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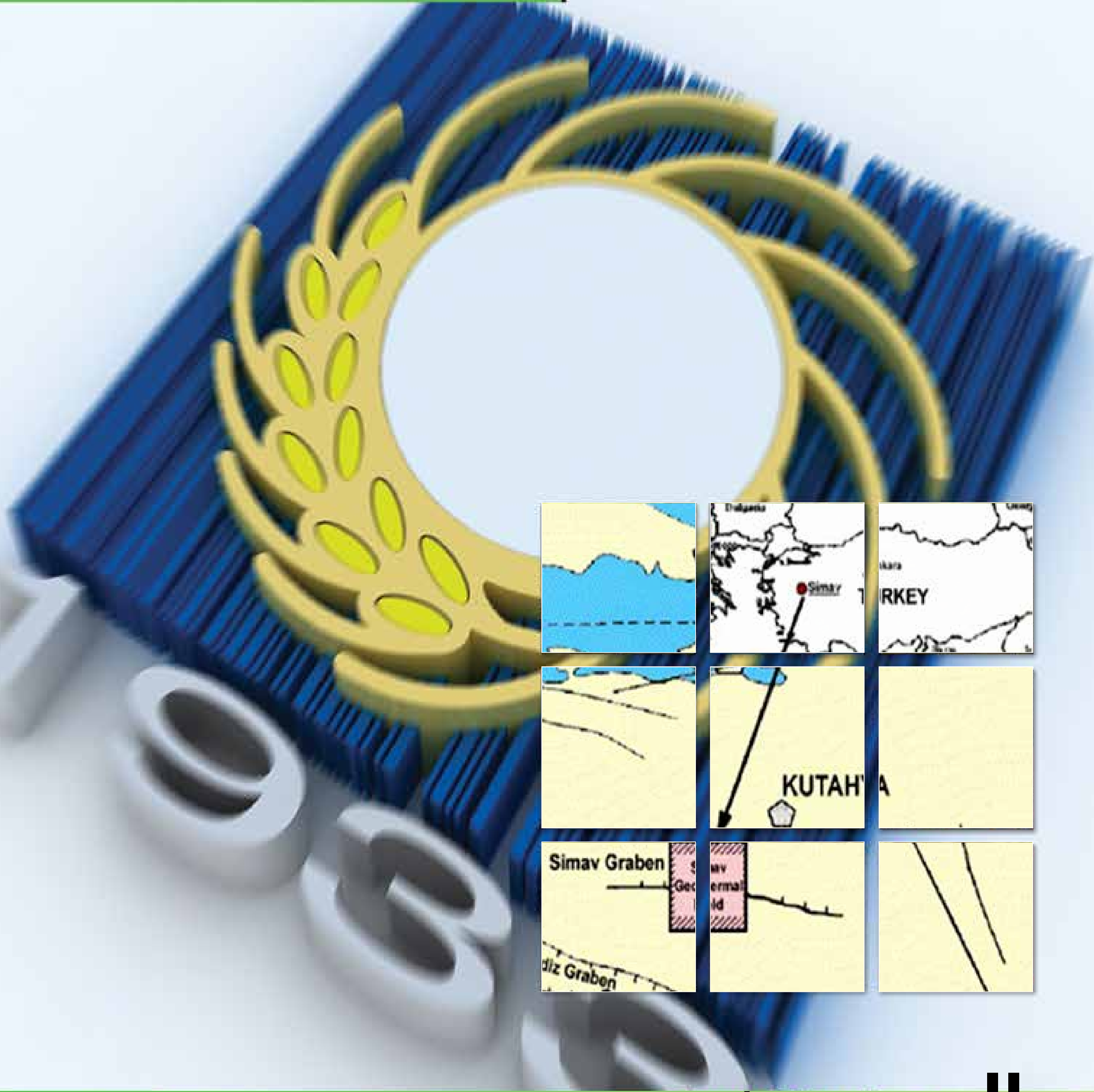

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Economical Analysis of Chamomile *(Matricaria recutita* L.*)* Cultivars, Flower Yields Which are Obtained from Different Sowing Times and Row Spacing

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ABSTRACT

This study was conducted with split-split plots experimental design based on four replications in the ecological conditions of Yalova in the area of Ataturk Central Horticultural Research Institute in growing seasons of 2008-2009 and 2010-2011.

In the study three chamomile (*Matricaria recutita* L.) cultivars (Bona, Bodegold and Zloty Lan) and a genotype, which was collected from the flora of Yalova Province, were used as material. There were four cultivars/genotypes, three sowing time (early November, early October, end of October) and four rows spacing (15 cm, 30 cm, 45 cm, 60 cm). Sowing times were constructed to main plots, cultivars/genotypes to sub plots and row spacing to sub-sub plots.

