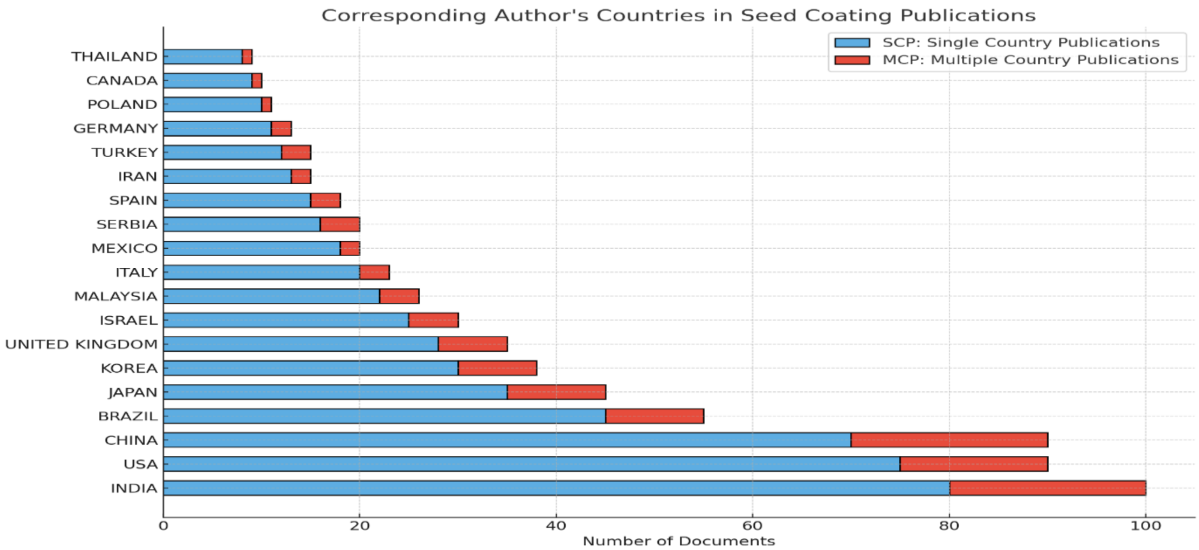

Figure 6. Distribution of authors with scientific publications on vegetable seed coating by country.

In terms of countries with corresponding authors of scientific publications on vegetable seed coating, India is the country with the most publications, with 109 articles (MCP: 17, SCP: 92). After India, USA with 102 articles, MCP: 15, SCP: 87, China with 87 articles, MCP: 21, SCP: 66 and Brazil with 29 articles, MCP: 5, SCP: 24 are the leading countries that have the highest number of scientific studies in this area. China has the highest internationally collaborative publications with 21 joint articles. Türkiye has 10 scientific papers on vegetable seed coating, of which 0 is at the level of MCP and 10 is on the level of SCP. It is worth mentioning that Türkiye is the only one among the top 20 countries with the largest number of publications in this field and does not have an internationally collaborative publication. This suggests a relatively more inward-looking nature for research in vegetable seed coating. Figure 7 shows the ranking of the first 10 countries that publish scientific studies on vegetable seed coating.

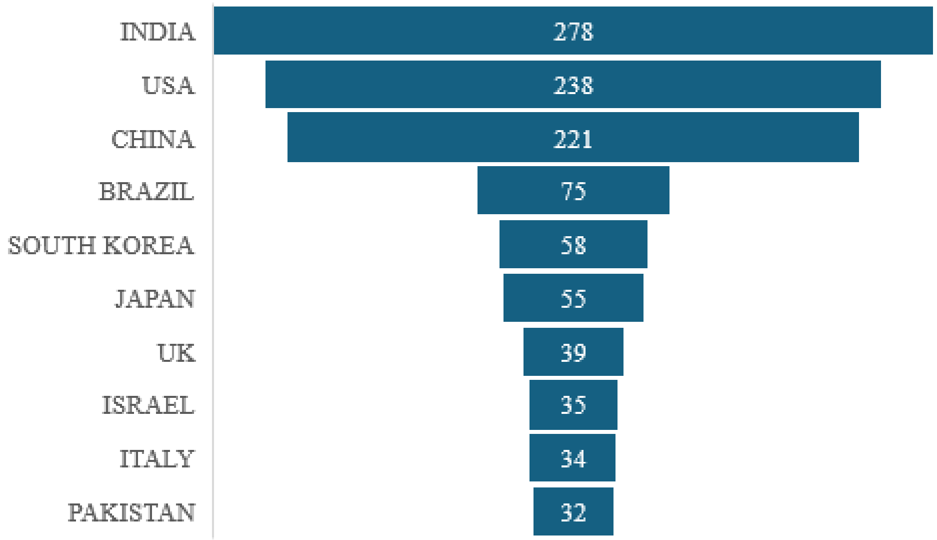

Figure 7. Top 10 countries by number of publications on vegetable seed coating

It places India in the lead with 278 scientific studies, followed by the USA with 238, China with 211, and Brazil with 75 scientific publications. Pakistan is ranked at the tail among the top ten countries, with a total of 32 studies. With 24, Türkiye doesn't make the Top 10. Considering scientific studies in number, the Asian and American continents are shown to be the leaders on vegetable seed coating studies. European countries conduct fewer vegetable seed coating studies compared to Asian and American countries. This cannot be overemphasized in European countries for any better approach to this aspect driven by a general lack in scientific studies regarding vegetable seed coatings. The 10 most important keywords in scientific studies on vegetable seed coating are shown in Figure 8.

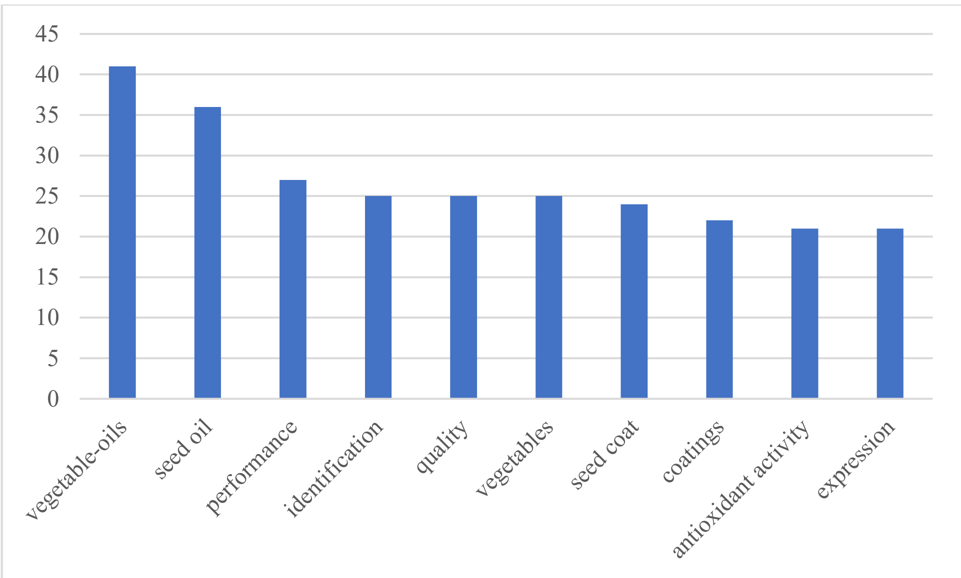

Figure 8. The most important keywords in scientific studies on vegetable seed coating.

Scientific studies on vegetable seed coating have focused on certain key terms. Among these studies, "vegetable-oils" (41) is the most frequently used keyword. Other commonly used keywords include "seed oil" (36), "performance" (27), "identification" (25), "quality" (25), "vegetables" (25), and "seed coat" (24). The other keywords are less frequently used, examples being "antioxidant activity", which occurs 21 times, and "expression" also occurring 21 times. From the keywords, it would appear that research into this area involves the application of seed oils as coating materials for the enhancement of germination rates, the selection of appropriate materials to use as coating agents for seeds, the assessment and evaluation of seed quality and shelf life, and the prevention of diseases in vegetable seeds using coating agents that have antioxidant properties. Figure 9 shows the word cloud created from these keywords.

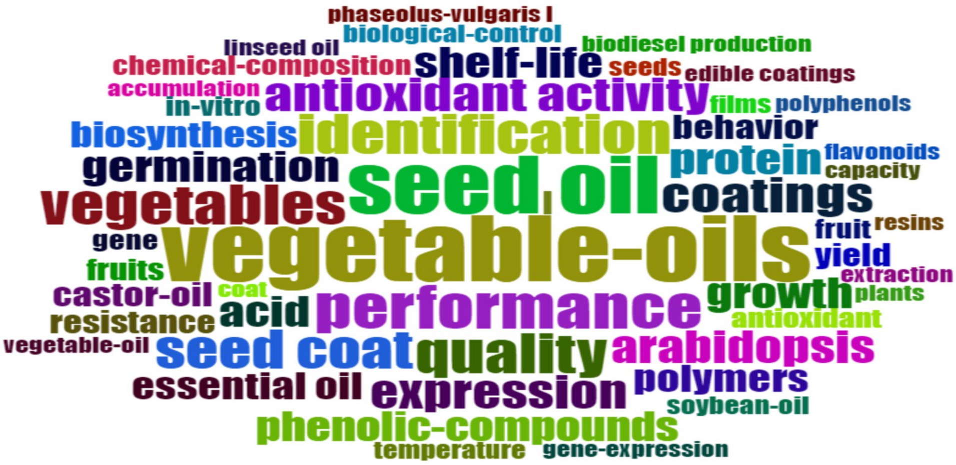

Figure 9. Word cloud generated from the most common keywords in the field of vegetable seed coating.

The relationships between the keywords used in scientific studies on vegetable seed coating are shown in Figure 10.

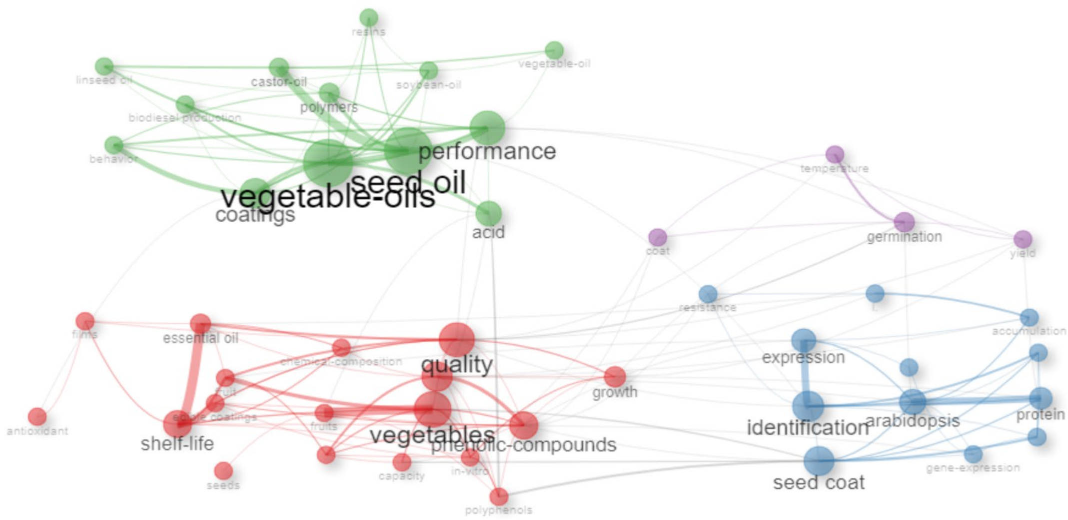

Figure 10. Relationships between keywords in scientific studies on vegetable seed coating.

Upon examining Figure 9, the keywords are organized into four groups, each represented by a different color. The first group is centered around the keywords "vegetable-oils" and "seed oil," which show a strong connection with the other terms in the group. The majority fall into this cluster probably due to studies focusing on the use of seed oils as coating materials. The second cluster combines words such as "vegetables" and "quality," showing some relationship between the studies of growth, coating, capacity, and chemical composition. The third cluster contains the words "germination," "temperature," "yield," and "coat," which represent studies on the influence of temperature on germination, the impact of germination on yield, and the improvement in the rate of seed germination. The fourth cluster, represented in blue, includes the words "identification," "expression," and "seed coat," which are related to each other. This association may relate to the studies of proper selection of the materials used in coating seeds and their processing for quality control.

4. Conclusions

The study presents a wide bibliometric analysis of vegetable seed coating research produced within the period of 1980-2024, hence highlighting advances, trends, and gaps in this important area of research. The results of the present study have evidenced the continuous rise in scientific publications, especially in the last decade, reflecting growing interest in seed coating technologies as basic tools to improve agricultural productivity and sustainability. This reflects more the importance of the area to handle issues concerning germination, development of seedlings, management of pests and diseases, and increasing crop yield in a changing environment; as can be seen in the increased number of publications and citation rates.

Despite these advances, standardized methodology, interdisciplinary collaboration, and the search for innovative materials for seed coating remain a number of the key challenges facing the area. Future studies will be directed toward the improvement of performance and scalability of the coatings by incorporating advanced technologies, such as nanomaterials and bio-based polymers. Besides, filling up the gaps in data sharing will add more strength to the foundation of this field.

The present study, while achieving remarkable successes in the field of seed coating, has tried to enhance the contribution of our country, which has a relatively modest share in the global research landscape, and raise awareness about the importance of seed coating technologies. Closing existing gaps and developing much more collaboration will go a long way in helping the contribution of seed coating technologies toward the achievement of betterment in sustainable agriculture, improving global food security, and answering the challenges brought forward by the changing climate.

Author Contributions: Conceptualization: S.E and Y.A.; methodology, S.E and Y.A, analysis of data, production of figures: S.E. Y.A. and A.K.C.T; writing, review, and editing: S.E., Y.A., A.K.C.T and J.K.A.; supervision of research, structuring the paper, writing the original draft, and draft preparation: S.E. and Y.A. All authors have read and agreed to the published version of the manuscript.

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References

Afzal I, Javed T, Amirkhani M, Taylor AG (2020). Modern seed technology: Seed coating delivery systems for enhancing seed and crop performance. *Agriculture,* 10(11): 526. https://doi.org/10.3390/agriculture10110526.

Altay Y, Kaplan S (2023). Bibliometric analyzes of some major effect genes associated with meat yield traits in livestock. *Selcuk Journal of Agriculture and Food Sciences,* 37(3): 608-617. https://doi.org/10.15316/SJAFS.2023.058 e.

Aria M, Cuccurullo C (2017). Bibliometrix: An R-tool for comprehensive science mapping analysis. *Journal of Informetrics,* 11(4): 959-975. https://doi.org/10.1016/j.joi.2017.08.007.

Çelik Ş (2020). Bibliometrics analysis and a husbandry practice. *Journal of Multidisciplinary Engineering Science Studie*s (JMESS), 6(12): 3632-3641. ISSN: 2458-925X.

Gullino ML, Gilardi G, Garibaldi A (2014). Chemical and non-chemical seed dressing for leafy vegetable crops. In: *Global Perspectives on the Health of Seeds and Plant Propagation Material,* p. 125-136. https://doi.org/10.1007/978-94-017-9389-6_9.

Kaplan S, Altay Y (2023). Bibliometric analysis of next-generation sequence applications in livestock. *Black Sea Journal of Agriculture,* 6(5): 485-491. https://doi.org/10.47115/bsagriculture.1296263.

Lamichhane JR, Debaeke P, Steinberg C, You MP, Barbetti MJ, Aubertot JN (2018). Abiotic and biotic factors affecting crop seed germination and seedling emergence: a conceptual framework. *Plant and Soil,* 432: 1-28. https://doi.org/10.1007/s11104-018-3780-9.

Onder H, Tırınk C (2022). Bibliometric analysis for genomic selection studies in animal science. *Journal of the Institute of Science and Technology,* 12(3): 1849-1856. https://doi.org/10.21597/jist.1133397

Pedrini S, Bhalsing K, Cross AT, Dixon KW (2018). Protocol Development Tool (PDT) for seed encrusting and pelleting. *Seed Science and Technology,* 46(2): 393-405. https://doi.org/10.15258/sst.2018.46.2.12.

Pedrini S, Balestrazzi A, Madsen MD, Bhalsing K, Hardegree SP, Dixon KW, Kildisheva OA (2020). Seed enhancement: getting seeds restoration-ready. *Restoration Ecology,* 28(S1): S266-S275. https://doi.org/10.1111/rec.13173.

R Core Team (2024). R: A Language and environment for statistical Computing. Vienna, Austria: *R Foundation for Statistical Computing*. https://doi.org/10.1890/0012-9658(2002)083[3097:CFHIWS]2.0.CO;2.

Taylor, A.G. (2003). Seed treatments. In *Encyclopedia of Applied Plant Sciences*, (eds. B.D.J.538 Thomas and B.G. Murphy), pp. 1291-1298, Elsevier Academic Press, Cambridge.

Taylor AG (2020). Seed storage, germination, quality, and enhancements. In: The Physiology of Vegetable Crops (pp. 1-30). Wallingford, UK: CABI. https://doi.org/10.1079/9781786393777.000.

Taylor AG, Eckenrode CJ, Straub RW (2001). Seed coating technologies and treatments for onion: challenges and progress. *HortScience,* 36(2): 199-205. https://doi.org/10.21273/HORTSCI.36.2.199

Taylor AG, Harman GE (1990). Concepts and technologies of selected seed treatments. *Annual Review of Phytopathology*, 28: 321-339. https://doi.org/10.1146/annurev.py.28.090190.001541.

Westbrook AS, Amirkhani M, Taylor AG, Loos MT, Losey JE, DiTommaso A (2023). Multi-Seed Zea Pellets (MSZP) for increasing agroecosystem biodiversity. *Weed Science,* 71(2): 160-171. https://doi.org/10.1017/wsc.2023.16.

Villalobos FJ, Orgaz F, Fereres E (2016). Sowing and planting. In: Principles of Agronomy for Sustainable *Agriculture,* 217-227. https://doi.org/10.1007/978-3-319-46116-8_16.

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Research Article

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Efficacy of the Seed Powder and Oil of *Jatropha Curcas* in the Control of *Acanthoscelides obtectus* in Stored Cowpea

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HIGHLIGHTS

- It is important to protect and preserve the cowpea for human and animal consumption.
- It is important to control post-harvest losses caused by cowpea weevil using non synthetic chemicals.

Abstract

A study was carried out to evaluate the efficacy of seed powder and oil of *Jatropha curcas* in controlling post-harvest losses caused by cowpea weevil, *Acanthoscelides obtectus* (Say) on cowpea. 100 g of *J. curcas* powder were put in 300 ml of n- hexane used as the solvent. The Jatropha seed powder was tested at dosages of 0.1g, 0.2g and 0.3g and Jatropha seeds oil at the rate of 0.05ml, 0.1ml and 0.2ml were applied to 20g cowpea seeds in petri dishes. The different rates of treatment recorded significant differences (P=0.05) in causing adult mortality compared to the untreated control. The different rates of treatment also recorded significant differences (P=0.05) in oviposition of each treatment compared to the control. The percentage weight loss and seed damage were also suppressed as a result of treatment with the plant material compared to the untreated control. However, among the treatments Jatropha seeds oil at 0.2ml/20g cowpea recorded the highest adult mortality rate and lowest oviposition while control had the lowest mortality rate and highest oviposition. The study shows that Jatropha had bio pesticides effect in controlling *A. Obtectus.*

Keywords: Soxhlet apparatus; *Acanthoscelides obtectus; Jatropha curcas;* cowpea; n-hexane; pirimiphos methyl; oviposition

1. Introduction

Cowpea (*Vigna unguiculata* (L.) Walp) originated in West and Central Africa before spreading to Latin America and Southeast Asia (Edeh and Igberi 2012; Karadaş and Ceyhan 2023). As a legume, cowpea plays a crucial role in nutrient cycling due to its tolerance to drought and soil acidity, as well as its ability to fix

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nitrogen from the air. This makes it particularly well-suited for regions experiencing declining soil fertility and drought (Sert and Ceyhan 2012; Ibrahim et al. 2016; Ceyhan and Karadaş 2023). Cowpea seeds serve as a major source of plant proteins and vitamins, animal feed, and a source of income (Ibrahim et al. 2016; Jou-Nteufa and Ceyhan 2022). In Nigeria, cowpea (*Vigna unguiculata*) is an important source of dietary protein, particularly because its edible seeds are a cost-effective alternative to more expensive protein sources (Adedire et al. 2011; Jou-Nteufa and Ceyhan 2025). Cowpeas are also a valuable addition to the diets of resource-poor farmers, providing supplementary nutrition alongside their primary staples, such as roots, tubers, and cereals (Ileke et al. 2013). Cowpea is a significant staple food and cash crop in Nigeria, with the potential to greatly reduce malnutrition among resource-poor farmers while improving food security, productivity, and the sustainability of crop-livestock systems (IITA 2009). A study by Ibrahim et al. (2016) found that low-income individuals tend to rely more on cowpea-based food products as their primary source of protein, rather than animal protein. Research conducted in Nigeria by Dugje et al. (2009) highlighted the economic benefits for farmers who cut and store cowpea fodder for sale during peak dry seasons, which can increase their annual income by 25%. When incorporated into crop rotation, cowpea contributes nitrogen to the soil, benefiting cereal crops such as maize, millet, and sorghum, particularly in areas struggling with poor soil fertility (Tijjani et al. 2015).

However, cowpea production faces numerous challenges, including damage from insect pests, parasitism by Striga gesnerioides, diseases, drought, low and unpredictable rainfall, and extended dry seasons (Singh et al. 2002). *Acanthoscelides obtectus* (Say) (Coleoptera: *Chrysomelidae*: *Bruchinae*) originates from Mesoamerica and is a significant post-harvest and field pest of both wild and cultivated common beans (*Phaseolus vulgaris* L., *Fabaceae*) in tropical regions (Paul et al. 2010; Thakur 2012). *Acanthoscelides obtectus* and *Zabrotes subfasciatus* (Boheman) are two species of beetles that often coexist in bean storage areas, sharing the same habitat and geographical range. Among these two species, *Acanthoscelides obtectus*, which has a wide distribution in eastern and southern Africa (Mutungi et al. 2015), is economically significant. Many small-scale farmers in this region rely on bean production and sales as a crucial source of household income. When infested, these farmers often sell their beans immediately after harvest, when market prices are at their lowest. Infestations can lead to substantial dry weight losses of 10% to 40% in less than six months, with damage rates potentially reaching up to 70% in the same time frame (Paul et al. 2009). The post-harvest damage caused by *A. obtectus* results in significant financial losses for African small-scale farmers; Mishili et al. (2011) estimated a 2.3% decrease in price for each insect emergence hole in 100 beans. All larval instars of *A. obtectus* are voracious feeders, consuming legume proteins, often reducing heavily infested beans to empty shells. *A. obtectus* has a short life cycle of three to four weeks and a high reproductive potential, allowing for the emergence of multiple generations per year under favorable conditions (Soares et al. 2015). Female *A. obtectus* lay eggs in clusters inside pods in the field or on shelled stored bean seeds (Godrey and Long 2008). Although typically one larva infests each seed, multiple infestations can occur, with late instar larvae entering the same seed through the burrow made by the first. The final instars then excavate a chamber just below the seed testa for pupation. The presence of a final instar or pupa can be visually detected by a small window in the seed coat. After emergence, the adult chews a hole in the seed coat to exit and is ready to mate. As with many other post-harvest pests of stored grain, *A. obtectus* infestations begin in the field, where adults lay eggs in dried bean pods. By late harvest, damage to the beans can be so severe that there might be little to no harvest at all (Velten et al. 2007). There is an urgent need to educate farmers about safer and effective pest control measures that have minimal side effects.

2. Materials and Methods

2.1. Study Area

This study was conducted in the Department of Crop Protection laboratory of the University of Ilorin. The study location falls within the southern Guinea Savannah agro-ecological zone of Nigeria between latitude 8°25'N and longitude 4°67'E.

2.2. Source of Seeds

The cowpea seed variety used for this study is RSH 256 and was sourced from the International Institute of Tropical Agriculture (IITA), Ibadan, Oyo State, Nigeria. The seeds were wrapped in a polyethylene bag and were kept in the freezer at the Crop Protection laboratory, University of Ilorin, to kill any immature stages of insects. The seeds were removed 10 days after freezing and then spread on a laboratory desk to unthaw.

2.3. İnsect Culture

A culture of cowpea weevil, *A. obtectus,* was prepared at laboratory ambient temperature (30 ± 3°C) and relative humidity (68 ± 3%) respectively. The weevils were collected from already existing stock in the Crop Protection laboratory, University of Ilorin, Nigeria. Fifty (50) unsexed adults of *A. obtectus* were used to infest cowpea seeds in a transparent plastic container which was covered with muslin cloth held tightly by perforated lid to ensure aeration and prevent escape of the insects. Freshly emerged adults were used for the study.

2.4. Apparatus and Equipment

The apparatus, equipment and materials used include transparent plastic containers, cowpea seeds, water, n-hexane, measuring cylinder, conical flasks, *Jatropha Curcas* seeds, muslin cloth, a pair of scissors, a razor blade, paper tape, a hairbrush, digital weighing balance, spatula, Soxhlet apparatus, a plastic sieve, electric blender, rubber band, laboratory desk, freezer, petri dish, foil paper and beakers.

2.5. Collection and Preparation of Plant Material

Jatropha Curcas seeds were collected from the parent plant at the University of Ilorin Jatropha plot between latitude 8°29′N and longitude 4°35′E in the morning. *J. Curcas* seeds were cracked and the shells were carefully removed to get the kernel. The kernels were dried at 30^0C to get constant moisture content after which they were ground with the aid of an electric blender, and sieved to obtain uniform particles. The powder was stored in a vial until required for use.

2.6. Soxhlet Extraction

100 g of *J. Curcas* powder was wrapped in a muslin cloth and tied with a rubber band before introducing into the thimble of a Soxhlet extractor. 300 ml of n-hexane was introduced into a round bottom flask. The electric cooker served as the source of heat (b.p. 68^0C) which lasted for 6-7hrs. The excess solvent was separated from the oil with the aid of rotary evaporator and was collected in the receiving flask.

2.7. Experimental Procedure

Experiment was conducted on four (4) main areas, namely Toxicity bioassay, Oviposition deterrence, Seed damage and Germination test

2.7.1. Toxicity bioassay

In this experiment, Jatropha seed powder at the rates of 0.1g, 0.2g and 0.3g and Jatropha oil at the rates of 0.05ml, 0.1ml and 0.2ml were applied to 20g cowpea seeds in petri dishes. A single rate of Pirimiphos methyl 0.4ml was added as a check. An untreated control was also added as part of the treatments making a total of eight (8) treatments. Cowpea seeds treated with Jatropha oil and pirimiphos methyl were air dried. Ten (10) unsexed *A. obtectus* were introduced into the treated cowpea seeds. All treatments were replicated three times. Mortality count was taken after 1, 3 and 7 days.

2.7.2. Ovipositon deterrence

This experiment was conducted in two phases namely: Choice test and No Choice test.

Choice Test: For this setup, 20g cowpea seeds in petri dishes were treated with highest and lowest concentration of Jatropha seed powder 0.3g and 0.1g, highest and lowest concentration of Jatropha seed oil and Pirimiphos methyl 0.4ml and untreated control was also added as part of the treatments making a total of six (6) treatments. After air drying, one seed from each treated cowpea seeds was picked and placed inside glass petri dish which contained melted paraffin wax to hold the seed in place. There were three (3) replicates for the experiment. The number of eggs laid on each cowpea seed was counted after the death of the insects.

No Choice test: For this setup, 20g cowpea seeds in petri dishes were treated with only highest concentration of Jatropha seed powder 0.3g and highest concentration of Jatropha seed oil 0.2ml and Pirimiphos methyl 0.4ml and untreated control were added as part of the treatments making a total of four (4) treatments. Six (6) seeds were selected at random from the treated cowpea seeds and placed carefully inside glass petri dishes which contained melted paraffin wax to hold the seed in place. A pair of adult day old of *A. obtectus* was introduced. The insects were not given choice of which treated cowpea seeds to lay their eggs as only seeds of the same treatment were represented in each petri dish. The treatments were replicated three (3) times, the number of eggs laid was counted after the death of the insects.

2.7.3. Seed damage

In this experiment, 20g of cowpea seeds were treated with 0.1g, 0.2g and 0.3g of Jatropha seed powder and Jatropha seed oil of 0.05ml, 0.1ml and 0.2ml and Pirimiphos methyl 0.4ml in transparent plastic containers. An untreated control was also added as part of the treatments making a total of eight (8) treatments in all. Cowpea seeds treated with Jatropha seed oil and pirimiphos methyl were air dried after which two (2) pairs of adult *A. obtectus* were introduced. Treatments were replicated three times. The plastic containers were covered with muslin cloth and the setup was placed in the laboratory for 45 days after which the number of undamaged seed (seeds without holes) and number of damaged seeds (seeds with holes) was counted and recorded. The weight of seeds in each cup was also taken and recorded.

2.7.4. Germination test

In this experiment, 20g of cowpea seeds were treated with 0.1g, 0.2g and 0.3g of Jatropha seed powder and Jatropha seed oil of 0.05ml, 0.1ml and 0.2ml and Pirimiphos methyl 0.4ml in transparent plastic containers. An untreated control was also added as part of the treatments making a total of eight (8) treatments in all. Cowpea seeds treated with Jatropha seed oil and pirimiphos methyl were air dried. Treatments were replicated three times and no insect was introduced to the seeds placed in a transparent plastic containers covered with muslin cloth and the setup was placed in the laboratory for three (3) months after which ten (10) seeds were randomly selected from each plastic container and placed in a petri dishes whose inside have been laid with tissue paper. Distilled water was used to moisten the tissue paper. Observations were taken on the following

I. The number of seeds with radicles at 24, 48 72 and 96 hours after set up
II. The number of seeds with radicles and root hairs at 96 hours after set up
III. The number of seeds with hypocotyl at 96 hours after set up
IV. The number of seeds with primary leaves at 96 hours after set up

2.8. Data Collection

Data were collected on various parameters including adult mortality, Oviposition, Seed damage and Germination. The percentage seed mass loss was computed following the method of Haines (1991) as follows:

$$\frac{initial\ weight - final\ weight}{Initial\ weight} X100 \qquad (1)$$

2.9. Statistical Analysis

All data collected were subjected to analysis of variance and in a Completely Randomized Design (CRD) model using GenStat 2017 version and significant differences. Treatment means were separated using Duncan`s Multiple Range test (DMRT) at 5% level of significance.

3. Results and Discussion

The results of this experiment shows that the various treatments used in this study had effects on mortality with increase of dosages of Jatropha seed oil caused adult mortality of *A. obtectus* at the high rates of 0.2ml after 1 day, 3 days and 7 days when compared to the untreated control. (Table 1) after the same period. This indicates that Jatropha seed powder and Jatropha seed oil has the ability to cause mortality of *A. obtectus* when compared with Pirimiphos methyl that was added as check. This support the findings of Babatunde and Musa. (2020) who reported that *E. globulus* plant extracts can be another source of insecticides/pesticides against stored grain pests.

There was considerable mortality of *A. obtectus* in cowpea seeds treated with the highest rate of (Table 1). Jatropha seed powder of 0.3 g which could be compared with the lowest concentration of Jatropha seed oil 0.05 ml.

Table 1. Effects of different rates of treatments on percentage mortality of adult

Percentage Adult mortality of *A. obtectus* (DAT)			
Treatments	**Day 1**	**Day 3**	**Day 7**
Jatropha powder 0.1g	0.0a	16.7a	93.3a
Jatropha powder 0.2g	10.0ab	20.0a	93.3a
Jatropha powder 0.3g	36.7bc	76.7b	100.0a
Jatropha Oil 0.05ml	63.3cd	90.0b	100.0a
Jatropha Oil 0.1ml	86.7de	100.0b	100.0a
Jatropha Oil 0.2ml	100.0e	100.0b	100.0a
Untreated control	0.0a	13.3a	90.a
Pirimiphos methyl 0.4ml	100.0e	100.0b	100.0a
SEM	8.9	9.6	3.9

Values with the same letter in the same column are not significantly different at P=0.05

For the Choice test, no egg was laid on cowpea seeds treated with Pirimiphos methyl, Jatropha seed oil both at lowest 0.05ml and highest 0.2ml rates compared with untreated control (Table 2a). This can be attributed to the fact that the introduced insects died within 24hrs after treatment suggesting that the treatment had effect on contact with the insects thereby hindering egg laying. This was in agreement with (Babatunde et al., 2021) who indicated that cashew nut extract reduced progeny emergence as well as reducing the damage on cowpea seeds by *C. maculatus*. For no choice test, eggs were laid on the cowpea with seeds treated with pirimiphos methyl when compared with untreated control (Table 2b). In the no choice test, eggs were laid on cowpea seeds treated with Jatropha seed oil 0.2ml but not as much as Jatropha seed powder 0.3g and untreated control (Table 2b).

Table 2a. Effects of different rates of treatments on oviposition of *A. obtectus* choice test

Treatments	**Mean number of eggs Laid**	**Mean number of seeds oviposited**
Jatropha powder 0.1g	1.7b	0.7b
Jatropha powder 0.3g	1.0ab	0.7b
Jatropha Oil 0.05ml	0.0a	0.0a
Jatropha Oil 0.2ml	0.0a	0.0a
Untreated control	1.7b	1.0b
Pirimiphos methyl 0.4ml	0.0a	0.0a
SEM	0.5	0.2

Values with the same letter in the same column are not significantly different at P=0.05

Table 2b. Effects of different rates of treatments on oviposition of *A. obtectus* (no choice test)

Treatments	**Mean number of eggs Laid**	**Mean number of seeds oviposited**
Jatropha powder 0.3g	38.0bc	6.0a
Jatropha Oil 0.2ml	14.3ab	4.0a
Untreated control	52.7c	6.0a
Pirimiphos methyl 0.4ml	0.0a	0.0a
SEM	9.2	0.8

Values with the same letter in the same column are not significantly different at P=0.05

Table 3a shows that cowpea seeds treated with pirimiphos methyl 0.4ml and Jatropha seed oil 0.2ml had the highest percentage of undamaged seeds (zero damage) when compared with untreated control. Jatropha powder also offered some measure of protection against *A. obtectus* on stored cowpea seeds as Jatropha seed powder 0.3g had the lowest percentage number of seeds with holes and therefore offered the highest protection followed by as Jatropha seed powder 0.2g and 0.1g as compared with control. This indicates that at high rate, Jatropha seed powder was more effective and reduction in damage may be due to fact that fewer eggs were laid on cowpea seeds giving a few population of next generation, hence damage was reduced. This corroborate (Musa and Sulyman 2014) who reported aqueous peel extracts of grapefruit and lime reduced number of eggs, progeny emergence and the seeds were not riddled with holes and also believe that applications demonstrated inhibitory effects on *C. maculatus* development.

There was weight reduction in all cowpea seeds treated with the three rates of Jatropha powder 0.1g, 0.2g and 0.3g and their weight were compared with control which shows that Jatropha powder was not as effective as Jatropha oil in protecting cowpea seeds against *A. obtectus.*

There was no weight reduction for the cowpea seeds treated with Jatropha oil as compared with control indicating that Jatropha oil was very effective in protecting cowpea seeds against damage (Table 3b)

Table 3a. Effects of different rates of treatments on seed damage on percentage seeds with and without hole.

Percentage Number of seeds with and without hole					
Treatments	**0 Hole**	**1 Hole**	**2 Holes**	**3 Holes**	**4 Hole and above**
Jatropha Powder 0.1g	53.5a	29.8b	11.3bc	4.3b	1.1a
Jatropha Powder 0.2g	72.2a	19.8ab	5.1ab	1.8ab	1.1a
Jatropha Powder 0.3g	74.0a	18.4ab	5.8ab	1.5ab	0.4a
Jatropha Oil 0.05ml	99.6b	0.4a	0.0a	0.0a	0.0a
Jatropha Oil 0.1ml	100.0b	0.0a	0.0a	0.0a	0.0a
Jatropha Oil 0.2ml	100.0b	0.0a	0.0a	0.0a	0.0a
Untreated Control	53.1a	27.3ab	13.9c	3.6a	2.1a
Pirimiphos Methyl 0.4ml	100.0b	0.0a	0.0a	0.0a	0.0a
S.E	6.3	3.5	2.1	1.1	0.7
Control S.E	7.8	4.1	2.6	1.4	0.8

Values with the same letter in the same column are not significantly different at P=0.05

Table 3b. Effects of different rates of treatments on weight of cowpea seeds after damage by *C. maculatus*

Treatments	**Initial Weight**	**Current Weight**	**% Weight Loss/Weight Gain**
Jatropha Powder 0.1g	20.0	17.7ab	11.5
Jatropha Powder 0.2g	20.0	16.9a	15.5
Jatropha Powder 0.3g	20.0	16.9a	15.5
Jatropha Oil 0.05ml	20.0	21.3c	6.7
Jatropha Oil 0.1ml	20.0	22.0c	10.0
Jatropha Oil 0.2ml	20.0	20.1bc	0.5
Untreated Control	20.0	17.0a	15.0
Pirimiphos Methyl 0.4ml	20.0	20.0abc	0.0
S.E	0.0	0.9	
Control S.E	0.0	1.1	

Values with the same letter in the same column are not significantly different at P=0.05

On the whole, germination was hindered for Pirimiphos methyl and Jatropha oil 0.2ml indicating that Jatropha oil to be used in preservation must be of low concentration because high concentration may lead to reduced germination. Jatropha powder did not affect germination of seeds. Germination was prolonged for the oils than the powder and more prolonged for the powder than the untreated control (Table 4a and 4b).

Table 4a. Effects of different rates of treatments on germination of cowpea seeds after 3 months of storage

Percentage Number of Seeds with Radicle				
Treatments	**@ 24 Hours**	**@ 48 Hours**	**@ 72 Hours**	**@ 96 Hours**
Jatropha powder 0.1g	6.7ab	43.3cd	83.3c	93.3cd
Jatropha powder 0.2g	6.7ab	33.3bcd	76.7bc	96.7d
Jatropha powder 0.3g	3.3a	40.0cd	66.7bc	86.7bcd
Jatropha Oil 0.05ml	0.0a	23.3abc	56.7b	66.7b
Jatropha Oil 0.1ml	3.3a	30.0bc	56.7b	70.0bc
Jatropha Oil 0.2ml	0.0a	6.7a	53.3b	66.7b
Untreated control	16.7b	53.3d	83.3c	96.7d
Pirimiphos methyl 0.4ml	0.0a	16.7ab	30.0a	33.3a
S.E	3.3	7.1	7.2	8.0

Values with the same letter in the same column are not significantly different at P=0.05

Table 4b. Effects of different rates of Treatments on Germination of Cowpea Seeds after 3 months of Storage

Percentage Number of Seeds with Radicle, Root Hairs, Hypocotyl and Primary leaves @ 96 Hours			
Treatments	**Percentage Radicle and Root Hairs**	**Percentage Hypocotyl**	**Percentage Primary leaves**
Jatropha powder 0.1g	60.0bc	60.0bc	16.7bc
Jatropha powder 0.2g	66.7bc	53.3abc	3.3ab
Jatropha powder 0.3g	53.3bc	53.3abc	6.7bc
Jatropha Oil 0.05ml	53.3bc	50.0abc	10.0abc
Jatropha Oil 0.1ml	53.3bc	53.3abc	10.0abc
Jatropha Oil 0.2ml	43.3ab	36.7ab	0.0a
Untreated control	76.7c	76.7c	20.0c
Pirimiphos methyl 0.4ml	23.3a	23.3a	0.0a
S.E	8.5	9.1	4.1

Values with the same letter in the same column are not significantly different at P=0.05

4. Conclusions

Numerous researchers have undertaken investigations to identify plants with insecticidal properties that can effectively manage pests impacting food crops. This approach seeks to reduce dependence on synthetic insecticides, which may contribute to pest resistance, pose health risks, and lead to environmental degradation. One study specifically assessed the efficacy of seed powder and oil from Jatropha curcas in controlling post-harvest losses caused by the cowpea weevil, Acanthoscelides obtectus. The findings revealed that both Jatropha curcas seed powder and oil significantly reduce populations of Acanthoscelides obtectus. When applied at higher doses—0.3 grams for the seed powder and 0.2 milliliters for the oil—these treatments notably diminished beetle infestation and seed damage, demonstrating effectiveness comparable to that of the synthetic insecticide Pirimiphos-methyl. Consequently, products derived from Jatropha curcas offer a sustainable and environmentally friendly alternative for protecting stored cowpea from insect pests. The study recommends further research into the formulation and large-scale application of these treatments.

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References

Adedire CO, Olusola OM, Akinkurolere RO, Oduleye SO (2011). Response of *Callosobruchus maculatus* (Coleoptera: Chrysomelidae: Bruchinae) to extracts of cashew kernels. *Journal of Plant Diseases and Protection*, 118: 75–79.

Babatunde SF, Musa AK (2020). Effect of Tasmanian blue gum (*Eucalyptus globulus* Labill.) leaf extract oncowpea weevil (*Callosobruchus maculatus* [Fabricius, 1775], Coleoptera: Chrysomelidae). *Acta agriculturae Slovenica*, 116(2): 351–356

Babatunde SF, Musa AK, Suleiman AF, Gambari LI (2021). Effect of cashew (*Anacardium occidentale)* nut shell stored and fresh extracts on cowpea bruchid, *Callosobruchus maculatus* (Fabricius.) (Coleoptera: Chrysomelidae). *Acta Fytotechnica et Zootechnica*, 24(2): 124–128.

Ceyhan E, Karadaş S (2023). Determination of biological yield and some agronomic characteristics of newly developed forage cowpea pure lines. In: *Proceedings* of *Sivas II. International Conference on Scientific and Innovation Research,* 15-17 September 2023; Sivas, Türkiye, pp. 942-949.

Dugje LO, Omoigui LO, Ekeleme F, Kamara AY, Ajeigbe H (2009). Farmers' Guide to Cowpea Production in West Africa. IITA, Ibadan, Nigeria. P 3.

Ekesi S, Dike MS, Ogunlana MO (1996). Relationship between planting dates and damage by the legume pod-borer, *Maruca testularis* (Geyes) (Lepidoptera: Pyralidae) on cowpea, (*Vigna unguiculata*) L. (Walp.) in Nigeria. *InternationalJournal of Pest Management,* 42:*315-316.*

Edeh HO, Igberi CO (2012). Assessment of Vegetable Cowpea Production among Smallholder Farmers in Ebonyi State, Nigeria. *ARPN Journal of Agricultural and Biological Science*, 7(3): 215–222.

Godrey LD, Long RF. (2008). Dry Bean Weevils. UC IPM Pest Management Guidelines: Dry Beans. University of California Agriculture and Natural Resources Publication 3446, http://ipm.ucanr.edu/PMG/r52300411.html

Haines, C.P. (1991). Insect and Arachnids of tropical Stored Products*: Their biology and identification. A laboratory manual (second edition).* Storage Department, Tropical Development and Research Institute, London. p. 286

Ibrahim AA, Waba SY, Mohammed N, Mustapha SB (2016). Factors influencing the level of adoption of cowpea production technologies in Askira/Uba Local Government Area of Borno State, Nigeria. *International Academic Journal of Innovative Research,* 3(9):15-23.

International Institute of Tropical Agriculture (IITA). (2009). Crops. Obtainable at http:// www.iita.org. (Accessed date: January 13, 2010).