The highest gross profit was calculated as 8818.33 € ha^{-1} in first time of Zloty Lan cultivar at 15 cm row spacing. Although the Yalova genotype has the lowest cost, the gross profit among the cultivars/genotypes has been the lowest genotype.

Keywords: Chamomile; *Matricaria recutita* L.; Economical analysis; Gross profit; Sowing time; Row spacing

1. Introduction

Medicinal and aromatic plants are used as healing, medicine, food and condiment for centuries. For this reason, the cultivation of certain plants such as cumin, poppy, anise have been ongoing from prehistoric ages. Essential oils and aromatic extracts were used widely as the source of aromatic chemicals, as the starting synthesis material of natural identical or semi-synthesized aromatic chemicals, as perfumes by smell and flavor industries, for the composition of food additives, cleaning products, cosmetics and pharmaceuticals (Güngör et al 2005; Bayram et al 2010).

Although more than 40% of drugs, which were listed in the beginning of the 20th century, were herbal origin, this ratio was decreased further than 5% in the middle of the 1970s. However, especially after the 1990s, the discovery of new areas of medicinal and aromatic plants, and the increasing demand for natural products increased the volume of use of these plants day by day (Bayram et al 2010). One of the plants, chamomile (*Matricaria recutita* L*.), (*Syn. *M. chamomilla),* has been using as folk medicine in Turkey and the World since ancient times. Thoughts on the power of treating are based on phytotherapy and ancient scientific studies, which was described by Hippocrates, Plinius, Dioscorides and Galen. These were also known by "Chamaemelon" who was known as Plinius, Dioscorides and Arabian doctors. The chamomile oil which was known as blue oil and important even today was discussed first time in 1588 (Gül 1995; Ceylan 1996). A famous Slovak proverb says: "An individual should always bow before the curative powers of the chamomile flower tea" (Salamon 2004).

Despite the growth of the world market, it is difficult to find chamomile's production values worldwide. Because, chamomile is cultivated on a small scales and statistics are not given by separating other medicinal and aromatic plants. Today these plants are produced in countries with low labor costs and are exported to industrialized countries.

Although there have been about agronomic studies of chamomile on agriculture, there has not been much work on the economic analysis of camomile farming.

In this study, the effects of different sowing times and different row spacing on total cost, gross profit and net profit were investigated in chamomile cultivars/genotypes and the result was tried to determine the highest gross profit. Thus, it is aimed to contribute to world literature on economical analysis of chamomile farming.

2. Material and Methods

2.1. Material

In this study, the data obtained from the study which was carried out according to with split-split plots experimental design based on four replications in the ecological conditions of Yalova in the area of Atatürk Central Horticultural Research Institute in growing seasons of 2008-2009 and 2010-2011 were used as material.

2.2. Method

The data obtained from the field research below is analyzed according to the gross margin analysis method.

The field research was conducted with split-split plots experimental design based on four replications in the ecological conditions of Yalova in the area of Ataturk Central Horticultural Research Institute in growing seasons of 2008-2009 and 2010-2011. In the study, there were four cultivars/genotypes, three sowing time and four rows spacing. Sowing times were constructed to main plots, cultivars to sub plots and row spacing to sub-sub plots.

The groups were classified as; Cultivars/genotypes: Bona, Bodegold, Zloty Lan, Yalova

Sowings times are done in three different times. Sowing dates are presented as in following:

Sowing time	2008-2009 growing season	2010-2011 growing season
1. Sowing time	10 November 2008	10 November 2011
2. Sowing time	02 December 2008	30 November 2011
3. Sowing time	26 December 2008	23 December 2011

It was planned 15 cm, 30 cm, 45 cm and 60 cm as row spacing.

The trail was established in 2008-2009 and 2010-2011 growing seasons. Sowing lines was prepared in a way to prevent seeds from flying from the winds. 1.5 g seeds were used in each parcel by calculating 278 g seeds in each ha^{-1}. All seed quantities were calculated one by one for all distances between rows. The amount of seed in each rows seed was weighted in 0.0001 sensitivities due to the number of sowing rows. The weighed seeds were mixed with fine wood flour and then packed ready for planting. In both two seasons, 60 kg N, 40 kg P_2O_5 and 40 kg K_2O were given to ha^{-1} according to Johri et al (1991). Also, fertilizers including phosphorus and potassium was given to soil before sowing. Nitrogenous fertilizer was given 40 kg ha^{-1} before sowing and remaining amount was given one month after the emergence. 26% ammonium nitrate was used for the remaining 20 kg ha^{-1} nitrogenous fertilizer. The necessary maintenance and irrigation procedures from the planting stage have been carried out until harvesting time. Any disease was not observed in the trail; however, aphid pest was found at the beginings of april and may in both two seasons, against to this, a chemical struggle was carried out with an agricultural drug including diazinon-acting substance. With this chemical struggle, plant was prevented from negative effects of this pest.

Gross profit, which is suitable to compare profitability of companies, was used to make economic analysis. Gross profit was calculated by using the formula;

Gross profit= Gross production value-Variable costs (1)

Gross margin= Total production-Indirect costs (2)

When the sum of the gross margins of the company's production is equal to the indirect costs, it is in dead or is operating breakeven or zero point (Karagölge 2001). Gross profit in the short-term value of agriculture products can be an important benchmark criterion. The data related to four chamomile cultivar/genotypes was coded into computer in terms of production cost, row spacing and cultivars to analyse them with different time and sowing distances. Then variable costs (seed bed preparation, sowing, irrigation, weed control, pest control, fertilizer, labor costs) and total production costs were calculated with respect to groups. In addition to gross margin analysis techniques, simple percent calculations were done during analysis. The gross profit per hectare has been calculated by multiplication of yield by price by attempting to sell wholesale to the herbalists in Turkey. Kruskal Wallis test was also used to identify statistically different groups by using some sample of data. The Kruskal-Wallis test is a non-parametric method for testing whether samples originate from the same distribution. It is used for comparing two or more independent samples of equal or different sample sizes (Kruskal & Wallis 1952).

3. Results and Discussion

Table 1 demonstrates the data obtained from different chamomile cultivar productions used in the study. The costs related to tillage, fertilizing, spraying and watering for all independent variables which were chamomile cultivars, sowing times, and row spacing were calculated equal to each other. Additionally; variable expenses, which were sowing costs and seed expenses for Bodegold, Bona and Zloty Lan, were 769.23 € ha^{-1} for each one. Seed expense for Yalova genotype population was 316.74 € ha^{-1}. The seed costs of foreign cultivars were higher than that of domestic genotype, because foreign cultivars of chamomile seed were imported from outside countries and they include Chamazulene in essential oil.

According to Table 1, the minimum maintenance costs are seen with 4.42% at second sowing time of Bodegold cultivar at 60 cm row spacing, and maximum one is seen with 17.27% at second sowing time of Yalova genotype at 15 cm row spacing. Another cost, harvesting and marketing costs, for first sowing time of Bodegold cultivar at 60 cm row spacing were found as minimum with 13.26%, while the maximum one was second sowing time of Yalova population at 15 cm row spacing with 25.90%. In this study, all of the harvests were made by hand. Manual harvesting is more costly in terms of time and energy than machine harvesting (Stričík & Salamon 2007). But for one ha and less, manual harvest is more profitable than machine harvest. On the other hand, for higher production areas, machine harvest is more profitable than manual harvest (Ivanović at al 2014). It is thought that the ratio of

Table 1- Variable costs and ratios for groups (€ ha^{-1}, %)