Ileke KD, Odeyemi OO, Ashamo MO (2013). Response of Cowpea Bruchid, *Callosobruchus maculatus* (Fabr.) [Coleoptera: Chrysomelidae] to Cheese Wood, *Alstonia boonei* De Wild Stem Bark oil extracted with different solvents. *Archives Phytopathol. Plant Protect*, 46(11): 1357 – 1370.

Jou-Nteufa C, Ceyhan E (2022). Determination of combining ability and heredity through diallel analysis method in F_2 populations of cowpea. *Selcuk Journal of Agriculture and Food Sciences,* 36(3): 299–311. https://doi.org/10.15316/SJAFS.2022.039

Jou-Nteufa C, Ceyhan E (2025). Genetic analysis of seed yield and some traits in cowpea using diallel analysis. *Turkish Journal of Agriculture and Forestry*, 48: 278-293. https://doi.org/10.55730/1300-011X.3180

Karadaş S, Ceyhan E (2023). Determination of grain yield and some agronomic characteristics of newly developed cowpea pure lines. In: *Proceedings* of *Sivas II. International Conference on Scientific and Innovation Research,* 15-17 September 2023; Sivas, Türkiye, pp. 932-941.

Mishili F, Temu A, Fulton J, Lowenberg-DeBoer J (2011). Consumer preferences as drivers of the common bean trade in Tanzania: a marketing perspective. *Journal of International Food and Agribusiness Marketing*, 23: 110–127.

Musa AK, Sulyman A (2014). Bioefficacy of Single and Mixed Applications of *Citrus paradisi* Maef and *Citrus aurantifolia* Swingle Peel Extracts against Seed Beetle, *Callosobruchus maculatus* (F.) (Coleoptera: Chrysomelidae). *Discourse Journal of Agriculture and Food Sciences*, 2(2): 85-90.

Mutungi C, Affognon HD, Njoroge AW, Manono J, Baributsa D, Murdock LL (2015). Triple-layer plastic bags protect dry common beans (*Phaseolus vulgaris*) against damage by *Acanthoscelides obtectus* (Coleoptera: Chrysomelidae) during storage. *Journal of Economic Entomology*, 108: 2479–2488.

Oliveira MRC, Corrêa AS, de Souza GA, Guedes RNC, de Oliveira LO (2013). Mesoamerican origin and pre- and post-Columbian expansions of the ranges of *Acanthoscelides obtectus* Say, a cosmopolitan insect pest of the common bean. *PLoS One*, 8: e70039.

Paul UV, Hilbeck A, Edwards PJ (2010). Preharvest infestation of beans (*Phaseolus vulgaris* L.) by *Acanthoscelides obtectus* Say (Coleoptera: Bruchidae) in relation to bean pod maturity and pod aperture. *International Journal of Pest Management*, 56: 41–50.

Sert H, Ceyhan E (2012). The effects of seed yield and some agricultural characters of different plant density on cowpea (*Vigna sinensis* (L.) Savi) in Hatay ecological conditions. *Selcuk Journal of Agriculture and Food Sciences*, 26(1): 34 -43.

Singh BB (2002). Recent genetic studies in cowpea. In: Fatokun C.A., Tarawali, S.A., Singh, B.B., Kormawa, P.M., Tamo, M. (eds). *Challenges and Opportunities for Enhancing Sustainable Cowpea Production.* International

Soares MA, Quintela ED, Mascarin GM, Arthurs SP (2015). Effect of temperature on the development and feeding behavior of *Acanthoscelides obtectus* (Chrysomelidae: Bruchinae) on dry bean (*Phaseolus vulgaris* L.). *Journal of Stored Products Research,* 61: 90–96.

Thakur DR (2012). Taxonomy, distribution and pest status of Indian biotypes of *Acanthoscelides obtectus* (Coleoptera: Chrysomelidae: Bruchinae)—a new record. *Pakistan Journal of Zoology*, 44: 189–195.

Tijjani AR, Nabinta RT, Muntaka M (2015). Adoption of innovative cowpea production practices in a rural area of Katsina State, Nigeria. *Journal of Agricultural and Crop Research,* 3(4):53-58.

Velten G, Rott AS, Cardona C, Dorn S (2007). Effects of a plant resistance protein on parasitism of the common bean bruchid *Acanthoscelides obtectus* (Coleoptera: Bruchidae) by its natural enemy *Dinarmus basalis* (Hymenoptera: Pteromalidae). *Biological Control,* 43: 78–84.

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Research Article

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Influence of Topography on Selected Pedological Properties of Soils Formed on Basement Complex in the Upland Areas of Rainforest Southwest Nigeria

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HIGHLIGHTS

- With the current reports of increased food insecurity in sub-Saharan Africa associated with improper and unconventional land use/ management practices among farmers that has adversely hindered food production, the study examined, classified and recommend various land management strategies that will improve soil health and enhance agricultural production in the studied area and in the areas with similar soil catena.

Abstract

The study investigates the impacts of topography on the formation and distribution of soils within the rainforest region of southwestern Nigeria, establishes their taxonomic classes (USDA & FAO/UNESCO), and recommends appropriate management practices that promote conservation. Five profile pits established and described at different physiographic positions were considered for this study. Soil samples were collected for physical, chemical, and mineralogical analysis, and rock samples were also collected for thin sectioning under a petrographic microscope. Fine sand fractions were separated into heavy and light minerals with bromoform. Correlation coefficients and simple regression analysis between the selected soil properties were calculated. The results revealed that the soils are derived from fined-grained biotite gneisses and schist; the clay content increased with increasing depth while the sand content decreased. Organic matter and available P content were relatively low, with values ranging from (0.2 – 1.35 %) and (1.29 – 5.40 ppm) respectively. The pH, exchangeable cations, and exchangeable acidity values fluctuate across the pedons. The crystalline oxides of Fe and Al were low, with no definite distribution pattern, indicating a highly weathered soil. The soils are predominantly ultisols and are placed in the ustults suborder (USDA), which was equated as Luvisols (FAO-UNESCO). The correlation between slope position and chemical properties showed that no singular property consistently showed the same significance level on the entire slope. For effective management of the soils, conservation management practices of economic importance, such as the usage of vegetal cover, should be adopted to assist in preventing rapid soil degradation across the landscape.

Keywords: Basement complex; taxonomic; mineralogy; bromoform; biotite gneisses

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1. Introduction

The weathering of rocks or other materials that have been deposited by water, gravity, or wind results in the formation of soil which qualities are determined by the interactions of climate and organisms acting on parent materials as conditioned by relief/ topography over time (Jenny 1941; Ukwuoma-Okolo 2021). One of the five primary soil-forming factors is topography which was described by Jenny (1941); Ben Mahmoud and Zurqani (2021) as an independent soil-forming factor which its contribution to soil formation can be considered on its own. Incidentally, most topographic information considers runoff and erosion to slope, which deals with soil removal and destruction. However, topography as a factor of soil formation on the other hand, influences radiation i.e., the amount of heat that penetrates the soil through sunlight and other objects that emits heat and the amount of water that penetrates the soil, resulting in leaching and the redistribution of elements and soil components (Hook and Burke 2000). It has a strong influence in determining the differences between soil types and their nutritional condition. Brandy and Weil (1999) define soil catena or topo sequence as soils developed as a result of topography in a specific area. They appear as a series of soils with close relationships to landscape positions that may differ in morphological, chemical, and fertility condition (Begna 2020). Topography is therefore essential in biogeochemical processes that influences important environmental, economic, and social activities (Griffiths et al. 2009; Bingqin et al. 2019).

Several studies reported changes in soil properties as a result of topographic local effects, which accounted for between 26 and 64% of overall variation in soil parameters and moisture Bockheim (2005), Cox et al. (2002), and Wilson et al. (2004). Moorman (1981) reported a strong relationship between topographic positions and soil genesis on basement complex in Southern Nigeria. However, topographic variability in crop production according to Dinaburga et al. (2010) is thus, a synthesis of soil attributes and factors influencing agricultural productivity via its effects on soil physicochemical characteristics, biomass production, incoming solar radiation, and precipitation, all of which have an impact on crop yield in the long run and hence confirms the facts that variation in crop production is a combined reflection of soil attributes and factors influencing agricultural productivity (Dinaburga *et* al. 2010). A soil therefore forms an integral part of the land surface and any variations in geomorphology which may influence hydrologic processes will affect the pedogenic processes through the effect of differential distribution of water, sediments and dissolved materials (Brunner *et* al. 2004 and Van der Meij et al. 2018).

The primary goal of agriculture is to produce enough food to feed the world's population, as well as enough raw materials to meet the demands of our industries and earn foreign exchange for the country through agricultural exports. In terms of food production, the soil thus represents everyone's hope. Given these facts, it becomes clear that the soil requires special care in terms of management in order to fulfil agricultural goals and ensure sustainability of the resources. Proper soil management, which entails managing a piece of land in such a way that it can be expected to yield at optimal level over time, is one of the pivots around which modern agriculture revolves.

Varied landforms may occur in a particular location with varied soils, and their capability/suitability potential for crop production may also vary, necessitating special land use management. This study is designed to aid in the provision of accurate information on the influence of topography in the study area's land quality. This will help soil users most especially farmers and the government to improve agricultural output in the study area and other areas with similar composition.

2. Materials and Methods

2.1. The Study Area

The studied area lies between latitudes 7° 32'N and 7° 33'N and longitudes 4° 39'E and 4° 40'E within southwestern Nigeria's schist belt (Rahaman 1988). The site is about 2.5 km away from Kajola village, a suburb of the Obafemi Awolowo University (O.A.U.) Teaching and Research Farm (T&R-F), located within the schist belt of southwestern Nigeria (Rahaman, 1988). The location is in the tropical rainforest agroecological zone, which has a hot, humid tropical climate with distinct dry and rainy seasons. The average annual rainfall is about 1527 mm, and the average monthly air temperature is around 31 °C, which is moderately high throughout the year, with a slight variation between the monthly mean minimum and maximum temperatures. The region also records the following average monthly data: humidity 73.8 %, and sunshine 6.6 hours. The wind speed was 114.6 km d^{-1} while potential evaporation was 4.36 mm d^{-1} (Meteorological data bank, T&R-F, O.A.U., Ile-Ife 2020). The site is an extensive farmland with many human activities, primarily farming and occasional overgrazing by animals, with some parts currently under fallow. According to Smyth and Montgomery (1962) and Rahaman (1988), the area is underlain by the Precambrian rocks which are part of what is collectively referred to as the basement complex of southwestern Nigeria. Previous research (Boesse and Ocan 1988) identified the underlying rock in the study region as fine-grained biotite gneisses and schist, which are part of the Precambrian basement complex rocks that are widespread in southwestern Nigeria.

2.2. Field Work (Sampling Procedure and Laboratory Analysis)

Soil sampling units was selected based on the physiographic positions of soils on the landscape. A slightly undulating toposequence that has relatively flat top was selected for the study with soil profile pits established at each physiographic position (Crest, upper slope, sedentary, hill-wash and valley bottom) as observed along the toposequence. Five soil profile pits were established along the toposequence, with a total of twenty four (24) soil samples collected from the identified genetic horizons. The repeated subsampling method was used to ensure that the samples taken from a given horizon for laboratory analyses were representative, starting with the lowest genetic horizon to the uppermost to avoid cross contamination from the horizons above (Smeck and Wilding 1980). Core samples were taken from each horizon for bulk density determination and rock samples were collected for thin section preparation and primary mineral identification.

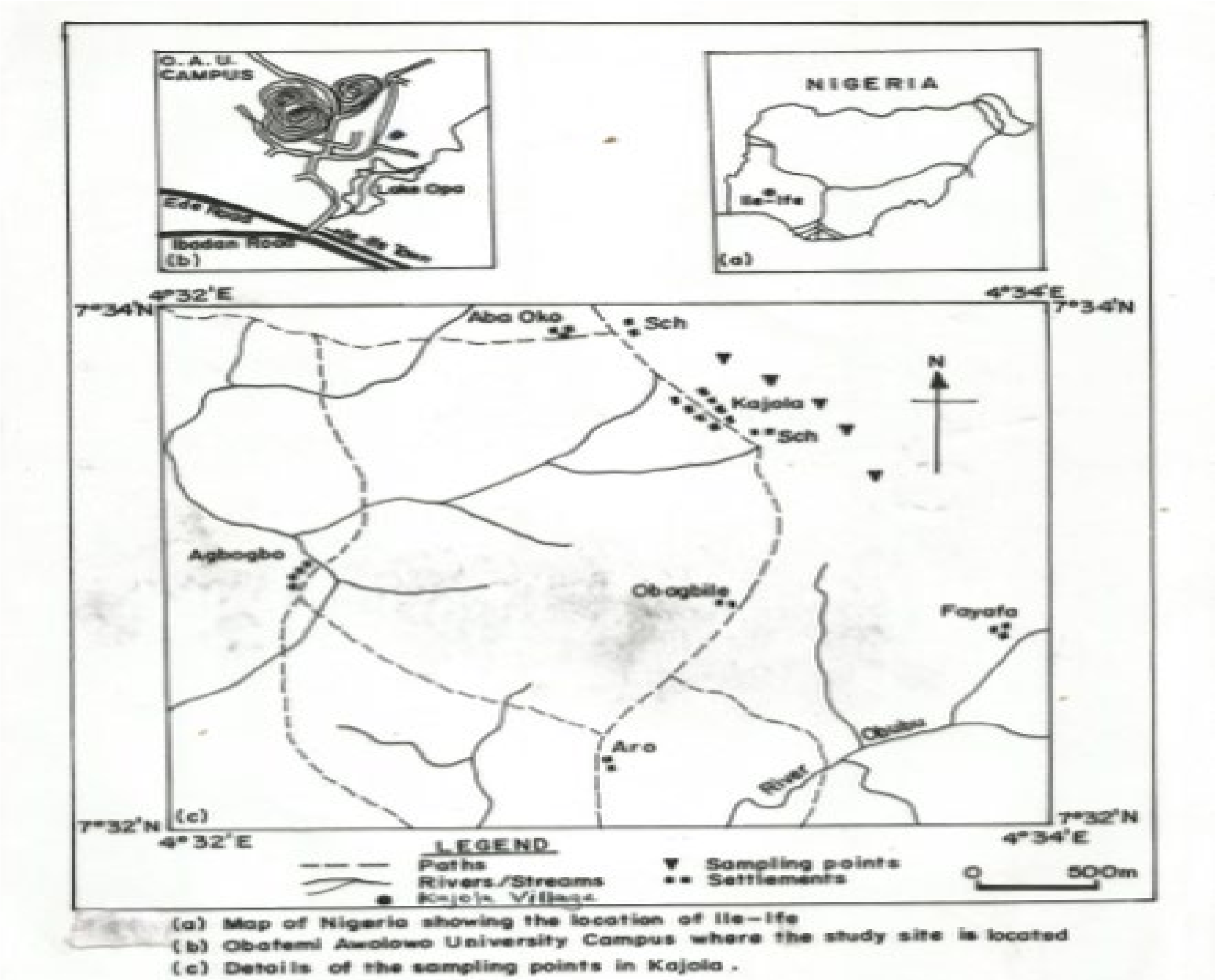

Figure 2. (**a**) Map of Nigeria showing the location of Ile Ife; (**b**) Obafemi Awolowo University Campus where the study site was located; (**c**) Details of the sampling site at Kajola.

The soil samples collected were air-dried, gently crushed with mortar and pestle and sieved with a 2 mm sieve to separate gravel content from the soil component. The less than 2 mm fraction was retained for physical, chemical and mineralogical analyses other than the bulk density determination.

2.3. Physical Parameters Assessed

The bulk density, gravel content, and particle-size distribution of the soil samples were examined. The percentage of soil retained by the 2 mm sieve, reported as a percentage of the total weight of the soil, was used to calculate the gravel content. The bulk density was determined by the core method as reported by (Blake and Hartge 1986), while the particle size distribution was evaluated by the modified Bouyoucos hydrometer method as reported by (Gee and Or,2002) using 5% w/v sodium hexametaphosphate (calgon) as the dispersing agent. Particle fractionation into very coarse sand (VCS), coarse sand (CS), medium sand (MS), fine sand (FS) and very fine sand (VFS) was carried out with the use of a set of sieves (1.0, 0.5, 0.25, 0.100 and 0.05 mm) representing 1000, 500, 250, 100 and 50 μm respectively arranged in decreasing order of sieve sizes as listed on the United State Department of Agriculture (USDA) particle size scale (Buol et al. 1997). Each of the sand fractions was weighed and expressed as a percentage of the total sand.

2.4. Chemical Parameters Assessed

The soil pH was determined in 1.0 M KCl (1:1 soil: solution ratio) with a glass electrode pH meter (Kent model 720) after equilibration for 30 minutes (Thomas, 1996). Exchangeable cations (Ca, Mg, K and Na) were extracted with 1.0 M ammonium acetate (NH_40AC) solution at pH 7.0 (Thomas and Throp 1985). Calcium (Ca^{2+}), sodium (Na^+), and potassium (K^+) ions in the extract were determined with the use of flame photometer (Gallenkamp Model FH 500), while magnesium (Mg^{2+}) ion in the extract was determined by titration. The exchangeable acidity was determined by extraction with 1.0 M KCl solution and titrated with NaOH and HCl solutions to measure total acidity (Al^{3+} and H^+) concentrations, respectively as reported by (Bertsch and Bloom, 1996). The available phosphorous was determined by Bray No. 1 method (Kuo, 1996). The organic carbon was determined by the Walkley Black method (Allison, 1965) as reported by (Darrell *et al.,* 1994). The effective cation exchange capacity (ECEC) was calculated as the summation of exchangeable cations and exchangeable Al (Sumner and Miller 1996). The free iron (Fe), aluminum (Al) and manganese (Mn) were determined by the dithionite citrate bicarbonate (DCB) method of Mehra and Jackson (1960). The ions extracted were determined with the use of atomic absorption spectrophotometer (AAS). The total elemental analysis was carried out by digesting the powdered soil samples with aqua regia and hydrofluoric acid under a fume chamber (Bernas 1968; Jackson 1958). Elements in the digest were determined by the atomic absorption spectrometer (AAS).

2.5. Mineralogical Analyses

2.5.1. Thin sectioning of the rock sample

A rectangular block small size of 3mm in diameter was cut from the rock sample collected from the field with a diamond saw, one side of this block was ground and polished to produce a flat, smooth surface free of scratches or imperfections, the block was carefully cleaned and cemented to a clean microscope slide with Canada balsam and a cover glass was cemented in place to produce the thin section (Cady et al. 1986). At this thickness (3mm diameter), it has been established that rocks behave like a transparent medium allowing the passage of light (Kerr 1977). Hence, the study of optical properties of mineral components of the rock was enhanced. This thin section of the rock samples was produced and mounted on a glass slide for mineralogical identification under a petrographic microscope as described by (Simpson 1986). Optical observations were made in both plane polarized light and cross nicol (Adetayo et al. 2013).

2.5.2. Sand mineralogy

The fine sand fraction was separated into light and heavy mineral fractions using bromoform (s.g. = 2.89). The light fraction in the sample stayed afloat while the heavy mineral fractions went down the separating funnel under the influence of gravity. The light and heavy mineral fractions were separated and the mineral constituents in each separate was examined with the use of a drop of Canada balsam (R.I. = 1.54) placed on a glass slide and enough sub-sample was taken with a micro spatula to ensure uniform coverage of an area about 22-mm square on a glass slide. This was then covered with a cover slip. Identification of mineral grains was made according to their optical properties under plain polarized light and in crossed polar. Mineral properties such as colour, relief, pleichroism, birefrigence, interference colour and extinction were used for the identification of the minerals. The relative amount of individual mineral present was determined by counting with the use of cross wire method (Wilding and Drees 1983).

Correlation coefficients and simple regression analysis between the selected soil properties were calculated to determine the relationships between the soil parameters assessed and the influence of topography on its distribution across the slope. All statistical analyses were carried out using SAS 9.1 version (2002-2004) software programme.

3. Results

3.1. Physical Properties of the Soils

Table 1 shows the particle size distribution and bulk density data of the soils along the toposequence under study. The gravel content varied from 8 to 68%, with pedons from the summit, upper, and middle slopes having relatively high values of 22 to 68%, while the lower slope and valley bottom pedons have lower values, 8 to 32%, probably owing to sorting. The gravel content generally increased from the A-horizon to the B-horizon and then decreased significantly from the B to the C-horizon, except at the lithologic break. This increase in gravel content from A to B horizon is a characteristic of the upland pedons (Fasina et al. 2005), which is as a result of several pedogenic and geomorphic processes, among which are eluviation, selective particle removal, the nature of the parent material, erosional processes, and landscape positioning (McAuliffe et al. 2018). These factors collectively shape the vertical distribution of soil particles, leading to the observed pattern of increasing gravel content with depth in the study area. However, in pedons at the lower slope position, there were no particular patterns of gravel distribution. This gravel accumulation was a characteristic property of soils formed in the upland portion of the landscapes derived from the granitic gneiss metamorphic rock complex of central western Nigeria (Smyth and Montgomery 1962; Okusami and Oyediran 1985). These have not obstruct root proliferation since roots are found beyond the gravel horizons.

The soil texture varied from sandy loam to sandy clay loam for surface horizons except in Pedon 05 which has clay texture. The B and C-horizons have clay loam texture except in Pedons 03 and 04 that were more clayey in the B and BC horizons. The sand content ranged from 29 to 67% and decreased with increasing depth except at certain depths where the BC-horizon contained more of sand, as encountered in Pedons 03 and 04. The silt content ranged from 11 to 25%, although the value fluctuated within all the pedons with increasing depth. The soils have low to moderate silt content at the surface irrespective of their location on the topography. A characteristic that distinguished the soils of granitic-gneiss rock complex origin from other sandy soils of southwestern Nigeria (Fasina 2001 and 2002; Fasina et al. 2007). The clay values ranged from 18 to 59%. The clay content increased generally with increasing depth to a maximum (probably due to illuviation/ eluviation interplay or possibly clay migration) and then decreased in the BC horizons. Similar trend was observed by Ojanuga (1978) in soils of Ife and Ondo areas of southwestern Nigeria. Generally, soils in the middle and lower slope positions have higher clay content than those that occupy the valley bottom position. For Pedons 02, 03, and 04, the particle size distribution of the sub-soil horizon of the soils suggests that the B-horizons were influenced more by eluviation – illuviation processes. The high clay content in the deeper horizons of some of the soils occasioned by clay dispersion, translocation, and accumulation, coupled with some morphological properties such as the colour, texture, consistence and plasticity in the profile description (Fasina et al. 2005) formed the basis for the recognition of argillic horizons in some of the soils. The lower clay content in the surface horizons could be due to the sorting of soil materials by biological and/or agricultural activities, clay migration or surface erosion by run-off or a combination of these (Ojanuga 1978; Ogban et al. 1999).

The bulk density values obtained ranged from 0.74 g cm^{-3} in the Ap horizons to 1.73 g cm^{-3} in the Bt horizons. The higher values in surface soils (e.g. pedons 02 and 03) are due to compaction from grazing animals and occasional movement of machines such as tractors (Kumar et al. 2018). Usually, soils with low bulk density are known to be associated with high total porosity, while root penetration becomes a problem when bulk density exceeds 1.6 g cm^{-3} (Payne 1988). Generally, the bulk density value increased with increasing

Table 1. Physical properties of the soils on the toposequence that was studied.

Horizon	Depth (cm)	>2000mm (% of the whole soil)	Very coarse sand (1000-2000µm)	Coarse sand (500-1000µm)	Medium Sand (250-500µm)	Fine Sand (50-100µm)	Very fine sand (0.05-0.50 µm)	Total Sand	Silt	Clay	Bulk density g/cm^3	Textural class
		%										
Profile 01 Egbeda series												
Ap	0-18	35	10	13	10	9	7	49	21	30	1.06	Sandy loam
AB	18-24	61	10	12	10	8	7	47	25	28	1.54	Sandy clay loam
B	24-51	68	7	8	7	5	4	31	11	58	1.62	Sandy clay
BC	51-70	50	7	7	6	5	4	29	13	58	1.59	Sandy clay
Profile 02 Olorunda series												
Ap	0-18	41	10	13	12	10	10	55	17	28	1.01	Sandy clay loam
AB	18-28	68	10	12	11	10	8	51	11	38	1.57	Sandy clay loam
B21	28-72	64	10	8	7	7	6	39	13	48	1.63	sandy clay
B22	72-132	56	11	4	7	7	5	33	13	54	1.68	sandy clay
BC1	132-185	33	9	7	7	7	6	35	17	48	1.73	Sandy clay
BC2	185-210	22	9	9	9	8	8	43	15	42	1.48	Sandy clay
Profile 03 Makun series												
Ap	0-18	43	12	13	11	11	9	55	17	28	1.34	Sandy clay loam
BA	18-33	42	11	12	10	10	8	51	15	34	1.42	Clay loam
B21	33-65	39	12	8	7	4	3	34	13	53	1.45	Sandy clay
B22	65-120	47	12	6	6	4	2	30	11	59	1.65	Sandy clay
BC	120-200	23	11	8	10	5	5	39	15	46	1.40	Sandy clay
Profile 04 Oba series												
Ap	0-20	8	11	15	13	11	8	57	15	28	1.54	Sandy clay loam
BA	20-40	32	10	12	7	7	2	39	13	48	1.48	Sandy clay loam
B1	40-71	25	8	11	5	5	3	31	11	58	1.30	Sandy Clay
2BtC1	71-115	27	8	11	4	5	3	31	13	56	1.71	Sandy Clay
2BC2	115-170	15	10	13	7	7	2	39	13	48	1.40	Sandy clay
Profile 05 Jago series												
Ap	0-18	26	16	15	14	13	10	67	15	18	0.74	Sandy clay
AB	18-40	19	14	13	13	13	12	65	13	22	1.21	Sandy clay
B	40-60	18	14	17	15	12	10	67	11	22	1.31	Sandy clay
Btg	60-75	24	13	14	13	13	12	65	15	20	1.13	Sandy clay

depth to a maximum and then declined with increasing soil depth. The exception to this trend was observed in Pedon 04, where their values fluctuated. However, the higher values at depth have not hindered plant root penetration, as evidenced by the deep rooting of plants into greater depth in Pedon 02.

3.2. Chemical Properties of the Soils

Table 2 shows the chemical properties of the pedons studied. The soils fall within the neutral to very strongly acid class (Soil Survey Staff, 2003), with pH (H_2O) values ranging from 5.6 to 7.0. The pH decreased with increasing soil depth except in Pedon 03. The pH (1M KCl) ranged from 4.4 to 5.7. The value also decreased as the soil depth increased, except in Pedon 03, where no definite pattern was observed. Generally, the surface horizons of the pedons were medium to slightly acid (pH 5.2 - 5.7), while B and C-horizon were strong to very strong acid with pH values ranging from 4.4 - 5.7. The acid nature of the soil can be ascribed to high rate of leaching of bases which is prevalent in the humid tropics, and the acidic nature of the parent rock (granite-gneiss) (Ojetade et al. 2022). The higher pH values observed at the soil surface horizons might be due to liming effect of bush burning and bio cycling of nutrients (Fasina et al. 2005). The pH in 1M KCl was lower than the pH in water (H_20), thus the difference in soil pH values between the pH in KCl and H_20 (as expressed by $\Delta pH = pH (KCl) - pH (H_20)$) were all negative ranging from -0.9 to -1.5. This suggests the dominance of silicate clay minerals over oxides (Van Raij and Michael 1972).

Generally, there was higher accumulation of bases in the surface horizons 6.59 - 12.57 cmol(+)kg^{-1} of the soil, and the total exchangeable bases decreased with soil depth except in some cases owing to nutrient biocycling (Ajiboye and Ogunwale 2010), and could also be due to differential weathering that had taken place or as a result of plant uptake and leaching losses. The exchangeable sites of the soils studied were dominated by exchangeable calcium and magnesium a common occurrence in most tropical soils. Exchangeable sodium (Na^+) and potassium (K^+) are low with values ranging from 0.08 to 0.26 cmol(+)kg^{-1} and 0.15 to 0.30 cmol(+)kg^{-1} soil respectively. These low values indicated that the soils under investigation developed from materials that are either low in K^+ and Na^+ content or have been exhausted by plant uptake or leaching due to their mobility within the soil. The higher values obtained at the surface horizon of the pedons could be attributed to higher organic matter content (Ano, 1991). However, the values fluctuated irregularly down the soil profile.

Exchangeable acidity values (Al^{3+} and H^+) ranged from 0.3 to 1.0 cmol (+) kg^{-1} soil. All the pedons examined showed little variation in the exchangeable acidity and the values were almost uniform with soil depth. Exchangeable Al^{3+} accounted for a greater percentage of the total acidity. Effective cations exchange capacity (ECEC) was generally low with values ranging from 3.73 to 14.26 cmol (+) kg^{-1} soil. There were higher values in the surface horizons of all the soils examined than in the sub-soil, probably due to the influence of organic carbon on the exchange sites of the soils. However, in those profiles where higher values were noticed in the sub-soil as in Pedons 02 (B22) and 03 (BC) with more of clay content, this could be due to the process of pedoturbation either by fauna or flora. Kadeba and Benjaminsen (1976) observed that low ECEC values were consistent with low organic carbon content of soils, especially in the B-horizons, and probably with the kaolinitic nature of the soils. The ECEC values decreased with increasing soil depth in all the pedons examined.

The organic matter content of the surface horizons of the pedons ranged from 1.54 to 2.55% and decreased with increasing soil depth. The sub-soil horizons were generally lower in organic carbon than the surface horizons of all the pedons examined, probably due to the fact that the surface horizons are the points where decomposition and humification of organic materials take place. The organic matter content of the entire soils studied was generally low, mostly less than 2%, except in the surface horizon of Pedon 01. The low organic matter obtained may be partly due to the effect of high temperature and relative humidity, which favor rapid mineralization of organic matter (Fasina et al. 2005), the degradative effect of cultivation, and other land use and management activities. In all the pedons examined, the exchangeable bases, ECEC, percent base saturation and organic matter contents were slightly higher in the surface horizons than in the sub-soils in general. Probable reason is that the surface horizons, although the most exposed to leaching and runoff, are indeed continuously recharged by phytocycling (Amusan and Ashaye 1991).

Available phosphorous (P) contents of the soils varied from 2.6 to 11.2 ppm in all the horizons in the profiles with the highest values at the surface horizons, The relatively high concentration of the available P and organic carbon in the surface horizons may imply significant organic or biocycled P in the soils and also an indication that organic matter contributes significantly to the available phosphorus in these soils. These values i.e. available phosphorous (2.6 to 11.2 ppm) and organic matter content (1.54 to 2.55%) were rated low to medium depending on the soil horizons (Esu et al. 1987). The available P values are considered low at some horizons as they were below or only slightly above the 10 ppm critical limit recommended for most commonly cultivated crops in the area (Uponi and Adeoye 2000; Aduayi et al. 2002; Obigbesan 2009). The values generally decreased with increasing depth, though the pattern is irregular in all the pedons that were examined. The low value of available P might be due to the fixation of phosphorus by iron and aluminum sesquioxides under well drained and acidic conditions of the soils (Onyekwere et al. 2001; Uzoho and Oti 2004). Further, Jubrin et al. (2000) noted that deficiency of P may occur in soils due to the strong adsorption of this nutrient by the soil colloids. Pedons 04 and 05 exhibit an unusually high concentration of available P in the sub-surface horizon, 8.4 and 8.9 ppm, respectively. This could probably be due to the effect of farming activities, especially decomposed cocoa pod residue, because the pedons are located within a cocoa farm or deposition of P in the valley bottom soils of the toposequence through erosion.

Profile distribution of crystalline Fe and Al oxides values and weathering ratios shows the percentage of dithionite extractable Fe and Al in all the soil profiles examined. The value of DCB- Fe (Fe_d) ranged from 1.46 to 3.44% and did not follow any pattern as the soil depth increased. In all the soil profiles examined, the highest Fe_d value occurred in the B-horizon except in Profiles 3 and 5, where the highest value of Fe_d was observed at the lowest horizon. The quantity of Al_d, that is Al substitution in Fe oxides and organic matter-bound Al (Parfitt and Childs 1988) in the soil was lower than Fe_d and ranged from 0.09 to 0.70%. Its distribution pattern was like that of the Fe_d distribution, which does not follow any pattern as the soil depth increased.

The Fe_d/Clay ratios were calculated to determine whether the Fe_d was associated with the clay fraction (Blume and Schwertmann 1969; Rebertus and Buol 1985). This ratio ranged from 0.03 to 0.11 and did not follow any pattern as the soil depth increased. The same pattern was observed in the Al_d/clay ratio with values ranging from 0.003 to 0.022. There is no obvious regular pattern in the distribution of the free Fe in the profiles.

Generally, it could not be established that all the B-horizons have higher Fe_d. This may be since iron movement is partially independent of clay movement. A similar observation was reported by Ogunsola et al. (1989) in soils overlying limestone areas in Nigeria. The means of the Fe_d/Clay ratios were low and were less than unity at the top sequence under study, indicating that basement complex topo sequence could be highly weathered (Enya et al. 2011). There is high negative significant correlation between Fe_d/clay and clay content in the soils studied (r= -0.82) (Table 9). At the same time, it was not significant when Fe_d/clay was correlated with silt (r=0.31), indicating the probable nonexistence of co-migration of clay and silt with depth, respectively (Enya et al. 2011).

3.3. Total Elemental Analysis of the Soil Samples and Saprolites

The total element analytical results are presented in Table 3. The concentration of Fe and Cu ranged from 100.5 to 523.01 ppm and 307 to 528 ppm, respectively. The highest values were observed at the surface of all the pedons examined. The values, especially Fe, do not follow a particular trend except in Profile 02, where it seems to reduce with increasing soil depth. In Profile 01, the values were reduced with increasing depth except at the last horizon, where the value suddenly increased, which may be a result of illuviation, parent material composition, redox processes, lateralization, capillary rise, or residual accumulation of Fe minerals. The dominant process depends on factors like climate, drainage, and soil formation history. In other Profiles 03, 04, and 05, the values were erratic. This could be due to the mineral composition of the underlying rock and/ or the transported materials, uptake of essential nutrients by plants, leaching of exchangeable cations through heavy rainfall or by erosion, or a combination of these factors. The higher values observed could be responsible for the occlusion of some of the plant's nutrients, like phosphorus, thereby making it unavailable for plant uptake, and could also be responsible for the lower exchangeable cations recorded from the soil under examination. However, this can be taken care of with an adequate soil management system. Apart from these two elements, the soil is presumed to have an acceptable level of ZnO (0.11- 3.00 ppm), CaO (2.00- 19.00 ppm), MgO (0.73- 14.06 ppm) and MnO_2 (3.90- 14.66 ppm) for plant growth. However, human activities also influence the composition of the minerals such as copper and calcium in the soil surface as some of the profile pits are located within cocoa plantations, and the fungicides that are commonly used in disease control are rich in Cu and Ca, hence there higher content compared with the rest.

Table 2. Chemical properties of the soil studied.

Horizon	**Depth (cm)**	**pH (H_20)**	**pH KCl**	**ΔpH**	**Exchangeable Bases** (CmolKg$^-$)				**Exchangeable Acidity** (CmolKg$^-$)		**Sum of Bases**	**ECEC** (CmolKg$^-$)	**Base sat. (%)**	**Al. Sat. (%)**	**O.M. (%)**	**Avail. P. (ppm)**	**Crystalline oxide (%)**		**Fe_d/Clay**	**Al_d/Clay**
					Ca^{2+}	**Mg^{2+}**	**Na^+**	**K^+**	**Al^{3+}**	**H^+**							**$Fe_{(d)}$**	**$Al_{(d)}$**		
Profile 01 Egbeda series																				
Ap	0-18	6.9	6.0	-0.9	7.2	4.86	0.21	0.30	0.4	0.2	12.57	12.97	97	3	2.55	11.2	2.08	0.58	0.07	0.019
AB	18-24	6.8	5.4	-1.4	6.6	4.86	0.26	0.26	0.2	0.3	11.98	12.18	98	2	1.61	7.4	2.14	0.61	0.08	0.022
B	24-51	6.5	5.0	-1.5	6.7	4.10	0.25	0.30	0.3	0.3	11.30	11.60	97	3	1.21	3.4	1.95	0.70	0.03	0.012
BC	51-70	6.0	4.6	-1.4	5.8	3.20	0.20	0.24	0.7	0.3	9.44	10.10	93	7	1.07	3.2	1.70	0.68	0.03	0.012
Profile 02 Olorunda series																				
Ap	0-18	6.5	5.3	-1.2	5.3	4.86	0.19	0.24	0.4	0.2	10.58	10.98	96	4	1.68	6.3	1.92	0.58	0.07	0.021
AB	18-28	6.4	5.0	-1.4	4.9	4.05	0.19	0.22	0.4	0.3	9.35	9.75	96	4	1.14	8.2	2.10	0.51	0.06	0.013
B21	28-72	6.2	5.0	-1.2	5.5	1.62	0.21	0.28	0.1	0.4	7.69	7.79	99	1	0.87	4.1	1.46	0.44	0.03	0.009
B22	72-132	6.1	5.0	-1.1	5.3	5.67	0.19	0.26	0.4	0.3	11.42	11.82	97	3	0.67	3.3	3.44	0.60	0.07	0.011
BC1	132-185	5.9	4.9	-1.0	5.3	4.05	0.20	0.26	0.1	0.3	9.81	9.91	99	1	0.60	3.0	2.40	0.51	0.05	0.011
BC2	185-210	5.6	4.8	-0.8	5.0	4.86	0.17	0.26	0.2	0.2	10.29	10.49	98	2	0.07	2.6	1.86	0.40	0.04	0.011
Profile 03 Makun series																				
Ap	0-18	6.8	5.6	-1.2	4.0	7.29	0.14	0.22	0.4	0.3	11.64	12.04	97	3	1.54	8.4	2.08	0.32	0.07	0.011
BA	18-33	6.7	5.5	-1.2	4.9	4.05	0.19	0.24	0.3	0.3	9.38	9.68	97	3	0.94	10.5	2.85	0.63	0.08	0.019
B21	33-65	6.6	5.5	-1.1	5.3	1.62	0.21	0.30	0.2	0.2	7.44	7.64	97	3	0.94	7.0	2.66	0.37	0.05	0.007
B22	65-120	6.4	5.5	-0.9	4.1	6.48	0.14	0.20	0.2	0.3	10.91	11.11	98	2	0.87	5.8	3.12	0.28	0.05	0.005
BC	120-200	6.7	5.7	-1.0	3.1	10.53	0.11	0.22	0.3	0.2	13.96	14.26	95	2	0.40	3.2	3.20	0.24	0.07	0.005
Profile 04 Oba series																				
Ap	0-20	6.4	5.2	-1.2	4.7	4.05	0.23	0.24	0.2	9.22	0.9	9.92	93	7	1.74	7.7	3.07	0.09	0.11	0.003
BA	20-40	6.2	4.8	-1.4	3.2	5.67	0.12	0.22	0.2	9.21	0.3	9.31	99	1	0.93	8.4	3.20	0.19	0.07	0.004
BC1	40-71	6.0	4.8	-1.2	2.2	5.67	0.08	0.24	0.2	8.19	0.3	8.29	99	1	0.67	5.8	3.07	0.26	0.05	0.004
2BtC1	71-115	5.8	4.6	-1.2	2.1	4.05	0.10	0.24	0.3	6.49	0.4	6.59	99	2	0.60	6.2	2.02	0.37	0.04	0.007
2BC2	115-170	5.6	4.4	-1.2	2.8	3.24	0.16	0.26	0.2	6.46	0.3	6.56	99	2	0.40	4.5	2.28	0.35	0.05	0.007
Profile 05 Jago series																				
Ap	0-18	7.0	6.7	-1.3	1.5	4.86	0.08	0.15	0.2	6.59	0.6	6.99	94	6	1.74	6.9	1.84	0.09	0.10	0.005
AB	18-40	6.6	5.2	-1.4	1.9	4.86	0.08	0.24	0.3	7.08	0.6	7.38	96	4	0.74	8.9	1.76	0.22	0.08	0.010
B	40-60	6.5	5.0	-1.5	0.6	2.43	0.08	0.22	0.2	3.33	0.6	3.73	89	11	0.40	5.4	2.01	0.24	0.09	0.010
Btg	60-75	6.3	5.0	-1.3	1.5	2.43	0.08	0.21	0.2	4.22	0.6	4.62	91	9	0.13	5.2	2.02	0.09	0.10	0.005

Table 3. Total elemental analysis of the soils studied.

Horizon	Depth (cm)	(Fe_2O_3)	(CaO)	(MgO)	(MnO_2)	(ZnO)	(CuO)
		ppm					
Profile 01 Egbeda series							
Ap	0-8	396.12	19.00	14.06	12.60	1.80	393.05
AB	8-24	296.48	17.00	12.32	8.70	3.00	436.06
B	24-51	292.02	3.00	5.34	9.66	1.20	517.27
BC	51-70	342.02	14.00	5.68	10.21	3.00	500.61
R	Above 70	396.95	10.00	1.19	13.11	ND	474.38
Profile 02 Olorunda series							
Ap	0-18	523.01	10.00	4.71	14.66	0.60	399.82
AB	18-28	506.81	14.00	4.41	16.49	1.70	528.1
B21	28-72	298.59	8.00	2.65	9.68	1.30	456.88
B22	72-132	274.69	6.00	2.44	7.64	0.40	448.77
BC1	132-185	179.70	1.00	1.36	6.20	0.30	457.3
BC2	185-210	100.50	1.00	2.50	3.90	1.20	449.8
Profile 03 Makun series							
Ap	0-20	322.64	10.00	3.00	12.11	2.00	333.15
BA	20-40	329.81	8.00	2.22	11.90	3.00	432.73
B21	40-71	288.22	6.00	2.32	9.21	1.10	422.73
B22	71-115	216.43	4.00	2.40	7.08	1.50	516.86
BC	115-170	124.00	2.00	1.21	5.33	ND	452.3
Profile 04 Oba series							
Ap	0-20	338.26	19.00	5.78	11.89	0.50	383.58
BA	20-40	332.22	11.00	4.21	10.24	0.22	398.34
B1	40-71	298.72	11.00	3.00	9.24	0.32	444.24
2BtC1	71-115	226.12	5.00	1.22	10.1	0.11	498.22
2BC2	115-170	308.92	1.00	2.08	10.98	ND	504.78
Profile 05 Jago series							
Ap	0-18	257.28	18.00	5.45	9.56	ND	332.36
AB	18-40	212.44	18.00	5.55	6.22	ND	311.22
B	40-60	135.40	18.00	6.29	4.39	0.80	307.37
Btg	60-75	190.60	2.00	0.73	6.11	0.20	459.80

3.4. Bedrock of the Soils Studied

Thin section of the rock sample was prepared for examination under the petrographic microscope to determine the mineralogical composition of the rock. Viewing the plates under the microscope, garnets and biotite were the major minerals observed under the plain polarized light (PPL), while quartz and plagioclase (feldspar) were the major elements observed under the cross polar (CP). These minerals were also accompanied by others identified as follow: staurolite, andalusite, kyanite, graphite, hornblende, tourmaline, rutile, ilmenite, haematite and pyrites, although in smaller quantities. However, considering the mineralogical composition of the fine sand fraction of the soils studied with the rock samples thin section under petrographic examination, it can actually be concluded that the soils along the toposequence studied were derived from the weathering of the fine grained biotite gneisses and mica schist. It had earlier been reported that mica schist was the common rock mineral found in the area (Smyth and Montgomery 1962).