Sowing time	*Cultivars*	*Row spacing (cm)*	*Maintenance costs*		*Harvesting and marketing costs*		*Other variable costs*		*Total variable costs*	
			EUR	*%*	*EUR*	*%*	*EUR*	*%*	*EUR*	*%*
I. Time	Bodegold	15	361.99	13.72	542.99	20.58	1733.71	65.70	2638.69	100.00
		30	271.49	11.12	452.49	18.53	1717.42	70.35	2441.40	100.00
		45	181.00	8.07	361.99	16.13	1701.13	75.80	2244.12	100.00
		60	90.50	4.42	271.49	13.26	1684.84	82.31	2046.83	100.00
	Bona	15	361.99	13.72	542.99	20.58	1733.71	65.70	2638.69	100.00
		30	271.49	11.12	452.49	18.53	1717.42	70.35	2441.40	100.00
		45	181.00	8.07	361.99	16.13	1701.13	75.80	2244.12	100.00
		60	90.50	4.42	271.49	13.26	1684.84	82.31	2046.83	100.00
	Zloty Lan	15	361.99	13.72	542.99	20.58	1733.71	65.70	2638.69	100.00
		30	271.49	11.12	452.49	18.53	1717.42	70.35	2441.40	100.00
		45	181.00	8.07	361.99	16.13	1701.13	75.80	2244.12	100.00
		60	90.50	4.42	271.49	13.26	1684.84	82.31	2046.83	100.00
	Yalova	15	361.99	17.27	542.99	25.90	1191.18	56.83	2096.15	100.00
		30	271.49	14.30	452.49	23.83	1174.89	61.87	1898.87	100.00
		45	181.00	10.64	361.99	21.27	1158.60	68.09	1701.58	100.00
		60	90.50	6.02	271.49	18.05	1142.31	75.94	1504.30	100.00
II. Time	Bodegold	15	361.99	13.72	542.99	20.58	1733.71	65.70	2638.69	100.00
		30	271.49	11.12	452.49	18.53	1717.42	70.35	2441.40	100.00
		45	181.00	8.07	361.99	16.13	1701.13	75.80	2244.12	100.00
		60	90.50	4.42	271.49	13.26	1684.84	82.31	2046.83	100.00
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		30	271.49	11.12	452.49	18.53	1717.42	70.35	2441.40	100.00
		45	181.00	8.07	361.99	16.13	1701.13	75.80	2244.12	100.00
		60	90.50	4.42	271.49	13.26	1684.84	82.31	2046.83	100.00
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		30	271.49	11.12	452.49	18.53	1717.42	70.35	2441.40	100.00
		45	181.00	8.07	361.99	16.13	1701.13	75.80	2244.12	100.00
		60	90.50	4.42	271.49	13.26	1684.84	82.31	2046.83	100.00
	Yalova	15	361.99	17.27	542.99	25.90	1191.18	56.83	2096.15	100.00
		30	271.49	14.30	452.49	23.83	1174.89	61.87	1898.87	100.00
		45	181.00	10.64	361.99	21.27	1158.60	68.09	1701.58	100.00
		60	90.50	6.02	271.49	18.05	1142.31	75.94	1504.30	100.00
III. Time	Bodegold	15	361.99	13.72	542.99	20.58	1733.71	65.70	2638.69	100.00
		30	271.49	11.12	452.49	18.53	1717.42	70.35	2441.40	100.00
		45	181.00	8.07	361.99	16.13	1701.13	75.80	2244.12	100.00
		60	90.50	4.42	271.49	13.26	1684.84	82.31	2046.83	100.00
	Bona	15	361.99	13.72	542.99	20.58	1733.71	65.70	2638.69	100.00
		30	271.49	11.12	452.49	18.53	1717.42	70.35	2441.40	100.00
		45	181.00	8.07	361.99	16.13	1701.13	75.80	2244.12	100.00
		60	90.50	4.42	271.49	13.26	1684.84	82.31	2046.83	100.00
	Zloty Lan	15	361.99	13.72	542.99	20.58	1733.71	65.70	2638.69	100.00
		30	271.49	11.12	452.49	18.53	1717.42	70.35	2441.40	100.00
		45	181.00	8.07	361.99	16.13	1701.13	75.80	2244.12	100.00
		60	90.50	4.42	271.49	13.26	1684.84	82.31	2046.83	100.00
	Yalova	15	361.99	17.27	542.99	25.90	1191.18	56.83	2096.15	100.00
		30	271.49	14.30	452.49	23.83	1174.89	61.87	1898.87	100.00
		45	181.00	10.64	361.99	21.27	1158.60	68.09	1701.58	100.00
		60	90.50	6.02	271.49	18.05	1142.31	75.94	1504.30	100.00

the cost of harvest in total variable costs may be lower when the harvest is mechanized.

In the study, the production values were calculated separetely for different sowing times (I., II., III.), cultivars/genotype (Bodegold, Bona, Zloty Lan and Yalova) and row spacing (15, 30, 45 and 60 cm).

According to Table 2, for all three cultivars except Yalova genotype, the total production and variable costs were found to be the highest at the row spacing of 15 cm in every three-sowing time. While the production cost per unit was considered the most cost was 44.62 € ha^{-1} at third sowing time of Bodegold cultivar at 30 cm row spacing, the lowest one was 18.10 € ha^{-1} at first sowing time of Zloty Lan cultivar at 15 cm row spacing.

When the differences among cultivars/genotypes were examined at the end of the statistical analysis; while there was no statistical significance between sowing time and total variable costs, there was significance of production cost per unit in terms of sowing times. When the cultivars/genotype were evaluated in terms of the differences on costs; the differences among total variable costs, total production costs and production cost per unit was found as statistically significant. When row spacing among groups were examined; total variable costs and total production costs were found as significance, but production costs per unit was not found significant (Table 2). While analysing gross margins; sale prices as flos chamomillae of Bodegold, Bona, Zloty Lan was 6.79 € kg^{-1}, that of Yalova genotype was 4.53 € kg^{-1}. The main reason of that price difference is cultivars that used as a material in this study include chamazulene inside, but Yalova population do not contain this matter. This situation results with lower selling price for Yalova genotype relatively. When the efficiency, gross production value, gross profit and net profit were examined; the maximum result for these variables was found for first sowing time of Zloty Lan cultivar at 15 cm row spacing. The minimum efficiency, gross profit and net profit were calculated for third sowing time of Bodegold cultivar at 30 cm row spacing. The minimum gross production value was 3900.45 € ha^{-1} for third sowing time of Yalova genotype at 30 cm row spacing.

In the statistical analysis to identify the differences between group for yield and production value; the differences among all sowing times were found as significant (Table 3). Additionally, the statistical analysis for cultivars/genotype were also significant. According to Table 3 statistical analysis for row spacing demonstrated that just there is a significant difference for yield and no significant difference was found for gross production value, gross profit and net profit. As a conclusion, there were four cultivars/genotype, three sowing time (early November, early October, end of October) and four rows spacing (15 cm, 30 cm, 45 cm, 60 cm) in the study and the gross margin analyses were done and the most important costs in variable expenses were found as seed cost, maintenance costs and harvesting costs. The important reason of why variable costs was important was different row spacing. As decrease the row spacing had increased both yield and maintanence costs in this study.

While other researchers (Kwiatkowski 2015) obtained the highest yield at 35 and 45 cm row spacing, the highest yield in our study was obtained from the lowest row spacing of 15 cm. It is presumed that this is caused by the fact that Kwiatkowski (2015)'s study is different in sowing time (in April) and different cultural practices (different growth stimulators).

Finally, although the differences among groups were not high ratios and statistically significant, the maximum gross profit was calculated for first sowing time of Zloty Lan cultivar at 15 cm row spacing and the minimum one was found for third sowing time of Bodegold cultivar at 30 cm row spacing.

4. Conclusions

In this study, three of the four different types of chamomile used as material are imported. The Yalova genotype is domestic. Therefore, the seed costs of imported varieties are higher than those of the domestic genotype. As a result of the statistical

Table 2- Variable costs, total production costs and production cost per unit for groups

Sowing time	*Cultivars*	*Row spacing (cm)*	*Variable cost (€ ha^{-1})*	*Total production cost (€ ha^{-1})*	*Production cost per unit (€ ha^{-1})*
I. Time	Bodegold	15	2638.69	3057.19	25.34
		30	2441.40	2854.03	30.00
		45	2244.12	2650.81	28.46
		60	2046.83	2447.60	28.14
	Bona	15	2638.69	3057.19	21.09
		30	2441.40	2854.03	22.81
		45	2244.12	2650.81	24.84
		60	2046.83	2447.60	21.13
	Zloty Lan	15	2638.69	3057.19	18.10
		30	2441.40	2854.03	25.57
		45	2244.12	2650.81	22.49
		60	2046.83	2447.60	24.48
	Yalova	15	2096.15	2498.42	24.07
		30	1898.87	2295.20	18.46
		45	1701.58	2091.99	22.35
		60	1504.30	1888.78	18.73
II. Time	Bodegold	15	2638.69	3057.19	20.68
		30	2441.40	2854.03	23.94
		45	2244.12	2650.81	26.20
		60	2046.83	2447.60	26.24
	Bona	15	2638.69	3057.19	28.42
		30	2441.40	2854.03	27.69
		45	2244.12	2650.81	24.21
		60	2046.83	2447.60	22.35
	Zloty Lan	15	2638.69	3057.19	22.26
		30	2441.40	2854.03	27.78
		45	2244.12	2650.81	26.15
		60	2046.83	2447.60	21.22
	Yalova	15	2096.15	2498.42	24.25
		30	1898.87	2295.20	26.47
		45	1701.58	2091.99	21.18
		60	1504.30	1888.78	19.28
III. Time	Bodegold	15	2638.69	3057.19	37.29
		30	2441.40	2854.03	44.62
		45	2244.12	2650.81	33.35
		60	2046.83	2447.60	37.33
	Bona	15	2638.69	3057.19	27.60
		30	2441.40	2854.03	33.85
		45	2244.12	2650.81	39.00
		60	2046.83	2447.60	32.62
	Zloty Lan	15	2638.69	3057.19	28.73
		30	2441.40	2854.03	25.38
		45	2244.12	2650.81	22.31
		60	2046.83	2447.60	25.75
	Yalova	15	2096.15	2498.42	22.94
		30	1898.87	2295.20	26.61
		45	1701.58	2091.99	23.67
		60	1504.30	1888.78	20.45

Table 3- Yield, gross production value, gross profit and net profit values for groups