3.5. Fine Sand Mineralogy

Table 4 shows the results of the mineralogical analysis of the soil's fine sand fraction (100 – 250 μm). Heavy liquid analysis showed that the light mineral fraction (specific gravity < 2.89) ranged from 23 to 84% while 16 to 77% constituted the proportion of the heavy mineral (s.g. > 2.89) fraction in the fine sand fraction. In majority of the pedons examined, the percentage of light mineral was greater than that of the heavy mineral fraction. This can be attributed to several pedogenic, geologic, and environmental factors, among which are parent material composition, intense weathering, selective transport, and deposition, leaching, and soil texture characteristics**.** These factors collectively influence the mineralogical composition of the soil, favoring the accumulation of more resistant light minerals over time. However, the composition did not follow a specific trend in all the soil profiles. In some, the percentages of both light and heavy minerals increased with an increase in soil depth, while in some, the values fluctuated with soil depth. The dominant light minerals found in the fine sand fraction were quartz, with values ranging from 86 to 98%, and feldspar (plagioclase), with values ranging from 02 to 04%, as shown in Table 5. The dominant heavy minerals were opaque in all the pedons observed, and they were about 62 to 92%.

Table 4. Percentage light and heavy mineral composition of the fine sand fraction of the pedons.

Horizon	Depth (cm)	Light mineral (s.g <2.89)	Heavy mineral (s.g > 2.89)
		← %	→
		Profile 01 Egbeda series	
Ap	0-18	48	52
AB	18-24	51	49
B	24-51	53	47
BC	51-70	55	45
		Profile 02 Olorunda series	
Ap	0-18	58	42
AB	18-28	59	41
B21	28-72	63	37
B22	72-132	67	33
BC1	132-185	71	29
BC2	185-210	79	21
		Profile 03 Makun series	
Ap	0-18	28	72
BA	18-33	32	68
B21	33-65	29	71
B22	65-120	23	77
BC	120-200	41	59
		Profile 04 Oba series	
Ap	0-20	64	36
BA	20-40	66	34
B1	40-71	72	28
2BtC1	71-115	68	32
2BC2	115-170	67	33
		Profile 05 Jago series	
Ap	0-18	79	21
AB	18-40	77	23
B	40-60	82	18
Btg	60-75	84	16

Brown staurolite, pinkish andalusite and gray kyanite were the predominant heavy minerals. However, garnet, rutile, ilmenite, hematite, zircon and apatite also occurred in some of the pedons examined in varying proportion considering the richness of the parent rock in ferromagnesian minerals.

Table 5. Percentage of light and heavy mineral composition of the fine sand fraction of the soils studied.

Horizon	Depth (cm)	Quartz	Feldsper	Opaque	Staurolite	Andalusite	Kyanite	Garnet	Rutile	Zircon	Ilmenite	Apatite	Hematite
		Light mineral composition		**Heavy mineral composition**									
							Profile 01 Egbeda series						
Ap	0-18	94	06	88	03	02	02	-	01	-	01	02	01
AB	18-24	96	04	92	01	02	01	01	-	-	02	01	-
B	24-51	95	05	78	05	05	02	02	02	01	-	-	05
BC	51-70	96	04	74	10	-	05	-	04	02	02	03	-
							Profile 02 Olorunda series						
Ap	0-18	98	02	62	05	10	-	05	05	08	-	05	-
AB	18-28	98	02	88	02	-	-	02	02	04	01	-	01
B21	28-72	98	02	84	-	04	02	02	-	02	03	01	02
B22	72-132	96	04	66	10	02	03	05	02	02	-	05	05
BC1	132-185	94	06	78	05	05	05	-	01	-	02	02	01
BC2	185-210	88	12	75	05	-	05	03	02	05	-	-	05
							Profile 03 Makun series						
Ap	0-18	86	14	84	04	02	-	-	02	03	01	-	04
BA	18-33	88	12	82	05	02	04	02	02	01	-	01	01
B21	33-65	88	12	78	05	02	03	05	-	-	03	03	01
B22	65-120	90	10	76	02	02	03	02	02	04	02	05	-
BC	120-200	90	10	88	-	-	04	-	-	-	04	-	04
							Profile 04 Oba series						
Ap	0-20	98	02	88	02	02	-	-	02	02	-	02	02
BA	20-40	96	04	92	-	-	02	-	02	01	01	01	01
B1	40-71	92	08	90	04	02	-	-	01	-	02	01	-
2BtC1	71-115	98	02	64	05	05	04	02	-	05	04	05	06
2BC2	115-170	97	03	76	04	05	-	01	04	02	-	05	03
							Profile 05 Jago series						
Ap	0-18	98	02	78	04	04	08	-	02	01	01	-	02
AB	18-40	98	02	78	03	05	08	-	02	01	-	01	02
B	40-60	98	02	88	-	02	02	-	02	02	-	02	02
Btg	60-75	98	02	90	01	-	02	04	-	01	02	-	-

The occurrence of staurolite, zircon and rutile in the heavy mineral fraction and the abundance of quartz in the light mineral fraction of the soils examined pointed to the possibility that the soils developed from metamorphosed argillaceous sedimentary rocks (staurolite-bearing schists). The presence of feldspars which are weatherable minerals in the soils indicated that the soils have some inherent nutrient reserve, while the presence of resistant minerals like zircon and staurolite attested to a highly weathered environment that had concentrated these resistant minerals into appreciable proportions within the soil (Adegbite and Ogunwale 1994).

3.6. Statistical Analyses

The results of correlation analysis of some selected soil physical and chemical properties are shown in the correlation matrix (Table 6). Clay content shows a significance positive correlation with pH (H_2O), Na and H^+ (r= 0.99, 0.93 and 0.96) which suggests that the contribution of clay to this parameters in the soils is high. Also, a positive correlation was observed between the clay content and the bulk density of the soils examined. This is an evidence of the prominent role that clay content plays in the soil bulk density compared with other parameters listed above.

The soils' pH (KCl) significantly correlated with the sum of bases (r= 0.82) and ECEC (r= 0.92). This indicated that the soil pH affected the cation exchange in the soils examined i.e. the higher the pH to a certain level, the greater the amount of bases exchanged within the soil solumn and the higher the nutrients available for plant uptake. The ECEC correlated positively and significantly at all levels with organic carbon (r= 0.88) and available P (r= 0.99). This observation is similar to the report of Kadeba and Benjaminsen (1976) that between 56 and 83% of the variation in CEC of tropical soils is accounted for by the organic matter content of the soil. Further, organic carbon content showed a good correlation with the available P (r= 0.83). This indicated the importance of organic matter to the process of ion exchange in soils, and shows that organic P is a major source of P in the soils.

3.6. Taxonomic Classification

All the pedons observed showed increasing trend in clay content with soil depth to a certain level, a kind of trend that was indicative of argillic horizon. Low level of fertility as observed from the organic matter content and other soil mineral composition to the extent that they cannot be used to grow crops economically unless fertilizers are used to supply nutrients (Soil Survey Staff 2003). These are the two most important differentiating characteristics of the Ultisols. The pedons are mineral soils with ochric epipedon, low in organic matter, high in colour values and chromas. The soils are dry for more than 90 cumulative days but less than 180. The upland soils of southwestern Nigeria is primarily under ustic moisture regime (Periaswamy and Ashaye 1982), therefore, the soils are in Ustults suborder. Pedon 05 qualifies as Aqults because of the hydromorphic properties right from the soil surface and the gleyed subsurface horizons. The presence of Kandic horizons are established in most pedons because they meet the following requirements: coarse textured surface horizon over vertically continuous sub-surface horizons; ECEC values within the sub-surface B-horizons that are less than 12cmol (+)/kgclay; a regular decrease in organic carbon contents with increasing soil depth (Soil Survey Staff, 2003). Soils of Pedons 01, 02, 03 and 04 have no evidence of hydromorphic properties within 150 cm of the mineral soil surface but have clay distribution such that the percentage clay decreased from its maximum by 20% or more within 150 cm of the mineral soil surface. These soils therefore, classify as Typic Kanhaplustults, they have ECEC of less than 12 cmol/kg soil. Soils of pedon 05 show evidence of redox depletion within 75 cm of the mineral soil surface and therefore, qualify as Aquic Haplustults**.** In the FAO-UNESCO soil legend, all the pedons under consideration qualify as Luvisols because of the presence of argillic horizon and humus surface horizon that is separated from the mineral horizon (Bruand et al. 2004), a horizon eluviated of clay minerals and a horizon of at least 5 cm. thick with illuvial clays (Bruand et al. 2004). The soils of pedon 03 and 04 classify as Plinthic Luvisols because of the presence of indurated coherent plinthite within 100 cm of the soil mineral surface. Soil of pedon 05 classifies as Gleyic Luvisols because of evidence of gleyic properties within 100 cm of the soil surface. The soils of pedon 01 and 02 classify as Eutric Luvisols because of the high base saturation (IUSS 2006).

Table 6. Pearson correlation coefficients of the soils' physical and chemical properties at the studied area.

	Total sand (%)	Silt (%)	Clay (%)	Bd g/cm3	pH (H20)	pH KCl	Ca^{2+}	Mg^{2+}	Na^{+}	K^{+}	Al^{3+}	H^{+}	Sum of Bases	ECEC	Base sat. (%)	Al. sat (%)	O M (%)	Avail P. (ppm)	$Fe_{(d)}$	$Al_{(d)}$	$Fe_{(d)}$/Clay	$Al_{(d)}$/Clay
Total sand (%)	1																					
Silt (%)	0.97	1																				
Clay (%)	0.13	0.36	1																			
Bd g/cm3	0.03	0.94	0.99	1																		
pH (H20)	0.29	0.51	0.99	0.19	1																	
pH KCl	0.89	0.75	-0.34	0.93	-0.18	1																
Ca^{2+}	0.17	0.4	0.98	0.07	0.99	-0.3	1															
Mg^{2+}	0.99	0.99	0.28	0.97	0.43	0.808	0.317	1														
Na^{+}	0.49	0.69	0.93	0.4	0.98	0.034	0.942	0.62	1													
K^{+}	0.78	0.91	0.72	0.72	0.82	0.41	0.745	0.87	0.93	1												
Al^{3+}	-0.07	-0.31	-0.99	0.03	-0.97	0.398	-0.995	-0.2	-0.9	-0.67	1											
H^{+}	0.41	0.62	0.96	0.31	0.99	-0.05	0.968	0.54	0.99	0.89	-0.94	1										
Sum of Bases	0.99	0.99	0.25	0.97	0.41	0.82	0.293	0.99	0.6	0.86	-0.19	0.52	1									
ECEC	0.99	0.94	0.03	0.99	0.19	0.92	0.071	0.97	0.4	0.72	0.03	0.32	0.975	1								
Base sat. (%)	-0.85	-0.95	-0.63	-0.8	-0.75	-0.52	-0.659	-0.9	-0.87	-0.99	0.58	-0.83	-0.91	-0.8	1							
Al sat. (%)	-0.45	-0.65	-0.94	-0.4	-0.99	0.013	-0.957	-0.6	-0.99	-0.91	0.92	-1	-0.56	-0.36	0.85	1						
O M (%)	0.92	0.99	0.51	0.87	0.64	0.635	0.544	0.97	0.79	0.97	-0.46	0.74	0.962	0.876	-0.99	-0.76	1					
Avail P. (ppm)	0.98	0.91	-0.06	0.99	0.11	0.958	-0.017	0.94	0.32	0.65	0.12	0.23	0.951	0.996	-0.74	-0.27	0.83	1				
$Fe_{(d)}$	-0.32	-0.54	-0.98	-0.2	-0.99	0.152	-0.988	-0.5	-0.98	-0.84	0.97	-0.99	-0.44	-0.22	0.77	0.99	-0.67	-0.14	1			
$Al_{(d)}$	-0.34	-0.55	-0.98	-0.2	-0.99	0.133	-0.985	-0.5	-0.99	-0.85	0.96	-0.99	-0.45	-0.24	0.78	0.99	-0.68	-0.16	0.99	1		
$Fe_{(d)}$/Clay	0.06	-0.19	-0.98	0.16	-0.94	0.507	-0.975	-0.1	-0.84	-0.58	0.99	-0.89	-0.07	0.152	0.48	0.87	-0.34	0.24	0.93	0.92	1	
$Al_{(d)}$/Clay	-0.08	-0.32	-0.99	0.02	-0.98	0.389	-0.996	-0.2	-0.91	-0.68	0.99	-0.94	-0.2	0.02	0.59	0.93	-0.47	0.11	0.97	0.97	0.99	1

4. Conclusions

The soils under investigation are predominantly ultisols with sandy top and clayey subsoils and are placed in ustults suborder. Pedological properties of the soils examined together with a low silt/clay ratio (<1.0) pointed to the fact that the soils were highly-weathered and intensely-leached. The soils are generally deep and well drained except at the crest and valley bottom due to the peculiarity of their locations on the topography. Quartz constituted the major primary mineral composition in the light mineral fraction with smaller amount of feldspathic minerals while opaque minerals dominated the heavy mineral portion with other minerals such as staurolite, Andalusite, kyanite e.t.c. that are present in smaller proportions in the fine sand fractions. The properties of the soils studied down the toposequence do indicate that chemical weathering and therefore, mineralogical alteration have proceeded translocation or possibly comminution. This is equally true of pedons that demonstrates the clay distribution of an in situ formed soil, presumably because the colluvium materials on top of the saprolite belonged to the same parent rock as the saprolite.

This will definitely influence management requirements because of the differences in the intensity of the soil properties down the topography and hence requires different management practices such as contour farming, terracing, strip cropping, usage of vegetal cover of economic importance and other soil conservation practices that suit each soil type at the different physiographic positions as evidenced by the taxonomic classes, for maintenance of soil health, preventing erosion, and enhancing productivity thereby assist in preventing rapid soil degradation across the landscape. In general, more research is required over time to determine the effectiveness of the recommended strategies and its impact on crop yields and soil characteristics in field experiments.

Author Contribution: Fawole OA. The work is a sole authorship, carved out of my Doctoral thesis

Data Availability Statement: All the data presented in this work are from the results generated during the Field work, Laboratory and statistical analysis carried out during the research. They can only be found in my field data logbook.

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References

Adegbite KA, Ogunwale JA (1994). Morphological, chemical and mineralogical properties of the soils of Abugi, Nigeria and their agricultural potential. *Pertanlka Journal of Tropical Agricultural Science 17 (3): 191-196.*

Adetayo F, Ayolabi EA, Ariyo SO (2013). Geological mapping, petrological study and structural analysis of precambrian basement complex rocks in part of Ago-Iwoye, Southwestern Nigeria. *International Research Journal of Geology and Mining (IRJGM)*, 3(1): 19-30.

Aduayi EA, Chude VO, Adebusiyi BA, Olayiwola SO (2002). Fertilizer use and management. Edition Produced by FFP, FMARD, Abuja. pp.188.

Ajiboye GA, Ogunwale JA (2010). Characteristics and classification of soils developed over talc at Ejiba, Kogi State, Nigeria. *Nigeria Journal of Soil Science,* 20 (1): 184- 206

Allison LE (1965). Organic Carbon. Pp 1367-1378. In C.A. Black *et al*., (edition). Methods of soil analysis. Part 2. Agronomy Monograph 9 ASA. Madison, WI.

Amusan AA, Ashaye TI (1991). Granitic-gneisis derived soils in humid forest tropical South Western Nigeria I: Genesis and classification. *Ife Journal of Agriculture*, 13: 1-10

Ano AA (1991). Potassium status of the Nigerian coastal plain sands. *Journal of Potassium Research,* 7 (4): 247-254.

Begna M (2020). Review on effect of toposequence on soil physicochemical properties. *Ethiopian Journal of Environmental Studies & Management*, *13*(4): 452 – 464.

Ben Mahmoud KR, Zurqani HA (2021). Soil forming factors and processes. *The Soils of Libya*, Springer International Publishing, USA, pp.33-48.

Bernas B (1968). A new method for the decomposition and comprehensive analysis of silicates by atomic absorption spectrometry: *Analytical Chemistry*, 40 (11): 1682- 1686

Bertsch PM, Bloom PR (1996). Aluminum. In *it'Iethods of soil anal irispart 3: Chemical methods* (SSSA Book Series No. 5). ed. D. L. Sparks. Madison, Wise.: SSSA/ASA.

Blake GR, Hartge KH (1986). Bulk density. In: Klute, A., Ed., Methods of Soil Analysis, Part 1—Physical and Mineralogical Methods, 2nd Edition, Agronomy Monograph 9, American Society of Agronomy—Soil Science Society of America, Madison, pp. 363-382.

Blume HP, Schwertmann U (1969). Genetic evaluation of distribution of Al, Fe and Mn Oxides. *Proceedings of Soil Science Society of America,* 33: 438 – 444.

Bockheim JG (2005). Soil endemism and its relation to soil formation theory. *Geoderma,* 129, 109 – 124.

Boesse JM, Ocan OO (1988). Geology and evolution of the ife-ilesha schist belt, South West Nigeria. Proceedings of Benin – Nigeria Geotraverse International Meeting on Proterozoic Geology and Tectonics of High Grade Terrains. Nov. 28- 30th, Obafemi Awolowo University, Ile-Ife, Nigeria, pp. 200-225.

Brandy NC, Weil RR (1999). The nature and properties of soils. (12th edition), by Prentice- Hall, Inc. Pearson Education Upper Saddle River, New Jersey. pp. 321 – 766.

Bruand AC, Hartmann S, P Sindtusen, R Poss, M Hardy (2004). Composition, fabric and porosity of an Arenic Haplustalf of Northeast Thailand in Relation to penetration resistance. *Soil Science Society of America Journal,* 48: 185-193.

Brunner AC, Park SJ, Ruecker GR, Dikaun R, Vlek PG (2004). Catenary soil development sinfluencing erosion susceptibility along a hillslope in Uganda. Elsevier Catena, 58(1), 23 September 2004, pp. 1-22

Buol SW, FD Hole, RJ Mc Cracken Southard RJ (1997). Soil genesis and classification. 4th Edition, Iowa State University Press, Ames.

Cox SB, Willing MR, Scatena FN (2002). Variation in nutrient characteristics of surface soils from Luquillo experimental forest of Puerto Rico. A multivariate perspective. *Plant and Soil*, 247: 189 – 198.

Darrell T, Maes P, Blumberg B, Pentland AP (1994). A Novel Environment for Situated Vision and Behavior. MIT Media Laboratory, Perceptual Computing Section Technical Report. No. 261.

Dinaburga G, Lapins D, Kopmanis J (2010). Differences of soil agrochemical properties in connection with altitude in Winter Wheat. *Engineering for Rural Development*, 79-84.

Enya OO, Omueti JA, Akinbola GE (2011). Particle size and free iron oxide distribution along two toposequence in South Western Nigeria. *Continental Journal of Agromony*, 5(2): 22-31

Esu IE, Ibanga IJ, Lombin G (1987). Soil landscape relationships in the Keffi plains of Northen Nigeria. *Samaru Journal of Agricultural Research*, 5 (1&2): 109-123.

Fasina AS (2001). Physical properties of soils of Lagos in relation to land use. *Annals of Agricultural Sciences*, 2(2): 1-3

Fasina AS (2002). Variability of maize yield and some soil properties in an exhaustively cultivated field in school of agriculture, Ikorodu. *Pertanika Journal of Tropical Agricultural Science*, 25: 11-18.

Fasina AS, Aruleba JO, Omolayo FO, Omotoso SO, Shittu OS, Okusami TA (2005). Properties and classification of five soils formed on granitic parent material of humid southwest Nigeria. *Nigeria Journal of Soil Science*, 15: 21-29.

Fasina AS, SO Omotoso, OS Shittu, Adenikinju AP (2007). Properties, classification and suitability evaluation of some selected cocoa soils of southwestern Nigeria. *American-Eurasian Journal of Agricultural and Environmental Science*, 2 (3): 311-317.

Gee GW, Or D (2002). Particle size analysis. In: Dane JH, Topp GC (eds) Methods of soil analysis part 4, Physical methods. Soil Science. Society of America. Book series no.5, ASA and SSSA, Madison WI, pp. 255-293.

Griffiths RP, Madritch MD, Swanson AK (2009). The effects of topography on forest soil characteristics in the Oregon Cascade Mountains (USA): Implications for the effects of climate change on soil properties. *Forest Ecology and Management*, 257: 1-7. http://dx.doi.org/10.1016/j.foreco.2008.08.010

Hook PB, Burke IC (2000). Biogeochemistry in short grass landscape: control by topography, texture and microclimate. *Ecology*, 81(10) 2686 – 2703.

IUSS Working Group WRB (2006). World Reference Base for Soil Resources 2006, 2nd edition. World Soil Resources Report 103, FAO, Rome. E.U. pp. 145.

Jackson ML (1958). Soil chemical analysis. Prentice-Hall, Inc., Englewood Cliffs, NJ.

Jenny H (1941). Factors of soil formation; systems of quantitative pedology. Dover publications Inc. New York, pp. 89.

Jubrin JM, Chude VO, Host WJ, Amagu IY (2000). The response of 10 leguminous cover crop and mage native and applied phosphate. *Proceedings of the 26th Annual Conference of Soil Science. Society of Nigeria*. Nigeria.

Kadeba O, Benjaminsen JN (1976). Contribution of organic matter and clay to the cation exchange capacity of soils in the savanna zone of Nigeria. *Communications in Soil Science and Plant Analysis*, 7:129–144.

Kerr Paul, F (1977). Optical Mineralogy, 4th Ed., New York, McGraw-Hill, [10]

Kumar V, Butter TS, Samanta A, Singh G, Kumar M, Dhotra B, Choudhary R (2018). Soil compaction and their management in farming systems: A review. *IJCS*, *6*(3): 2302-2313.

Kuo S (1996). Phosphorus. In: Sparks DL ed., Methods of soil analysis. Part 3. Chemical methods. SSSA Book Series. 5. SSSA and ASA, Madison, WI., pp. 869–919. doi:10.2136/sssabookser5.3.c32

McAuliffe JR, McFadden LD, Hoffman MT (2018). Role of aeolian dust in shaping landscapes and soils of arid and semi-arid South Africa. *Geosciences*, *8*(5): 171.

Mehra OP, Jackson ML (1960). Iron oxides removal from soils and Days. Dithionite-citrate stems buffered with sodium bicarbonate. *Clays and Clay Minerals,* 7: 313 – 327.

Moorman FR (1981). Representative of toposequence of soils in southern Nigeria and their pedology. In characterization of soils in relation to their classification and management for crop production. D. J. Greenland (ed) Clarendo press Oxford, pp. 29.

Neil E, Smeck LP, Wilding (1980). Quantitative evaluation of pedon formation in calcareous glacial deposits in Ohio. *Geoderma*, 24(1), pp. 1-16.

Obigbesan GO (2009). Impact of Liebig's reseach on the development of Agriculture in Africa. A paper presented at the *33nd Conference of the Soil Science*. Society. of Nigeria at Ado-Ekiti., pp.51.

Ogban PI, Babalola O, Okoji AM (1999). Profile characteristics of a typical toposequence in Southern Nigeria. *Africa Soils*, 28, pp 147 – 165.

Ogunsola OA, Omueti JA, Olade O, Udo JE (1989). Free oxide status and distribution in soils overlying limestone area in Nigeria. *Soil Science Society of Nigeria journal*, 147(4): 245-251.

Ojanuga AG (1978). Genesis of soils in the metamorphic forest & region of southwestern Nigeria *Pedolomorphic Pedologie*, 1: 105-117.

Ojetade JO., Olasoji, HO., Muda, SA., Amusan, AA (2022). Characterisation, classification and suitability assessment of soils formed in granite and gneiss in humid area of southwestern Nigeria for cacao (*Theobroma cacao*) production. *Tropical Agriculture*, *99*(1): 11-21.

Okusami TA, Oyediran GO, (1985). Slope-Soil relationship on an Aberrant Toposequence in Ife area of southwestern Nigeria; Variabilities in soils properties. *Ife Journal of Agriculture.* 7 (1&2) 1-15.

Onyekwere I, N Akpan Idiok AU, Amalu UC, Asawalam DO, Eze PC (2001). Constraints and opportunities in Agricultural utilization of some wetland soils in Akwa Ibom State. In: management of wetland soils for sustainable agriculture and environment. *Proceedings of the 27th Annual Conference of Soil Society of Nigeria* (SSSN), pp 139-149.

Parfitt RL, Childs CW (1988). Estimation of forms of Fe and Al: A review and analysis of contrasting soils by dissolution and Moessbauer methods. *Australian Journal of Soil Resources*, 26: 121-144

Payne D (1988). Soil structure, tilth and mechanical behaviour. *In* A Wild (ed.) Russell's soil conditions & plant growth. Longman Scientific & Technical, Burnt Mill, England, pp. 378–411.

Periaswamy SP, Ashaye TI (1982). Updated classification of some southwestern Nigeria soils. *Ife Journal of Agriculture* 4: 25-41.

Rahaman MA (1988). Recent advances in the study of the basement complex of Nigeria. *In: Geological Survey of Nigeria (Ed) Precambrian Geol Nigeria*, pp. 11–43.

Rebertus RA, Buol SW (1985). Iron distribution in a developmental sequence of soils from mica gneiss and schist. *Soil Science Society of America Journal,* 49:713-720

Simpson C (1986). Determination of movement sense in mylonites. *Journal of Geological Education*, 34: 246- 261.

Smyth AJ, Montogomery RF (1962). Soils and land use in central western Nigeria. Western Nigeria Government, Ibadan, pp. 265.

Soil Survey Staff (2003). Keys to taxonomy, Ninth Edition USDA/NRCS, Washington D.C., pp.332.

Sumner ME, Miller WP (1996) Cation exchange capacity and exchange coefficients. In: Sparks, D.L., Ed., Methods of soil analysis Part 3: Chemical methods, SSSA Book Series 5, Soil Science Society of America, Madison, Wisconsin, pp. 1201-1230.

Thomas GW (1996). Soil pH and soil acidity. In: Sparks, D.L., Ed., Methods of soil analysis: Part 3—Chemical methods, Book Series No. 5, SSSA and ASA, Madison, WI, pp. 475-489.

Thomas MF, Throp MB (1985). Environmental change and episodic ectoplantation in the humid tropics of sierra leone: The koidu etchplain: In Douglas, and Spencer, T. (ed.) Environmental Change and Tropical Geomorphology. George Allen and Unwin, London, pp. 239-267.

Ukwuoma-Okolo A (2021). Introduction to soil science. *Agricultural Technology for Colleges*, 38.

Uponi JI, Adeoye GO (2000). Soil testing and plant analysis: An overview. Agronomy in Nigeria, pp.177.

Uzoho BU, Oti NN (2004). Phosphorus adsorption characteristics of selected southwestern Nigeria soils. *Proceedings of the 29th Annual Conference of the Soil Science Society of Nigeria*, Abeokuta, Nigeria, pp. 121-130.

Van der Meij WM, Temme AJ, İn HS, Gerke HH, Sommer M (2018). On the role of hydrologic processes in soil and landscape evolution modeling: concepts, complications and partial solutions. *Earth-Science Reviews*, 185: 1088-1106.

Van Raij Bernardo, Michael Peech (1972). Electrochemical properties of some oxisols and alfisols of the tropics. *Soil Science Society of Nigeria Journal*, 36: 587-593.

Wilding LP, LR Dress (1983). Spatial Variability in Pedology, pp 83-113

Wilson DJ, Western AW, Grayson RB (2004). Identifying and quantifying sources of variability in temporal and spatial soil moisture observations. *Water Resources Research* 40(2): W02507. https://doi.org/10.1029/2003WR002306

Zhao B, Zhang L, Xia Z, Xu W, Xia L, Liang Y, Xia D (2019). Effects of rainfall ıntensity and vegetation cover on erosion characteristics of a soil containing rock fragments slope, *Advances in Civil Engineering,* 7043428: 14. https://doi.org/10.1155/2019/7043428

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Yield Stability and Agronomic Performance of Late-Maturing Pro-Vitamin A Maize in Southwest Nigeria Using GGE Biplot Analysis

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HIGHLIGHTS

- Variations in environmental conditions lead to significant annual fluctuations in maize yield performance.
- GGE biplot analysis was effectively used to assess genotype stability and performance.

Abstract

Maize is one of the most essential cereal crops grown across the major agroecological zones of Nigeria, both in terms of production and consumption. However, significant differences in soil properties and climatic conditions have led to wide fluctuations in the yield performance of maize genotypes annually, making the study of genotype by environment interactions crucial for plant breeders. Given the crop's importance as a staple food in Sub-Saharan Africa, this study evaluated the yield performance and stability of open-pollinated maize genotypes (OPVs) across diverse agroecological environments in Nigeria over two cropping seasons (2019 and 2020). In 2019 and 2020, fourteen open-pollinated cultivars were examined in three sites utilizing a Randomized Complete Block Design (RCBD) with three replications. The commercial check entry (3.73 t ha^{-1}) and the local check variety (3.68 t ha^{-1}) yielded far less than the genotype with the maximum grain yield, which produced 4.24 t ha^{-1}. The tested genotypes and the commercial check exhibited greater stability than the local check. Results indicated that the genotypes AFLATOXIN SYN-YF2 and PVA SYN 13 demonstrated both high yield and stability, making them valuable resources for breeding programs focused on developing improved maize varieties for small-scale farmers in Nigeria, many of whom have limited access to commercial maize hybrids. Additionally, locations - Ibadan'19 and Orin-Ekiti'20 emerged as the most discriminating and representative test environments for maize selection. The study underscores the need for breeding programs to prioritize genotypes with short anthesis-silking intervals (ASI), moderate plant and ear heights, and strong disease resistance for enhanced yield stability. These findings provide critical insights for maize breeders and small-scale farmers, promoting sustainable maize production in Nigeria amid changing climatic conditions.

Keywords: Genotype by environment (G x E); environments; open pollinated variety (OPV); stability; *Zea mays* L.

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1. Introduction

Maize is a major food security crop in Sub-Saharan Africa (SSA) and other developing world regions, providing sustenance for millions of people (FAOSTAT 2021). The significance of maize in the diet of Nigerians has compelled researchers to focus more on enhancing the productivity of new maize varieties through breeding and agronomic evaluation under a wide range of climatic variables, including moisture stress. This effort is to generate high-yielding, stable varieties, and nutritionally acceptable maize genotypes for farmers with limited resources to adopt, for enhanced yield and improved standard of living.

Maize (*Zea mays* L.) is a critical component of Nigeria's food security and economy, with production reaching 12.75 million metric tons in 2021 (Statista 2023). Although maize production grew at an annual rate of 4.2% between 2010 and 2020, yields have remained stagnant at 1.7 t/ha since 2017, significantly lower than those of South Africa (4.9 t/ha) and Ethiopia (4.2 t/ha) (Wossen et al. 2023). This yield gap underscores the need for improved agricultural practices and innovations to enhance productivity. Furthermore, conventional maize varieties often lack sufficient micronutrient content, which is critical for human health, especially in rural regions where food supplements are scarce. By raising the amounts of vital vitamins and minerals in staple crops, biofortification can assist in solving this issue in a more sustainable and cost-effective manner(Avnee et al. 2023). Notably, provitamin A maize can provide more than 50% of the recommended dietary vitamin A intake (Goredema-Matongera et al. 2021). Furthermore, studies indicate that its consumption effectively improves body vitamin A stores, similar to supplementation (Gannon et al. 2014). However, the adoption of biofortified crops depends on several factors, including government policies and resource availability (Onyeneke et al. 2019).

Vitamin A is essential for vision, immune function, and growth (Huang et al. 2018). Its deficiency leads to severe health consequences, such as night blindness, increased susceptibility to infections, and stunted growth (Song et al. 2023; Villamor et al. 2000). Additionally, a study in Uganda found a significant correlation between VAD and growth failure in preschool children (Ssentongo et al. 2020), reinforcing the importance of adequate vitamin A intake in early childhood development. In many resource-poor regions, micronutrient deficiencies are exacerbated by chronic gastrointestinal infections, which impair vitamin A absorption. This issue is further compounded by zinc deficiency, as zinc plays a crucial role in vitamin A absorption and transport (Hotz et al. 2012; Rahman et al. 2002). About 30% of children under five suffer from VAD, which is still a serious public health problem worldwide and accounts for 2% of these children's fatalities (Hodge and Taylor 2023; Wirth et al. 2017). Therefore, vitamin A-biofortified maize in Nigeria has the potential to significantly decrease vitamin A deficiency, improving overall health and nutrition for communities vulnerable to micronutrient deficiencies.

Despite the success of maize hybrids in Nigeria, their adoption by smallholder farmers remains limited due to high production costs, technical complexity, and the need for substantial inputs, which contrast with the affordability and ease of managing open-pollinated varieties (Eze et al. 2020). However, enhanced maize populations and the hybrids derived from them offer valuable alternatives to traditional single-cross hybrids and serve as crucial resources for developing new inbred lines (Kutka 2011). These improved populations provide a broad genetic base that can be harnessed to create inbred lines with desirable traits, contributing to sustained genetic gains in maize breeding (Yong et al. 2019). This genetic diversity is essential for addressing environmental stresses and evolving pest and disease pressures. Additionally, population-derived hybrids demonstrate better adaptation to specific agroecological conditions, with some outperforming commercial hybrids in challenging environments, presenting viable alternatives for improving productivity and resilience (Eze et al. 2020).

Finding stable, high-yielding maize hybrids is challenging due to the significant influence of genotype-environment interaction (GEI), which is magnified by environmental changes (Akcura et al. 2011). Grain yield, a crucial agronomic and economic trait, typically shows GEI, complicating efforts to achieve yield stability (Fan et al. 2007; Bocianowski et al. 2024). GEI plays a significant role in genotype performance, particularly in environments with variable climatic conditions (Sibiya et al. 2012). Shrestha (2013) examined the stability of six high-quality protein maize (QPM) varieties over two years at eleven sites. The results showed that environmental factors had a highly significant impact, while GEI was not significant in the Combined

ANOVA. Similarly, Abakemal et al. (2016) evaluated the yield stability of QPM single-cross hybrids in tropical highlands, finding significant differences among hybrids and environments, with some hybrids showing stable performance across diverse conditions. Dosho et al. (2022) assessed QPM hybrids under varying soil nitrogen conditions, revealing significant GEI effects, which helped identify stable genotypes. Bankole et al. (2023) found that environmental factors strongly influenced grain yield, with certain QPM hybrids demonstrating superior stability and adaptability under rainfed conditions.

It's vital to introduce, assess, and modify potential germplasm or hybrids to satisfy the expanding need for maize to meet its growing demand. The GGE biplot is a valuable tool for predicting increased performance and grain yield stability across new situations in this respect (Daemo and Ashango 2024). The GGE biplot analysis is beneficial as it enables the prediction of average genotype yield in a specific environment and helps identify the most stable genotype for a given location (Yan 2014). G x E studies can maximize grain yield potential while lowering production costs by identifying the optimal site for each genotype (Oyekunle et al. 2017). Research by Kumar et al. (2024) showed that GGE biplot analysis effectively evaluates yield stability and adaptability of various maize hybrids, allowing for better prediction of their performance across different environmental conditions. This is because maize cultivars vary greatly in yield depending on the cultivation area (Olakojo and Iken 2001; Guo et al. 2022). As a result, maize varieties must be assessed for their ability to adapt to different agroecological zones and production potential (Olaoye 2009). To better adapt germplasm to varied production environments, it's crucial to understand the link between yield testing locations (Trethowan et al. 2001; Bassa and Goa 2016). This study looked at the yield performance and stability of 14 open-pollinated varieties in South-west Nigeria, and the connections between test environments in various agro-ecologies. Late-maturing maize genotypes were favored for this research for their higher yield potential. Their extended growth phases enhance biomass accumulation and optimize resource use, leading to greater grain yield.

2. Materials and Methods

Field trials were conducted in 2019 and 2020 at the Teaching and Research Farms of the Institute of Agricultural Research and Training (IAR&T), Ibadan (7°23'10''N, 3°50'21''E; and 1250 – 1500 mm annual rainfall), Eruwa (7°53'04''N, 3°41'70''E; and 1200 – 1300 mm annual rainfall), and Orin-Ekiti (7°49'48''N, 5°14'24''E; and 1333.2 - 1400 mm annual rainfall) in Nigeria. Thirteen elite open-pollinated maize varieties derived from late-maturing pro-vitamin A breeding populations of IITA were evaluated. A total of fifteen entries, comprising thirteen OPV maize genotypes, one commercial check, and one farmer’s check, were assessed for grain yield performance across six different environments under optimal growing conditions in 2019 and 2020.

The six environments were characterized by variations in total rainfall and planting periods (Appendix 1). Trials conducted under optimal conditions were established during the main maize planting season, coinciding with adequate rainfall. The environmental conditions varied significantly concerning water availability, while differences in seasonal rainfall distribution contributed to location-specific variations. The general strategy of the conducted trial series was to assess genotype performance across multiple environments within a short period, thereby capturing agroecological variability that would typically require multi-year testing.

The experiment was laid out in a Randomized Complete Block Design (RCBD) with three replications per location. Each experimental unit consisted of two-row plots, each measuring 5 meters in length with a row spacing of 0.75 m and an intra-row spacing of 0.50 m. Three seeds were initially sown per hill, and approximately two weeks after emergence, the seedlings were thinned to two per hill to achieve a final plant population density of about 53,333 plants ha^{-1}. Weed control was achieved through the application of pre-and post-emergence herbicides. Paraquat (0.75 kg a.i. ha^{-1}) and Atrazine (2.5 kg a.i. ha^{-1}) were applied immediately after planting. Additional hand weeding was carried out as necessary to ensure effective weed management throughout the growing season. The fertilizer application followed a two-step process. A basal application of NPK 20-10-10 was carried out at a rate of 80 kg N ha^{-1}, 40 kg P_2O_5 ha^{-1}, and 40 kg K_2O ha^{-1} three weeks after planting. This was followed by a top-dressing of Urea (46-0-0) six weeks after planting to ensure adequate nitrogen availability for optimal crop growth and grain development.

2.1. Collection of Agronomic and Yield Data

A scale of 1 to 5 is used to rate diseases, and the data collected includes days to 50 % anthesis, days to 50 % silking, plant and ear heights (cm), root lodging (number of plants leaning more than 30^{o} from vertical), stalk lodging (stalks broken at or below highest ear node), ear aspect, and number of plants harvested. All the data and scoring in this study were according to Badu-Apraku et al. 2012.

Table 1. Description of Open-pollinated genotypes tested across three locations in 2019 and 2020.

Code	Entry Name	Source
1	ACR.91 SUWAN 1-SR C1	IITA
2	AFLATOXIN SYN-YF2	IITA
3	F2SCA1413-12	IITA
4	F2SCA1413-36	IITA
5	F2TWLY100121	IITA
6	F2TWLY100123	IITA
7	F2TWLY131211	IITA
8	F2TWLY131228	IITA
9	F2TWLY13124	IITA
10	Local Check	Farmer’s Variety
11	PVA SYN HGA C2	IITA
12	PVA SYN HGB C0	IITA
13	PVA SYN HGB C2	IITA
14	Sammaz 52(PVA SYN 13)	IITA
15	PVA SYN-6	IITA

2.2. Statistical Analysis

To ascertain if the G x E interaction effects were significant, the data were first examined independently for each site before being merged and examined for grain yield across locations using PROC GLM in SAS (SAS version 9.2). The standard error of the mean (S.E.) at P <0.05 and the New Duncan Multiple Range Test at P <0.05 were used to differentiate the means. The grain yield data were then subjected to GGE biplot analysis to ascertain the stability of grain yield as well as the genotype response pattern assessed in each of these environments.

2. Results

3.1. Analysis of Variance

The combined ANOVA showed that genotypes performed differently (P <0.05) from one environment to another, showing the diversity of the test environments (E). For grain yield, however, the associations of genotype x location x year were not significant. Except for root lodging, all fifteen of the maize genotypes showed highly significant (P <0.01) location impacts on grain yield and agronomic traits (Table 2). Genotypes also varied significantly (P <0.01) for grain yield, husk cover, anthesis-silking interval, and stalk lodging. Similarly, days to silking, plant height, and stalk lodging were significant at P <0.05. On the contrary, there were no significant differences among the genotypes for days to pollen shed, plant height, ear height, plant and ear aspect, ear per plant, root lodging, ear rot, blight syndrome rating, and *Curvularialunata* disease syndrome rating. Genotype x location effects were not significantly different except for husk cover, rust, and curvularia in this study (Tables 2 and 3). Furthermore, genotype x year interaction was not significant for all measured traits except for plant height and curvularia leaf spot ratings which differed significantly at P <0.01. On the other hand, location x year interaction was significant for all measured traits (P <0.01) except for ear aspect and ear rot. Genotype x location x year interactions were however observed to be highly significant for plant height and curvularia. This suggests that climatic variables of the test environment in the two years had a pronounced effect only on the expression of most of the traits studied (Tables 2 and 3), while the interactive effects of G x Y were significant for plant height and curvularia leaf spot.

3.2. Performance of the Maize Genotypes for Grain Yield and Agronomic Traits in 2019 and 2020

The grain yield of open-pollinated maize genotypes (OPVs) varied significantly across Eruwa, Ibadan, and Orin-Ekiti during the 2019 and 2020 cropping seasons, largely influenced by prevailing weather conditions (Appendix I). Mean grain yield was higher in 2019, with Ibadan recording the highest yield (4.14 t ha^{-1}), followed by Eruwa (3.77 t ha^{-1}) and Orin-Ekiti (3.27 t ha^{-1}) (Table 4). However, yields declined in 2020, with the sharpest reduction in Eruwa (2.80 t ha^{-1}), followed by Ibadan (3.78 t ha^{-1}) and Orin-Ekiti (3.69 t ha^{-1}). These decreases corresponded with reduced annual rainfall across all locations. The highest yielding genotypes recorded 4.24 and 4.23 t ha^{-1} for F2TWLY100123 and ACR.91 Suwan 1-SR C1 respectively. The two highest-ranking genotypes were statistically different from the checks (Commercial and local check). The highest-yielding check was the commercial check PVA SYN-13 (Sammaz 52) with a mean grain yield of 3.73 t ha^{-1} while the local check had a mean grain yield of 3.68 t ha^{-1}. The highest mean yields across the three locations were from F2TWLY100123 and ACR.91 Suwan 1-SR C1 with 4.24 and 4.23 t ha^{-1} respectively while the lowest yielding genotype was PVA SYN HGB C0 with 2.89 t ha^{-1}.