Sowing time	*Cultivars*	*Row spacing (cm)*	*Yield (ton ha^{-1})*	*Gross production value (€ ha^{-1})*	*Gross profit (€ ha^{-1})*	*Net profit (€ ha^{-1})*
I. Time	Bodegold	15	1.21	8192.31	5553.62	5135.11
		30	0.95	6454.75	4013.35	3600.72
		45	0.93	6325.79	4081.67	3674.98
		60	0.87	5904.98	3858.14	3457.38
	Bona	15	1.45	9848.42	7209.73	6791.22
		30	1.25	8497.74	6056.33	5643.71
		45	1.07	7242.08	4997.96	4591.27
		60	1.16	7866.52	5819.68	5418.91
	Zloty Lan	15	1.69	11457.01	8818.33	8399.82
		30	1.12	7581.45	5140.05	4727.42
		45	1.18	7995.48	5751.36	5344.66
		60	1.00	6787.33	4740.50	4339.73
	Yalova	15	1.04	4692.31	2596.15	2193.89
		30	1.24	5628.96	3730.09	3333.76
		45	0.94	4235.29	2533.71	2143.30
		60	1.01	4561.09	3056.79	2672.31
II. Time	Bodegold	15	1.48	10031.67	7392.99	6974.48
		30	1.19	8090.50	5649.10	5236.47
		45	1.01	6861.99	4617.87	4211.18
		60	0.93	6332.58	4285.75	3884.98
	Bona	15	1.08	7303.17	4664.48	4245.97
		30	1.03	6990.95	4549.55	4136.92
		45	1.10	7432.13	5188.01	4781.31
		60	1.10	7438.91	5392.08	4991.31
	Zloty Lan	15	1.37	9325.79	6687.10	6268.60
		30	1.03	6970.59	4529.19	4116.56
		45	1.01	6875.57	4631.45	4224.75
		60	1.15	7827.15	5780.32	5379.55
	Yalova	15	1.03	4660.63	2564.48	2162.22
		30	0.87	3923.08	2024.21	1627.87
		45	0.99	4466.06	2764.48	2374.07
		60	0.98	4429.86	2925.57	2541.09
III. Time	Bodegold	15	0.82	5565.61	2926.92	2508.42
		30	0.64	4343.89	1902.49	1489.86
		45	0.80	5395.93	3151.81	2745.11
		60	0.66	4452.49	2405.66	2004.89
	Bona	15	1.11	7513.57	4874.89	4456.38
		30	0.84	5721.72	3280.32	2867.69
		45	0.68	4615.38	2371.27	1964.57
		60	0.75	5090.50	3043.67	2642.90
	Zloty Lan	15	1.06	7221.72	4583.03	4164.52
		30	1.13	7635.75	5194.34	4781.72
		45	1.19	8063.35	5819.23	5412.53
		60	0.95	6454.75	4407.92	4007.15
	Yalova	15	1.09	4932.13	2835.97	2433.71
		30	0.86	3900.45	2001.58	1605.25
		45	0.88	4000.00	2298.42	1908.01
		60	0.92	4176.47	2672.17	2287.69

analysis, it is meaningful that significant differences were found between product and total variable costs. The reason for the high gross profit and net profit obtained from the import varieties used as the material in the study is that the selling price of the product is higher than the domestic genotype. The fact that imported varieties can be sold at 50% more price than the Yalova genotype is the most important reason for this difference. This result supports the fact that the difference between the groups in terms of yield and production values is significant in terms of statistical analysis.

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Mathematical Modelling of Crop Water Productivity for Processing Tomato

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ABSTRACT

Crop water productivity models (CWPMs) are of great importance in evaluating different irrigation programs. The mean goal of the study was to evaluate the performance of the Jensen, Minhas, Blank, Stewart and Rao CWPMs in predicting fruit yield of processing tomato. Field experiments were conducted for two consecutive growing seasons. The soil water stress sensitivity indices of the CWPMs were determined using experimental data from the second crop growing season. Yields simulated by the CWPMs were compared with the experimental data for the first season. The sensitivity indices for the crop growth stages were taken into account as appropriate weights of the soil water sensitivity of the vegetative, flowering, yield formation and ripening stages of the processing tomato crop. The results give evidence that processing tomato is much more sensitive to soil water stress during flowering and yield formation stages whereas the adverse impact of water stress on yield is very limited at vegetative stage. The highest modelling efficiency (0.96) between field-measured and simulated yield by the model, the lowest arithmetic mean of errors (0.04), mean absolute deviation (0.07), mean square error (0.02), absolute percentage error (12.76), root mean square error (0.15) and coefficient of residual mass (0.05) were achieved by Minhas model and followed by Rao model based on same parameters of statistical analyses. Both the Minhas and the Rao models with their soil water stress sensitivity indices generated for the different growth stages obtained in this study are recommended for the processing tomato in the sub-humid environments.

Keywords: Deficit irrigation; Relative evapotranspiration; Relative yield; Stress sensitivity indices

1. Introduction

Crop-water productivity model (CWPM) which is known as the relationship between seasonal crop evapotranspiration (ET) and fresh or dry yield is of great interest among scientists who work on soil, plant and water. As it was stated in Kipkorir et al (2002) and Igbadun et al (2007); CWPM can be mainly divided into two parts; one relates yield to seasonal ET (e.g., Stewart & Hagan 1973; Doorenbos & Kassam 1979; Hanks 1983), another relates yield reduction to water deficit at some crop growth stages (Jensen 1968; Minhas et al 1974; Sudar et al 1981).

Based on Igbadun et al (2007); dependent variables associated with water may be expressed in two types: additive and multiplicative (Tsakiris 1982). The multiplicative-type assumes that water deficit in two or more crop growth stages reduces yield in a multiplicative way (Jensen 1968; Minhas et al 1974; Bernardo et al 1988), while additive-type predicts that crop yield may be reduced by water deficit in two or more crop growth stages in an additive way (Stewart et al 1977; Bras & Cordova 1981).

CWPMs are crucial for irrigation water management. Irrigation water management aims to accomplish optimal crop production and higher water use efficiency or a reliable, continuous, and equitable irrigation water supply to water users (Tarjuelo & de Juan 1999).

Reliability and practicability over 300 CWPMs were tested by Clumpner & Solomon (1987). They found major differences on growing season-to-season and site-to-site basis as well as the impacts of crop growth stages (Al-Jamal et al 2000; Igbadun et al 2007). In as much as dependent and/or independent variables of CWPMs are strongly influenced by crop characteristics and environmental conditions, there is no universal CWPM for all crops, growth stages and climates (Rhenals & Bras 1981). For that reason, performance evaluation should be carried out for different crops and location before using CWPMs in irrigation water management and in developing water management strategies (Igbadun et al 2007).

In this study, a comparative analysis of various additive and multiplicative type CWPM models which relates crop yield to relative evapotranspiration (ETa/ETm) considering crop growth stages was carried out under sub-humid climatic conditions. The aim of this study is to test the model performance in predicting the fruit yield of processing tomato in the sub-humid climate conditions.

2. Material and Methods

2.1. Crop water production models

In this study, 5 CWPMs related to relative ET or relative ET deficit, developed by the various researchers were used for the predicting relative yield or relative yield decrease of a processing tomato crop (Table 1).

In Table 1, *Ya* actual yield (t ha^{-1}) from the plot with soil water stress during the growing season (called as fruit yield in this study); *Ym* (t ha^{-1}) is the maximum yield from the plot without water stress during the growing season; ETa_i actual evapotranspiration (mm) from the plot with water stress during the growing stage *i*; ETm_i maximum evapotranspiration (mm) from the plot without

Table 1- Crop water production functions

Source	*Crop water production function*	*Type/Independent variable*
Jensen (1968)	$\frac{Ya}{Ym}=\prod_{i=1}^{n}\left(\frac{ETa_i}{ETm_i}\right)^{\lambda_i}$	Multiplicative/Relative evapotranspiration (ET)
Minhas et al (1974)	$\frac{Ya}{Ym}=\prod_{i=1}^{n}\left[1-\left(1-\frac{ETa_i}{ETm_i}\right)\right]^{\delta_i}$	Multiplicative/Relative ET deficit
Blank (1975)	$\frac{Ya}{Ym}=\sum_{i=1}^{n}A_i\left(\frac{ETa}{ETm}\right)_i$	Additive/Relative ET
Stewart et al (1977)	$\left(1-\frac{Ya}{Ym}\right)=\sum_{i=1}^{n}ky_i\left(1-\frac{ETa}{ETm}\right)_i$	Additive/Relative ET deficit
Rao et al (1988)	$\frac{Ya}{Ym}=\prod_{i=1}^{n}\left[1-K_i\left(1-\frac{ETa_i}{ETm_i}\right)\right]$	Multiplicative/Relative ET deficit

water stress during the growing stage *i*; *n* the number of crop development stages; ∏ multiplicative sign; Σ additive sign and, λ_i, δ_i, A_i, ky_i *and* K_i sensitivity indices of the crop to water stress during the growing stage *i*.

2.2. Field experiments and irrigation treatments

Irrigation experiments were carried out on the experimental farm of Mustafakemalpasa Vocational School of Bursa Uludağ University, Turkey (40°02′N, 28°23′E). Average rainfall amounts were 121 and 52 mm, mean temperatures were 25.3 and 23.8 ºC, and the relative humidity were 64 and 66% for both growing seasons of experimental years, respectively. The experimental site has a clay-loam Entisol soil. Soil samples were taken from each 0.30 m layers of 0-1.2 m soil profile prior to irrigation treatments. Based on results of soil samples analyses, electrical conductivity, lime content, pH and the available water holding capacity (the difference between the water content at FC and PWP) were 0.02-0.04 dS m^{-1}, 4-11%, 7.7-8.0 and 183 mm/0.90 m. A total of 180 kg N ha^{-1} and 120 kg P_2O_5 ha^{-1} fertilizer was applied. All agricultural inputs (fertilizer, pesticide etc.) other than water were assumed constant.