The intricate interplay between genotype and environment on the grain yield of the examined maize varieties, as illustrated in Table 4, reveals a striking truth: all genotypes exhibited superior yields in the year 2019 when juxtaposed with their performance in 2020. This divergence can likely be attributed to the protracted moisture stress and the erratic distribution of rainfall that marred the agricultural landscape of 2020 across various locations. In Eruwa, rainfall dropped from 1,777.39 mm in 2019 to 1,013.50 mm in 2020, while Ibadan experienced a decline from 1,774.61 mm to 1,104.50 mm, and Orin-Ekiti from 2,099.94 mm to 1,497.97 mm. The most pronounced reduction in Eruwa aligned with its largest yield drop, indicating that moisture stress played a critical role in limiting productivity. The temperature remained relatively stable, with minor fluctuations. In Eruwa, the mean temperature was 27.92°C in 2019 and 27.42°C in 2020, while Ibadan recorded 27.17°C in 2019 and 27.08°C in 2020. Orin-Ekiti had the lowest temperatures, at 25.67°C in 2019 and 25.42°C in 2020. However, relative humidity (RH) increased in 2020 despite lower rainfall, rising from 71.17% to 76.25% in Eruwa and from 73.08% to 77.17% in Ibadan. This higher RH may have elevated disease pressure, particularly fungal infections like maize rust and gray leaf spot, contributing to further yield reductions. Wind speed also increased slightly in 2020. In Orin-Ekiti, it rose from 7.46 kmph in 2019 to 7.60 kmph in 2020, while in Eruwa, it changed from 7.42 kmph to 7.27 kmph. Stronger winds during the flowering and grain-filling stages may have led to lodging and reduced pollination efficiency, further impacting yields.

Varietal performance varied across locations. ACR.91 SUWAN 1-SR C1 recorded the highest mean yield (4.23 t ha^{-1}), performing exceptionally well in Eruwa (6.04 t ha^{-1} in 2019). However, its yield declined significantly in 2020, particularly in Eruwa (3.78 t ha^{-1}) and Orin-Ekiti (2.99 t ha^{-1}). F2TWLY100123 also performed well, averaging 4.24 t ha^{-1} across locations. In contrast, PVA SYN HGB C0 had the lowest mean yield (2.89 t ha^{-1}), suggesting lower adaptability to environmental fluctuations, particularly under drier conditions. The local check variety exhibited moderate performance (3.68 t ha^{-1}), indicating adaptation to local conditions but without significant yield advantages over improved OPVs.

Table 5 presents the mean performance of the maize genotypes across locations for the measured traits. Days to Anthesis ranged from 56.22 to 58.28 days, while days to silking ranged between 59.06 and 61.17 days, resulting in an anthesis-silking interval (ASI) of 1.78 to 3.89 days. The shortest ASI of 1.78 days was observed in PVA SYN HGB C2. Conversely, F2TWLY100123 exhibited the longest ASI of 3.89 days. Significant variation in plant and ear height was observed among genotypes. ACR.91 SUWAN 1-SR C1 attained the tallest stature at 155.89 cm, with the highest ear placement of 69.78 cm. In contrast, PVA SYN HGB C2 recorded the shortest plants at 140.28 cm. The lowest ear height was noted in PVA SYN-6 at 58.06 cm, which may confer better lodging resistance. Plant aspect (PA) and ear aspect (EA) scores varied across genotypes, with lower values indicating more desirable traits. PA scores ranged from 1.94 to 2.56, while EA scores ranged from 1.89 to 2.44.

Table 2. Mean Square values from the combined analysis of Variance (ANOVA) of grain yield and yield-related traits of fifteen OPVs evaluated across three locations in 2019 and 2020

Source of Var.	Df	Days to pollen	Days to Silking	ASI	Plant Height	Ear Height	Husk Cover	Plant Aspect	Ear Aspect	EPP	Grain Yield
Rep	2	4.25	1.05	5.61	2964.28	338.54	0.09	1.30	0.68	0.001	1.82
Year	1	2632.03**	2011.74**	41.61**	87480.00**	39240.83**	22.53**	4.80**	0.004	0.023**	6.23**
Genotype	14	5.53	7.31*	6.33**	512.89	147.80	0.22**	0.47	0.45	0.001	2.50**
Location	2	817.60**	341.14**	102.78**	6920.78**	1231.30**	27.91**	7.78**	21.54**	0.064**	11.00**
Gen. x Loc.	28	3.64	3.56	3.49	336.55	179.99	0.17*	0.51	0.52	0.002	1.13
Gen. x Year	14	4.81	4.01	3.83	701.02**	199.17	0.15	0.26	0.22	0.002	1.07
Loc. x Year	2	489.43**	201.16**	69.00**	5503.01**	404.68*	17.63**	2.80**	0.18	0.066**	11.04**
G x L x Y	28	2.93	6.73*	4.13	464.55*	181.88	0.13	0.53	0.36	0.002	0.95
Error	178	3.23	4.12	2.93	300.92	132.27	0.10	0.45	0.44	0.01	0.91
Means		57.40	60.36	2.96	141.54	62.10	1.68	2.27	2.14	0.99	3.58
CV (%)		3.13	3.36	57.76	12.26	18.52	18.46	29.71	31.00	4.57	26.66

* = P*<0.05, *** = P*<0.01, ASI = Anthesis-Silking-Interval, EPP = Number of ears per plant respectively*

Table 3. Mean Square values from the combined analysis of variance of lodging and disease reaction of fifteen OPVs evaluated across three locations in 2019 and 2020

Source of Variation	df	Root Lodging	Stalk Lodging	Ear Rot	Streak	Rust	Blight	Curvularia
Rep	2	0.03	0.07	0.41	0.05	0.01	0.29	0.01
Year	1	0.37	0.37	0.30	0.37	0.45**	1.07**	9.26**
Genotype	14	0.09	0.31**	0.58	0.26**	0.13**	0.20	0.09
Location	2	0.08	3.13**	9.14**	1.66**	1.08**	3.21**	16.48**
Gen. x Loc.	28	0.18	0.15	0.48	0.14	0.09*	0.25	0.08*
Gen. X Year	14	0.27	0.08	0.48	0.16	0.07	0.21	0.12**
Loc. x Year	2	3.51**	1.08**	1.03	2.50**	1.78**	3.61**	6.71**
G x L x Y	28	0.15	0.19*	0.62	0.15	0.10**	0.13	0.11**
Error	178	0.22	0.13	0.56	0.11	0.06	0.17	0.05
Means		1.20	1.21	1.46	1.19	1.10	1.31	1.53
CV (%)		39.14	29.46	51.45	28.21	21.52	31.61	14.92

* = P <0.05, ** = P <0.01 *respectively*

Table 4. Mean grain yield (t ha^{-1}) of the OPVs maize in 2019 and 2020 cropping season

	Eruwa		**Ibadan**		**Orin-Ekiti**		
Varieties	**2019**	**2020**	**2019**	**2020**	**2019**	**2020**	**Mean**
ACR.91 SUWAN 1-SR C1	6.04	3.78	4.27	4.33	3.98	2.99	4.23
AFLATOXIN SYN-YF2	3.62	3.34	4.00	3.35	3.61	4.05	3.66
F2SCA1413-12	3.55	2.29	3.60	4.55	2.67	3.26	3.32
F2SCA1413-36	3.17	2.36	3.79	3.74	3.48	4.55	3.52
F2TWLY100121	3.79	2.42	5.38	4.03	3.29	4.21	3.85
F2TWLY100123	3.67	3.78	5.97	3.98	3.34	4.68	4.24
F2TWLY131211	3.60	3.05	4.48	3.53	3.98	2.80	3.57
F2TWLY131228	3.50	2.68	4.53	3.51	3.03	4.66	3.65
F2TWLY13124	3.74	2.83	3.67	3.28	3.42	4.98	3.65
Local Check	4.31	3.12	4.45	3.61	3.44	3.13	3.68
PVA SYN HGA C2	3.54	2.64	3.02	4.42	2.68	3.30	3.27
PVA SYN HGB C0	3.17	2.25	3.25	3.32	2.27	3.04	2.89
PVA SYN HGB C2	3.03	2.17	2.99	3.61	3.31	3.59	3.12
Sammaz 52	4.07	2.52	5.50	3.94	3.49	2.86	3.73
PVA SYN-6	3.75	2.72	3.24	3.55	3.06	3.32	3.27
Mean	3.77	2.80	4.14	3.78	3.27	3.69	3.58
CV (%)	30.68		28.38		30.29		30.68
S.E (0.05)	0.11		0.12		0.11		0.07

Table 5. Mean performance of fifteen (15) genotypes for grain yield and agronomic traits across three locations over two (2) years.

Entry Name	Grain Yield (t ha $^{-1}$)	Days to Anthesis (days)	Days to Silking (days)	ASI (days)	Plant height (cm)	Ear Height (cm)	Plant Aspect (1-5)	Ear Aspect (1-5)	Husk cover (1-5)	Streak (1-5)	Blight (1-5)	Curv. (1-5)	Rust (1-5)
ACR.91 SUWAN 1-SR C1	4.23a	56.22d	59.06d	2.83abcde	155.89a	69.78a	1.94c	1.89b	1.56cd	1.11cde	1.33abcd	1.50bc	1.22a
AFLATOXIN SYN-YF2	3.66abc	56.83cd	59.67bcd	2.83abcde	144.56ab	61.89b	2.22abc	2.06ab	1.78ab	1.00e	1.28abcd	1.50bc	1.00b
F2SCA1413-12	3.32bcd	57.72abc	61.06a	3.33abc	138.28bc	61.89b	2.28abc	2.06ab	1.72bc	1.22bcd	1.33abcd	1.61ab	1.06b
F2SCA1413-36	3.52bc	57.78abc	60.83ab	3.06abcd	142.44bc	59.06b	2.33abc	1.89b	1.72bc	1.06de	1.50a	1.56bc	1.06b
F2TWLY100121	3.85ab	57.56abc	61.00ab	3.44ab	142.44bc	64.00ab	2.22abc	2.11ab	1.61bcd	1.28abc	1.44ab	1.50bc	1.11ab
F2TWLY100123	4.24a	57.00bcd	60.89ab	3.89a	141.56bc	62.83ab	2.11bc	1.89b	1.67bcd	1.11cde	1.39abc	1.44c	1.06b
F2TWLY131211	3.57bc	57.28abcd	60.61abc	3.33abc	131.67c	58.22b	2.44ab	2.22ab	1.72bc	1.11cde	1.11d	1.56bc	1.06b
F2TWLY131228	3.65abc	57.83abc	61.17a	3.33abc	143.61b	64.22ab	2.39ab	2.28ab	1.72bc	1.33ab	1.39abc	1.50bc	1.00b
F2TWLY13124	3.65abc	57.33abcd	60.61abc	3.28abc	141.94bc	62.89ab	2.06bc	2.06ab	1.67bcd	1.17bcde	1.22bcd	1.44c	1.06b
Local Check	3.68abc	58.06ab	60.33abcd	2.28cde	145.94ab	61.50b	2.39ab	2.22ab	1.94a	1.44a	1.28abcd	1.72a	1.22a
PVA SYN HGA C2	3.27bcd	57.83abc	60.44abc	2.61bcde	141.22bc	60.72b	2.11bc	2.06ab	1.67bcd	1.11cde	1.17cd	1.50bc	1.11ab
PVA SYN HGB C0	2.89d	57.28abcd	59.28cd	2.00de	142.28bc	60.83b	2.56a	2.44a	1.61bcd	1.22bcd	1.33abcd	1.56bc	1.11ab
PVA SYN HGB C2	3.12cd	58.28a	60.06abcd	1.78e	140.28bc	63.56ab	2.39ab	2.28ab	1.56cd	1.17bcde	1.22bcd	1.50bc	1.22a
Sammaz 52	3.73abc	57.28abcd	60.17abcd	2.89abcde	138.11bc	62.06b	2.28abc	2.06ab	1.78ab	1.33ab	1.33abcd	1.56bc	1.00b
PVA SYN-6	3.27bcd	56.67cd	60.22abcd	3.56ab	135.78bc	58.06b	2.28abc	2.22ab	1.500d	1.11cde	1.39abc	1.56bc	1.22a
Mean	3.58	57.40	60.36	2.96	141.54	62.10	2.27	2.14	1.68	1.19	1.31	1.53	1.10
S.E. (0.05)	0.07	0.29	0.24	0.13	1.71	1.06	0.05	0.05	0.04	0.02	0.03	0.03	0.02
CV (%)	30.68	8.34	6.61	73.44	19.80	28.07	32.81	35.67	35.67	33.63	36.58	34.02	27.32

ACR.91 SUWAN 1-SR C1 recorded the lowest EA score of 1.89, suggesting superior ear quality. Husk cover scores varied between 1.50 and 1.94, with PVA SYN-6 exhibiting the lowest score of 1.50, indicative of better husk protection against pest damage and grain deterioration.

Genotypic differences in disease resistance were also observed. Rust, blight, and streak scores ranged from 1.00 to 1.50, with most genotypes exhibiting moderate resistance. Notably, the local check variety had higher blight and streak scores, indicating greater susceptibility compared to improved open-pollinated varieties (OPVs). AFLATOXIN SYN-YF2 displayed the best resistance to maize streak virus, with a streak score of 1.00. Blight scores ranged from 1.11 to 1.50, with the lowest susceptibility recorded in F2TWLY131211. Rust resistance scores varied between 1.00 and 1.22, while curvularia leaf spot scores ranged from 1.44 to 1.72, indicating varying levels of tolerance among the genotypes. Grain yield performance varied significantly among genotypes across three locations over two years. The mean grain yield ranged from 2.89 t ha^{-1} (PVA SYN HGB C0) to 4.24 t ha^{-1} (F2TWLY100123), with an overall mean of 3.58 t ha^{-1}. The highest-yielding genotypes were F2TWLY100123 (4.24 t ha^{-1}) and ACR.91 SUWAN 1-SR C1 (4.23 t ha^{-1}). Conversely, PVA SYN HGB C0 exhibited the lowest yield at 2.89 t ha^{-1}. The coefficient of variation (CV) for grain yield was 30.68%, indicating moderate variability among genotypes.

3.3. GGE Biplot Analysis

The environment-vector view of the GGE biplot showing similarities among test environments is presented in Figure 1. The environment vectors and the cosine of the angle between the vectors show a strong crossover GE and explain a very large GE considering the wide degree existing between Environment E and F. Dissimilarity in discriminating the genotypes is measured by the distance between two environments and this has successfully regrouped 6 test environments into 3. Environment A, C, D, and E formed a group, while Environment B and F are separately each in a class of its own.

Figure 2 shows the biplot of grain yield performance of 15 OPV maize tested in six environments. Varieties, ACR.91 SUWAN 1-SR C1, F2SCA1413-36, F2TWLY100123, F2TWLY131228, PVA SYN HGA C2, PVA SYN HGB C0, and PVA SYN HGB C2 were located at the vertex cultivar and represent the best-performing genotypes in terms of grain yield at the environments that fall within their sectors (Yan et al.2005). They are equally considered to be highly responsive having the farthest distance from the biplot origin. However, genotypes positioned within the polygon are therefore considered less responsive (Yan and Rajcan, 2002).

Figure 3 shows the discrimination and representativeness view of the GGE biplot showing discriminating ability and representativeness of test environments. It was observed that Orin-Ekiti'20 and Ibadan'19 both had a long vector length followed by Eruwa'19, while Orin-Ekiti'19, Eruwa'20, and Ibadan'20 had a short vector length. Moreover, Ibadan'19 has the smallest angle with the Average Environment Axis (AEA), followed by Orin-Ekiti'19 and Eruwa'20. (In Figure 3, It should be explained what the long or short vectors and the wide or narrow angles mean). The biplot's computational framework facilitated the visualization of environment vector lengths, which are directly proportional to the standard deviation within each environment and reflect the environment's ability to distinguish genotypes. As a result, test environments with longer vectors demonstrated a stronger capacity for genotype differentiation, whereas shorter vectors provided minimal or no useful information regarding genotype variations (Yan et al. 2007). The mean coordinates of all test environments were represented within the mean environment, depicted as a small circle at the tip of the arrow along the line (Fig. 4). The axis extending from the biplot's origin to the mean environment is referred to as the axis of the mean environment (AME). Test environments with shorter angles relative to the AME were considered the most representative (Yan and Tinker, 2006).

The average-environment coordination (AEC) view showing the mean performance and stability of 15 PVA maize is presented in Figure 4. The circle surrounding the image represented genotype yield and stability, assessed using the mean environment coordination (MEC) approach. The axis of the ideal environment was defined by a line passing through both the ideal environment and the biplot's origin. The optimal environment was identified by analyzing the principal component scores of each habitat, with the arrow pointing toward the highest genotypic value. The coordinate axis, perpendicular to the abscissa, indicated genotypes with lower stability, primarily influenced by genotype-environment interactions. Additionally, this axis differentiated genotypes with performance records above and below the average (Yan and Tinker, 2006). Results show that F2TWLY100123 followed by ACR.91 SUWAN 1-SR C1, PVA SYN-13, and F2TWLY100121 were the highest-yielding varieties. However, genotypes PVA SYN HGB C0, PVA SYN HGB C2, and PVA SYN HGA C2 performed below average with PVA SYN HGB C0 being the least yielding variety. In addition, lines PVA SYN HGB C2, PVA SYN HGB C0, AFLATOXIN SYN-YF2, and PVA SYN-13? had a short length of projection while ACR.91 SUWAN 1-SR C1, F2SCA1413-36, F2TWLY100123, F2TWLY131228 and F2TWLY13124 had a long length.

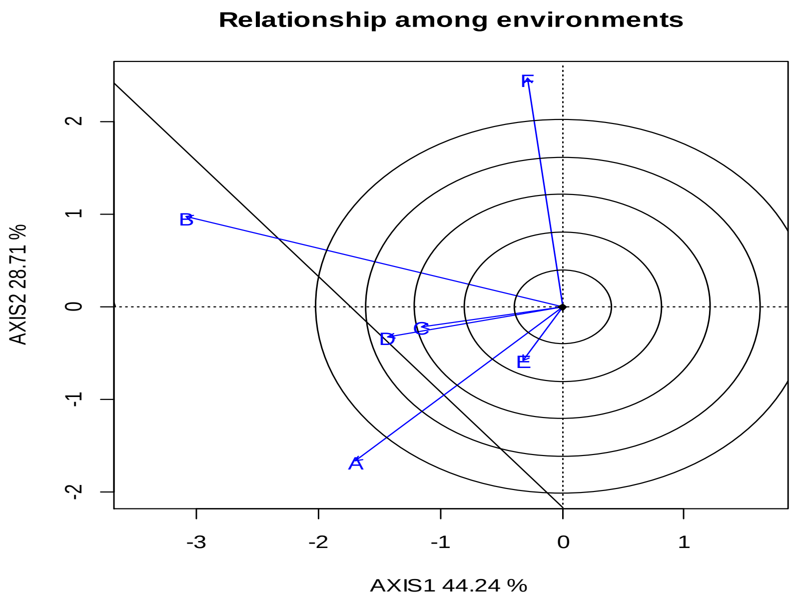

Figure 1. The environment-vector view of the GGE biplot showing similarities among test environments in discriminating PVA maize. Where A – Eruwa 2019; B – Ibadan 2019; C – Orin Ekiti 2019; D – Eruwa2020; E – Ibadan 2020; F - Orin Ekiti 2020

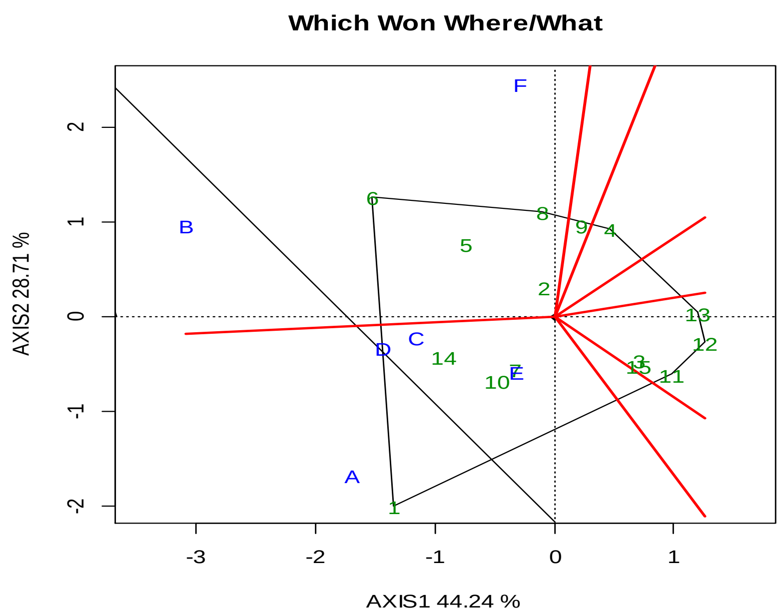

Figure 2. GGE biplot representing the which-won-where graph indicating the yield rankings of 15 OPVs. Where A – Eruwa 2019; B – Ibadan 2019; C – Orin Ekiti 2019; D – Eruwa2020; E – Ibadan 2020; F - Orin Ekiti 2020

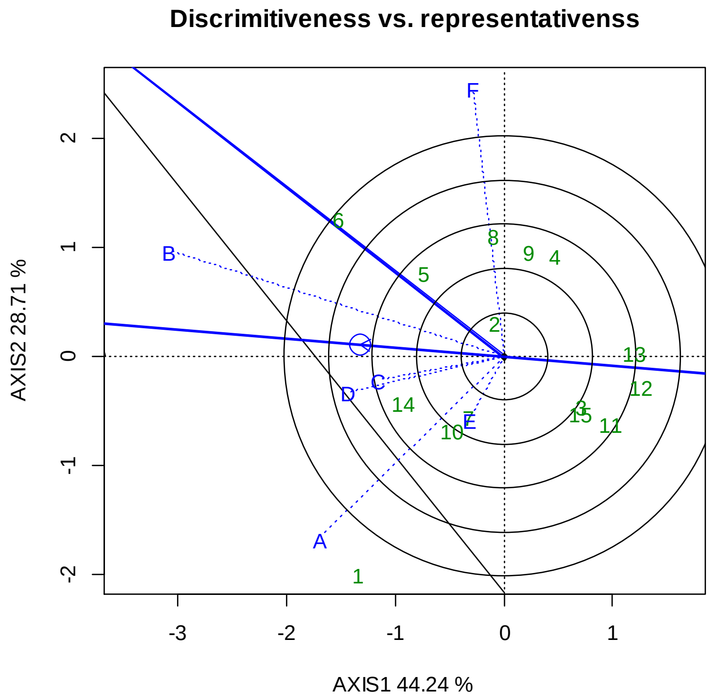

Figure 3. GGE biplot comparing 15 PVA OPVs evaluated according to the discriminating and representativeness of environments for grain yield (t ha -1). Where A – Eruwa 2019; B – Ibadan 2019; C – Orin Ekiti 2019; D – Eruwa 2020; E – Ibadan 2020; F - Orin Ekiti 2020

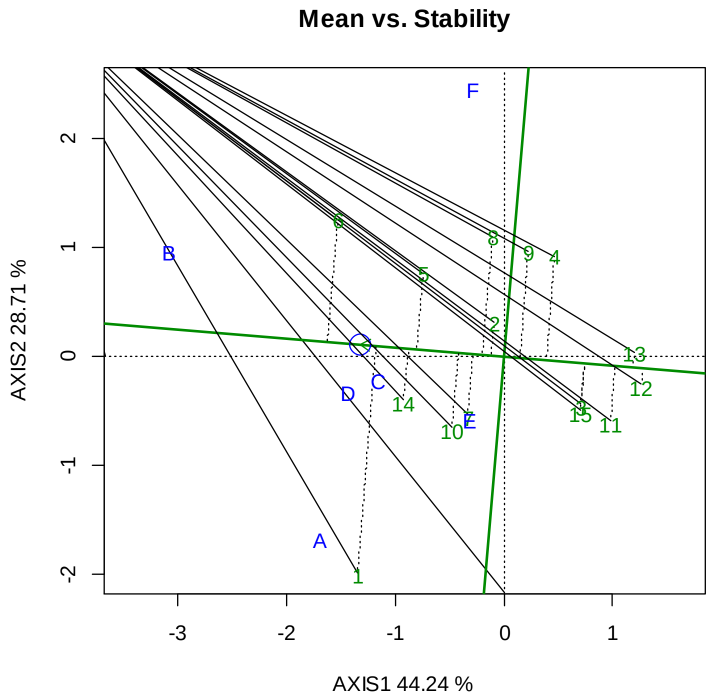

Figure 4. GGE biplot representing the 'mean vs. stability' indicating the yield rankings of 15 PVA OPVs and their respective production stabilities. Where A – Eruwa 2019; B – Ibadan 2019; C – Orin Ekiti 2019; D – Eruwa2020; E – Ibadan 2020; F - Orin Ekiti 2020;

3. Discussion

Comparing Nigeria's average maize production (1.7 t ha $^{-1}$) for the same period of years (1986–2011) with that of the United States (9.3 t ha^{-1}), it is relatively low (Olaniyan 2015; IITA 2020; Mani et al.2022). Since maize is an important staple food crop for the people of Sub-Saharan Africa, this has inspired breeding for high-yielding varieties of the crop (IITA 2009). Additionally, it is a widely distributed crop that is farmed by small-scale rural farmers. The goal of the current study was to assess maize varieties which are promising alternatives that were high-yielding, easily accessible, and affordable for use by small-scale farmers compared to expensive hybrid seeds.

The ongoing effects of climate change in the areas where subsistence farmers in Nigeria grow their crops have raised significant concerns, as the ideal conditions for production may increasingly be disrupted by erratic climate variations (Gaudin et al. 2015). The study was conducted in a range of environments that differed in terms of growing conditions, rainfall patterns, and various geographical and climatic factors. Assessing the yield stability of crops in diverse agroecosystems with high accuracy was highly emphasized by Mühleisen et al. (2014) and, therefore became imperative for optimum yield.

Different varieties of maize respond variably to environmental conditions, making it essential to improve stability in maize performance and select optimal genotypes for adaptation (Campos et al. 2006). Identifying ideal testing sites for superior and stable genotypes has become a key priority for maize breeders (Badu-Apraku et al. 2015a). The non-significant mean squares for grain yield in this study indicate remarkable stability in this trait across diverse testing environments. This stability suggests that a genotype showing promise in one locale is likely to perform well in other areas with similar agroecological characteristics. These findings align with Adu et al. (2013), who observed minimal genotype-by-environment (G × E) interactions among 54 maize genotypes across three sites in Ghana. Similarly, Ma et al. (2024) noted that while environmental factors significantly impacted yields, G × E interactions were not the dominant factor, emphasizing the need to select stable hybrids. Alam et al. (2022) echoed these conclusions, finding that environmental effects were notable, but G × E interactions were insignificant, indicating that certain hybrids perform consistently across various conditions.

The location's substantial mean square indicated that environmental factors significantly influenced the genetic potential of the genotypes. Koroma et al. (2017) similarly found that yield-limiting factors like minimum temperature, relative humidity, moisture stress, pests, and diseases largely impacted the genotype × environment interaction on maize yield. Do Couto et al. (2023) also observe that environmental variables accounted for a large portion of yield variation, emphasizing their influence on the genotypes' performance. Likewise, Ljubičić et al. (2023) reported that environmental factors predominantly determined yield variation, showing that maize genotype performance is highly dependent on the environmental context. These studies collectively highlight the substantial effect of environmental conditions on maize yield.

The anthesis-silking interval (ASI) is a critical trait in maize, as a prolonged ASI often leads to poor pollination and reduced kernel set, ultimately affecting grain yield (Bolaños and Edmeades 1996). The study revealed variations in ASI among genotypes indicating efficient pollination and a reduced risk of yield loss under stress conditions. On the other hand, long ASI could negatively impact grain filling. Similar findings by Araus et al. (2012) suggest that selecting genotypes with a short ASI enhances reproductive efficiency, particularly in drought-prone regions. Additionally, Tao et al. (2023) study demonstrated that overexpressing the ZmEXPA5 gene in maize reduced the ASI and improved grain yield under both drought and well-watered conditions.

Plant and ear heights influence lodging resistance and ease of mechanical harvesting (Xue et al. 2020). The observed differences in plant height among genotypes suggest varying genetic potentials for vegetative growth. The moderate ear heights observed in other genotypes suggest a balance between grain-filling potential and lodging resistance (Zhang et al. 2023). Taller plants have an advantage in capturing sunlight due to their elevated canopy, which can lead to increased photosynthesis and potentially higher yields. However, this increased height can make them more susceptible to lodging which is particularly problematic in high-wind environments (Stubbs et al. 2023). Conversely, shorter plants generally exhibit greater lodging resistance due to their reduced leverage and sturdier stature (Xue et al. 2020). This characteristic makes them more suitable for high-density planting systems, as they can maintain structural integrity even when planted closely together

Plant aspect, ear aspect, and husk cover are critical traits influencing maize grain quality and storability. Husk integrity is particularly vital in protecting ears from environmental stresses and biological threats. Drought and high temperatures before tasseling can cause ear extension beyond the husk, increasing susceptibility to mold, insect damage, and bird predation, which degrade grain quality. Thus, selecting maize varieties with well-filled ears and tight husk cover is essential for maintaining grain integrity (Jiang et al. 2020). Disease resistance is a key factor in maize production, especially in regions prone to fungal and bacterial infections. The local check variety exhibited higher disease scores, indicating greater susceptibility than improved OPVs. Breeding for disease resistance enhances yield stability and reduces reliance on chemical control measures (Badu-Apraku and Akinwale 2011). Abera et al. (2021) demonstrated that certain OPVs exhibit superior grain yield and resistance to Turcicum leaf blight, highlighting the potential of breeding programs in developing resilient varieties. Berger et al. (2020) reported yield losses of 36% to 72% in susceptible hybrids due to Northern Leaf Blight (NLB), whereas resistant hybrids showed no significant reductions, underscoring the success of local breeding efforts. Moreover, utilizing diverse germplasm and advanced breeding technologies can mitigate yield losses and reduce dependency on chemical controls (Garoma et al. 2024). This study reinforces the importance of breeding for disease resistance to ensure yield stability and promote sustainable agricultural practices.

The significant variation in grain yield among the open-pollinated maize genotypes (OPVs) across the three locations (Eruwa, Ibadan, and Orin-Ekiti) and the two cropping seasons (2019 and 2020) underscores the crucial influence of environmental factors, particularly rainfall distribution. The higher mean grain yield recorded in 2019 compared to 2020 suggests that adequate and well-distributed rainfall played a pivotal role in supporting maize productivity (Lobell et al. 2011; Obour et al. 2022). Liu et al. (2021) study emphasized that both the amount and distribution of rainfall are crucial for optimizing grain yield and water use efficiency in maize cultivation. Furthermore, Mekonnen et al. (2023) study investigating the effects of planting dates and environmental factors on maize grain yield reported that erratic and inconsistent rainfall distribution poses a significant threat to maize production. Ibadan consistently recorded the highest yields across both years, possibly due to relatively favorable climatic conditions, including adequate soil moisture retention and stable temperatures (Kamara et al. 2020).

The sharp reduction in grain yield in 2020, particularly in Eruwa, aligns with the pronounced decline in rainfall, emphasizing moisture stress as a key limiting factor in maize productivity. The relationship between rainfall and yield decline corroborates findings from previous studies, which established that prolonged moisture stress at critical growth stages, such as flowering and grain filling, significantly reduces maize yield potential (Bänziger et al. 2000). The drastic decline in Eruwa's annual rainfall corresponds with its substantial yield reduction, further validating the assertion that water availability is a fundamental determinant of maize performance (Campos et al. 2004; Bagula et al. 2022; Șimon et al. 2023).

Genotypic performance varied significantly across environments indicating that these genotypes possess favorable traits that enhance yield resilience under fluctuating climatic conditions (Badu-Apraku et al., 2015). Conversely, the lowest-yielding genotype exhibited lower adaptability, particularly under moisture-stressed conditions, suggesting that its genetic makeup may not confer sufficient drought tolerance (Edmeades et al., 1999). The commercial check (PVA SYN-13) and local check mean grain yield across the years indicate that while improved varieties offer superior yield advantages, locally adapted genotypes may still perform moderately under prevailing conditions. This agrees with Ficiciyan et al. (2018) research which found that while modern varieties typically yield more, their performance declines under harsh conditions. In contrast, traditional landraces remain resilient and widely cultivated by small-scale farmers, especially in nutrient-poor soils.

Moreover, besides rainfall, relative humidity (RH) and wind speed also influenced yield performance. The unexpected rise in RH in 2020, despite lower rainfall, may have increased disease pressure, particularly fungal infections like maize rust and gray leaf spot, which thrive in high humidity potentially reducing maize yields. Likewise, stronger winds, especially in Orin-Ekiti, may have caused lodging and reduced pollination efficiency, negatively impacting grain development (Tollenaar and Wu 1999; Akintibu et al. 2023). Although temperature fluctuations were minor, their interaction with other climatic factors likely influenced yield outcomes, highlighting the complexity of genotype-environment interactions (Chapman and Edmeades 1999; Hudson et al. 2022).

Close association existing among environments within the same group implies that similar results could be obtained from other environments within the group as regards the genotypes. In other words, since test environments A, C, D, and E are considered to be correlated, therefore one of these environments can ably represent the others without losing much information which could result in a possible reduction in the cost of testing material across environments. The long vector length observed for Orin-Ekiti'20 and Ibadan'19 positions these two environments as the most discriminating while Orin-Ekiti'19, Eruwa'20, and Ibadan'20 were the least discriminating as a result of short distances to the biplot origin. This suggests that less discriminating environments (Orin-Ekiti'19, Eruwa'20, and Ibadan'20) are not strongly associated with those with long vector test environments (Orin-Ekiti'20 and Ibadan'19) and should be treated as independent test environments (Badu-Apraku et al. 2011). Similarly, Yan and Tinker (2006) discussed the utility of GGE biplot analysis in evaluating test environments and genotypes. They noted that environments with longer vector lengths in the biplot are more discriminating, whereas those with shorter vectors are less discriminating and may not effectively distinguish between genotypes. Including such environments in multi-environment trials might not contribute valuable insights into genotype performance. Orin-Ekiti'20 and Ibadan'19 being the most discriminating environments are also said to be most informative among other environments. The small angle observed between environments (Ibadan'19, Orin-Ekiti'19, and Eruwa'20) and AEA shows that they can be regarded as the most representative of the six tested environments (Yan et al. 2007). This implies that any of these three test environments can represent the other.

Yan and Tinker (2006) suggest that test environments that are both discriminating and representative are effective for selecting genotypes that are generally well-adapted. By focusing on such environments, breeders can identify genotypes that not only perform well in specific conditions but also exhibit stable performance across diverse environments. Similarly, Yan et al. (2007) emphasize the importance of selecting test environments that combine high discriminating power with representativeness. They argue that this approach enhances the efficiency of breeding programs by ensuring that selected genotypes are likely to perform consistently across various target environments.

The selection of genotypes that are precisely suited, on the other hand, may benefit from environments that are discriminating but not representative, especially if the target environments can be divided into discrete mega-environments (Yan and Tinker 2006). Nonetheless, it can also help remove unstable genotypes in a single environment; it should not be utilized for selection. Consequently, PVA maize cultivars that are especially suited to this climate may be chosen using Orin-Ekiti'20, which is discriminating but non-representative. Moreover, Ibadan'19 which appears to be both discriminating and representative, is indicative of its effectiveness as a test environment for selecting varieties that are generally adaptable.

Genotypes PVA SYN HGB C2, PVA SYN HGB C0, AFLATOXIN SYN-YF2, and PVA SYN-13 were the most stable as a result of the short length of their projection (Yan et al. 2005). While ACR.91 SUWAN 1-SR C1 and F2TWLY100123 were high-yielding but appeared less stable. In this study, PVA SYN-13 and AFLATOXIN SYN-YF2 are regarded as stable and high-yielding PVA maize varieties due to their short projection distance and above-average mean performance.

4. Conclusions

The findings of this study emphasize the importance of developing and promoting maize genotypes with enhanced drought tolerance and disease resistance to mitigate yield losses caused by erratic weather patterns.

The absence of significant genotype-by-environment (G × E) interactions for grain yield suggests that well-performing genotypes at one site are likely to be suitable for cultivation in other locations within the same agroecological zone. However, environmental factors still played a crucial role in genotype performance, underscoring the need for adaptive breeding strategies. Among the evaluated genotypes, AFLATOXIN SYN-YF2 and PVA SYN 13 demonstrated both high yield potential and stability, making them ideal for cultivation across multiple locations, including Eruwa, Ibadan, and Orin-Ekiti. Additionally, F2TWLY100123 and ACR.91 SUWAN 1-SR C1 exhibited strong adaptability and superior grain yield, particularly in moisture-limited environments, highlighting their potential for further breeding and large-scale adoption. Genotypes such as PVA SYN HGB C2 and F2TWLY131211, characterized by short anthesis-silking intervals (ASI) and moderate plant height, offer valuable traits for breeding drought-resilient maize varieties. To enhance maize productivity under changing climatic conditions, agronomic interventions such as moisture conservation practices, timely planting, and integrated disease management strategies should be prioritized. Future research should focus on the long-term impact of climatic variables on maize performance and explore breeding strategies that incorporate drought and disease resistance traits for sustainable production.

Furthermore, the observed variation in grain yield and agronomic traits provides valuable insights for breeding programs targeting high-yielding and stress-tolerant maize varieties. Given their promising performance, the evaluated genotypes warrant further testing for potential release to resource-limited farmers in southwestern Nigeria, contributing to improved food security and agricultural resilience in the region.

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References

Abakemal D, Shimelis H, Derera J (2016). Genotype-by-environment interaction and yield stability of quality protein maize hybrids developed from tropical-highland adapted inbred lines. *Euphytica,* 209: 757–769. https://doi.org/10.1007/s10681-016-1673-7

Abera N, Odong T, Kasozi, LC (2021). Phenotypic diversity and correlation coefficient analysis of open-pollinated maize varieties in Uganda. *Global Journal of Agricultural Research, 9*(1): 36–47. https://doi.org/10.37745/gjar.2013

Adu GB, Akromah R, Abdulai MS, Obeng-Antwi K, Kena AW, Tengan KML, Alidu H (2013). Assessment of genotype by environment interactions and grain yield performance of extra-early maize (*Zea mays* L.) hybrids*. Journal of Biology, Agriculture and Healthcare,* 3(12): 7-15.

Akcura M, Taner S, Kaya Y (2011). Evaluation of bread wheat genotypes under irrigated multi-environment conditions using GGE biplot analyses. *Agriculture,* 98: 35–40.

Akintibu TS, Oluwaranti A, Fakorede MAB (2023). The impacts of climatic variability on the grain yield performance of open-pollinated varieties (OPV) of maize in a rainforest location of Nigeria. *Journal of Agriculture and Ecology Research International, 24*(5): 28–36. https://doi.org/10.9734/JAERI/2023/v24i5539

Alam MA, Rahman M, Ahmed S, Jahan N, Khan MA, Islam MR, Alsuhaibani AM, Gaber A, Hossain A (2022). Genetic variation and genotype by environment interaction for agronomic traits in maize (*Zea mays* L.) hybrids. *Plants,* 11: 1-16. https://doi.org/10.3390/plants11111522

Araus JL, Serret MD, Edmeades GO (2012). Phenotyping maize for adaptation to drought. *Frontiers in Physiology, 3*: 305. https://doi.org/10.3389/fphys.2012.00305

Avnee S, Sood S, Chaudhary DR, Jhorar P, Rana RS (2023). Biofortification: An approach to eradicate micronutrient deficiency. *Frontiers in Nutrition,* 10: 1233070. https://doi.org/10.3389/fnut.2023.1233070

Badu-Apraku B, Fakorede M, Oyekunle M, Akinwale R (2015). Genetic gains in grain yield under nitrogen stress following three decades of breeding for drought tolerance and *Striga* resistance in early maturing maize. *The Journal of Agricultural Science, 1*(4): 1–15. https://doi.org/10.1017/S0021859615000593

Bankole FA, Olajide OO, Olaoye G (2023). Performance and yield stability of quality protein maize (*Zea mays* L.) hybrids under rainfed condition. *Agriculture* (Poľnohospodárstvo), 69(2): 66–76. https://doi.org/10.2478/agri-2023-0006

Bassa D, Goa Y (2016). Performance evaluation and adaptation of improved maize (*Zea Mays* L) varieties for Highland of Alicho, Silti and Analemo Districts of Southern Ethiopia. *Journal of Natural Sciences Research,* 6(15): 28–32.

Badu-Apraku B, Akinwale R (2011). Identification of early-maturing maize inbred lines based on multiple traits under drought and low N environments for hybrid development and population improvement. *Canadian Journal of Plant Science, 91*(5), 931–942. https://doi.org/10.4141/cjps2011-024

Badu-Apraku B, Lum AF, Akinwale RO, Oyekunle M (2011a). Biplot analysis of diallel crosses of early maturing tropical yellow maize inbreds in stress and non-stress environments. *Crop Science,* 51: 173-188.

Badu-Apraku B, Fakorede MAB, Menkir A, Sanogo D (2012). Conduct and management of maize field trials. IITA, Ibadan, Nigeria. pp. 1-59.

Badu-Apraku B, Annor B, Oyekunle M, Akinwale RO, Fakorede MAB, Talabi AO, Akaogu IC, Melaku G, Fasanmade Y (2015a). Grouping of early maturing quality protein maize inbreds based on SNP markers and combining ability under multiple environments. *Field Crops Research,* 183: 169–183.

Bagula EM, Majaliwa JGM, Basamba TA, Mondo JGM, Vanlauwe B, Gabiri G, Tumuhairwe B, Mushagalusa GN, Musinguzi P, Akelo S (2022). Water use efficiency of maize (*Zea mays* L.) crop under selected soil and water conservation practices along the slope gradient in Ruzizi Watershed, Eastern D.R. Congo. *Land, 11*(10): 1833. https://doi.org/10.3390/land11101833

Bänziger M, Edmeades GO, Beck D, Bellon M (2000). Breeding for drought and nitrogen stress tolerance in maize: From theory to practice. *CIMMYT.*

Berger DK, Mokgobu T, de Ridder K, Christie N, Aveling TA (2020). Benefits of maize resistance breeding and chemical control against northern leaf blight in smallholder farms in South Africa. *South African Journal of Science, 116*(11–12), 1–7. https://doi.org/10.17159/sajs.2020/8286

Bocianowski J, Nowosad K, Rejek D (2024). Genotype-environment interaction for grain yield in maize (*Zea mays* L.) using the additive main effects and multiplicative interaction (AMMI) model. *Journal of Applied Genetics*,65: 653-664. https://doi.org/10.1007/s13353-024-00899-4

Bolaños J, Edmeades GO (1996). The importance of the anthesis-silking interval in breeding for drought tolerance in tropical maize. *Field Crops Research, 48*(1): 65–80. https://doi.org/10.1016/0378-4290(96)00036-6

Bouis HE, Saltzman A (2017). Improving nutrition through biofortification: a review of evidence from HarvestPlus, 2003 through 2016. *Global Food Security,* 12: 49–58. https://doi.org/10.1016/j.gfs.2017.01.009

Campos H, Cooper M, Habben JE, Edmeades GO, Schussler JR (2004). Improving drought tolerance in maize: A view from industry. *Field Crops Research, 90*(1): 19-34.

Campos H, Cooper M, Edmeades GO, Löffler C, Schussler JR, Ibañez M (2006). Changes in drought tolerance in maize associated with fifty years of breeding for yield in the U.S. Corn Belt. *Maydica,* 51: 369-381.

Chapman SC, Edmeades GO (1999). Selection improves drought tolerance in tropical maize populations: II. Direct and correlated responses among secondary traits. *Crop Science, 39*(5): 1315-1324.

Daemo BB, Ashango Z (2024). Application of AMMI and GGE biplot for genotype by environment interaction and yield stability analysis in potato genotypes grown in Dawuro Zone, Ethiopia. *Journal of Agriculture and Food Research,* 18(2): 1-9. https://doi.org/10.1016/j.jafr.2024.101287

Do Couto DP, Oliveira WB, de Oliveira JS, Guilhen JH, Bernardes CD, Posse SC, Ferreira MF, Ferreira A (2023). Analysis of the effect of the interaction of genotype and environment on the yield stability of maize varieties. genetic resources for breeding. *Agronomy*, 13(8): 1-15. https://doi.org/10.3390/agronomy13081970

Dosho BM, Ifie BE, Asante IK, Danquah EY, Zeleke H (2022). Genotype-by-environment interaction and yield stability for grain yield of quality protein maize hybrids under low and optimum soil nitrogen environments. *Journal of Crop Science and Biotechnology,* 25: 437–450. https://doi.org/10.1007/s12892-022-00143-7

Ebbisa A (2022). Mechanisms underlying cereal/legume intercropping as nature-based biofortification: A review. *Food Production, Processing and Nutrition,* 4(19): 1-17. https://doi.org/10.1186/s43014-022-00096-y

Edmeades GO, Bolaños J, Chapman SC, Lafitte HR, Bänziger M (1999). Selection improves drought tolerance in tropical maize populations: Gains in biomass, grain yield, and harvest index. *Crop Science, 39*(5): 1306-1315.

Eze CE, Akinwale RO, Michel S, Burstmayr H (2020). Grain yield and stability of tropical maize hybrids developed from elite cultivars in contrasting environments under a rainforest agro-ecology. *Euphytica,* 216: 1-13. https://doi.org/10.1007/s10681-020-02620-y

FAOSTAT (2021). FAO Stat. FAO, Rome. http://www.fao.org/faostat (access date: 12.04.2022)

Fan XM, Kang MS, Chen H, Zhang Y, Tan J, Xu C (2007). Yield stability of maize hybrids evaluated in multi-environment trials in Yunnan, China. *Agronomy Journal,* 99: 220-228.

Ficiciyan A, Loos J, Sievers-Glotzbach S, Tscharntke T (2018). More than yield: Ecosystem services of traditional versus modern crop varieties revisited. *Sustainability*, *10*(8), 2834. https://doi.org/10.3390/su10082834

Gannon B, Kaliwile C, Arscott SA, Schmaelzle S, Chileshe J, Kalungwana N, Mosonda M, Pixley K, Masi C, Tanumihardjo SA (2014). Biofortified orange maize is as efficacious as a vitamin A supplement in Zambian children even in the presence of high liver reserves of vitamin A: A community-based, randomized placebo-controlled trial. *The American Journal of Clinical Nutrition, 100*(6): 1541–1550. https://doi.org/10.3945/ajcn.114.087379

Garoma B, Yadessa L, Tilahun B, Asefa G (2024). Comprehensive breeding approach for resistance to disease in crops: Research review. *Journal of Soil Science and Plant Physiology, 6*(3): 1-8. https://doi.org/10.36266/JSSPP/181

Gaudin ACM, Tolhurst TN, Ker AP, Janovicek K, Tortora C, Martin RC, Deen W (2015). Increasing crop diversity mitigates weather variations and improves yield stability. *PLoS ONE* 10(2): 1-20. https://doi.org/10.1371/journal.pone.0113261

Goredema-Matongera N, Ndhlela T, Magorokosho C, Kamutando CN, van Biljon A, Labuschagne M (2021). Multinutrient biofortification of maize (*Zea mays* L.) in Africa: Current status, opportunities, and limitations. *Nutrients, 13*(3): 1039. https://doi.org/10.3390/nu13031039

Guo S, Liu Z, Zhou Z, Lu T, Chen S, He M, Zeng X, Chen K, Yu H, Shangguan Y, Dong Y, Chen F, Liu Y, Qin Y (2022). Root System architecture differences of maize cultivars affect yield and nitrogen accumulation in Southwest China. *Agriculture,* 12: 1-14. https://doi.org/10.3390/agriculture12020209.

Hotz C, Chileshe J, Siamusantu W, Palaniappan U, Kafwembe E (2012). Vitamin A intake and infection are associated with plasma retinol among pre-school children in rural Zambia. *Public Health Nutrition, 15*(9): 1688–1696. https://doi.org/10.1017/S1368980012002731

Hodge C, Taylor C (2023). Vitamin A deficiency. In *StatPearls* [Internet]. StatPearls Publishing. Retrieved from https://www.ncbi.nlm.nih.gov/books/NBK567789/

Huang Z, Liu Y, Qi, G, Brand D, Zheng SG (2018). Role of vitamin A in the immune system. *Journal of Clinical Medicine, 7*(9): 258. https://doi.org/10.3390/jcm7090258

Hudson AI, Odell SG, Dubreuil P, Tixier MH, Praud S, Runcie DE, Ross-Ibarra J (2022). Analysis of genotype-by-environment interactions in a maize mapping population. *G3: Genes, Genomes, Genetics, 12*(3): jkac013. https://doi.org/10.1093/g3journal/jkac013

IITA (2009). Crops. http://www.iita.org (Access date: 01.05.2014).

IITA (2020). IITA-BIP sets record for maize production per hectare in Nigeria. https://www.iita.org/news-item/iita-bip-sets-record-for-maize-production-per-hectare-in-nigeria/ (access date:02.12.2021)

Koroma MS, Swaray M, Akromah R, Obeng-Antwi K (2017). Genotype by environment interaction and stability of extra-early maize hybrids (*Zea mays* L.) for yield evaluated under irrigation. *International Journal of Environment, Agriculture and Biotechnology,* 2: 2573-2580. https://doi.org/10.22161/ijeab/2.5.39.

Kutka F (2011). Open-Pollinated vs. hybrid maize cultivars. *Sustainability* 3: 1531–1554. https://doi.org/10.3390/su3091531

Kumar B, Choudhary M, Kumar P, Kumar S, Sravani D, Vinodhana NK, Kumar GS, Gami R, Vyas M, Jat BS, Dagla MC, Rakshit S (2024). GGE biplot analysis and selection indices for yield and stability assessment of maize (*Zea mays* L.) genotypes under drought and irrigated conditions. *Indian Journal of Genetics and Plant Breeding,* 84(02): 209–215. https://doi.org/10.31742/ISGPB.84.2.8

Jiang S, Zhang H, Ni P, Yu S, Dong H, Zhang A, Cao H, Zhang L, Ruan Y, Cui Z (2020). Genome-wide association study dissects the genetic architecture of maize husk tightness. *Frontiers in Plant Science,* 11:861. https://doi.org/10.3389/fpls.2020.00861

Kamara AY, Kamai N, Omoigui LO, Togola A, Onyibe JE (2020). *Guide to maize production in northern Nigeria*. International Institute of Tropical Agriculture (IITA).

Liu Y, Hou P, Huang G, Zhong X., Li, H, Zhao J, Li S, Mei X (2021). Maize grain yield and water use efficiency in relation to climatic factors and plant population in northern China. *Journal of Integrative Agriculture, 20*(12): 3156–3169. https://doi.org/10.1016/S2095-3119(20)63428-1

Ljubičić N, Popović V, Kostić M, Pajić M, Bude M, Gligorević K, Dražić M, Bižić M, Crnojević V (2023). Multivariate interaction analysis of *Zea mays* L. genotypes growth productivity in different environmental conditions. *Plants,* 12(11): 1-25.

Ma C, Liu C, Ye Z (2024). Influence of Genotype × environment interaction on yield stability of maize hybrids with AMMI model and GGE biplot. *Agronomy,* 14: 1-16. Doi: 10.3390/agronomy14051000

Mani JR, Issah FO, Abdussalam Z, Damisa MA (2022). Factors influencing farmer participation in maize production in Kaduna State, Nigeria. *Journal of Agriculture and Environment* 18(1): 1–11.

Mekonnen TW, van Biljon A, Ceronio G Labuschagne M (2023). Effects of planting date, environments, and their interaction on grain yield and quality traits of maize hybrids. *Heliyon, 9*(11): e21660. https://doi.org/10.1016/j.heliyon.2023.e21660

Mu̇hleisen J, Piepho H-P, Maurer H-P, Longin CFH, Reif JC (2014a). Yield stability of hybrids versus lines in wheat, barley and triticale. *Theoretical and Applied Genetics,* 127: 309–316.

Obour PB, Arthur IK, Owusu K (2022). The 2020 Maize production failure in Ghana: A case study of ejura-sekyedumase municipality. *Sustainability*, *14*(6): 3514. https://doi.org/10.3390/su14063514

Olakojo SA, Iken JE (2001). Yield performance and stability of some improved maize varieties. *Moor Journal of Agricultural Research,* 2: 21-24.

Olaoye G, Menkir A, Ajala SO, Jacob S (2009). Evaluation of local maize (*Zea mays* L.) varieties from Burkina Faso as source of tolerance to drought. *Journal of Applied Bioscience,* 17: 887-898.

Olaniyan AB (2015). Maize: Panacea for hunger in Nigeria. *African Journal of Plant Science,* 9(3): 155–174.

Onyeneke RU, Amadi MU, Anosike FC (2019). Biofortification in Nigeria: A systematic review. *AIMS Agriculture and Food, 4*(4): 892–906. https://doi.org/10.3934/agrfood.2019.4.892

Oyekunle M, Haruna A, Badu-Apraku B, Usman IS, Mani H, Ado SG, Olaoye G, Obeng-Antwi K, Abdulmalik RO, Ahmed HO (2017). Assessment of early-maturing maize hybrids and testing sites using GGE biplot analysis. *Crop Science,* 57: 2942-2950. https://doi.org/10.2135/cropsci2016.12.1014

Prasanna BM, Palacios-Rojas N, Hossain F, Muthusamy V, Menkir A, Dhliwayo T, Ndhlela T, San Vicente F, Nair SK, Vivek BS, Zhang X, Olsen M, Fan X (2020). Molecular breeding for nutritionally enriched maize: status and prospects. *Frontiers in Genetics,* 10: 1-16. https://doi.org/10.3389/fgene.2019.01392

Rahman MM, Wahed MA, Fuchs GJ, Baqui AH, Alvarez JO (2002). Synergistic effect of zinc and vitamin A on the biochemical indexes of vitamin A nutrition in children. *American Journal of Clinical Nutrition, 75*(1), 92–98. https://doi.org/10.1093/ajcn/75.1.92

Shrestha J (2013). Genotype by environment interaction and yield stability analysis of quality protein maize genotypes in the Terai region of Nepal. *International Journal of Applied Sciences and Biotechnology,* 1(2): 75–79.

Sibiya J, Tongoona P, Derera J, Rij N (2012). Genetic analysis and genotype by environment (G x E) for grey leaf spot disease resistance in elite African maize (*Zea mays* L.) germplasm. *Euphytica,* 185: 349–362.

Șimon A, Moraru PI, Ceclan A, Russu F, Chețan F, Bărdaș M, Popa A, Rusu T, Pop AI, Bogdan I (2023). The impact of climatic factors on the development stages of maize crop in the Transylvanian plain. *Agronomy*, *13*(6): 1612. https://doi.org/10.3390/agronomy13061612

Song P, Adeloye D, Li S, Zhao D, Ye X, Pan Q, Qiu Y, Zhang R, Rudan I, Global Health Epidemiology Research Group (GHERG) (2023). The prevalence of vitamin A deficiency and its public health significance in children in low- and middle-income countries: A systematic review and modelling analysis. *Journal of Global Health, 13*: 04084. https://doi.org/10.7189/jogh.13.04084

Ssentongo P, Ba DM, Ssentongo AE, Fronterre C, Whalen A, Yang Y, Ericson JE, Chinchilli VM (2020). Association of vitamin A deficiency with early childhood stunting in Uganda: A population-based cross-sectional study. *PLoS ONE, 15*(5): e0233615. https://doi.org/10.1371/journal.pone.0233615

Statista (2023). Production quantity of maize in Nigeria. Available at https://www.statista.com/statistics/1300743/production-volume-of-maize-in-nigeria/ (access date: 05.03.2025).

Stubbs CJ, Kunduru B, Bokros N, Verges V, Porter J, Cook DD, DeBolt S, McMahan C, Sekhon RS, Robertson DJ (2023). Moving toward short stature maize: The effect of plant height on maize stalk lodging resistance. *Field Crops Research, 300*:109008. https://doi.org/10.1016/j.fcr.2023.109008

Tao K, Li Y, Hu Y, Li Y, Zhang D, Li C, He G, Song Y, Shi Y, Li Y, Wang T, Lu Y, Liu X (2023). Overexpression of *ZmEXPA5* reduces anthesis-silking interval and increases grain yield under drought and well-watered conditions in maize. *Molecular Breeding, 43*(12): 84. https://doi.org/10.1007/s11032-023-01432-x

Tollenaar M, Wu J (1999). Yield improvement in temperate maize is attributable to greater stress tolerance. *Crop Science, 39*(6): 1597-1604.

Trethowan R, Crossa J, van Ginkel M, Rajaram S (2001). Relationships among bread wheat international yield testing locations in dry areas. *Crop Science 41*: 1461–1469. https://doi.org/10.2135/cropsci2001.4151461x.

Villamor E, Fawzi WW (2000). Vitamin A supplementation: Implications for morbidity and mortality in children. *The Journal of Infectious Diseases, 182*(Suppl_1): S122–S133. https://doi.org/10.1086/315921

Wirth JP, Petry N, Tanumihardjo SA, Rogers LM, McLean E, Greig A, Garrett GS, Klemm RD, Rohner F (2017). Vitamin A supplementation programs and country-level evidence of vitamin A deficiency. *Nutrients, 9*(3): 190. https://doi.org/10.3390/nu9030190

Wossen T, Menkir A, Alene A, Abdoulaye T, Ajala S, Badu-Apraku B, Gedil M, Mengesha W, Meseka S (2023). Drivers of transformation of the maize sector in Nigeria. *Global Food Security, 38*: 100713. https://doi.org/10.1016/j.gfs.2023.100713

Xue J, Ming B, Xie R, Wang K, Hou P, Li S (2020). Evaluation of maize lodging resistance based on the critical wind speed of stalk breaking during the late growth stage. *Plant Methods, 16*, 148: 1-12. https://doi.org/10.1186/s13007-020-00678-0

Yan W, Rajcan I (2002). Biplot evaluation of test sites and trait relations of soybean in Ontario. *Crop Science* 42: 11–20.

Yan W, Tinker NA, Falk D (2005). QTL identification, mega-environment classification, and strategy development for marker-based selection using biplots. *Journal of Crop Improvement* 14: 299–324.

Yan W, Tinker NA (2006). Biplot analysis of multi-environment trial data: Principles and applications. *Canadian Journal of Plant Science* 86(3): 623–645. https://doi.org/10.4141/P05-169

Yan W, Kang MS, Ma B, Woods S, Cornelius PL (2007). GGE biplot vs. AMMI analysis of genotype-by-environment data. *Crop Science* 47: 643–655.

Yan W (2014). Crop variety trials: data management and analysis. Chichester: John Wiley and Sons.

Yong H, Zhang F, Tang J, Yang Z, Zhao X, Li M, Zhang D, Hao Z, Weng J, Li X (2019). Breeding potential of inbred lines derived from five maize (*Zea mays* L.) populations. *Euphytica*, 215(1): 1–12. https://doi.org/10.1007/s10681-018-2319-8.

Zhang P, Gu S, Wang Y, Xu C, Zhao Y, Liu X, Wang P, Huang S (2023). The relationships between maize (*Zea mays* L.) lodging resistance and yield formation depend on dry matter allocation to ear and stem. *The Crop Journal, 11*(1): 258–268. https://doi.org/10.1016/j.cj.2022.04.020

APPENDIX 1. Summary of Weather Parameters for Ibadan, Eruwa, and Orin-Ekiti (2019 & 2020)

Month	**Ibadan 2019**				**Ibadan 2020**				**Eruwa 2019**				**Eruwa 2020**				**Orin-Ekiti 2019**				**Orin-Ekiti 2020**			
Parameter	**RF (mm)**	**T (°C)**	**WS (kmph)**	**RH (%)**	**RF (mm)**	**T (°C)**	**WS (kmph)**	**RH (%)**	**RF (mm)**	**T (°C)**	**WS (kmph)**	**RH (%)**	**RF (mm)**	**T (°C)**	**WS (kmph)**	**RH (%)**	**RF (mm)**	**T (°C)**	**WS (kmph)**	**RH (%)**	**RF (mm)**	**T (°C)**	**WS (kmph)**	**RH (%)**
Jan	24.00	30	5.6	50	0.90	30	5.2	51	26.10	31	6.6	49	0.50	30	5.5	51	12.80	28	5.9	41	1.60	28	5.7	36
Feb	40.14	30	6.7	55	1.50	31	5.4	59	28.58	31	8.0	51	1.00	31	6.0	58	53.61	28	6.8	50	0.90	29	5.8	45
Mar	77.97	30	7.9	63	84.71	29	8.6	72	62.3	31	9.2	59	100.90	30	9.5	69	105.77	28	8.8	67	106.98	27	9.5	73
Apr	96.11	29	8.3	68	71.49	29	8.4	76	93.63	30	9.5	64	61.32	29	9.2	73	107.03	27	9.7	75	90.69	27	9.7	79
May	166.3	28	8.2	69	150.44	27	7.3	82	143.5	29	9.0	66	135.91	28	7.6	81	251.33	26	10.4	80	168.67	26	8.3	85
Jun	216.66	25	7.3	87	186.19	25	7.3	86	245.36	26	7.7	85	187.72	26	7.7	87	268.39	24	8.6	89	199.18	24	8.6	90
Jul	226.44	24	7.0	90	197.82	24	7.7	89	252.37	25	7.4	90	204.58	24	8.0	89	279.75	23	8.1	91	235.91	22	9.0	92
Aug	230.89	24	7.6	90	60.2	24	8.1	84	217.4	25	8.2	89	41.94	24	8.5	85	214.76	23	8.7	90	83.97	22	9.7	88
Sep	276.22	25	6.5	90	245.56	24	7.0	91	288.54	25	7.2	89	171.05	24	7.4	91	371.16	24	7.1	90	393.58	22	8.1	93
Oct	348.22	25	5.8	89	93.09	26	5.3	86	381.13	25	6.0	88	97.02	26	5.8	87	380.47	23	6.1	89	205.79	24	5.8	86
Nov	40.26	27	4.6	77	9.40	28	4.7	77	36.08	28	5.1	75	10.46	28	5.3	75	53.77	26	4.9	71	9.60	27	4.6	70
Dec	1.40	29	4.3	49	3.20	28	6.0	73	2.40	29	5.1	49	1.10	29	6.7	69	1.1	28	4.4	33	1.10	27	6.4	68
Total	1774.61	326	79.80	877	1104.50	325	81.00	926	1777.39	335	89.00	854	1013.50	329	87.20	915	2099.94	308	89.50	866	1497.97	305	91.20	905
Mean	145.38	27.17	6.65	73.08	92.04	27.08	6.75	77.17	148.12	27.92	7.42	71.17	84.46	27.42	7.27	76.25	175.00	25.67	7.46	72.17	124.83	25.42	7.60	75.42

Note: RF – Rainfall (mm), T – Temperature (°C), WS – Wind Speed (kmph), RH – Relative Humidity (%)

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Research Article

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Antioxidant Capacity and Quality Parameters of Early Maturing Soybean Genotypes Under Sivas Ecological Conditions

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HIGHLIGHTS

- Twenty-five soybean genotypes and five registered soybean varieties (Arısoy, Traksoy, Samsoy, Soyanam and Ataem-7) were used as materials.
- This study aimed to evaluate the antioxidant capacity, moisture, and ash content of early-maturing soybean genotypes cultivated under the Sivas ecological conditions.

Abstract

Soybean *(Glycine max* L.) is a versatile crop characterized by its cholesterol- and saturated fat-free composition and high-quality protein content. Evaluating parameters such as antioxidant capacity, moisture, and ash content is crucial for assessing the nutritional quality of soybeans, which are widely consumed both globally and in Türkiye. Antioxidants are compounds that mitigate or neutralize the harmful effects of free radicals in the body. Dietary natural antioxidants are among the most critical factors for enhancing the body's antioxidant defense system. This study aimed to evaluate the antioxidant capacity, moisture, and ash content of early-maturing soybean genotypes cultivated under the Sivas ecological conditions. The highest antioxidant activity using the ABTS method was observed in the ÜNV-2 genotype (11.82 µmol TE/g dw), while the DPPH method revealed the ÜNV-15 genotype as the highest (4.03 µmol TE/g dw). The moisture content of the soybean genotypes and varieties used in this study ranged from 8.75% to 12.34%, while the ash content varied between 2.86% and 4.05%. Differences in all investigated traits among the samples were statistically significant at the 1% level. As a result, the ÜNV-2 and ÜNV-15 genotypes were prioritized due to their relatively higher antioxidant activity, and the Ataem-7 variety was preferred for its lower moisture content.

Keywords: *Glycine max* L.; DPPH; ABTS; quality traits

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1. Introduction

Soybean (*Glycine max* L.) is a plant that stands out for its rich protein and oil content, yet its antioxidant potential remains an area ripe for exploration. Antioxidants are compounds that reduce or neutralize the harmful effects of free radicals in the body. Dietary natural antioxidants play a pivotal role in enhancing the body's antioxidant defense system (Seeram et al. 2006; Yu et al. 2006). Bioactive components in foods directly influence human health. Scientific studies have shown that polyphenolic antioxidant compounds derived from plants mitigate the effects of unstable and harmful substances known as free radicals. Free radicals can arise from enzymatic reactions and environmental factors such as cigarette smoke, air pollution, UV radiation, and radiation exposure (Young and Woodside 2001). These radicals interact with lipids, proteins, and DNA, disrupting their normal functions. Damage caused by free radicals is suggested to be at the root of various health conditions, including heart disease, diabetes, cancer, and aging (Akkuş 1995; Bayram et al. 2019). Antioxidants prevent cellular damage by scavenging existing radicals (Kahkönen et al. 1999).

Research indicates that increasing antioxidant intake through daily consumption of plant-based foods is essential (Prior and Cao 2000; Kılınç and Kılınç 2002; Arbos et al. 2008). Factors that determine the significance of antioxidants for human health include their chemical structures, solubility, structure-activity relationships, and natural availability (Kaur and Kapoor 2001). In addition to antioxidants, parameters like moisture and ash content are widely used as indicators of food quality. The ash content of a food item refers to the inorganic residue left after the combustion of organic matter, which results in the formation of water and carbon dioxide, leaving behind a mineral-rich inorganic fraction. Plants are the primary source of minerals for humans, who obtain them through plant-based foods, water, and animal-derived products. Most minerals in foods are bound to organic compounds such as proteins, fats, and carbohydrates. Ash determination is a crucial method for assessing the quality of food products. Moisture determination is among the most fundamental analyses in food processing and quality control. The water content of food significantly affects its shelf life, as dry matter and water content are inversely proportional. As water content increases, dry matter decreases, and vice versa (Karaköy et al. 2012, Soretire and Olayinka 2013; Güler and Emeksiz 2014, Çilesiz et al. 2023).

This study aimed to evaluate the antioxidant capacity, moisture, and ash content of early-maturing soybean genotypes grown under the Sivas ecological conditions.

2. Materials and Methods

This study utilized 25 soybean genotypes and five registered soybean varieties (Arısoy, Traksoy, Samsoy, Soyanam, and Ataem-7) as plant material. Arısoy; Thanks to its high adaptability, it has a high yield potential in fields with different soil structures. In Arısoy soybean seeds, the average first pod height is 15 cm over the years, preventing harvest losses. It is suitable for both main and second crop cultivation in Çukurova, Aegean, and Southeastern Anatolia regions, as well as for main crop cultivation in the Black Sea and Central Anatolia regions. As Arısoy soybean belongs to the medium-early maturity group, it provides earliness. It has high tolerance to whitefly and high tolerance to pod blight disease. It also exhibits tolerance to charcoal rot disease. In the second crop, sowing should be performed at a rate of 8–10 kg of seeds per decare. Arısoy soybean is suitable for both main and second crop soybean farming under Çukurova conditions. It is also appropriate for main crop cultivation in Southeastern Anatolia and the Black Sea regions. Samsoy; Developed through the selection breeding method, this variety belongs to the medium-early maturity group (143 days) and has a determinate growth habit. The plant exhibits a semi-erect structure with an average height of 115 cm, a first pod height of 15 cm, and white flowers. The seeds are round-flat in shape and light brown in color. The plant produces an average of 90 pods, with a thousand-seed weight of 202 g and a yield of 435 kg/da. It has no lodging or seed shattering issues. The protein content is 42.83%, while the oil content is 21.7%. Recommended cultivation areas include the Black Sea, Marmara, and transitional regions as a main crop, and the Mediterranean and Southeastern Anatolia regions as a second crop. Traksoy; Developed through hybridization (NE 3399 × Iroquois S01-02-0S-6T-1T-2T-1T-0T) by the Trakya Agricultural Research Institute, this variety belongs to the second maturity group and has purple flowers. The pod color is dark gray (brownish), while the pubescence color is gray. The seed hilum is grayish-black. The plant height ranges from

100 to 110 cm and is resistant to lodging. It produces 60–80 pods per plant, with a first pod height of 12–14 cm. The thousand-seed weight is 180–190 g. The protein content is 39.3%, and the oil content is 22.0%. It can be cultivated as a main crop in all regions (Trakya-Marmara, Black Sea, Aegean, Mediterranean, and Southeastern Anatolia). Additionally, it is suitable for second-crop cultivation in the Çukurova and Trakya regions. Soyanam (Kristal22); Developed through hybridization by the Trakya Agricultural Research Institute, this variety belongs to the third maturity group and has purple flowers. The pod color is brown, while the pubescence color is light brown. The seed hilum is black. The plant height ranges from 100 to 120 cm and is resistant to lodging. It produces 80–90 pods per plant, with a first pod height of 13–15 cm. The thousand-seed weight is 140–160 g. In the main crop, it has a yield potential of approximately 500 kg per decare. It can be cultivated as a main crop in all regions (Trakya-Marmara, Black Sea, Aegean, Mediterranean, and Southeastern Anatolia). Additionally, it is suitable for second-crop cultivation in the Çukurova and Trakya regions. As a main crop, it should be sown between April 20 and May 10, while for the second crop, sowing should be done between June 20 and June 30, and in the Trakya region, until July 10–12. The recommended seed rate is 6–8 kg per decare. Ataem7; Developed by the Western Mediterranean Agricultural Research Institute, this variety belongs to the third maturity group and has white flowers. The pod color is brown, while the pubescence color is light brown. The plant height ranges from 100 to 120 cm and is resistant to lodging. It produces 70–80 pods per plant, with a first pod height of 15–18 cm. The thousand-seed weight is 140–160 g. In the main crop, it has a yield potential of approximately 480 kg per decare. It can be cultivated as a main crop in the Mediterranean, Aegean, Çukurova, and Southeastern Anatolia regions. Additionally, it is suitable for second-crop cultivation in the Çukurova and Mediterranean regions.

The experiment was conducted during the 2023 soybean growing season at the Agricultural R&D Center trial fields of Sivas University of Science and Technology. The field experiment was arranged in a randomized block design with three replications. Each genotype was sown in 5-meter-long plots with four rows per plot. The row spacing was 70 cm, and the intra-row spacing was 10 cm.

At the time of sowing, 6 kg/da nitrogen (N) and 8 kg/da phosphorus (P_2O_5) were applied. Sowing was carried out on May 16, 2023, considering the climatic conditions of the region. During the growing season, all necessary agronomic practices, including weed control, irrigation, and other maintenance, were performed meticulously based on the needs of the plants and prevailing weather conditions. A drip irrigation system was used for watering. The soil properties of the trial field at the Agricultural R&D Center of Sivas University of Science and Technology are presented in Table 1.

Table 1. Soil properties of the Agricultural R&D Center of Sivas Science and Technology University, where the research was conducted.

Depth	Texture	pH	Lime (% $CaCO_3$)	Salt (%)	Phosphorus (P_2O_5 kg/da)	Potassium (K_2O kg/da)	Organic Matter Content (%)
0-30 cm	Silty clayey	7.28	19.6	0.33	3.40	93.59	1.7

The soil at the research location is classified as silty clay loam, with a pH value of 7.28. It is characterized by low organic matter content (1.7%), high potassium levels (93.59 kg/da), low phosphorus (3.40 kg/da), significant lime content (19.6%), and low salinity (0.33%) (Table 1). During the study, no issues with groundwater were encountered, and the field was adequately drained.

Climate data for the 2023 growing season and long-term averages for Sivas province are presented in Table 2. Sivas has a continental climate, characterized by hot and dry summers and cold, snowy winters. Key climatic values for the study period, such as total precipitation, average temperature, and average relative humidity, are summarized in Table 2. During the experimental period in 2023, the lowest total precipitation was recorded in July (3.0 mm), while the highest was observed in April (74.8 mm). The lowest average temperature was recorded in April (9.1 °C), whereas the highest average temperature occurred in August (23.4 °C). Regarding relative humidity, were the lowest average value was observed in September (72.3%), while the highest was recorded in June (95.3%).

Table 2. Climatic data for the 2023 growing season and long-term data of Sivas Province *

	Total Precipitation (mm)		Average Temperature (˚C)		Average Relative Humidity (%)	
Months	**2023**	**Long Term**	**2023**	**Long Term**	**2023**	**Long Term**
April	74.8	33.7	9.1	8.9	92.8	62.3
May	56.4	54.7	13.0	13.5	93.6	61.1
June	51.4	43.4	17.3	17.0	95.3	58.3
July	3.0	6.2	20.1	20.0	82.8	54.0
August	3.6	4.5	23.4	20.3	76.6	53.0
September	4.3	17.8	19.2	16.3	72.3	62.0
October	7.6	36.8	18.4	10.9	74.5	64.0
Total/Average	**201.1**	**197.1**	**17.21**	**15.27**	**83.99**	**59.24**

* Sivas Provincial Directorate of Meteorology.

2.1. Moisture

Moisture determination was performed according to the TS EN ISO 712 standard (TSE, 2012). Harvested seeds were first dried and then ground for analysis. A 3 g sample was weighed and placed into a moisture container. The samples were then transferred to a preheated oven at 130 °C and held for 2 hours. After drying, the samples were weighed. The moisture content was calculated using the following formula:

$$\text{Moisture (\%)} = [(m2-m1)(m2-m3)]\times 100 \quad (1)$$

m1: Weight of the dried, empty moisture container and lid (g)

m2: Weight of the analysis sample plus the moisture container and lid (g)

m3: Weight of the moisture container and lid with the analysis sample after drying (g).

2.2. Ash Analysis

Samples ground from each seed were weighed (2 g) and placed into pre-weighed porcelain crucibles. The samples were combusted in an ash furnace at 650 °C for 3.5 hours. After combustion, the crucibles with the ash were weighed again. The weight of the porcelain crucibles was subtracted to determine the ash content of the samples.

$$\text{Ash} = [(\text{amount of ash}) / (\text{amount of sample})] * 100\ (\%) \quad (2)$$

2.3. Antioxidant Analysis

DPPH Assay: The DPPH radical (2,2-diphenyl-1-picrylhydrazyl) is an artificially stable radical widely used as a standard for determining antioxidant activity. The free radical scavenging activity of soybean samples was measured using the DPPH radical method as described by Blois (1958). For the assay, 0.1 mL of the sample was added to 2 mL of DPPH solution (0.025 g/L). The resulting mixture was incubated in the dark for 30 minutes. Absorbance was recorded at 517 nm. Results were expressed as μmol Trolox equivalents (TE) per gram of dry sample.

ABTS Assay: The ABTS assay was performed using the spectrophotometric method developed by Re et al. (1999). To prepare the stock solution, ABTS (7 mM) and potassium persulfate (2.45 mM) were each dissolved in 50 mL of distilled water. A mixture of 10 mL of the prepared ABTS solution and 10 mL of potassium persulfate solution was used to generate the ABTS radical solution. This mixture was allowed to stand in the dark at room temperature for 16 hours to form the ABTS radical solution. The working solution was prepared by diluting the stock solution to achieve an absorbance value of 0.700 ± 0.02 at 734 nm, establishing the initial absorbance value. For the assay, 100 μL of the extract was added to 3 mL of the working solution (ABTS + potassium persulfate), mixed thoroughly, and incubated at room temperature in the dark for 10 minutes.

Absorbance values were then recorded at 734 nm. Trolox was used as the standard, and results were expressed as Trolox equivalents (TE).

2.4. Statistical Analysis

The observed data were subjected to variance analysis using the MSTATC statistical software. Differences among the means were grouped using the Least Significant Difference (LSD) test at a significance level of p≤0.05.

3. Results and Discussion

The lower the absorbance of the mixture formed between antioxidants and the reagents (DPPH or ABTS), the higher the antioxidant's free radical scavenging activity. As the antioxidant concentration increases, the absorbance value decreases. The decrease in absorbance is due to the reaction between the radical and antioxidant molecules, resulting in the binding of hydrogen and the scavenging of the radical. The DPPH assay is commonly used to determine the degree to which a molecule acts as a free radical scavenger or hydrogen donor. The ABTS radical is soluble in both aqueous and organic solvents, making it suitable for measuring the antioxidant capacity of both lipophilic and hydrophilic compounds (Wong et al. 2006; Osman et al. 2006; Reis et al. 2011). The antioxidant capacity values determined by the DPPH method are presented in Table 3.

Table 3. Variance analysis results for the antioxidant capacity (DPPH method) values of the soybean genotypes

Genotypes	DPPH	Genotypes	DPPH
Arısoy	1,53±0,17	Ünv-3	3,25±0,12
Traksoy	3,94±0,04	Ünv-4	2,65±0,11
Samsoy	2,95±0,08	Ünv-5	1,37±0,24
Soyanam	2,32±0,17	Ünv-6	3,37±0,11
Ataem-7	2,51±0,11	Ünv-7	3,82±0,16
4	3,27±0,10	Ünv-8	2,93±0,08
5	2,63±0,17	Ünv-11	3,95±0,09
6	3,41±0,26	Ünv-12	3,51±0,11
7	2,34±0,22	Ünv-13	2,88±0,08
8	2,25±0,13	Ünv-15	4,03±0,18
9	3,49±0,31	Ünv-16	3,15±0,12
10	2,85±0,11	Ünv-17	1,88±0,13
11	3,05±0,10	Ünv-18	3,06±0,20
12	2,37±0,06	Ünv-19	3,32±0,19
Ünv-2	2,88±0,13	Ünv-20	3,36±0,10

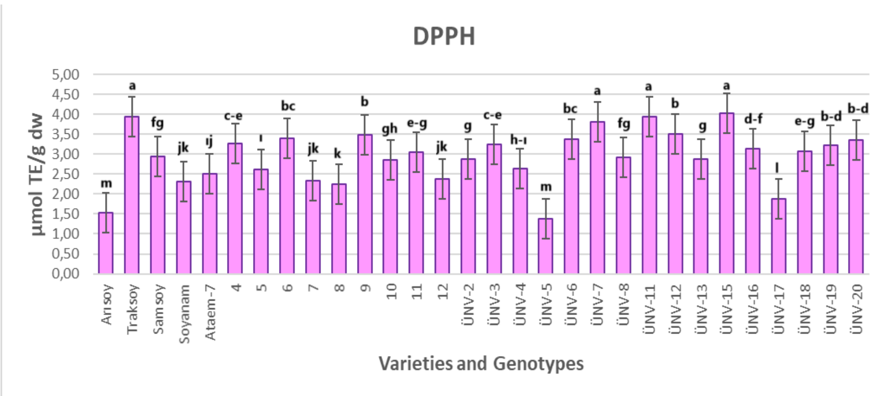

Figure 1. Different lowercase letters indicate significant differences between the samples (p < 0.01). LSD: 0.219. (dw: dry weight)

In the DPPH assay, the differences among the genotypes for this trait were statistically significant at the 1% level. The highest antioxidant activity was obtained from the ÜNV-15 genotype, with a value of 4.03 μmol TE/g dry weight (dw). This was followed by the ÜNV-11 genotype with 3.95 μmol TE/g dw and the Traksoy variety with 3.94 μmol TE/g dw. It was determined that the antioxidant activities of the Traksoy variety and ÜNV-11/15 genotypes were nearly identical. The lowest antioxidant activity was found in the ÜNV-5 genotype, with a value of 1.37 μmol TE/g dw. The differences among the genotypes for this trait were statistically significant at the 1% level (Table 3; Figure 1). Soedarjo et al. (2020), in their study using the DPPH method, reported that some developed soybean varieties, such as Detap 1 (29.66%), Anjasmoro (29.13%), and Dena 2 (28.35%), exhibited higher antioxidant activity compared to imported soybeans (27.75%). Zaini et al. (2024), in their study using the DPPH method, found that soybean samples showed a free radical scavenging activity of 38.92%, with an antioxidant activity of 3.47 mg AAE/100 g and a phenolic content of 14.75 mg GAE/100 g.

Table 4. Variance analysis results for the antioxidant capacity (ABTS method) values of the soybean genotypes

Genotypes	ABTS	Genotypes	ABTS
Arısoy	11,02±0,98	Ünv-3	8,83±0,27
Traksoy	8,65±0,36	Ünv-4	9,05±0,61
Samsoy	8,26±0,26	Ünv-5	8,26±0,43
Soyanam	6,98±0,57	Ünv-6	8,79±0,62
Ataem-7	7,65±0,60	Ünv-7	11,49±0,39
4	10,57±0,45	Ünv-8	10,22±0,42
5	8,95±0,55	Ünv-11	9,20±0,18
6	9,23±0,47	Ünv-12	10,54±0,70
7	7,74±0,57	Ünv-13	8,62±0,15
8	10,81±0,52	Ünv-15	9,44±0,07
9	9,25±0,51	Ünv-16	8,44±0,07
10	8,95±0,80	Ünv-17	10,34±0,23
11	9,45±0,42	Ünv-18	9,09±0,45
12	10,69±0,56	Ünv-19	8,11±0,13
Ünv-2	11,82±0,61	Ünv-20	8,25±0,11

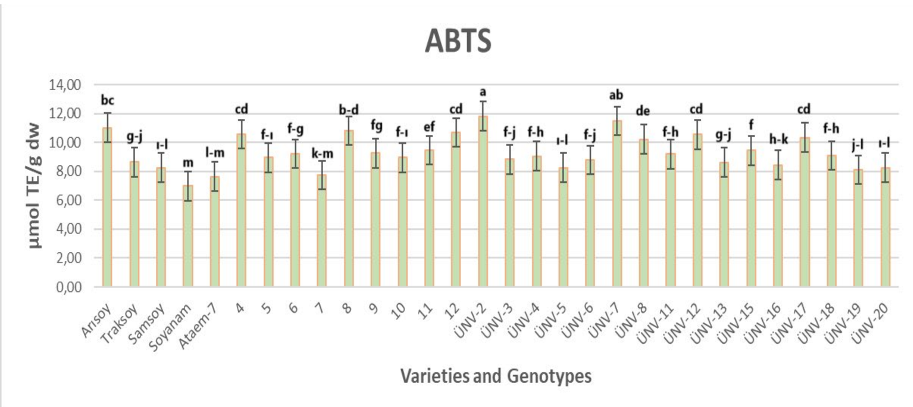

Figure 2. Different lowercase letters indicate significant differences between the samples (p < 0.01). LSD: 0.777. (dw: dry weight)

In the ABTS assay, the differences among the genotypes for this trait were statistically significant at the 1% level. The highest antioxidant activity was obtained from the ÜNV-2 genotype with a value of 11.82 μmol TE/g dry weight (dw). This was followed by the ÜNV-7 genotype with 11.49 μmol TE/g dw and the Arısoy variety with 11.02 μmol TE/g dw. The lowest antioxidant activity was found in the Soyanam variety, with a value of 6.98 μmol TE/g dw (Table 4). The differences among the genotypes for this trait were statistically significant at the 1% level (Table 4; Figure 2). In similar studies, Juliana et al. (2020) determined the IC50 value of the black soybean extract's ABTS scavenging activity as 77.39 μg/mL. Soedarjo et al. (2020), using the ABTS method, reported that the Demas sample exhibited 62.26% antioxidant activity, while Tanggamus soybean had 51.15% inhibitory activity. Zaini et al. (2024), in their study using the ABTS method, found that soybean samples exhibited an activity of 18.76 mg AAE/100 g.

Table 5. Variance analysis results of the moisture content of soybean genotypes

Genotypes	Moisture	Genotypes	Moisture
Arısoy	9,67±0,37	Ünv-3	11,13±0,11
Traksoy	10,14±0,12	Ünv-4	11,56±0,08
Samsoy	9,13±0,16	Ünv-5	10,73±0,44
Soyanam	10,72±0,18	Ünv-6	11,07±0,12
Ataem-7	8,75±0,20	Ünv-7	10,71±0,13
4	10,75±0,13	Ünv-8	11,43±0,21
5	10,40±0,05	Ünv-11	11,94±0,04
6	9,81±0,14	Ünv-12	11,57±0,09
7	9,66±0,08	Ünv-13	11,46±0,07
8	9,70±0,04	Ünv-15	10,88±0,12
9	10,69±0,40	Ünv-16	11,20±0,17
10	11,50±0,22	Ünv-17	11,39±0,14
11	11,21±0,15	Ünv-18	10,75±0,11
12	11,58±0,31	Ünv-19	10,59±0,24
Ünv-2	10,42±0,42	Ünv-20	12,27±0,22

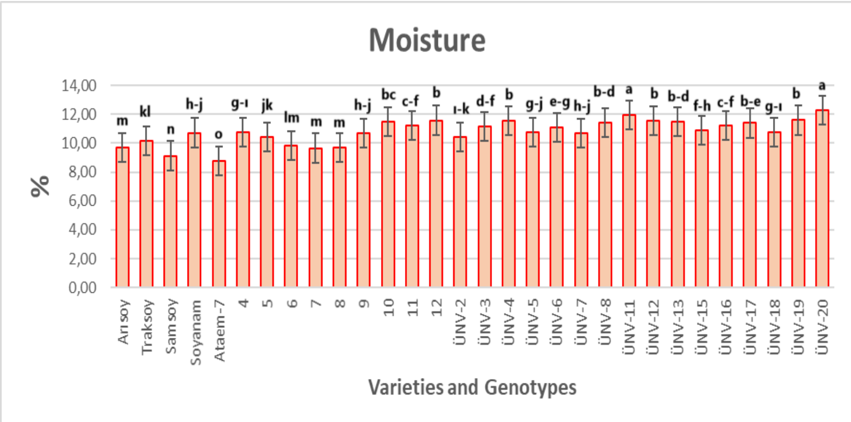

Figure 3. Different lowercase letters indicate significant differences between the samples ($p < 0.01$). LSD: 0.342.

The moisture content of the soybean genotypes and varieties used as material in this study ranged from 8.75 to 12.27%, and this difference was found to be statistically significant at the 1% level (Table 5; Figure 3). It was determined that the Soyanam variety had 10.72% moisture, while the Univ-19 and Univ-20 genotypes had 11.59% and 12.27% moisture, respectively. Ataem-7 and Samsoy varieties were found to have 8.75% and 9.13% moisture, respectively. Kolay (2007), in a study investigating the effects of different soil tillage methods on yield and some soil properties in second-crop soybean farming under Diyarbakır conditions, reported that the average moisture content of the seeds ranged between 7.06 and 7.13%. Furthermore, it was observed that the varieties had very similar values, and thus no statistical differences were found. The study also indicated that soil tillage methods did not significantly affect this trait (Kolay 2007).

Moisture content, one of the basic standard characteristics determined by TSE, is widely considered by all parties involved in buying and selling soybeans. However, industrial organizations involved in soybean imports mostly follow the official standards of the United States. In the TSE standard for soybeans, moisture is one of the key properties, and it has become an established rule in the market. Due to this characteristic, moisture content is one of the most important criteria for soybean marketing. The reason why moisture content is given considerable attention is that soybeans must contain low moisture to be harvested, stored, and processed properly (Güler and Emeksiz 2014).

Table 6. Variance analysis results of the ash content of soybean genotypes

Genotypes	Ash	Genotypes	Ash
Arısoy	2,91±0,06	Ünv-3	3,58±0,08
Traksoy	3,94±0,03	Ünv-4	3,24±0,11
Samsoy	3,76±0,05	Ünv-5	4,05±0,04
Soyanam	2,86±0,05	Ünv-6	3,71±0,05
Ataem-7	3,86±0,06	Ünv-7	3,51±0,15
4	2,98±0,08	Ünv-8	3,29±0,08
5	3,07±0,06	Ünv-11	3,65±0,21
6	3,17±0,16	Ünv-12	3,46±0,22
7	3,61±0,27	Ünv-13	3,55±0,12
8	3,96±0,06	Ünv-15	3,06±0,09
9	3,53±0,11	Ünv-16	3,40±0,23
10	3,13±0,12	Ünv-17	3,33±0,13
11	3,77±0,12	Ünv-18	3,36±0,23
12	3,88±0,08	Ünv-19	3,05±0,18
Ünv-2	3,22±0,11	Ünv-20	3,36±0,11

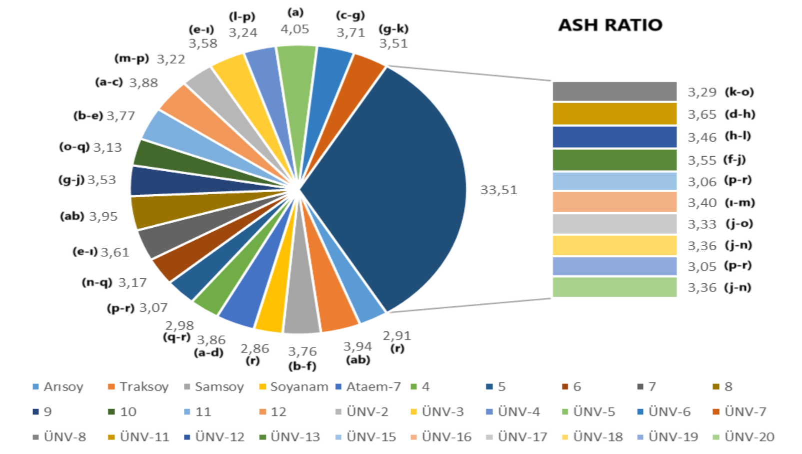

Figure 4. Different lowercase letters indicate significant differences between the samples. ($p < 0.01$). LSD: 0.219.

As seen in Table 6, the ash content in the genotypes ranged from 2.86 to 4.05%, with an average of 3.44%. The difference in the ash content values between the samples was found to be statistically significant at the 1% level (Table 6; Figure 4). Soyanam, Arısoy varieties and genotype number 4 had ash content of 2.86%, 2.91% and 2.98%, respectively. Öztürk et al. (2020) reported that the crude ash content in their study, which aimed to determine the silage quality of mixtures of hop with corn and feed soybean, ranged from 7.38 to 15%. Pejuhan (2018), in a study conducted over two years at different locations, determined the crude ash content of soybeans in the first year as 11.5% in Erzurum and 12.4% in Urumiye, and in the second year as 11.5% in Erzurum and 13.3% in Urumiye. Erekul (2020), in a study conducted in Aydın province, reported crude ash content values of 4.14 to 5.08% for soybeans in 2017-2018 field conditions. The values observed in these studies are higher than those found in the current study. This difference may be attributed to the genotypes used as material and the growing conditions.

4. Conclusions

Plants are powerful natural sources of antioxidants, exhibiting wide variability in chemical composition and biological properties. The antioxidant potential in some plants is based on the cumulative effects of various bioactive compounds. Therefore, it is essential to perform at least two different analytical methods simultaneously to obtain a reliable evaluation of antioxidant activity. In this regard, the DPPH analysis, which measures antioxidants with hydrogen-reducing capacity, and the ABTS analysis, which identifies both hydrophilic and hydrophobic antioxidants, were included in this study. The results revealed variability in antioxidant activity values among soybean seeds with different genetic backgrounds. Furthermore, since it is preferable for seeds to contain low moisture content for storage and processing, Ataem-7 was distinguished as the variety with the lowest moisture content. As a result, the ÜNV-2 and ÜNV-15 genotypes, with relatively higher antioxidant activity, and the Ataem-7 variety, with a low moisture content, were prioritized for selection based on these characteristics.

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References

Akkuş İ (1995). Free Radicals and Their Pathophysiological Effects. Mimoza Publications, Konya.

Arbos KA, Claro LM, Borges L, Santos C, Weffort-Santos AM (2008). Human erythrocytes as a system for evaluating the antioxidant capacity of vegetable extracts. *Nutrition Research*, 28(7): 457-463.

Bayram Y, Torlak Y, Sağdıç O (2019). Antioxidant activity of rowan fruit. *European Journal of Science and Technology*, (16): 933-939.

Blois MS (1958). Antioxidant determinations by the use of a stable free radical. *Nature*, 181(4617): 1199-1200.

Çilesiz Y, Nadeem MA, Gürsoy N, Kul R, Karaköy T (2023). Assessing the cooking and quality traits diversity in the seeds of faba bean germplasm. *Turkish Journal of Agriculture and Forestry*, 47(4): 448-466.

Erekul O (2020). International Innovation Cooperation Network for Increasing Soybean Production Due to Global Change (INNISOY), Aydın.

Güler D, Emeksiz F (2014). Soybean production, consumption and marketing in Türkiye. Master Thesis, Çukurova University(Unpublished), Türkiye.

Juliana C, Lister INE, Girsang E, Nasution AN, Widowati W (2020). Antioxidant and elastase inhibitor from black soybean (*Glycine max* L.) and its compound (daidzein). *Journal of Biomedicine and Translational Research*, 6(1): 11-14.

Kahkönen MP, Hopia AI, Vuorela HJ, Rauha JP, Pihlaja K, Kujala TS, Heinonen M (1999). Antioxidant activity of plant extracts containing phenolic compounds. *Journal of Agricultural and Food Chemistry*, 47(10): 3954-3962.

Karaköy T, Erdem H, Baloch FS, Toklu F, Eker S, Kilian B, Özkan H (2012). Diversity of macro-and micronutrients in the seeds of lentil landraces. *The Scientific World Journal*, (1): 710412.

Kaur C, Kapoor HC (2001). Antioxidants in fruits and vegetables–the millennium's health. *International Journal of Food Science and Technology*, 36(7): 703-725.

Kılınç K, Kılınç A (2002). Oxygen radicals as mediators of oxygen toxicity. *Hacettepe Medical Journal*, 33(2): 110-118.

Kolay B (2007). Effects of different tillage methods on yield and some soil properties in second crop soybean cultivation in Diyarbakır conditions. PhD Thesis, Harran University (Unpublished), Türkiye.

Osman AM, Wong KK, Hill SJ, Fernyhough A (2006). Isolation and the characterization of the degradation products of the mediator ABTS derived radicals formed upon reaction with polyphenols. *Biochemical and Biophysical Research Communications*, 340(2): 597-603.

Öztürk YE, Gülümser E, Mut H, Başaran U, Doğrusöz MÇ (2020). Determination of silage quality of hop mixtures with corn and feed soybean. *Harran Journal of Agricultural and Food Sciences*, 24(4): 440-446.

Pejuhan J (2018). Effects of foliar iron and zinc application with biological fertilizers on hay yield and yield components in forage soybean. PhD Thesis, Atatürk University, University of Science, Erzurum.

Prior RL, Cao G (2000). Analysis of botanicals and dietary supplements for antioxidant capacity. *Journal of AOAC International*, 83(4): 950-956.

Re R, Pellegrini N, Proteggente A, Pannala A, Yang M, Rice-Evans C (1999). Antioxidant activity applying an improved ABTS radical cation decolorization assay. *Free Radical Biology and Medicine*, 26(9-10): 1231-1237.

Reis FS, Heleno SA, Barros L, Sousa MJ, Martins A, Santos-buelga C, Ferreira I (2011). Toward the antioxidant and chemical characterization of mycorrhizal mushrooms from northeastern Portugal. *Journal of Food Science*, 76(6): 824–830.

Seeram NP, Zhang Y, Henning SM, Lee R, Niu Y, Lin G, Heber D (2006). Pistachio skin phenolics are destroyed by bleaching resulting in reduced antioxidative capacities. *Journal of Agricultural and Food Chemistry*, 54: 7036-7040.

Soedarjo M, Suhartina S, Nugrahaeni N, Wijanarko A, Putri DA, Fatmawati S (2020). The antioxidant activities and phenolic content of improved soybean seeds varieties of different grain sizes. *IPTEK The Journal for Technology and Science*, 31(1): 83-90.

Soretire AA, Olayinka A (2013). Response of soybean (*Glycine max* L.) to cow dung and wood ash application in tropical acid soils of South-Western Nigeria. *Nigerian Journal of Soil Science*, 23(2): 103-113.

TSE (2012). TS EN ISO 712, Grain and grain products - Determination of moisture content. Turkish Standard, Turkish Standards Institute, Ankara.

Wong SP, Leong LP, Koh JHW (2006). Antioxidant activities of aqueous extracts of selected plants. *Food Chemistry*, 99(4): 775-783.

Young IS, Woodside JV (2001). Antioxidants in health and disease. *Journal of Clinical Pathology*, 54(3): 176-186.

Yu J, Ahmedna M, Goktepe I, Dai J (2006). Peanut skin procyanidins: composition and antioxidant activities as affected by processing. *Journal of Food Composition and Analysis*, 19: 364-371.

Zaini NS, Karim R, Razis AFA, Zunairah W, Ibadullah W, Zawawi N (2024). Comparative analysis of ultra-high performance liquid chromatography-mass spectrometry and antioxidant properties between kenaf (*Hibiscus cannabinus* L.) seed and soybean (*Glycine max*) milk substitutes. In The 7th International Conference for Women in Science Without Borders, 2 April, Malaysia, p. 61.

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Research Article

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Lepidoptera Species in Selçuk University Alaeddin Keykubat Campus Area

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HIGHLIGHTS

- 37 species belonging to 11 families in the order Lepidoptera were identified in the Selçuk University.
- In Rhopalocera, butterflies, 16 genera and 18 species belonging to 5 families were identified.
- In Heterocera, moths, 17 genera and 19 species belonging to 6 families were identified.
- Nymphalidae (7 species) and Noctuidae (8 species) were the families with the highest number of species detected.

Abstract

This study was conducted in April-September 2023 to determine the Lepidoptera species in the Selçuk University Alaeddin Keykubat campus area. Four different methods were used to collect butterfly specimens. The first of these methods is the sweep net method, which allows catching Rhopalocera adults in April-September when they fly actively in nature, or on plants. Robinson-type light traps powered by a 125 W mercury vapor bulb were used as the second method for catching Heterocera species. In the third method, the specimens were captured in production areas such as vegetable fields, vine and apple orchards with species-specific delta-type sexual attractant pheromone traps in order to determine the presence of lepidopteran species specific to these areas. The last method is the butterfly walk method, which is based on the collection of eggs, larvae and pupae of pre-adult butterflies during field observation. The collected specimens were prepared for diagnosis in the laboratory and species identifications were made. As a result of the study, 37 species belonging to 11 families in the order Lepidoptera were identified in the Selçuk University Alaeddin Keykubat campus area. In Rhopalocera, butterflies, 16 genera and 18 species belonging to 5 families were identified. Among these families, the family Nymphalidae with 7 species was the family having the most species detected. In Heterocera, moths, 17 genera and 19 species belonging to 6 families were identified. Among these families, Noctuidae, with 8 species, was the family having the most species detected.

Keywords: Alaeddin Keykubat Campus; Butterfly; Konya; Lepidoptera; Selçuk University

1. Introduction

Insects have a very important place in human life due to their diversity, ecological role, agriculture and impact on human health (Rosenberg et al. 1986). In a popular sense, “insect” usually refers to familiar pests or disease carriers such as bedbugs, houseflies, clothes moths, aphids, mosquitoes, fleas, horseflies, etc. There are

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also many beneficial insects such as honeybees, silkworms and ladybirds (Gillott 2005). Insects belong to the class Insecta of the Arthropoda phylum of the animal kingdom. There are 26 orders of insects in the subclass Pterygota and the orders with the most species are Coleoptera, Lepidoptera, Hymenoptera and Diptera (Kansu 2000; Ülger 2022).

The order Lepidoptera, which includes butterflies and moths in the class Insecta, is second only to Coleoptera in terms of species richness. Lepidoptera is an insect order with more than 250,000 species and accounts for about a quarter of all known insect species (Grimaldi and Engel 2005). The order Lepidoptera is also known as the "Scalewings". The Lepidoptera is divided into two suborders: "butterflies (Rhopalocera)" and "moths (Heterocera)" (Kansu 2000; Gullan and Cranston 2014). Butterflies (Rhopalocera) are usually brightly colored, mostly diurnal, wings closed at rest, antennae with knobs, body slender and smooth, pupa in chrysalis structure, hard and silky, without frenulum, the structure that connects the forewing to the hindwing. Moths (Heterocera) are usually dull in appearance and nocturnal, their wings are open at rest, their antennae are hairy or thread-like, their bodies are more densely hairy, their pupae have a silky structure called cocoon, and they have a frenulum (Kansu 2000; Gullan and Cranston 2014).

Lepidoptera are an indicator of a healthy environment and ecosystem, and are a part of life on Earth and an important component of its rich biodiversity. They are also an important component of the food chain and are prey for birds, bats and other insectivores. These insects are widely used by ecologists as model organisms to study the impact of habitat loss, fragmentation and climate change (Ghazanfar et al. 2016; Ögür and Çetin 2018).

Lepidoptera have always attracted the attention of human beings due to both their elegance and visual appeal. For this reason, they are used in weddings, birthdays, education, academic studies, fashion and design (Isbill 2003; Rogers 2013; Padilla et al. 2021). In addition, there are species that cause significant economic losses in cultivated plants. For example, the larvae of *Cydia pomonella* (L., 1758) (Lepidoptera: Tortricidae) directly damage the fruit (Birgücü and Erol 2021). Another pest, *Lobesia botrana* (Denis & Schiffermüller 1775) (Lepidoptera: Tortricidae) is one of the main pests of grapevines (Venette et al. 2003). *Spodoptera littoralis* (Boisd., 1833) (Lepidoptera: Noctuidae) larvae feed on the leaves, flowers, and fruits of vegetables, causing quality and yield loss (Karlsson Green 2021). Another important pest, *Helicoverpa zea* (Boddie, 1850) (Lepidoptera: Noctuidae) causes great damage to more than 100 crop plants such as corn, sorghum, cotton, eggplant, pepper and tomato (Jackson et al. 2008).

Selçuk University Alaeddin Keykubat (SUAK) campus, where the study was carried out, is one of the leading campuses of our country with an area of 10,000 da. More than half of this area is vegetative landscape, with a total of 6,270 da designated as green space. This means that 62.7% of the campus consists of green areas. Selçuk University Faculty of Agriculture has a production area of approximately 120 da in which various vegetables (*Phaseolus vulgaris* L., *Solanum lycopersicum* L., *Daucus carota* L. and *Cucumis sativus* L.), field crops (*Zea mays* L., *Triticum aestivum* L., *Helianthus annuus* L. and medicinal aromatic plants) and various orchards (*Malus communis* L., *Pyrus communis* L., *Fragaria ananassa* Duch. and *Vitis vinifera* L.) are grown. In addition, various vegetables and fruits are grown in hobby gardens of 30 da within the campus.

In this study, it was aimed to determine the Lepidoptera species in the campus area of SUAK. Determination of Lepidoptera species in the research area is important for the conservation of butterfly species identified in this area and to determine the increase or decrease in species in future studies. In addition, it is important to determine the species that may cause damage to the landscape plants in the campus area and the cultivated plants in the production areas of the Faculty of Agriculture and to take necessary measures against the pests to be identified. The fact that no previous study has been conducted on the determination of Lepidoptera species in SUAK campus area reveals that this study is unique.

This study was carried out between April and October 2023. Field trips were organized in the campus area of SUAK, and light and pheromone traps were set, where diurnal and nocturnal lepidopteran specimens were collected. The species found were identified with the help of literature and artificial intelligence applications (Seek, Picture Insect, and Google Lens).

2. Materials and Methods

2.1. Capturing of Lepidoptera species

Adults were generally caught during the 6-7 month period from April to October, when lepidoptera began to fly actively, in the landscape areas of SUAK campus and in the production areas of Selçuk University Faculty of Agriculture, which were determined as study areas for capturing Lepidoptera species (Figure 1). There are approximately 250 types of ornamental plants in the campus landscape areas. The most common ones are; *Lavandula angustifolia* Mill., *Rosa* spp., *Platanus orientalis* L., *Anthemis nobilis* L. and *Picea pungens* Engelm. There are also a wide variety of vegetables (*Phaseolus vulgaris* L., *Solanum lycopersicum* L., *Daucus carota* L. and *Cucumis sativus* L.), field crops (*Zea mays* L., *Triticum aestivum* L., *Helianthus annuus* L. and medicinal aromatic plants), and various fruit gardens (*Malus domestica* L., *Pyrus communis* L., *Fragaria* spp. and *Vitis vinifera* L.) in the production areas of Selçuk University Faculty of Agriculture.

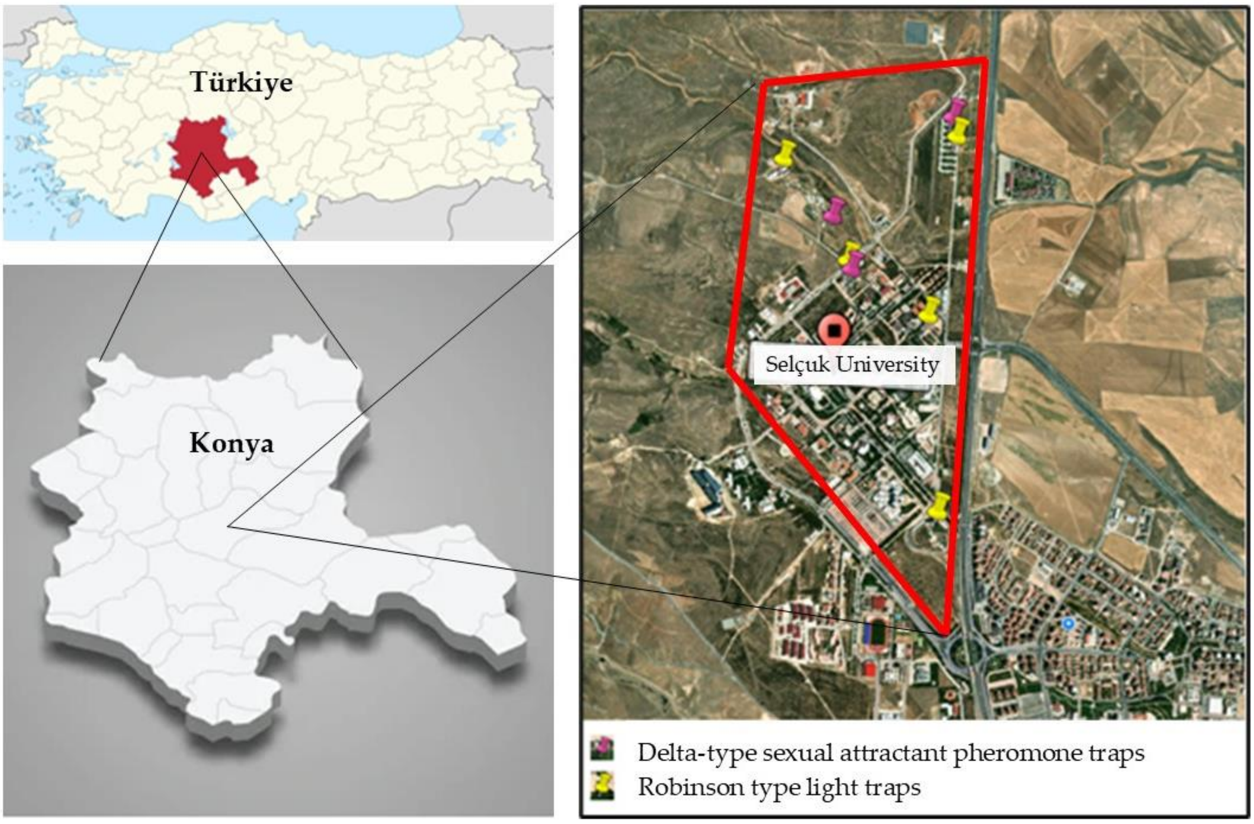

Figure 1. Map of the research area. Locations where the Robinson-type light traps and the delta-type sexual attractant pheromone traps were set up are pointed.

2.1.1. Butterfly walk method

The larvae collected during field observations were cultured on the plant they were found on and adults were obtained. The adults obtained were used for identification purposes. Some Archips larvae were collected by hand from cherry and maple trees and cultured in the laboratory to obtain adults.

2.1.2. Trapping with sweep net

A sweep net was used to collect diurnal butterflies. Trapping with a sweep net was done every day during the daylight hours from the end of April, when diurnal butterflies were actively flying in nature, until the end of September, 2023. The captured specimens were immediately placed in a killing bottle containing cyanide and taken to the laboratory on the same day for stretching and pinning (Perveen and Fazal 2013; Gibb 2014).

2.1.3. Trapping with light traps

Robinson-type light traps working with 125 W mercury vapor bulbs were used to catch the moths. Moths were collected with light traps in five different locations in the SUAK campus area from May to the end of September, 2023 and for two weeks in each location. Boxes containing DDVP impregnated sawdust were placed in the chamber section of the light trap to ensure that the butterflies that came to the trap died. In some cases, rehydration was performed to prevent the samples that were too dry from breaking during the stretching process (Perveen and Fazal 2013; Gibb 2014). The coordinates and dates of the light traps set up in the SUAK campus area are shown in Table 1.

Table 1. Coordinates and installation dates of Robinson-type light traps established in Selçuk University Alaeddin Keykubat campus area.

Trap name	Trap coordinates	Dates of trap installation
Trap 1 - Greenhouse	38°01'49''N 32°30'31''E	23.05.2023
Trap 2 - Farm	38°02'05''N 32°30'20''E	23.06.2023
Trap 3 - Landscape area	38°01'42''N 32°30'52''E	21.07.2023
Trap 4 - Tram	38°01'09''N 32°30'53''E	4.08.2023
Trap 5 – Hobby garden	38°02'12''N 32°30'52''E	18.08.2023

2.1.4. Trapping with pheromone traps

In order to determine the presence of pest species specific in production areas such as vine greenhouse and apple orchard, moth trapping was carried out with species-specific pheromone traps. For this purpose, delta-type sexual attractant pheromone traps were used against grapevine moth (*Lobesia botrana*) in vine greenhouses, tomato moth (*Tuta absoluta* (Meyrick, 1917) (Lepidoptera: Gelechiidae)) in hobby gardens, and codling moth (*Cydia pomonella*) in the apple orchard. These traps were established in production areas starting from the end of April and the first week of May, 2023 in accordance with the biology of the species. Observations were made weekly until adults were caught in the traps (Table 2). If necessary, pheromone capsules were replaced with new ones after 4-6 weeks.

Table 2. Coordinates and installation dates of delta-type sexual attractant pheromone traps established in Selçuk University Alaeddin Keykubat Campus Area.

Trap name	Trap coordinates	Dates of trap installation
Trap 1 - Apple orchard	38°01'57''N 32°30'33''E	28.04.2023
Trap 2 - Vine greenhouse	38°01'47''N 32°30'32''E	12.05.2023
Trap 3 - Hobby garden	38°02'12''N 32°30'52''E	26.05.2023

In addition to all these collection methods, lepidopter larvae found on plants were collected, reared in the laboratory, and adults were obtained.

2.2. Evaluation of Lepidoptera Species

After killing the captured lepidopterans, they were brought to the laboratory without drying and immediately stretched on stretching boards. The insect was positioned in a natural standing position on the stretching board, the samples were fixed in a horizontal position with the wings, legs, and antennae at body level and were left to dry at room temperature for at least 15 days and then removed from the stretching boards and labeled. The samples removed from the stretching boards were labeled with a label indicating the place of collection, date of collection and host plant information and were examined for identification. The collected samples were placed in storage boxes containing naphthalene to protect them from ants and museum insects (Gibb 2014).

2.3. Identification of Lepidoptera Species

Specimens collected in the study were identified according to their external morphological characters. All collected specimens were identified using the literature (Şengün and Güneyi 1968; Horak and Komai 2006; Bıçak 2007; Zobar and Genç 2008; Ioriatti et al. 2012; Koçak and Kemal 2012; Serik 2015; Parlak 2017; Mediouni Ben Jemâa et al. 2023) and verified by Prof. Dr. Levent ÜNLÜ. In addition, applications such as Seek, Picture Insect and Google Lens were also used for species identification.

3. Results

As a result of the study carried out to determine the Lepidoptera species in the SUAK campus area, a total of 37 species belonging to 33 genera from 11 families of the order Lepidoptera were identified. In the study, 1,871 individuals from 5 families, 16 genera and 18 species of diurnal butterflies were collected in SUAK campus area. Among these, the Nymphalidae with 7 species was the family having the most species detected within the Rhopalocera suborder. This was followed by Pieridae (4), Lycaenidae (4), Papilionidae (2) and Hesperiidae (1) families (Table 3). *Pieris brassicae* (L., 1758) (Lepidoptera: Pieridae) was found to be the most abundant species among the diurnal butterflies. It is thought that this species is polyphagous and its host plants are widespread in the campus area. The least abundant species was *Iphiclides podalirius* (L., 1758) (Lepidoptera: Papilionidae). Due to the fact that the study area was open to the prevailing winds, sometimes there were difficulties in capturing butterflies with the sweep net. This is one of the reasons for the low number of butterflies caught especially in the Papillionidae family.

Among the nocturnal butterflies, 2,971 individuals were collected from 17 genera and 19 species belonging to 6 families. Among these, the Noctuidae with 8 species was the family having the most species detected within the Heterocera suborder. This was followed by Sphingidae (4), Tortricidae (3), Erebidae (2), Gelechiidae (1) and Lasiocampidae (1) families (Table 3). It was determined that *Agrotis crassa* (Hübner, 1803) (Lepidoptera: Noctuidae) was the most abundant nocturnal butterfly species and *Leucania loreyi* (Duponchel, 1827) (Lepidoptera: Noctuidae) was the least abundant species. In order for the Robinson-type light traps used to catch nocturnal butterflies to be more effective, areas with high plant diversity and dark areas were preferred in the campus area. However, the scarcity of dark areas in the campus area, the difficulty in reaching the electricity source in the dark areas and the disturbing behaviors of some animals (dogs, etc.) while going to check the traps caused difficulties in sampling.

4. Discussion

This study is unique as it is the first study to determine the Lepidoptera species in SUAK campus area. In such faunistic studies, unless the study area is the same, comparison with the results of previous studies on the subject is not very accurate. However, the results obtained from this study are similar to those of some previous studies. This study revealed that the Nymphalidae with 7 species was the family having the most species detected within the Rhopalocera suborder and followed by Pieridae, Lycaenidae, Papilionidae and Hesperiidae families. Among the diurnal butterflies, *Pieris brassicae* was the most abundant species. Similar results were obtained in the study conducted by Zobar and Genç (2008) to determine the species belonging to the order Lepidoptera in Terzioglu Campus of Çanakkale Onsekiz Mart University. As a result of the study, Nymphalidae family was determined as the family with the highest number of species in Rhopalocera suborder. This was followed by Pieridae, Hesperiidae, Papillionidae and Lycaenidae. As determined in our study, *Pieris brassicae* was the most frequently observed species in the campus area. In some other studies (Hüseyinoğlu and Atay 2017; Arslangündoğdu et al. 2018; Atay et al. 2019) conducted to determine Lepidoptera species in different parts of our country, it was determined that Nymphalidae was the family with the highest number of species in the Rhopalocera suborder. Not only in our country, but also in studies conducted on university campuses abroad, similar results have been found. Sebua and Nuñeza (2023) conducted a study to determine the species diversity of Lepidoptera in Western Mindanao State University (Philippines) and found that the family Nymphalidae was dominant, most abundant, and had the highest

Table 3. Suborder, family, species names, date of capture and capture method of individuals belonging to the order Lepidoptera detected in Selçuk University Alaeddin Keykubat Campus area

Suborder	Family	Species	Type of capture	Date of capture
Rhopalocera	Hesperiidae	*Thymelicus sylvestris* (Poda, 1761)	Sweep net	28.07.2023
	Pieridae	*Pieris brassicae* (Linnaeus, 1758)	Sweep net	28.07.2023
		Pieris rapae (Linnaeus, 1758)	Sweep net	28.07.2023
		Colias crocea (Fourcroy, 1785)	Sweep net	15.07.2023
		Pontia edusa (Fabricius, 1777)	Sweep net	16.07.2023
	Lycaenidae	*Aricia agestis* (Denis & Schiffermüller, 1775)	Sweep net	16.07.2023
		Eumedonia eumedon (Esper, 1780)	Sweep net	29.07.2023
		Lycaena alciphron (Rottemburg, 1775)	Sweep net	29.07.2023
		Polyommatus icarus (Rottemburg, 1775)	Sweep net	02.08.2023
	Nymphalidae	*Brintesia circe* (Fabricius, 1775)	Sweep net	02.08.2023
		Chazara briseis (Linnaeus, 1764)	Sweep net	15.08.2023
		Hyponephele lupina (Costa, 1836)	Sweep net	15.08.2023
		Hyponephele lycaon (Rottemburg, 1775)	Sweep net	15.08.2023
		Melanargia larissa (Geyer, 1828)	Sweep net	02.08.2023
		Melitaea trivia (Lederer, 1861)	Sweep net	01.07.2023
		Vanessa cardui (Linnaeus, 1758)	Sweep net	01.07.2023
	Papilionidae	*Iphiclides podalirius* (Linnaeus, 1758)	Sweep net	01.07.2023
		Papilio machaon (Linnaeus, 1758)	Sweep net	01.07.2023
Heterocera	Noctuidae	*Agrotis crassa* (Hübner, 1803)	Light trap	14.07.2023
		Agrotis segetum (Denis & Schiffermüller, 1775)	Light trap	24.07.2023
		Acontia lucida (Hufnagel, 1766)	Sweep net	23.07.2023
		Leucania loreyi (Duponchel, 1827)	Light trap	03.08.2023
		Mythimna unipuncta (Haworth, 1809)	Light trap	06.08.2023
		Noctua pronuba (Linnaeus, 1758)	Light trap	03.08.2023
		Spodoptera exigua (Hübner, 1803)	Light trap	10.08.2023
		Tyta luctuosa (Denis & Schiffermüller, 1775)	Light trap	03.08.2023
	Erebidae	*Grammodes stolida* (Fabricius, 1775)	Light trap	10.08.2023
		Psalis pennatula (Fabricius, 1793)	Light trap	10.08.2023
	Gelechiidae	*Tuta absoluta* (Meyrick, 1917)	Pheromone trap	12.08.2023
	Lasiocampidae	*Malacosoma neustria* (Linnaeus, 1758)	Light trap	17.08.2023
	Sphingidae	*Hyles euphorbiae* (Linnaeus, 1758)	Light trap	21.08.2023
		Hyles nicaea (de Prunner, 1798)	Light trap	18.08.2023
		Agrius convolvuli (Linnaeus,1757)	Light trap	18.08.2023
		Macroglossum stellatarum (Linnaeus, 1758)	Light trap	12.08.2023
	Tortricidae	*Archips rosana* (Linnaeus,1758)	Larvae culture	28.07.2023
		Cydia pomonella (Linnaeus,1758)	Pheromone trap	01.05.2023
		Lobesia botrana (Denis & Schiffermüller, 1775)	Pheromone trap	22.05.2023

species richness. The results of some other studies (Khan et al. 2007; Ilhamdi et al. 2023; Ruales et al. 2023) also showed that the most abundant family was Nymphalidae. These results should not be surprising because, with approximately 7,080 known species worldwide, the Nymphalidae are the largest butterfly family (Heppner 2008). On the other hand, Perveen and Fazal (2013) studied butterflies of Hazara University, Garden Campus, Mansehra, Pakistan and found that Pieridae was the dominant family and followed by Nymphalidae and Papilionidae.

Noctuidae is one of the largest families of Lepidoptera order (Ribas-Marques et al. 2022). In our study the Noctuidae with 8 species was the family having the most species detected within the Heterocera suborder and followed by Sphingidae, Tortricidae, Erebidae, Gelechiidae and Lasiocampidae families. Hüseyinoğlu and Atay (2017) carried out a study to determine the Lepidoptera fauna in Darboğaz, Bolkar Mountains (Niğde) and detected that the highest number of genera in moths, belonged to Noctuidae, followed by Geometridae, Crambidae and Pyralidae. Seven and Çakır (2019) studied the Heterocera fauna of eastern Türkiye and determined that most of the identified species belonged to the family Noctuidae. By contrast, Sebua and Nuñeza (2023) did not find any Noctuidae in Western Mindanao State University.

Türkiye is one of the few countries in the world with continental characteristics in terms of biological richness (Can 2010). While there are 482 lepidopter species in the European continent, there are 400 species only in Türkiye and 45 of them are endemic. However, these species, which are an important part of biodiversity today, are rapidly decreasing and this decrease may occur as a result of natural effects as well as human effects (Çelik and Topsakal 2017). Lepidoptera have been recognized by experts as indicators of biodiversity (Ghazanfar et al. 2016). Their fragile and sensitive structure causes them to be unable to respond quickly to changes in their environment. Therefore, their struggle for survival is a serious warning about our environment (Ögür and Çetin 2018).

5. Conclusions

Abnormal weather events caused by climate change, which have become more pronounced in recent years, excessive chemical spraying in agricultural areas, soil, water and air polluted as a result of human activities rapidly affect lepidopter species, which are very sensitive to changes in their environment. For this reason, it is necessary to protect and increase natural environments as much as possible by avoiding all kinds of interventions that may harm the ecosystem. Although there are some species (*T. absoluta*, *C. pomonella*, *L. botrana*, etc.) that can be defined as pests among the species we detected, it should not be forgotten that they have a very important place in the ecosystem due to some other functions. All these species are very valuable for the biodiversity of the fauna of Türkiye and necessary measures should be taken for their conservation.

Selçuk University Alaeddin Keykubat campus area is one of the green campuses of our country and more than half of this area (62.7%) consists of landscape areas. Approximately 250 plant species in herbaceous, shrub and tree form are used in landscape areas. Diversity and richness in plant species affect the diversity and richness in insect species. It is thought that the diversity of plant species has an effect on the high number of species obtained as a result of the study. The determination of Lepidoptera species in SUAK is important for the conservation of the species to be identified in this area and to determine the increase or decrease in species in future studies. This study provided a basis for future studies on the species identification of Lepidoptera in Konya province. However, such studies should be continued in detail in order to determine the entire fauna of the region and contribute to the determination of the fauna of Türkiye.

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References

Arslangündoğdu Z, Bacak E, İhsanoğlu MA (2018). İstanbul-rumelikavağı'nın gündüz kelebekleri. *Turkish Journal of Bioscience and Collections*, *2*(1): 1-11.

Atay E, Yıldırım H, Tatlı M (2019). Bursa ili Rhopalocera (*Lepidoptera)* faunası ve yeni kayıtlar. *Journal of Anatolian Environmental and Animal Sciences*, *4*(2): 70-75. https://doi.org/10.35229/jaes.539213

Bıçak G (2007). Istanbul Belgrad ormanı Lepidoptera faunası. Master Thesis, Istanbul University (Unpublished), Türkiye.

Birgücü AK, Erol AB (2021). Isparta ili elma bahçelerinde *Cydia pomonella* (L.) (Lepidoptera: Tortricidae)'nın larva popülasyonun ve vuruklu meyve oranının belirlenmesi. *Türk Tarım ve Doğa Bilimleri Dergisi*, *8*(2): 453-462.

Can F (2010). Evaluation of morphological characters and male genitalia features of emerald moths (Lepidoptera: Geometridae, Geometrinae) from Turkey. *African Journal of Agricultural Research*, *5*(9): 867-873.

Çelik P, Topsakal Y (2017). Butterfly watching in Turkey within the scope of alternative tourism. Innovation and Global Issues in Social Science, 27 - 29 Nisan 2017,Antalya, Türkiye, pp. 100-105.

Ghazanfar M, Malik MF, Hussain M, Iqbal R, Younas M (2016). Butterflies and their contribution in ecosystem: A review. *Journal of Entomology and Zoology Studies*, *4*(2): 115-118.

Gibb TJ (2014). Contemporary insect diagnostics: the art and science of practical entomology. Academic Press, Oxford- England, p. 1-336.

Gillott C (2005). Entomology. Springer Science & Business Media. Springer Dordrecht. p. 1-832 https://doi.org/10.1007/1-4020-3183-1

Grimaldi D, Engel MS (2005). Evolution of the Insects. Cambridge University Press, USA. p. 1-755.

Gullan PJ, Cranston PS (2014). The insects: an outline of entomology. Wiley-Blackwell, USA. p. 1-624.

Heppner JB (2008). Butterflies (Lepidoptera: Rhopalocera). In: Capinera, J.L. (eds) Encyclopedia of Entomology. Springer, Dordrecht. pp. 623-626 https://doi.org/10.1007/978-1-4020-6359-6_497

Horak M, Komai F (2006). Olethreutine Moths of Australia:(Lepidoptera: Tortricidae). Csiro Publishing, Australian. p. 1-521.

Hüseyinoğlu Y, Atay E (2017). A faunistic study of diurnal and nocturnal Lepidoptera in Darboğaz, Bolkar Mountains (Niğde, Turkey). *Journal of the Kansas Entomological Society*, *90*(1): 44-53.

Ilhamdi ML, Al Idrus A, Santoso D, Hadiprayitno G, Syazali M, Hariyadi I (2023). Abundance and diversity of butterfly in the Lombok Forest Park, Indonesia. *Biodiversitas Journal of Biological Diversity*, *24*(2). https://doi.org/10.13057/biodiv/d240205

Ioriatti C, Lucchi A, Varela LG (2012). Grape berry moths in western European vineyards and their recent movement into the New World. *Arthropod management in vineyards: pests, approaches, and future directions*, 339-359. https://doi.org/10.1007/978-94-007-4032-7_14

Isbill J A (2003). The continuing influence of René Lalique in jewelry design. California State University, Dominguez Hills, California, USA. p. 1-13.

Jackson RE, Bradley JR, Van Duyn J, Leonard BR, Allen KC, Luttrell R, Head G (2008). Regional assessment of *Helicoverpa zea* populations on cotton and non-cotton crop hosts. *Entomologia Experimentalis et Applicata*, *126*(2), 89-106. https://doi.org/10.1111/j.1570-7458.2007.00653.x

Kansu IA (2000). Genel entomoloji (Dokuzuncu Baskı). Birlik Matbaacılık-Yayıncılık, Ankara, Türkiye. p. 1-425.

Karlsson Green, K (2021). The effects of host plant species and larval density on immune function in the polyphagous moth *Spodoptera littoralis*. *Ecology and Evolution*, *11*(15), 10090-10097. https://doi.org/10.1002/ece3.7802

Khan MR, Rafi MA, Munir M (2007). Biodiversity of butterflies from districts Kotli, Mipur and Azad Kashmir. *Pakistan Journal of Zoology*, 39, 27–34.

Koçak A, Kemal M (2012). Iğdır Kelebekleri, Centre for Entomological Studies, Ankara, Türkiye, p. 253

Mediouni Ben Jemâa J, Soltani A, Djebbi T, Mejri I, Kanyesigye D, Otim MH (2023). The maize caterpillar *Mythimna* (= Leucania) *loreyi* (Duponchel, 1827)(Lepidoptera: Noctuidae): Identification, distribution, population density and damage in Tunisia. *Insects*, *14*(10): 786. https://doi.org/10.3390/insects14100786

Ögür E, Çetin H (2018). Kentsel alanların biyoindikatörleri böcekler. Uluslararası Yeşil Başkentler Kongresi, 8-11 Mayıs 2018, Konya, Türkiye, pp. 884-896.

Padilla JJ, Collado JL, García CGG, Collado CJL (2021). Spatial and economic assessment of butterfly-based handicrafts as a tourism service provided by rural populations. *Cuadernos de desarrollo rural= International journal of rural development*, *18*(1): 6. https://doi.org/10.11144/Javeriana.cdr18.seab

Parlak NN (2017). Istanbul-Büyükada'nın Lepidoptera türleri. Yüksek Lisans, Istanbul Üniversitesi, Türkiye.

Perveen F, Fazal F (2013). Biology and distribution of butterfly fauna of Hazara university, garden campus, Mansehra, Pakistan. *Open Journal of Animal Sciences,* 3(2): 28-36. https://doi.org/10.4236/ojas.2013.32A004

Rosenberg DM, Danks HV, Lehmkuhl DM (1986). Importance of insects in environmental impact assessment. *Environmental management*, 10: 773-783. https://doi.org/10.1007/BF01867730

Ribas-Marques E, Díaz-Calafat J, Boi M (2022). The role of adult noctuid moths (Lepidoptera: Noctuidae) and their food plants in a nocturnal pollen-transport network on a Mediterranean island. *Journal of Insect Conservation*, 26(2): 243-255. https://doi.org/10.1007/s10841-022-00382-7

Rogers L (2013). All about professional butterfly farming. https://www.butterflyboutique.net/ebooks/11aapbf84612/en/All_About_Professional_Butterfly_Farming.pdf (access date: 4 February 2024).

Ruales JJJ, Demetillo MT, Along AA, Mohagan AB, Jumawan JH (2023). Diversity and status of true butterflies (Lepidoptera: Papilionoidea) in two ecological parks of Butuan City, Agusan del Norte, Philippines with new locality record. *Species,* 24: 1-12. https://doi.org/10.54905/disssi/v24i73/e46s1539

Sebua CMD, Nuñeza OM (2020). Species diversity of Lepidoptera in Western Mindanao State University–experimental forest area, Zamboanga City, Philippines. *Entomology and Applied Science Letters*, *7*(1): 33-43.

Serik T (2015). Moldova'nın gündüz kelebekleri (Lepidoptera, Rhopalocera) faunası. PhD Thesis, Istanbul University (Unpublished), Turkey Türkiye.

Seven E, Çakır A (2019). A Contribution to the moth fauna (lepidoptera, heterocera) of Elazığ Province, Turkey. *Bitlis Eren University Journal of Science and Technology*, *9*(1): 18-21.

Şengün A, Güneyi N (1968). İstanbul gündüz kelebekleri, Fen Fakültesi Basımevi, İstanbul, Türkiye, p.103.

Ülger G (2022). Kahramanmaraş ili ve çevresinde yer alan Pterygota (*Arthropoda Insecta*) taksonları üzerine ekolojik ve faunistik araştırma. PhD Thesis, Nevşehir Hacı Bektaş Veli University (Unpublished), Türkiye.

Venette RC, Davis EE, DaCosta M, Heisler H, Larson M (2003). Mini risk assessment grape berry moth, *Lobesia botrana* (Denis & Schiffermuller)[Lepidoptera: Tortricidae]. *USDA CAPS PRA, Department of Entomology, University of Minnesota, St. Paul, Minnesota, USA*. p.129.

Zobar D, Genç H (2008). Çanakkale Onsekiz Mart Üniversitesi Terzioğlu yerleşkesi gündüz kelebekleri (Lepidoptera: Rhopalocera). *Tekirdağ Ziraat Fakültesi Dergisi*, *5*(3): 309-321.

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Smart Agriculture Blockchain Applications

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HIGHLIGHTS

- Blockchain can revolutionize agriculture by enhancing transparency, traceability, and efficiency in the food industry.
- Blockchain enables secure tracking of product movement, ensuring data and food safety, and boosting consumer confidence.
- Real-time data from blockchain helps farmers make informed decisions on crop yields, weather, soil health, and animal welfare.
- Smart contracts on blockchain can automate insurance payouts, optimize soil health, create decentralized markets, and support environmentally friendly farming practices.

Abstract

Smart Agriculture is a combination of AI, Cloud Computing, and IoT that revolutionizes farming efficiency and sustainability. It successfully addresses the developing worldwide request for food production through expanding crops, optimizing inventory management, minimizing food waste, and improving the safe consumption of food. Precision agriculture, facility agriculture, and order agriculture form the core components of smart agriculture. Each one of them concentrates on specific areas of farming processes, altogether contributing to enhancing farm productivity and efficiency. The integration of blockchain technology further amplifies these benefits. Blockchain's decentralized data storage ensures data integrity and accessibility while mitigating risks associated with centralized systems. In supply chain management, blockchain enhances logistics, quality control, and risk mitigation, while in livestock management, it facilitates welfare tracking, secure identification, and grazing oversight. This study presents an in-depth review of blockchain-based smart agriculture implementations, emphasizing areas such as supply chain management, food safety, traceability, and stock management. These domains have demonstrated substantial benefits from blockchain integration. While previous research has explored various aspects of smart agriculture, an increasing number of studies highlight the supply chain as a key area of focus. The paper also highlights emerging opportunities, including the development of hybrid blockchain models to balance transparency and scalability. Additionally, blockchain-based auditing systems are identified as a promising tool to promote environmentally sustainable farming practices. Addressing these advancements can ensure

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sustainable food production, improve data management, and foster eco-friendly agricultural methods. This research underscores the transformative potential of blockchain in smart agriculture and provides a roadmap for future exploration and innovation, paving the way for sustainable and efficient farming practices.

Keywords: Blockchain; Food safety; Food traceability; Smart agriculture; Supply chain

1. Introduction

Smart Agriculture is an emerging technology that involves the integration of Artificial Intelligence (AI), Cloud Computing and the Internet of Things (IoT) in farming processes to enhance both productivity and sustainability. The fast-growing global population has led to a surge in demand for food production, making smart agriculture crucial. According to the Food and Agriculture Organization (FAO), food production must increase about 70% to sustain more than 9.1 billion people by 2050. For example, the imports of cereal products into the developing countries must increase three times to reach 300 million tons by 2050, thus the production amount in those countries needs to raise by 100% (High-Level Expert Forum, 2009). Smart agriculture can help close this gap by increasing crop yields, reducing waste, improving efficiency, and enhancing food safety.

The World Resources Institute’s (WRI) research on sustainable food for the future, as outlined in research (Searchinger et al. 2018), highlights the need for the implementation of several solutions together to achieve food sustainability. Smart agriculture can offer multiple solutions presented in the WRI research. It can help reduce food loss and waste, avoid competition for crops and land, grow livestock and pasture productivity, manage water and soil, adapt to climate change, and improve manure management.

The components of smart agriculture include precision agriculture, livestock and free-range monitoring, smart irrigation, supply chain management, information storage, crop insurance and product distribution. These components constitute Cyber-Physical Systems (CPS), which integrate physical devices with computing systems to optimize farming processes.

The integration of blockchain technology into smart agriculture is a promising solution to address many problems in the agriculture industry. In the past decade, researchers have proposed many systems for smart agriculture that utilize blockchain technology. The goal behind integrating blockchain technology with smart agriculture is to create a transparent, secure, and tamper-proof system that can track the entire lifecycle of food production, from farm to table.

There are three main sectors—agriculture, food processing, and distribution— primarily responsible for gathering data on food items. Consumers increasingly seek traceability of food products throughout the supply chain, emphasizing food quality. Supply chain entities are now seeking to earn consumers' trust by providing accurate information, adhering to standards of credibility, integrity, and quality. Regulatory bodies have introduced standards to enhance transparency and traceability in the food supply chain. A shift from centralized to distributed systems is underway to leverage benefits such as fault tolerance, scalability, and improved storage (L.B. 2022).

In smart agriculture, many surveys have been conducted over the past decade. Krithika L.B. (L.B. 2022) concentrated on existing research within specific subfields of agriculture, including smart agriculture. Other surveys have examined technology acceptance in smart agriculture (Thomas et al. 2023). As this field continues to evolve, researchers are increasingly directing their focus towards the implementation of systems such as the Internet of Things (IoT) in shaping the future of smart agriculture (Ahmed et al. 2022; Quy et al. 2022; Shaikh et al. 2022; Sinha and Dhanalakshmi 2022). However, this integration raises significant security concerns, prompting researchers to delve into the associated security and privacy challenges in smart agriculture (Ahmadi 2023; Basharat and Mohamad 2022). Moreover, some researchers have explored the potential applications of blockchain in the context of smart villages (Kaur and Parashar 2022). This paper offers a comprehensive overview of the constituents of smart agriculture, with particular focus on the relationship between smart agriculture and blockchain technology. Furthermore, it highlights the advantages of integrating blockchain technology into smart agriculture and provides insights into its potential applications within the agricultural sector. The paper also classifies blockchain applications across all domains of smart agriculture and provides a detailed discussion of their implementation.

The rest of the paper is arranged as follows. Section 2 will provide an overview of the components of smart agriculture. Section 3 explores blockchain technology and its relationship with smart agriculture. Blockchain-based smart agriculture implementations are given in Section 4. Finally, Section 5 concludes the paper with a discussion of the potential future directions of smart agriculture and blockchain integration in agriculture.

2. Smart Agriculture

Among all industries, agriculture stands out as one of the most crucial. Its production is vital for the economy and plays a pivotal role in ensuring defense, nutrition, and health for populations. Moreover, agricultural production is indispensable for the planet's sustainability. With the world's population increasing daily, the demand for agricultural products is on the rise (Alam 2023).

Bogoviz et al. (Bogoviz et al. 2023) explained the disparities between traditional and smart agriculture. Farmers adhering to traditional methods for crop sowing, cultivation, and harvesting often struggle to efficiently utilize water resources and human labor. For instance, different crops require distinct watering schedules and methods. Advancing technologies, such as wireless sensor networks, enable the detection of water requirements, facilitating tailored water planning for different crops in a field. The utilization of microelectromechanical systems (MEMs) enables precise control over water amounts. These methodologies underscore the importance of integrating intelligent systems.

Smart agriculture offers real-time monitoring capabilities, reduces reliance on human labor, and automates the detection and provision of essential resources such as water, fertilizer, and sunlight. Smart agriculture systems, incorporating ubiquitous computing, wireless ad-hoc sensor networks, radio frequency identity detection, cloud computing, data analytics, remote sensing, context-aware computations, the Internet of Things and blockchain (Kumar and Dwivedi 2023), have already permeated our daily lives (Ojha et al. 2015).

Furthermore, these systems optimize various aspects including crop development, field monitoring, greenhouse gas tracking, production management, and crop protection. However, they operate with devices characterized by low power consumption, limited memory capacity, and modest computational capabilities (Atalay 2023).

Consequently, smart agriculture is the application of advanced technologies such as remote sensing, communication, and data processing in agriculture to improve productivity. It is a rapidly growing area that can revolutionize the way agriculture has been handled from field to consumer. Smart agriculture development approaches are examined under three main categories in the literature, considering the place of agriculture and the agricultural actions taken. These are precision agriculture, facility agriculture and order agriculture (Yang et al. 2021). In Europe, order agriculture closely resembles contract agriculture (Atalay 2023).

2.1 Precision Agriculture

To enhance soil quality and productivity, farmers can implement a range of targeted interventions, a practice commonly referred to as precision farming. This is made feasible by advancements in increasingly sophisticated technologies. The term "precision" is aptly used because these cutting-edge tools allow for precise interventions to be executed at the right location, at the right time, and with exceptional accuracy, tailored to the specific needs of individual crops and areas of land (Raj et al. 2021). The emergence of smart farming and precision agriculture represents a groundbreaking innovation within the agricultural sector. These technologies automate farming processes with the goal of achieving both high yield quantities and quality, thereby promoting food sustainability (Kwaghtyo and Eke 2023).

Precision farming is carried out in rural areas. Because agriculture is done outdoors it is affected by climatic conditions. This agricultural approach optimizes the timing and amount of water, fertilizer, seed, and pesticide with a focus on increasing the targeted product yield and protecting the agricultural ecological environment. Precision Agriculture utilizes data from multiple sources to optimize the farming process by utilizing sensors to keep track of any environmental changes, and then using machine learning algorithms to examine the data gathered. This provides valuable information on the condition of crops, helps determine the optimal approaches for planting, watering, and fertilizing, and reduces environmental impact.

Wireless sensor networks (WSNs) play a key role in this farming approach. The agricultural data (environmental data, crop development and health status data) collected from the different low-energy sensors lowers the risks in the precision farming process and enables the implementation of effective agricultural management (Srbinovska et al. 2015). Integrating wireless sensor networks lies at the core of precision farming, by facilitating real-time monitoring and data-driven decision-making in agricultural practices (Dangi 2004). In addition to remote sensing methods, Geographical Information Systems (GIS) and Global Positioning System (GPS) data are used extensively in the precision agriculture approach (Ferrag et al. 2020).

Data from the field, GIS and GPS are processed to obtain useful information. In the data processing, learning-based (Machine learning, Deep learning, etc.) methods are used (Yang et al. 2021). The information obtained because of the process is used in product and production management.

2.2 *Facility Agriculture*

Facility agriculture represents a novel production system that utilizes artificial technology to regulate the growth environment of crops, aiming to achieve efficient production (Bi and Liu 2023). In facility agriculture, the focus is on enhancing productivity within industrial settings. In these contexts, developmental areas oversee productivity across larger time intervals and broader plantations compared to precision agriculture. The main factors in facility agriculture include capital, technological resources, and workforce. Within these facilities, various environmental factors are artificially controlled. Synthetic processes can influence planting, cultivation, and other agricultural activities (Atalay 2023).

Facility agriculture is usually carried out in industrial areas close to the city. It aims to grow high quality products efficiently. To meet the demands and needs of people in the developing world, it makes it possible to grow the desired product in controlled environments without being affected by environmental and seasonal limitations (Vijay Hari Ram et al. 2020). As in precision agriculture, remote sensing systems are used extensively.

Horticultural and animal husbandry works done using similar technologies and systems suitable for facility agriculture are also evaluated within this scope. Any product can be grown in the facility by meticulous control of temperature and air pressure, illumination, irrigation, and fertilization, using forecast models developed using historical data of the product to be grown in facility agriculture.

The most typical example is smart greenhouses. They are commonly utilized for the cultivation of vegetables, fruits, and flowers. Currently, the main sensors used encompass environmental and plant sensors. Environmental sensors typically include temperature, humidity, soil moisture, and carbon dioxide sensors (Sun et al. 2023). This approach is also used in the fields of aquaculture, plant factory, poultry and livestock breeding with the help of different sensors and specialized control systems (Yang et al. 2021).

Smart Greenhouses are automated environments that use sensors and control systems to regulate temperature, humidity, light, and nutrients. This technology ensures that plants receive the optimal conditions for growth and reduces the need for manual labor.

Livestock Breeding and Monitoring uses technology to monitor livestock health and behavior. This involves using sensors attached to animals to monitor their movement and vital signs and analyze data to detect early signs of illness and optimize food and water consumption.

2.3 Order Agriculture

The order agriculture model is implemented to integrate technological innovations into human life. In this approach, agricultural infrastructure models tailored to specific geographical regions are strategized, utilized, monitored, and overseen. These region-specific agricultural activities can be optimized by effectively managing the supply chain, integrating appropriate technologies into relevant socio-economic units, and enhancing crop storage conditions.

Solutions for industrial applications are offered by order agriculture, derived from developments in precision and facility agriculture. For instance, when optoelectronic sensors are used to monitor and analyze weed and pest growth around a specific plant, it is categorized under precision agriculture. In facility agriculture, this mechanism is implemented and evaluated to routinely monitor a specific plantation. In order agriculture, the objective is to minimize pesticide use to mitigate damage to the ecosystem (Atalay 2023).

Although product efficiency can be increased using advanced technologies, this alone cannot gain commercial value (Dalohoun et al. 2009). To prevent unconscious production and to minimize production risks, an efficient commercial model is created with the help of order agriculture, which considers the external demand for the product (Bellemare and Bloem 2018). The agricultural supply chain developed in this process.

1. It provides agricultural product transparency from field to market.
2. It prevents information imbalance between farmers and suppliers.
3. It helps to ensure the supply-demand balance in agricultural products.

It has been proposed to use the increasingly popular blockchain technology to solve the trust problem in agricultural product supply chains (Hua et al. 2018).

To implement smart agriculture, several components and requirements are necessary. These include remote sensing tools for data collection, communication systems for data transmission, data storage, and control system. Figure 1 demonstrates the data flow in a smart agriculture system.

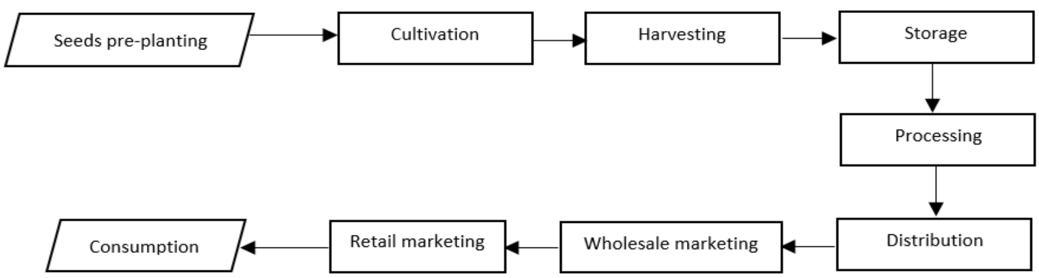

Figure 1. Data flow in food chain.

Additionally, farmers need specialized agricultural software to analyze the collected data to manage their farms depending on their needs. Smart Agriculture requires investment in technology, but it has the potential to increase crop yields and reduce resource usage.

3. Blockchain Basics

Blockchain is a distributed digital ledger technology that is designed to securely record transactions and maintain a tamper-proof record of information, which makes it resistant to fraud and hacking attempts. A blockchain consists of a series of blocks, where each block contains of a set of transactions that have been verified and recorded by the network. Figure 2 provides a simple explanation of how a transaction is added to the blockchain.

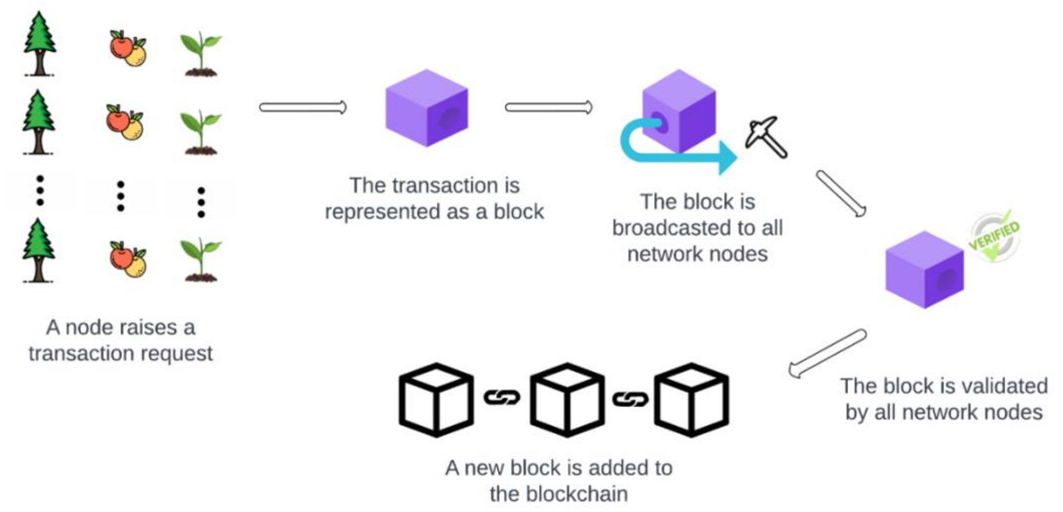

Figure 2. Transaction flow in a blockchain.

Blockchain technology serves as a robust tool for enhancing the efficiency of smart agriculture processes. In a blockchain system, users submit transactions, which are then grouped together to create a block. In a smart agriculture scenario, information about each agricultural resource, such as livestock, soil, water, plant, and seed, can be represented as a transaction. Single or multiple transactions can form a block. A block is created and verified, then it is connected to the existing chain, with the collaboration of independent parties called miners. A leader is chosen randomly among the miners to decide who will add the block to the chain. Selecting the leader can be achieved with the help of consensus algorithms. Additionally, miners must approve and accept block's proper generation before it is added to the chain.

A block includes information from the previous block to create connection between the blocks in the chain. This feature strengthens security, since modifying a single transaction would require updating all subsequent information in the chain, which is a challenging task.

Additionally, the blockchain can execute a task without third-party intervention. This is done using automated scripts called smart contracts. Smart contracts operate autonomously on the blockchain, which enables the seamless integration of these procedures during the design of the blockchain system.

Blockchain technology is highly secure and resistant to fraud. Consensus algorithms and smart contracts create a more trustworthy system by ensuring that all transactions are validated and recorded in a tamper-proof manner (Sakib 2024).

There are numerous benefits of using blockchain technology in smart agriculture. It increases transparency and immutability, improves supply chain management, and enhances food safety. Blockchain can help farmers and consumers track the origin and journey of food products from farm to consumer and ensure that the food is produced in a sustainable and ethical manner. Additionally, with its growing lists of securely linked blocks, blockchain can record sales and all data from seed planting to consumption (Patil et al. 2018).

Blockchain can help reduce fraud and corruption in the agricultural sector, leading to fair prices for farmers and improved access to markets.

4. Blockchain Platforms and Technologies

This paper explores the integration of blockchain in smart agriculture, highlighting specific platforms and technologies that address agricultural challenges.

4.1 Blockchain Platforms

1. Ethereum: Known for its smart contract functionality, Ethereum enables automated solutions for crop insurance (Omar et al. 2023), land registry (Shrivastava and Dwivedi 2023), and food traceability (Kechagias et al. 2023). This reduces dependency on intermediaries while enhancing transparency and efficiency.
2. Hyperledger Fabric: A permissioned blockchain platform, Hyperledger Fabric facilitates secure data sharing (Hu et al. 2024) and ensures transparency in supply chains. It has been widely adopted for improving accountability (Srikanth et al. 2024) and managing agricultural resources.
3. Hyperledger Sawtooth: a blockchain framework designed for enterprise use, brings transformative benefits to smart agriculture by enabling secure, decentralized, and efficient management of agricultural operations. It enhances supply chain traceability (Gkogkos et al. 2023), ensuring the authenticity and quality of agricultural products by recording every transaction on an immutable ledger. This provides transparency and builds trust among consumers, farmers, and stakeholders.
4. Multi-Chain: A blockchain platform designed to facilitate the deploying of customized permissioned blockchains. For example, the FoodFresh model (Stangl and Neumann 2023), which significantly enhances smart agriculture by enabling controlled data transparency and secure collaboration across agricultural supply chains. Multi-Chain can also help enhancing land record management in smart agriculture (Kumar et al. 2024).

4.1 Implementation Techniques

To harness blockchain effectively, the following implementation techniques have been adopted:

1. Smart Contracts: They automate processes like crop insurance payouts based on predefined conditions such as weather data or yield thresholds. For example, (Loukil et al. 2021) proposed CioSy, a blockchain-based collaborative insurance system that automates policy processing, claim handling, and payment through smart contracts, enabling peer-to-peer insurance while ensuring transparency and reducing operational costs.
2. IoT: IoT sensors combined with blockchain platforms monitor environmental conditions like temperature, humidity, and soil moisture (Ahmed et al. 2024). IoT-based aquaculture systems can leverage advanced monitoring technologies to track vital water quality parameters such as temperature and dissolved oxygen in real time, ensuring optimal fishpond conditions (Prapti et al. 2022).

5. Blockchain Applications on Smart Agriculture

Blockchain technology provides secure and temper-proof transactions without intermediaries and can transform various industries, including agriculture. There are many benefits to using blockchain technology in smart agriculture. These benefits include food safety, data transparency, traceability, and efficiency in supply chain (Yadav and Singh 2019). It also makes it possible to securely save information about the farms on blocks (Xiong et al. 2020).

In today's world, consumers are increasingly demanding more transparency in the food supply chain. They want to know where their food comes from, how it is produced, and whether it is safe to consume. However, the global food supply chain is complex, which makes it difficult to track and trace products. Blockchain technology provides a solution by creating a secure, transparent, and immutable ledger that can track the entire food supply chain. It can ensure that the food is produced and transported in compliance with regulations and ethical standards. Additionally, blockchain technology can improve farmers' livelihoods by giving them better access to sales, insurance, farm overseeing and other services.

Blockchain technology can provide transparency and traceability in the supply chain, so that farmers can track the movement of products from the farms to the consumers. This also provides consumers with information on the safety and quality of the food they are consuming, thus improving their confidence. In addition, blockchain helps farmers manage their farms more efficiently by providing them with real-time data on crop yields, weather conditions, and soil health. This data helps farmers make better decisions about crop selection, fertilization, and irrigation, and thus improving their productivity and profitability.

Accordingly, blockchain technology can be used in various areas of smart agriculture, including:

1. Production: It refers to the agricultural product that is produced using technology. Smart agriculture uses technology to produce more food and retain its sustainability.
2. Storage: It refers to the use of technology to optimize the storage of agricultural products. Smart storage can also involve optimizing inventory management and reducing waste.
3. Stock: It refers to the land, farm, livestock. Smart agriculture uses technology to manage these assets to improve productivity, sustainability, and profitability.

5.1 Production

Blockchain technology can be implemented in food supply chain and food traceability. Figure 3 shows where this technology can be implemented in food production.

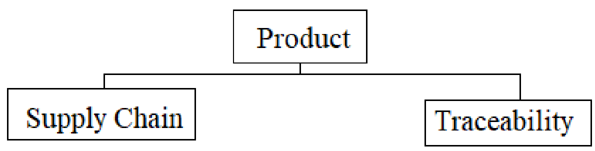

Figure 3. Areas where blockchain can be implemented in production.

5.1.1 Food Supply Chain

Supply chain is divided into two entities: organizations or individuals that are directly involved in products, services, finances, and data from seed planting to consumption (Mentzer et al. 2001). Food is one of the most important aspects that needs to be considered from a society perspective. It is the pillar of health, happiness, and economy. However, when it comes to delivering the product to consumer, the priority to consider is the security and personal safety of individuals ("Food Supply Chain Manag." 2007). Some blockchain applications in agri-food area are:

Chatterjee et al. (Chatterjee et al. 2023) explored the integration of Internet of Things (IoT) technology within Industry 4.0, underscoring its diverse applications across sectors like banking, manufacturing, healthcare, and government. Industrial IoT (IIoT) is specifically highlighted for its role in leveraging IoT services to link existing industrial processes with emerging technologies such as smart sensors, robotics, and artificial intelligence, with a particular focus on enhancing supply chain management. The rising consumer demand for organic food products has underscored the significance of traceability and transparency in food supply chains. Consequently, regulatory bodies are advocating for enhanced standards, prompting a shift from centralized to distributed systems to bolster fault tolerance and scalability. The design of food supply chain systems is examined through the lens of a hierarchical location problem, with recent research exploring optimization strategies. Additionally, they introduced a blockchain-based food supply chain system that was tailored for the agro-food industry. Their paper emphasized the system's advantages over traditional management methods, especially in terms of traceability and security. However, the proposed model does have some limitations, including scalability issues and a susceptibility to certain types of attacks. These limitations require future enhancements.

Mandela et al. (Mandela et al. 2023) introduced the complexities confronting agricultural supply chains in Andhra Pradesh, India, within the broader context of ensuring food safety and quality. They proposed using consensus algorithms to develop a blockchain-based query processing system, with the intention to augment transparency, accountability, traceability, and efficiency throughout the agricultural supply chain. Moreover, they underscored potential benefits, such as enhanced market access and sustainability. The study also covered precision agriculture, emphasizing its use of advanced technologies to enhance productivity and minimize waste, alongside the growing challenge of ensuring food safety. It highlights the significance of blockchain technology in addressing these challenges and bolstering food safety, traceability, and environmental sustainability. The authors identified consensus algorithms as pivotal for upholding the integrity and security of blockchain transactions, which would facilitate the establishment of dependable and transparent supply chains for smart agriculture.

In a study conducted by Leng et al. (Leng et al. 2018) they identified and discussed significant problems in the Chinese public service platform related to agriculture. and proposed a solution using public blockchain technology. This included problems such as a lack of resource matching mechanisms, low utilization rates, and suboptimal system performance. These issues all increased transaction costs and discouraged users from engaging with the platform. The authors also discussed concerns about transaction security, transparency, user privacy, and platform credibility. They suggested integrating a two-chain public blockchain system tailored for agricultural business resources into the existing platform. This integration would provide technical support, create a functional environment, and enhance the usage rate, credibility, and effectiveness of the platform while ensuring secure handling of diverse types of data.

One crucial procedure in Agri-food is quality measurement. Lucena et al. (Lucena et al. 2018) addressed the quality management of the grain throughout the transportation chain using blockchain to bring more efficiency and resilience to this process. They presented the Grain Exporters Business Network 'GEBN,' which is an enterprise that collects information from quality assurance processes and then provides data for diverse business partners of the Brazilian GEBN. It is made up of various stakeholders, such as grain producers, local credit unions, warehouse companies, trading exporters, agrochemical companies, freight forwarders, and ports authorities. The platform can assist producers to trace the products stored in warehouses.

Saberi et al. (Saberi et al. 2019) examined the application of blockchain technology for promoting sustainability within supply chains. With increasing pressure from both local and global governments and communities to achieve sustainability goals, there is a growing need to explore how blockchain can address supply chain sustainability. The authors proposed a transformation of traditional supply chains into blockchain-based systems, which involved four main entities: a registrar providing actor identification, a standards body defining blockchain guidelines and technical requirements, a certifier authorizing parties to participate in the supply chain, and several factors such as manufacturers, retailers, and customers (Project Provenance Ltd 2015). They also put forward future research suggestions to overcome barriers and promote the adoption of blockchain in supply chain management.

5.1.2 Traceability

Food traceability means following and tracing the documentation and linkage of all stages in the supply chain, both forwards and backwards, through which a food product and its ingredients move from production to distribution (FDA 2022). The utilization of blockchain in traceability systems ensures the reliability and authenticity of shared information (Tian 2016).

Researchers have been exploring the application of blockchain technology to enhance food safety. One notable study by Lin et al. (Lin et al. 2019) introduced a blockchain system to both prevent the tampering of food data and to address the shortcomings of the existing traceability systems. The authors developed a prototype system that combined blockchain and Electronic Product Code Information Services (EPICS) to trace the products. This distributed system allowed for the creation and sharing of visibility data, ensured the security of sensitive information through tamper-proof features while maintaining scalability. The integration of an enterprise-level smart contract ensured the confidentiality of business data and authenticated the identity of the enterprise. By leveraging the advantages of the EPCIS specification, which included ObjectEvent, AggregationEvent, QuantityEvent, and TransactionEvent, the proposed system demonstrated potential for effectively addressing food safety concerns.

Ferrández-Pastor et al. (Ferrández-Pastor et al. 2022) investigated the potential of integrating Internet of Things (IoT) facilities and ambient intelligence paradigms to optimize agronomic processes, with a focus on the hemp industry., Their goal was to enhance both traceability and security. They proposed a comprehensive model that amalgamates agricultural expertise, blockchain technology for value chain planning, and IoT protocols for digital traceability. The efficacy of the model was demonstrated through a proof-of-concept implementation, highlighting its ability to deliver tamper-proof and transparent traceability services. Additionally, the article underscored the significance of integrating information technologies to address consumer concerns regarding product safety, quality, and origin. Their proposed model offered numerous benefits, including the active engagement of agricultural experts, enhanced traceability, improved data security, process optimization, and cost savings through smart contracts. However, it also presented new challenges, such as ensuring seamless integration into existing systems, providing intuitive interfaces for farmers and technicians, and ensuring ongoing maintenance and updates. Overall, the article presented a promising approach to advancing traceability and resource optimization services in agricultural production processes, with the potential for future extensions and enhancements.

A study conducted by Sezer et al. (Sezer et al. 2022) discussed the critical aspects of traceability in supply chain management, underlining the pivotal role of customer trust. It pointed out the inadequacies of the existing frameworks in delivering efficient traceability, real-time data, and privacy safeguards. The article introduced a supply chain traceability framework that leveraged smart contracts to safeguard privacy from external entities. This framework incorporated a digital signature and verification mechanisms to uphold data integrity and authenticity. Thus, offering both anonymity and traceability according to user preferences. Furthermore, it discussed how blockchain technology enhances transparency and trust by securely storing transaction data in an immutable manner. The article also delved into the complexities of traceability in supply chains and stressed the need for systems that ensure product tracking while maintaining trust and privacy. The proposed framework, grounded in permissioned blockchain architecture, sought to strike a balance between anonymity and transparency while ensuring traceability and privacy through on-chain and off-chain smart contracts. Their experimental findings suggest that the framework presents a user-friendly and auditable model for supply chains. Future research endeavors include addressing space complexity using side chains, implementing lightweight blockchain architectures for IoT devices, and exploring real-world applications of the proposed framework.

Lin et al. (Lin et al. 2018) proposed a secure food traceability system that utilized blockchain technology and IoT devices to address food safety concerns. The system’s goal was to track and monitor the entire food production process, from seed cultivation to selling, and to reduce human intervention using IoT devices to record and verify. In their system, a combination of the traditional Enterprise Resource Planning (ERP) legacy system and a new IoT system was employed. Mobile phones served as a portal or blockchain thin node for farm companies, logistic companies, and customers to access the data stored in the chain.

Another study by Tse et al. (Tse et al. 2017) proposed blockchain technology as a solution to improve food safety standards in response to rising concerns in China. The current food safety systems failed to meet the required standards, which resulted in products classified as unsafe for trade. To address this problem, the authors suggested blockchain to secure information within the food supply chain. They applied tailored theoretical methods and conducted a market analysis to develop an efficient and reliable solution for managing agricultural product safety in China. Through the implementation of blockchain, the study sought to enhance the quality and safety of food products in the country, mitigating potential health risks and improving the overall quality of life.

Feng Tian (Tian 2016) proposed a food safety system that used blockchain technology to achieve transparency and openness in the food supply chain. This system tracked products in real-time using logistic companies, which eliminated the need for a centralized organization to oversee food safety information. All members of the system had access to an information platform, and blockchain ensured the integrity of the food data. The system focused on two categories of agri-food: fresh fruits and vegetables, and meat. It utilized RFID technology to acquire and share data throughout the entire production process, while blockchain certified the reliability of the information shared within the system. Overall, the system provided a secure and traceable platform for all members of the food supply chain.

Caro et al. (Caro et al. 2018) evaluated the effectiveness of AgrilockIoT, a system that utilized both IoT sensing devices and blockchain technology for agricultural traceability. The system generated digital values through the IoT devices and stored them securely in a blockchain, creating transparent and unchangeable records. The study found that using a permissioned blockchain, instead of a public blockchain, significantly improved the performance of the smart agriculture system. The permissioned blockchain had lower latency compared to the public blockchain, and thus offered faster and easier operation. Additionally, the public blockchain consumed approximately 50 percent more resources, resulting in poorer performance.

In a study conducted on using blockchain technology to track wood electronically throughout the supply chain. Figorilli et al. (Figorilli et al. 2018) implemented a blockchain architecture to enable traceability of the wood supply chain, simulating processes from tree cutting to the sawmill in Italy. Open source IoT devices collected data on tree species, date, position, number of logs, and commercial information, which was stored in a centralized database using a specific forest operations app. The blockchain system allows retrieval of historical information by tracing each tree's journey. Activation of the blockchain involved an activation code and data transmission, synchronized with a remote server. Two steps were required: authorization through Azure Blockchain workbench and data formatting, verification, and writing onto the blockchain. The application ensured that no unauthorized logs were inserted and provided progress feedback during the synchronization process.

Kumar and Iyengar (Kumar and Iyengar 2017) presented a case study of the application of blockchain technology to the rice supply chain. When participants in the chain registered in the system, they had unique identities and digital profiles which were stored in the blockchain. The rice supply chain has five phases: production, procurement, processing, distribution, and retailing. Blockchain was used to ensure traceability, combat fraud, and reduce errors by documenting all events that occurred in the chain. Each time the rice changed location, the information was recorded on the blockchain, creating a permanent record of its journey from the manufacturer to the consumer. The authors demonstrated how blockchain technology enhanced product safety and improved the efficiency of the rice supply chain through comprehensive traceability, event recording, and monitoring of rice quality and security.

5.2 Storage

Food products like rice and beans must go through a series of procedures such as production, grading, storage, and transportation before reaching the market. Throughout this process any fraudulent or tampered steps could pose significant risks to food safety. Utilizing IoT for the real-time tracking of these procedures can ensure that the entire process is traceable. However, it is important to note that traditional storage methods are vulnerable to manipulation or destruction of the stored data. To address this problem, blockchain technology offers a safe and decentralized database consisting of a series of secured blocks of data. Once a block has been confirmed and incorporated into the chain, it cannot be altered unless someone has control over more than half of the nodes simultaneously. This feature guarantees the reliability of the blockchain as data storage.

The agricultural process begins with seed storage, which can significantly impact seed quality and production. Thus, it is crucial to monitor seed storage. To ensure better control of the market price, the distribution and market prices must be properly monitored. To have effective monitoring, agriproducts must be traceable from seed storage to the consumer. The availability of verifiable data for the agriproducts not only increases transparency but also enables monitoring and manipulation of the system. Addressing these challenges is essential for ensuring a consistent supply of food products without any shortages.

There have been many studies conducted in agricultural data storage to investigate the potential applications and advantages of integrating blockchain technology. For instance, Xie et al. (Xie et al. 2017) designed a secure data storage scheme for food tracking that was based on blockchain technology. In this system, agricultural products were equipped with an IoT sensor module that acquired data in real-time and uploaded it to the server. The server used a double-chain storage structure consisting of two interconnected main and secondary blockchains to automatically store the data in the blockchain. This certified that agricultural tracking data could be stored securely using blockchain, thereby ensuring better food safety.

Zhang et al. (Zhang et al. 2023) introduced a blockchain-based traceability model for the grain and oil food supply chain, tailored for its inherent complexities and challenges. An outlier detection model was developed to ensure accurate data collection from IoT devices, which exhibited excellent performance in outlier detection. A storage model that combined blockchain with database and IPFS was introduced to improve storage efficiency, and to reduce pressure on the blockchain network. A data recovery mechanism was implemented to ensure the timely recovery of lost or damaged data, which enhanced the system fault tolerance. Experimental results demonstrated the effectiveness of the proposed model, with average query latencies indicating efficient data retrieval. Finally, a blockchain-based grain and oil food supply chain traceability system was designed and implemented using Hyperledger Fabric, which allowed for multi-source data uploading, lightweight storage, and data recovery in the supply chain.

To ensure the secure exchange and storage of vast amounts of remote sensing data, Zou et al. (Zou et al. 2023) presented a decentralized system utilizing blockchain technology. Because conventional centralized systems are susceptible to attacks, there is a risk of data loss. Distributed Ledger Technology (DLT) provides security, traceability, and tamper-proof capabilities suitable for remote sensing applications. However, there were some challenges integrating remote sensing data into blockchain networks due to its large volume and spatiotemporal characteristics. To address these challenges, they proposed a multi-level blockchain architecture. Remote sensing data was stored in the IPFS network, with its hash value uploaded to the Ethereum chain for public access. This distributed data storage approach improved security, facilitated information exchange, and increased data management efficiency. Additionally, the paper suggested a data storage security solution leveraging multiple blockchains to offer accountable and distributed storage. Utilizing the Ethereum blockchain platform, the prototype system was designed and simulated, displaying its effectiveness in terms of data storage speed, traceability, security, and availability. However, some challenges remained, such as the need to encrypt data for further security enhancement.

Lin et al. (Lin et al. 2017) proposed a storage security approach using blockchain. The continued progression of Moore's Law, Kryder's Law, and Nielsen's Law, which describe the increasing speed of data processing, decreasing costs of data storage respectively, combined with increasing bandwidth, suggests that blockchain technology can play a vital role in the agricultural industry. As a decentralized network, blockchain has the potential to democratize transactions and processes in the industry, making it the next evolutionary step for traditional Information and Communication Technology (ICT) based farming systems and e-agriculture initiatives. Through blockchain technology, agricultural and environmental monitoring data can be stored in a distributed cloud. This will lead to the achievement of sustainable agricultural development with the help of transparent data and ICT.

The proposed system was used for managing and sorting food monitoring information. They put forward a model of an ICT-based system which included a blockchain infrastructure. They suggested adding water quality monitoring data into the blockchain. Miner nodes in the network contributed its equipment to validate the blockchain and compile the complete water data.

Another study by Pranto et al. (Pranto et al. 2021) proposed the use of IoT to enhance the movement of vital data within the agricultural system. The IoT devices kept an eye on the quality and state of products stored in warehouses while providing information during the cultivation process. The monitored data was then securely saved in a blockchain, and a smart contract took care of automation, event triggering, and the enforcement of necessary terms and conditions for all parties involved.

5.3 Stock

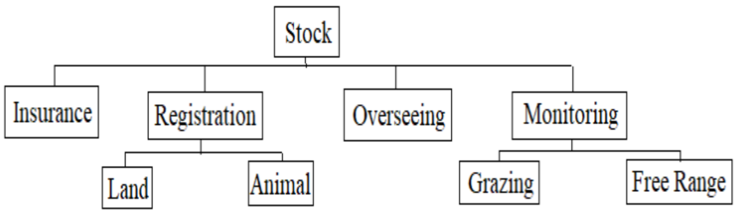

Figure. 4. Stock management in smart agriculture.

5.3.1 Crop Insurance

Farmers often do not trust insurance companies and fear delays or non-payment of claims, which makes crop insurance systems complex and challenging. However, some studies have suggested affordable and efficient crop insurance solutions that could benefit many farmers. One proposed solution was a blockchain-based crop insurance system, which provided a new infrastructure for storing, validating, and securely transferring data. Blockchain technology would ensure that the data is stored without manipulation and eliminates the need for third parties. This makes it a reliable solution for crop insurance.

Crop insurance is divided into two categories: yield protection insurance and calamity-based insurance. In yield protection insurance farmers can register a claim if their crop yield falls below a predetermined threshold. Calamity-based insurance automatically settles claims if a crop is destroyed due to a natural disaster. It does not require farmers to file a claim. Instead, when a natural disaster is recorded on the blockchain, a smart contract automatically initiates the payment process into the farmer's account.

Agricultural production is exposed to numerous uncontrollable risks. Most of the time, farmers have limited means to mitigate these risks. The most obvious being insurance, which is hindered by the lack of private sector involvement and by complicated claim procedures that discourage farmers from exploring new crops. In India, the current crop security system does not adequately address the escalating risks farmers face. To solve this problem, Iyer et al. (Iyer et al. 2021) proposed a decentralized peer-to-peer crop insurance framework to protect farmers' interests. This framework eliminated intermediaries and provided a secure, standardized, and transparent system that ensured all stakeholders had access to the necessary information. Through blockchain technology, the system promoted trust in a trustless environment and allowed for farmers and private investors to form a contract.

Omar et al. (Omar et al. 2023) presented an innovative approach to crop insurance using blockchain technology to tackle challenges like complexity, inflated costs, and a lack of trust in the conventional methods. Their blockchain-based crop index insurance solution recorded transactions and data exchanges on an immutable ledger, which ensured transparency, trust, and accountability. Smart contracts can streamline claim processing, reduce fraud and settlement delays, and lower costs by removing intermediaries. Their solution democratized crop insurance and expanded access to a wider range of farmers. It also included those in low-income countries. Their paper underscores the importance of tailoring the solution to meet the needs of smallholder farmers in developing nations and highlights the advantages of decentralization and blockchain in improving transparency and trust between farmers and insurers. The authors make their smart contract code openly available and outline plans for future work to automate additional insurance processes and tackle scalability and governance challenges within the blockchain landscape.

Jha et al. (Jha et al. 2021) proposed a decentralized platform for executing contracts and storing the results of crop insurance processes using smart contracts. This system helped insurance companies detect fraud and evaluate claims through smart contracts, which are more secure and less prone to exploitation than traditional insurance systems. Blockchain allowed for secure coordination between insurers, and records were stored on a secure distributed ledger. This made any malicious activity clear and then halted the transaction. The major stakeholders in this system included farmers, smart contracts, and insurance providers. The farmers provided personal data, crop information, and land coordinates for verification by insurance providers. Once verified, the insurance policy details were written on the blockchain as a smart contract hosted on a cloud platform, and insurance providers could verify weather conditions and determine the amount of money to be paid to the farmer in case of natural calamity.

The insurance industry is facing challenges related to the processing time, security, and settlement time of payments. However, blockchain technology can provide solutions to these problems. Bai et al. (Bai et al. 2022) proposed a novel approach that leveraged blockchain technology's immutability and integrity features to enhance the insurance process. They proposed a use case of blockchain and IoT technology to develop a transparent and secure framework that could improve processing time, security, and settlement time of payments. The proposed system sought to provide a more efficient and reliable insurance service through the reduction of the time delay in a claim settlement. The system assigned a unique ID to each participant, allowing them to read and write data on the blockchain. This made it possible for all stakeholders to upload all relevant information related to the insurance policy. To capture data from agricultural land, IoT devices and various sensors such as soil, moisture and fire sensors are then deployed. The use of IoT devices removes human interaction in the process.

5.3.2 Registration

Land registration is susceptible to tampering in various countries. Blockchain technology offers significant benefits when it comes to maintaining asset registers like property, vehicles, and contracts. Blockchain has specifically gained prominence in land registration due to its ability to address fraud concerns and provide a trusted and transparent system for storing and transferring data. By decentralizing and standardizing land registration records, blockchain reduced the need for intermediaries, enhanced trust in the transacting parties' identities, improved process efficiencies, and reduced the time and cost associated with registration (Deloitte 2018). Thus, technology must continue contributing to this field.

5.3.2.1 Land Registration

According to HernandoDe Soto out of the 7.3 billion people in the world, only around 2 billion possess legal and effective documentation confirming their ownership of an asset. The lack of legal record of ownership makes it impossible to use these assets as collateral to obtain credit or to transfer a portion of property as an investment. This implies that individuals cannot fulfill their potential to create credit, because their assets may be owned without proper documentation. Thus, HernandoDe Soto emphasizes the importance of effective land administration systems to ensure that property ownership is accurately recorded and recognized by the law (Barbieri and Gassen 2017).

In 2015, it was reported that the Government of Honduras was collaborating with Factom and Epigraph to create a land registry system using blockchain technology. This initiative sparked discussions and debates about the potential applications of blockchain in land administration (Anand et al. 2017). According to Anand et al. (Anand et al. 2017), some potential applications of blockchain technology included registering title deeds, creating time-stamped transactions, providing transparent governance tools for multiple parties, creating a tamper-proof recording system, establishing a disaster recovery system, and offering restitution and compensation in post-conflict zones.

Barbieri and Gassen (Barbieri and Gassen 2017) argue that the use of blockchain technology for land registers is still not fully understood. This is especially true when it comes to the well-established interplay between cadaster and land register, and the role of notary in the framework of preventive administration of justice. They suggested that advocates of blockchain solutions may not fully understand these aspects. Thus, from their perspective, blockchain technology appears to be useful only in the context of machine-to-machine communication at present.

Vos et al. (Vos et al. 2017) examined the potential of replacing an existing land administration system with a blockchain-based alternative. They highlighted the technical and administrative requirements that must be met for a blockchain-based land administration system to be successful. Blockchain technology can be used to archive transactions and to secure their content by storing transaction data on the blockchain. They suggested that blockchain technology could be useful in countries without a reliable electronic system for transferring ownership. However, they did note that traditional database systems may be sufficient in some cases.

In considering blockchain, several principles of Good Governance can be fulfilled. For example, the transparency and efficiency of transactions can be ensured, and a tamperproof history of transactions can be maintained. Blockchain-based land administration systems can function similarly to traditional land registry systems since they keep track of property ownership and transaction records and verify the authenticity of these records. Additionally, they can ensure the accuracy and timeliness of land transactions and prevent fraudulent activities.

5.3.2.2 Animal Registration

Cho and Lee (Cho and Lee 2019) introduced an animal administration system, that utilizes blockchain technology to distinguish between animals that are preregistered or not. This system can be implemented in various areas such as animal hospitals, pet stores, animal shelters, and pet insurance policies. N-printing technology, which is like how fingerprints are used to identify humans, can be used to identify animals. This authentication process uses nose-print recognition to identify animals and connect various clients through a blockchain network.

5.3.3 Farm Overseeing

Monitoring environmental information is vital for maintaining a healthy environment. Similarly, monitoring the agri-food environment is essential for ensuring food safety. Lin et al. (Lin et al. 2017) put forward an e-agriculture system model that utilized ICT and blockchain infrastructure for water distribution monitoring at the local and regional scale. The system added water quality monitoring data to the blockchain, which was then backed up and distributed across all nodes. Each node had a copy of the water data, and a query was created for reference purposes. The provider node could later cross-reference the data in the blockchain with the backed-up data if required.

The agricultural industry has started using IoT-based greenhouse technology that enabled remote monitoring and automation. However, the security concerns related to the large-scale and widely distributed network were still significant. To address these problems, Patil et al. (Patil et al. 2018) suggested utilizing blockchain technology to secure the emerging IoT-based greenhouse technology. The system model consisted of four groups; a smart greenhouse, an overlay network for reducing network overhead and delay, a cloud storage to store data from the greenhouse devices, and end users who own, control, and remotely monitor the greenhouse through their laptops or smartphones. The proposed architecture used a lightweight blockchain-based approach, which enhanced power consumption and benefits from private immutable ledgers. The IoT devices deployed in greenhouses act as a centrally managed blockchain, which further improved the security of the system.

Mujeye et al. (Mujeye et al. 2023) discussed the potential applications of 5G technology and IoT devices in agriculture and healthcare. Their focus was on addressing security and privacy concerns through the utilization of Blockchain technology. This study outlined a research endeavor involving Illinois State University and industry collaborators, who sought to deploy a private 5G network equipped with IoT sensors for monitoring soil moisture levels on a university farm. Additionally, the project's goal was to support underserved patients in managing chronic illnesses by remotely tracking vital signs through IoT devices. The study's primary objective was to evaluate the efficacy of Blockchain in securing communications between these sensors and healthcare providers. Furthermore, it emphasized the broader impacts of the research, including enhancing healthcare accessibility for marginalized communities and tackling agricultural issues such as soil nutrient depletion and water conservation. These efforts contributed to bolstering food security for the growing global population.

5.4.4 Monitoring

5.4.4.1 Livestock grazing

The swift expansion of blockchain technology in precision agriculture has resulted in the creation of several platforms with the potential to be used for a range of agricultural activities. For example, the AppliFarm platform, established by Neovia in 2017 (ADM 2023), enabled the provision of digital evidence for animal welfare and livestock grazing. The animal sector's livestock data could be tracked through the platform, which involved the use of linked tags placed around cows' necks to identify their grazing areas. This made it possible to collect enough data to ensure high-quality grazing for the livestock. In addition, their system could be used to ensure that monitored livestock farms adhere to animal welfare requirements. The animal welfare data is integrated into the system and accessible by stakeholders whenever they need access.

5.4.4.2 Free Range

Descovi et al. (Descovi et al. 2021) presented a case study based on blockchain of the health certification of poultry farms of breeding birds in the state of Rio Grande do Sul. They then mapped it with Business Process Model and Notation (BPMN). The presented blockchain system simplified the animal sanitary control of breeding birds and demonstrated the physical and digital flow of the process and how data is stored in the blockchain.

6. Discussion and Conclusion

Blockchain technology can revolutionize the agriculture industry by improving transparency, traceability, and efficiency of the food industry. In recent years, several studies have explored the applications of blockchain in smart agriculture, highlighting its potential to transform various areas such as product supply chain, product traceability, storage, farm, and stock management.

Agricultural data should be securely stored, if it is not protected from being maliciously tampered, many risks and drawbacks are encountered, such as affecting the smart agriculture efficiency and the potential danger of compromising data integrity. Since data is written in blocks unchangeably, blockchain enables stakeholders to securely track the movement of products from the farm to the consumer, which guarantees the safety of the data as well as the safety of the food. So, by providing consumers with information on the safety and quality of the food, blockchain can improve their confidence in smart agricultural products. Additionally, blockchain technology can be used to improve productivity and profitability. It can provide farmers with real-time data on crop yields, weather conditions, and soil health, helping them make better decisions. Blockchain could also be used to track animal health records, breeding information and genetic data, and monitor animal welfare and environmental impact. Regarding insurance, blockchain could be used to create smart contracts that automatically trigger insurance payouts when certain conditions are met, such as crop failure.

Table 1. Blockchain Applications in Smart Agriculture.

Study	Classification	Problem	Technologies Used Besides the Blockchain	The Proposed Technique	How Blockchain Becomes Beneficial
Chatterjee et al. (Chatterjee et al. 2023)	Product/ Supply Chain	The rising consumer demand for organic food products	IIoT	Blockchain-based solution for the food supply system	Data Integrity and Sharing: Immutability and Decentralization
Mandela et al. (Mandela et al. 2023)	Product/ Supply Chain	Fragmentation of land in Andhra Pradesh, India / lack of transparency, storage, and processing facilities	IoT	A blockchain-based query processing system for secure supply chain	Food Safety: Transparency, immutability, and improved traceability
Leng et al. (Leng et al. 2018)	Product/ Supply Chain	Lack of adaptive rent-seeking and matching mechanism / security and transparency of transactions and platform credibility	Double chain architecture	Public blockchain of agricultural supply chain system	Agricultural resource sharing: BC provided distributed rent-seeking and matching mechanism- improved the utilization rate of agricultural business resources and credibility of the public service platform
Lucena et al. (Lucena et al. 2018)	Product/ Supply Chain	Grain quality management	Smart contracts and IoT devices	GEBN Blockchain Business Network	Quality Management: Data integrity verification for quality management purposes / Efficiency and resilience of quality measurement in grains transportation
Ferrández-Pastor et al. (Ferrández-Pastor et al. 2022)	Product/ Traceability	Integration-related challenges in the implementation of facilities based on the IoT, embedded systems, and ambient intelligence paradigms in the hemp industry	IoT	IoT and Blockchain based model for traceability: Application in industrial hemp production	Data flow security
Sezer et al. (Sezer et al. 2022)	Product/ Traceability	Poor traceability, lack of real-time data, and lack of privacy in the supply chain	Cryptography and smart contract	TPPSUPPLY: A traceable and privacy-preserving blockchain system architecture	Real-time data, data privacy, and scalability
Lin et al. (Lin et al. 2019)	Product/ Traceability	Preventing sensitive information disclosure and data tampering	Enterprise-level smart contract / off-chain database	Food safety traceability system based on blockchain and EPCIS - management architecture of on-chain and off-chain data	Data Privacy and Integrity: Sensitive business information protection / enterprise identity verification / verification of data integrity
Lin et al. (Lin et al. 2018)	Product/ Traceability	Monitoring chemical use in farming, heavy metal contamination, low-quality raw materials, and excessive additives endanger food safety	IoT devices and smart contracts	Blockchain and LoRa IoT technology-based food traceability solution	Food Safety: Tracking and monitoring the entire food production process through verification mechanism and tamper-proof advantage
Tse et al. (Tse et al. 2017)	Product/ Traceability	Potential risks to people's health and limitations in precise traceability	N/A	Blockchain technology as a potential solution for enhancing information security	Food Safety: Immutable transaction records, automated monitoring and auditing system, and food safety and certification enhancement
Zhang et al. (Zhang et al. 2023)	Storage	The redundant data storage in the grain-and-oil-food-supply chain	IoT devices, MySQL database, machine learning, and IPFS	A blockchain-based traceability model for the grain-and-oil-food-supply chain	Enabling multi-source data uploading, lightweight storage, and data recovery

Zou et al. (Zou et al. 2023)	Storage	Incorporating the large volume and spatiotemporal characteristics of remote sensing data	IPFS	A remote sensing data storage model based on a multi-chain structure blockchain architecture	Data sharing and management
Lin et al. (Lin et al. 2017)	Storage	Agricultural data security and monitoring data sharing	ICT-based system	A model ICT e-agriculture system with a blockchain infrastructure	Data Integrity and sharing: Immutability and decentralization (instead of a centralized database)
Pranto et al. (Pranto et al. 2021)	Storage	Agricultural data sharing and monitoring	Smart contracts and IoT devices	A blockchain-based IoT and smart contracts	Data Integrity and sharing: Immutability, data availability, and transparency
Omar et al. (Omar et al. 2023)	Stock/ Crop Insurance	Complexity, excessive costs, and a lack of trust in conventional crop insurance methods	IPFS and Ethereum Smart contracts	Blockchain-based crop index insurance solution using Ethereum smart contract	Immutability and Transparency throughout the insurance ecosystem
Iyer et al. (Iyer et al. 2021)	Stock/ Crop Insurance	Farmers' risk vulnerability due to insurance gaps, private sector absence, crop security, and the need for innovative insurance products	Know Your Customer (KYC) verification and smart contract	A decentralized peer-to-peer crop insurance framework	Agricultural Insurance: Promotes trust in an untrusted environment by providing data integrity – The system enables the provision of insurance coverage for nonseasonal and unusually heavy rainfall.
Jha et al. (Jha et al. 2021)	Stock/ Crop Insurance	Cost of administering insurance and the great losses due to natural disasters	Smart contracts and a cloud environment	A blockchain based affordable crop insurance solution	Agricultural Insurance: Infrastructure for storing, validating and transfer of data securely
Vos et al. (Vos et al. 2017)	Stock/ Land Registration	Fraud, corruption, and lack of trust in classical land registration systems	Smart contracts	A comparison between a classical land registration and blockchain-based land registration system for good governance	Agricultural registration and record keeping: Ownership authentication, enhance trust, and reduce cost by eliminating intermediaries
Cho and Lee (Cho and Lee 2019)	Stock/ Animal Registration	Authenticate animals as specific preregistered entities in various contexts	Nose-print recognition	Nose-print based animal management system for animal registration and authentication	Authentication: Data integrity and accessibility
Mujeye et al. (Mujeye et al. 2023)	Stock/ Farm Overseeing	Failing to meet the quality of service (QoS)	5G and IoT sensors	Local 5G (L5G) network	Data sharing
Lin et al. (Lin et al. 2017)	Stock/ Farm Overseeing	Data security and monitoring data sharing	ICT-based system	A model ICT e-agriculture system with a blockchain infrastructure for water distribution monitoring	Data Integrity and sharing: Verify data integrity
Patil et al. (Patil et al. 2018)	Stock/ Farm Overseeing	Data sharing, irregular satellite monitoring, expensive security methods for energy consumption, and security in IoT	IoT devices	A lightweight blockchain based framework for smart greenhouse farming	Data Integrity and sharing: Data transparency, immutability, scalability, anonymity, distribution, and decentralization
Descovi et al. (Descovi et al. 2021)	Stock/ Monitoring/ Free Range	Exploring the application of blockchain in the health certification process for birds, and traceability of animal sanitary records	N/A	The integration of an animal sanitary control platform (PDSA-RS) with a private blockchain	Data integrity: Simplifying the animal sanitary control of breeding birds, traceability, immutability, transparency, anonymity, and auditability

Table 1 presents a range of blockchain applications within the context of smart agriculture, that address diverse problems, as outlined in the existing literature. The prevailing focus in many smart agriculture studies, as shown in table 1, revolves around data, particularly data integrity and its correlation with quality management standards and certifications. Various projects within the field require data integrity. Moreover, some projects employed secure data sharing underscoring the significance of data integrity as the primary challenge. In this regard, utilizing blockchain technology can potentially address these problems. For instance, a recent integration of blockchain technology within an IoT-based crop prediction system has ensured secure, tamper-resistant storage of sensor data and crop forecasts. This integration utilizes cryptographic techniques to enhance data integrity, decentralize data management, and establish trust among stakeholders (Sizan et al. 2023).

Blockchain's distributed nature ensures high availability, and its consensus algorithms provide immutability of records. Consequently, in literature, many blockchain applications leverage the integrity property of blockchain to provide services such as auditing insurance, bookkeeping, and secure data storage. These services are closely related to quality management, standardization, and certification purposes. There is also a common use in the market for blockchain technology, particularly when high availability is essential. This makes it a versatile tool within the field of smart agriculture.

In a distributed environment, blockchain can play a crucial role in maintaining data through its high availability and integrity verification. It can either serve as the primary repository for data or support any existing information services by ensuring the integrity of the stored data. Additionally, blockchain systems commonly integrate with IoT devices and systems, which generate data that can be automatically recorded in the blockchain. Through the application of blockchain, the integrity of data can be guaranteed, facilitating the automation of day-to-day monitoring operations in smart agriculture. For example, a study aimed at monitoring the impact of fertilizers on agricultural land and related parameters, such as increased temperature, reduced moisture, and light intensities, by recording data in a system using IoT sensors (Rehman et al. 2023).

Effective bookkeeping also plays a significant role in tracking land and animal information. In turn, this aids insurance systems and facilitates insurance audits, serving audit purposes. This process enhances the monitoring standards and certifications of insurance providers. Given the overarching emphasis on data integrity, blockchain technology has become widely adopted for verifying and ensuring the integrity of data across various applications.

Furthermore, it is noteworthy that blockchain is often employed in conjunction with IoT devices and systems to address different problems, automate non-traditional approaches, and devise innovative automated solutions. Many of the data management practices in smart agriculture involve information services, where data is stored in databases and accessible for querying. Blockchain can serve as the underlying database for such information services, and it can be employed to check if the information contained therein is still valid.

While the utilization of smart contracts in smart agriculture remains limited, they have found particular use in insurance and can also be applied to sales within the market. For instance, blockchain-based crop index insurance has been introduced as an innovative solution to overcome the complexities, high costs, and trust issues that plague traditional crop insurance methods. By utilizing blockchain technology, the solution ensures transparency, as every transaction and data exchange is recorded on an immutable ledger. Smart contracts enable autonomous claims processing, significantly reducing the risk of fraud and expediting payouts to farmers. This system also reduces administrative costs by eliminating intermediaries and streamlining processes, making crop insurance more affordable and accessible, especially for farmers in low-income regions (Omar et al. 2023).

Table 1 highlights different areas in agriculture where blockchain technology is applied to address various research problems and improve the overall agricultural processes. In the literature, studies regarding using blockchain in agriculture are divided into three main categories: production, storage, and stock. Blockchain is used in product supply chain as a solution to enhance its functionality, usage, and credibility. It also focuses on improving grain quality management throughout the transportation chain by leveraging blockchain technology and using the GEBN to enhance efficiency and resilience. Blockchain also addresses security concerns in supply chain information exchange and ensures data integrity and authentication, mitigating risks in the agricultural domain.

Studies in product traceability discuss food safety (Chatterjee et al. 2023; Tian 2016; Kumar and Iyengar 2017; Leng et al. 2018; Lin et al. 2018; Lin et al. 2019; Lucena et al. 2018; Mandela et al. 2023; Saberi et al. 2019; Snyder et al. 2017; Tse et al. 2017) and traceability (Ferrández-Pastor et al. 2022; Sezer et al. 2022); Caro et al. 2018; Tian 2016; Figorilli et al. 2018; Kumar and Iyengar 2017; Lin et al. 2018; Lin et al. 2019; Tse et al. 2017) in the agricultural domain. It addresses the problems of tampering with food data (Chatterjee et al. 2023; Mandela et al. 2023); Tian, 2016; Lin et al. 2018; Lin et al. 2019; Lucena et al. 2018) and the limitations of existing traceability systems (Caro et al. 2018; Leng et al. 2018; Tse et al. 2017). The studies also highlight concerns regarding food safety (Chatterjee et al. 2023; Mandela et al. 2023; Tian 2016; Kumar and Iyengar 2017; Leng et al. 2018; Lin et al. 2018; Lin et al. 2019; Lucena et al. 2018; Saberi et al. 2019; Tse et al. 2017) and emphasize that current food safety systems are inadequate to meet the required standards. Blockchain technology is proposed as a solution to enhance traceability in food supply chain, thereby improving food safety and consumer confidence.

Storing agricultural data is also a pressing concern in smart agriculture. The studies focus on some key challenges in the agricultural sector. For example, they address the difficulty of securely storing agricultural and environmental monitoring data (Zhang et al. 2023; Zou et al. 2023; Lin et al. 2017; Pranto et al. 2021; Xie et al. 2017). They also emphasize the movement of vital data (Lin et al. 2017; Pranto et al. 2021; Xie et al. 2017; Zhang et al. 2023; Zou et al. 2023) within the agricultural system. To overcome these challenges, studies (Zhang et al. 2023; Zou et al. 2023; Lin et al. 2017; Pranto et al. 2021; Xie et al. 2017) leverage using blockchain technology that offers a secure and decentralized solution for data storage and management, ensuring the availability and integrity of vital agricultural information.

The literature focuses on improving crop insurance using blockchain technology. Studies (Omar et al. 2023; Bai et al. 2022; Iyer et al. 2021; Jha et al. 2021) in this domain present some approaches such as a decentralized peer-to-peer crop insurance framework (Iyer et al. 2021) to protect farmers' interests and facilitate secure contracts with investors, and a decentralized platform with smart contracts (Jha et al. 2021) to detect fraud and coordinate insurers and farmers efficiently. Blockchain and IoT-based framework was also used in (Bai et al. 2022) to enhance insurance processing, security, and settlement by eliminating human interaction.

This paper covers two main topics in stock registration. Firstly, it discusses the potential applications of blockchain technology in land registration and administration (Anand et al. 2017; Barbieri and Gassen 2017; Vos et al. 2017), highlighting its advantages as well as addressing some concerns about the complexity of integrating it in existing land registry systems. Secondly, the paper presents an animal administration system research that utilizes blockchain for animal identification using nose-print recognition (Cho and Lee 2019), allowing for secure identification, and tracking of animals in various areas such as animal hospitals, pet stores, shelters, and pet insurance.

Blockchain can serve as a useful tool in farm overseeing. For example, Lin et al. (Lin et al. 2017) proposed an e-agriculture system model that utilizes ICT and blockchain for water distribution monitoring. Water quality data is added to the blockchain, ensuring its integrity and availability across all nodes. IoT devices can also be employed as a centrally managed blockchain, providing a more secure and efficient system for remote greenhouse monitoring and automation.

The literature includes some studies related to the application of blockchain technology in monitoring livestock grazing and free range. The AppliFarm platform (ADM 2023) enables digital evidence for animal welfare and livestock grazing through linked tags to track their grazing areas and ensure high-quality grazing for the livestock. Another case study on blockchain's application in the health certification of poultry farms was presented, demonstrating the simplification of animal sanitary control for breeding birds and data storage in the blockchain.

Blockchain technology in smart agriculture faces several technical challenges that need to be addressed to enable its broader adoption. One of the key issues is scalability. As the number of connected IoT devices increases, blockchain networks may struggle to handle the large volume of transactions generated by these devices, which can lead to slower processing times and higher operational costs. To address scalability and energy consumption challenges, alternative consensus mechanisms like Proof of Stake (PoS), used by Ethereum 2.0 and Cardano, provide energy efficiency while maintaining robust security. Additionally, Delegated Proof of Stake (DPoS), employed by platforms such as EOS and Tron, further improves scalability but comes with the trade-off of increased centralization (Alkhodair et al. 2023).

Another significant challenge is data privacy. Public blockchain platforms are transparent by design, which could expose sensitive agricultural data to unauthorized parties. To overcome this, permissioned blockchains can be implemented to restrict access, ensuring that only authorized participants can access certain data. Furthermore, techniques like Zero-Knowledge Proofs (ZKPs) (Zhang et al. 2023) can allow data to be verified without revealing the underlying information, ensuring privacy while maintaining transparency.

Another major limitation is the absence of standardized regulations and international frameworks, which restrict interoperability and delay widespread implementation. Connectivity issues in rural areas further complicate participation, as blockchain networks require reliable internet access. Additionally, the technology demands substantial IoT infrastructure to ensure accurate and trustworthy data input, creating logistical and financial hurdles. Uncertainty regarding the legal interpretation of smart contracts and governance frameworks adds to the complexity, posing legal and operational risks. Overcoming these regulatory, infrastructural, and accessibility challenges is essential for blockchain to achieve its transformative potential in enhancing transparency, efficiency, and sustainability in the agricultural sector (AGDAILY 2024).

Different blockchain systems may use incompatible data formats or communication protocols, making it difficult to integrate them with existing agricultural systems and it can arise communication issues. The development of universal standards for data exchange and protocol integration would facilitate smoother interoperability and greater collaboration across platforms, leading to more efficient blockchain-based solutions in agriculture. Addressing these technical limitations will be crucial in unlocking the full potential of blockchain to improve transparency, efficiency, and security in the agriculture sector.

Although there are significant studies have examined various aspects of smart agriculture, researchers are increasingly directing their focus toward the supply chain (Ahamed and Vignesh 2022; Ashfaq 2022; Chatterjee et al. 2023; Haji et al. 2022; Kumar and Dwivedi 2023; Mandela et al. 2023), encompassing the entirety of activities from crop cultivation to consumer. This shift is occurring because individuals are increasingly recognizing the importance of effectively managing this process to ensure that farming operations are conducted efficiently and with traceability. Delving into the supply chain, researchers aim to identify strategies for resource optimization, product enhancement, and waste reduction in agricultural operations. Additionally, the increasing consumer consciousness regarding food provenance and production methods underscores the need to ensure transparency within the supply chain. Technologies like blockchain help achieve this goal by enabling the enhanced tracking of food origins and the handling processes. Consequently, the exploration of the relationship between smart agriculture and the supply chain is emerging as a significant area of research, which will yield substantial improvements in agricultural practices for broader societal benefit.

Despite the numerous benefits of using blockchain in smart agriculture, there are still several open questions and areas that require further research. For instance, using blockchain to track soil quality metrics such as nutrient levels, pH, and organic matter content. This data could then be used to optimize soil health and productivity and identify areas requiring specific treatments. In smart agriculture, Blockchain technology can enable several transformative capabilities. For example, the creation of decentralized markets, and fostering direct connections between farmers and buyers. Additionally, blockchain can facilitate the development of auditing systems to monitor and support environmentally friendly farming practices. By utilizing smart contracts, the use of intermediaries can be minimized, which will lead to reduced food prices and improved efficiency in the agricultural supply chain. Finally, blockchain could be used to track research data, funding, and collaborations in agricultural research, enabling greater sharing and collaboration among researchers.

Feature studies on blockchain applications in agriculture should focus on several emerging areas. Developing standardized frameworks is essential to enable seamless integration of blockchain with IoT and AI technologies, fostering interoperability across industries. Studies are needed to assess the economic and operational viability of blockchain solutions, particularly in resource-constrained farming environments. Exploring hybrid blockchain models that combine public and private chains could help achieve a balance between transparency and scalability. Furthermore, addressing ethical considerations and socio-economic impacts is critical to ensure equitable benefits for smallholder farmers and reduce potential disparities. Finally, localized investigations should examine how blockchain can address region-specific challenges, such as enhancing drought resilience in arid regions or implementing disease tracking systems in livestock farming, tailoring solutions to the unique needs of different agricultural contexts.

In conclusion, this paper has highlighted the numerous applications of blockchain technology in smart agriculture, including product supply chain management, storage, farm, and stock management. By leveraging the power of blockchain, we can create a more transparent, sustainable, and equitable food system for all. However, further research is required to address open questions and areas to ensure the successful integration of blockchain into the agriculture industry. Overall, this paper contributes to the ongoing discussion on the potential of blockchain in smart agriculture and highlights the need for further research to realize its full potential.

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References

ADM (2023). Pioneering solutions in animal nutrition. https://www.adm.com/en-us/products-services/animal-nutrition/ (access date: 06.12.2024).

AGDAILY (2024). How blockchain technology is transforming the agricultural industry. https://www.agdaily.com/smartnews/blockchain-technology-transforming-agricultural-industry/ (access date: 06.12.2024).

Ahamed NN, Vignesh R (2022). Smart agriculture and food industry with blockchain and artificial intelligence. *Journal of Computer Science* 18(1): 1–17.

Ahmadi S (2023). A systematic literature review: security threats and countermeasures in smart farming. *Authorea Preprints*.

Ahmed A, Parveen I, Abdullah S, Ahmad I, Alturki N, Jamel L (2024). Optimized data fusion with scheduled rest periods for enhanced smart agriculture via blockchain integration. IEEE Access.

Ahmed RA, Hemdan EE, El-Shafai W, Ahmed ZA, El-Rabaie EM, Abd El-Samie FE (2022). Climate-smart agriculture using intelligent techniques, blockchain, and Internet of Things: concepts, challenges, and opportunities. *Transactions on Emerging Telecommunications Technologies* 33(11).

Alam S (2023). Security concerns in smart agriculture and blockchain-based solution. In: *2022 OPJU International Technology Conference on Emerging Technologies for Sustainable Development (OTCON),* 8 February 2023; IEEE, pp. 1-6.

Alkhodair AJ, Mohanty SP, Kougianos E (2023). Consensus algorithms of distributed ledger technology: a comprehensive analysis. *arXiv preprint arXiv*:2309.13498.

Anand A, McKibbin M, Pichel F (2017). Colored coins: Bitcoin, blockchain, and land administration. In: *Annual world bank conference on land and poverty*, 14 March 2016; pp. 20-24.

Ashfaq A (2022). Application of blockchain technology and IoT-based smart sensors in supply chain management via business development approach. Thesis of Masters, Polıtecnico di Torino, Italy.

Atalay M (2023). An overview of digital twin applications on smart agriculture. In: *2023 International Balkan Conference on Communications and Networking (BalkanCom),* 5 June 2023; IEEE, pp. 1-5.

Bai P, Kumar S, Kumar K (2022). Use of blockchain-enabled IoT in insurance: A case study of calamity-based crop insurance. In: *2022 Third International Conference on Intelligent Computing Instrumentation and Control Technologies (ICICICT)*, 11 August 2022; IEEE, pp. 1135-1141.

Barbieri M, Gassen D (2017). Blockchain: Can this new technology really revolutionize the land registry system. In: *Responsible land governance: towards an evidence based approach: proceedings of the annual World bank conference on land and poverty*, 20 March 2017, pp. 1-13.

Basharat A, Mohamad MM (2022). Security challenges and solutions for Internet of Things-based smart agriculture: a review. In: *2022 4th International Conference on Smart Sensors and Application (ICSSA),* 26 July 2022; IEEE, pp. 102-107.

Bellemare MF, Bloem JR (2018). Does contract farming improve welfare? A review. *World Development* 112: 259–271.

Bi J, Liu Y (2023). Research on agricultural machinery control system based on adaptive control. In: *2023 Asia-Europe Conference on Electronics, Data Processing and Informatics (ACEDPI)*, 17 April 2023; IEEE, pp. 45-49.

Bogoviz AV, Osipov VS, Vorozheykina TM, Yankovskaya VV, Sklyarov IY (2023). Food security in the digital economy: traditional agriculture vs. smart agriculture based on artificial intelligence. In: *Food Security in the Economy of the Future: Transition from Digital Agriculture to Agriculture 4.0 Based on Deep Learning,* 26 March 2023; pp. 59-74.

Caro MP, Ali MS, Vecchio M, Giaffreda R (2018). Blockchain-based traceability in agri-food supply chain management: a practical implementation. In: *2018 IoT Vertical and Topical Summit on Agriculture-Tuscany (IOT Tuscany),* 8 May 2018; Tuscany, Italy, pp. 1-4.

Chatterjee K, Singh A, Neha (2023). A blockchain-enabled security framework for smart agriculture. *Computers and Electrical Engineering* 106: 108594.

Cho J-Y, Lee S (2019). Animal administration system using nose-print recognition and blockchain network. *Journal of IKEEE* 23(4): 1477–1480.

Dalohoun DN, Hall A, Van Mele P (2009). Entrepreneurship as driver of a self-organizing system of innovation: The case of NERICA in Benin. *International Journal of Technology Management and Sustainable Development* 8(2): 87–101.

Dangi A (2004). Revolutionizing agriculture: wireless sensor network approaches for precision farming. *International Journal of Advance Scientific Research* 4: 1–7.

Deloitte (2018). Blockchain in public sector: Transforming government services through exponential technologies. https://www2.deloitte.com/in/en/pages/public-sector/articles/blockchain-in-public-sector.html (access date: 06.12.2024).

Descovi G, Maran V, Ebling D, Machado A (2021). Towards a blockchain architecture for animal sanitary control. In: *ICEIS (1)*, 2021; pp. 305-312.

FAO (2009). Global agriculture towards 2050. How to Feed the World 2050. https://www.fao.org/fileadmin/templates/wsfs/docs/Issues_papers/HLEF2050_Global_Agriculture.pdf (access date: 05.12.2024).

FDA (2022). Tracking and tracing of food. https://www.fda.gov/food/new-era-smarter-food-safety/tracking-and-tracing-food#:~:text=Food traceability is the ability,of food products and ingredients (access date: 06.12.2024).

Feng T (2016). An agri-food supply chain traceability system for China based on RFID and blockchain technology. In: *2016 13th international conference on service systems and service management (ICSSSM),* 24 July 2016; IEEE, pp. 1-6.

Ferrag MA, Shu L, Yang X, Derhab A, Maglaras L (2020). Security and privacy for green IoT-based agriculture: Review, blockchain solutions, and challenges. *IEEE Access* 8: 32031–32053.

Ferrández-Pastor F-J, Mora-Pascual J, Díaz-Lajara D (2022). Agricultural traceability model based on IoT and Blockchain: Application in industrial hemp production. *Journal of Industrial Information Integration* 29: 100381.

Figorilli S, Antonucci F, Costa C, Pallottino F, Raso L, Castiglione M, Pinci E, Del Vecchio D, Colle G, Proto A, Sperandio G, Menesatti P (2018). A Blockchain implementation prototype for the electronic open source traceability of wood along the whole supply chain. *Sensors* 18(9): 3133.

Gkogkos G, Lourenço P, Pechlivani EM, Encarnação L, Votis K, Giakoumoglou N, Tzovaras D (2023). Distributed ledger technologies for food sustainability indexing. *Smart Agricultural Technology* 5: 100312.

Haji M, Kerbache L, Al-Ansari T (2022). Food quality, drug safety, and increasing public health measures in supply chain management. *Processes* 10(9): 1715.

Hu Z, Kim B, Jeong J (2024). A Study on mechanization, data, and insurance challenges for agriculture based on hyperledger fabric. *IEEE Access.*

Hua J, Wang X, Kang M, Wang H, Wang F-Y (2018). Blockchain based provenance for agricultural products: a distributed platform with duplicated and shared bookkeeping. In: *2018 IEEE intelligent vehicles symposium (IV),* 26-30 June 2018; pp. 97-101.

Iyer V, Shah K, Rane S, Shankarmani R (2021). Decentralised peer-to-peer crop insurance. In: *Proceedings of the 3rd ACM International Symposium on Blockchain and Secure Critical Infrastructure* 24 May 2021, pp. 3-12.

Jha N, Prashar D, Khalaf OI, Alotaibi Y, Alsufyani A, Alghamdi S (2021). Blockchain based crop insurance: a decentralized insurance system for modernization of indian farmers. *Sustainability* 13(16): 8921.

Kaur P, Parashar A (2022). A systematic literature review of blockchain technology for smart villages. *Archives of Computational Methods in Engineering* 29(4): 2417–2468.

Kechagias EP, Gayialis SP, Papadopoulos GA, Papoutsis G (2023). An ethereum-based distributed application for enhancing food supply chain traceability. *Foods* 12(6): 1220.

Kumar A, Vishwakarma L, Das D (2024). LandChain: A multichain based novel secure land record transfer system. *IEEE Transactions on Reliability.*

Kumar D, Dwivedi RK (2023). Blockchain and IoT based smart agriculture and food supply chain system. In: *2023 International Conference on Intelligent and Innovative Technologies in Computing, Electrical and Electronics (IITCEE),* 27 January 2023; IEEE, pp. 755-761.

Kumar MV, Iyengar NCSN (2017). A Framework for blockchain technology in rice supply chain management plantation. *Advanced Science and Technology Letters* 146: 125–130.

Kwaghtyo DK, Eke CI (2023). Smart farming prediction models for precision agriculture: a comprehensive survey. *Artificial Intelligence Review* 56(6): 5729–5772.

LB K (2022). Survey on the applications of blockchain in agriculture. *Agriculture 12*(9), 1333.

Leng K, Bi Y, Jing L, Fu HC, Van Nieuwenhuyse I (2018). Research on agricultural supply chain system with double chain architecture based on blockchain technology. *Future Generation Computer Systems* 86(1): 641–649.

Lin J, Shen Z, Zhang A, Chai Y (2018). Blockchain and IoT based Food traceability for smart agriculture. In: *Proceedings of the 3rd International Conference on Crowd Science and Engineering,* 28 July 2018; pp. 1-6.

Lin Q, Wang H, Pei X, Wang J (2019). Food safety traceability system based on blockchain and EPCIS. *IEEE Access* 7: 20698–20707.

Lin YP, Petway J, Anthony J, Mukhtar H, Liao S-W, Chou C-F, Ho Y-F (2017). Blockchain: The evolutionary next step for ICT E-agriculture. *Environments* 4(3): 50.

Loukil F, Boukadi K, Hussain R, Abed M (2021). Ciosy: A collaborative blockchain-based insurance system. *Electronics* 10(11): 1343.

Lucena P, Binotto APD, Momo F da S, Kim H (2018). A case study for grain quality assurance tracking based on a blockchain business network. *arXiv preprint arXiv:*1803.07877.

Mandela S, Mohan RNVJ, Naik MC (2023). Blockchain-based consensus for a secure smart agriculture supply chain. *European Chemical Bulletin* 12(4): 8669–8678.

Mentzer JT, DeWitt W, Keebler JS, Min S, Nix NW, Smith CD, Zacharia ZG (2001). Defining supply chain management. *Journal of Business Logistics* 22(2): 1–25.

Mujeye S, Qaddour J, Ullah S, Calderon S, Rhykerd R, Edamala C, Kidwaro F (2023). A proposal on how to use blockchain to secure communications in the 5G ecosystem. *International Journal of Future Computer and Communication* 12(1): 14–18.

Ojha T, Misra S, Raghuwanshi NS (2015). Wireless sensor networks for agriculture: The state-of-the-art in practice and future challenges. *Computers and Electronics in Agriculture* 118: 66–84.

Omar IA, Jayaraman R, Salah K, Hasan HR, Antony J, Omar M (2023). Blockchain-based approach for crop index insurance in the agricultural supply chain. *IEEE Access,* 11: 118660–118675.

Patil AS, Tama BA, Park Y, Rhee K-H (2018). A framework for blockchain-based secure smart greenhouse farming. *Lecture Notes in Electrical Engineering* 474: 1162–1167.

Pranto TH, Noman AA, Mahmud A, Haque AB (2021). Blockchain and smart contract for IoT-enabled smart agriculture. *PeerJ Computer Science*, 7: e407.

Prapti DR, Mohamed Shariff AR, Che Man H, Ramli NM, Perumal T, Shariff M (2022). Internet of things (IoT)-based aquaculture: An overview of IoT application on water quality monitoring. *Reviews in Aquaculture* 14(2): 979–992.

Project Provenance Ltd. (2015). Blockchain: The solution for transparency in product supply chains. https://www.provenance.org/news-insights/blockchain-the-solution-for-transparency-in-product-supply-chains (access date: 06.12.2024).

Quy VK, Hau NV, Anh DV, Quy NM, Ban NT, Lanza S, Randazzo G, Muzirafuti A (2022). IoT-enabled smart agriculture: Architecture, applications, and challenges. *Applied Sciences* 12(7): 3396.

Raj EF, Appadurai M, Athiappan K (2021). Precision farming in modern agriculture. In: *Smart agriculture automation using advanced technologies: Data analytics and machine learning, cloud architecture, automation and IoT*, 1 January 2022; Singapore, pp. 61-87.

Rehman KU, Andleeb S, Ashfaq M, Akram N, Akram MW (2023). Blockchain-enabled smart agriculture: Enhancing data-driven decision-making and ensuring food security. *Journal of Cleaner Production* 427: 138900.

Saberi S, Kouhizadeh M, Sarkis J, Shen L (2019). Blockchain technology and its relationships to sustainable supply chain management. *International Journal of Production Research* 57(7): 2117–2135.

Sakib SMN (2024). Blockchain technology for smart contracts. In: *Achieving Secure and Transparent Supply Chains With Blockchain Technology* 2024; IGI Global, pp. 246–266.

Searchinger T, Waite R, Beringer T, Hanson C, Ranganathan J, Dumas P, Matthews E (2018). Creating a sustainable food future. *The World Resources Institute* pp. 1–96.

Sezer BB, Topal S, Nuriyev U (2022). TPPSUPPLY: A traceable and privacy-preserving blockchain system architecture for the supply chain. *Journal of Information Security and Applications*, 66: 103116.

Shaikh FK, Karim S, Zeadally S, Nebhen J (2022). Recent trends in internet-of-things-enabled sensor technologies for smart agriculture. *IEEE Internet of Things Journal,* 9(23): 23583–23598.

Shrivastava AL, Dwivedi RK (2023). Blockchain-based secure land registry system using efficient smart contract. In: *2023 International Conference on Intelligent Data Communication Technologies and Internet of Things (IDCIoT),* 5 January 2023; IEEE, pp. 165-170.

Sinha BB, Dhanalakshmi R (2022). Recent advancements and challenges of Internet of Things in smart agriculture: A survey. *Future Generation Computer Systems* 126: 169-184.

Sizan NS, Dey D, Mia MS, Layek MA (2023). Revolutionizing Agriculture: An IoT-Driven ML-Blockchain framework 5.0 for optimal crop prediction. In: *2023 5th International Conference on Sustainable Technologies for Industry 5.0 (STI),* 9 December 2023; IEEE, pp. 1-6.

Snyder D, Garcia-Romero D, Povey D, Khudanpur S (2017). Deep neural network embeddings for text-independent speaker verification. In: *Interspeech,* 20 August 2017; pp. 999-1003.

Srbinovska M, Gavrovski C, Dimcev V, Krkoleva A, Borozan V (2015). Environmental parameters monitoring in precision agriculture using wireless sensor networks. *Journal of Cleaner Production* 88: 297-307.

Srikanth M, Mohan RJ, Naik MC (2024). Agricultural supply chain efficiency and decreased risk through the use of hyperledger fabric and smart contracts. *Journal of Nonlinear Analysis and Optimization* Vol. 15, Issue. 1, No.6: 2024 ISSN: 1906-9685

Stangl P, Neumann CP (2023). Food fresh: Multi-chain design for an inter-institutional food supply chain network. *arXiv preprint arXiv:*2310.19461.

Sun N, Fan B, Ding Y, Liu Y, Bi Y, Seglah PA, Gao C (2023). Analysis of the development status and prospect of china's agricultural sensor market under smart agriculture. *Sensors* 23(6): 3307.

Thomas RJ, O'Hare G, Coyle D (2023). Understanding technology acceptance in smart agriculture: A systematic review of empirical research in crop production. *Technological Forecasting and Social Change* 189: 122374.

Tse D, Zhang B, Yang Y, Cheng C, Mu H (2017). Blockchain application in food supply information security. In: *2017 IEEE International Conference on Industrial Engineering and Engineering Management (IEEM)*, 10 December 2017; IEEE, pp. 1357-1361.

Vijay Hari Ram V, Vishal H, Dhanalakshmi S, Meenakshi Vidya P (2020). Regulation of water in agriculture field using Internet of Things. *Journal of Xidian University* 14(4): 112-115.

Vos J, Lemmen C, Beentjes B (2017). Blockchain-based land administration: Feasible, illusory or panacea. In: *18th Annual World Bank Conference on Land and Poverty 2017: Responsible Land Governance: Towards and Evidence Based Approach*, 2017; The World Bank.

Xie C, Sun Y, Luo H (2017). Secured data storage scheme based on block chain for agricultural products tracking. In: *2017 3rd International Conference on Big Data Computing and Communications (BIGCOM)*, 10 August 2017; IEEE, pp. 45-50.

Xiong H, Dalhaus T, Wang P, Huang J (2020). Blockchain technology for agriculture: Applications and rationale. *Frontiers in Blockchain* 3: 7.

Yadav VS, Singh AR (2019). A systematic literature review of blockchain technology in agriculture. In: *Proceedings of the International Conference on Industrial Engineering and Operations Management* 23 July 2019; Southfield, MI, USA, pp. 973-981.

Yang X, Shu L, Chen J, Ferrag MA, Wu J, Nurellari E, Huang K (2021). A survey on smart agriculture: development modes, technologies, and security and privacy challenges. *IEEE/CAA Journal of Automatica Sinica* 8(2): 273-302.

Zhang B, Xu J, Wang X, Zhao Z, Chen S, Zhang X (2023). Research on the construction of grain food multi-chain blockchain based on zero-knowledge proof. *Foods* 12(8): 1600.

Zhang Y, Wu X, Ge H, Jiang Y, Sun Z, Ji X, Jia Z, Cui G (2023). A blockchain-based traceability model for grain and oil food supply chain. *Foods* 12(17): 3235.

Zou Q, Yu W, Bao Z (2023). A blockchain based framework for remote sensing data management. *preprints.org*. Doi: 10.20944/preprints202308.0178.v1