The hybrid cultivar Shasta variety (Campbell's Seeds™ Inc, CA, USA) was planted in the growing seasons of 2010 and 2011. Each experimental plot was 5.10 m long by 5.60 m wide (28.56 m^2), with 4 rows per plot. A buffer zone spacing of 2.00 m was provided between the plots. The row spacing and plant–plant spacing were 1.40 and 0.30 m, respectively (Kuşçu et al 2014). Seedlings at the 3-4 true leaf stage were transplanted to the treatment plots, on 15 May 2010 and 20 May 2011. The irrigation experiments were conducted using randomized block design and repeated three times. Fifteen different irrigation treatments considering vegetative, flowering, yield formation, and the ripening stages of crop development were planned to assess the effects of water deficit in the soil (Table 2).

Irrigation interval was 3 days at all crop growth stages with irrigation (VFYR). Irrigation was applied once in every 3 days to the treatments specified as (+) symbol in Table 1. Irrigation at each growth stage was applied with the amount of

Table 2- Irrigation treatments

Treatments	*Crop development stages*			
	Vegetative (V)	*Flowering (F)*	*Yield formation (Y)*	*Ripening (R)*
VFYR	+[a]	+	+	+
FYR	–	+	+	+
VFY	+	+	+	–
VFR	+	+	–	+
VYR	+	–	+	+
VF	+	+	–	–
VR	+	–	+	–
VY	+	–	–	+
FY	–	+	+	–
FR	–	+	–	+
YR	–	–	+	+
V	+	–	–	–
F	–	+	–	–
Y	–	–	+	–
R	–	–	–	+

[a] (+), irrigation at specified crop development stages; (–), no irrigation at specified crop development stages; (V), vegetative stage; (F), flowering stage; (Y), yield formation stage; (R), ripening stage

irrigation water required to fill the moisture content of 0-90 cm soil layer to field capacity.

2.3. Soil moisture monitoring and evapotranspiration

The soil moisture was monitored in 0.3 m depth increments to 1.2 m prior to and after irrigation from each plot. Soil water content was gravimetrically determined. The soil water contents of 90 cm and 120 cm soil depth were used for determination of water amount applied in each irrigation and seasonal ET, respectively.

The actual crop evapotranspiration was calculated using a soil-water balance equation (Kuşçu et al 2014).

2.4. Fruit yield determination

When ripe fruit ratio was reached to 95%, all experimental plots were harvested by hand on 23 August 2010 and 28 August 2011, respectively. Tomatoes which were harvested from the two center rows were compared with total ground area as fruit yield (t).

2.5. Determination of sensitivity indices of the crop to water stress in the models

Since the rainfall amounts at the crop growth stages of 2011 was lower than those of 2010 (total rainfall amounts: 121 mm for 2010, 52 mm for 2011), sensitivity indices of the crop to water stress in the models were calculated more precisely by using data obtained from the experimental field at growth stages of 2011. All models were converted to multiple linear functions. While relative crop yield decrease (Y_a/Y_m) was taken as dependent variable, relative evapotranspiration deficit (ET_a/ET_m) was assigned as independent variable in this conversion (Igbadun et al 2007). Fruit yield obtained from the field and evapotranspiration associated with crop growth stages were described as relative yield (ratio of yield at some growth stages with no irrigation to yield at full irrigation treatment) and relative evapotranspiration (ratio of crop evapotranspiration at some growth stages with no irrigation to crop evapotranspiration of full irrigation treatment), respectively. In this study, relative yield and evapotranspiration data were used for solution of multiple regression equations for each model in determining the sensitivity indices of crop to water stress at four crop growth stages. The regression equations were realized by using SPSS 23 Statistical Program.

2.6. Model performance evaluation

The performance of the model for the prediction of relative fruit yield was tested by using relative evapotranspiration results obtained from the treatments at crop growth stages of 2010. Both graphical and statistical methods were employed for the assessment of the models. The rates of yield reduction were plotted for measured and simulated values at graphical method. The response of each model could be quantified by this method. In statistical analyses, various performance indicators were used to compare the data observed with the results estimated by the model (Loague & Green 1991; Hagi-Bishow & Bonnell 2000). The performance indicators were given at Equations 1-7.

$$\text{Arithmetic mean of the errors, } BIAS = \frac{\sum_{i=1}^{n}(O_i - P_i)}{n} \tag{1}$$

$$\text{Mean absolute deviation, } MAD = \frac{\sum_{i=1}^{n}|O_i - P_i|}{n} \tag{2}$$

$$\text{Mean square error, } MSE = \left[\frac{\sum_{i=1}^{n}(O_i - P_i)^2}{n}\right] \tag{3}$$

$$\text{Mean absolute percentage error, } MAPE = \frac{\sum_{i=1}^{n}\frac{|O_i - P_i|}{O_i} * 100}{n} \tag{4}$$

Root mean square error, $$RMSE = \left[\frac{\sum_{i=1}^{n}(P_i - O_i)^2}{n}\right]^{0.5} * \frac{100}{\bar{O}} \qquad (5)$$

Modeling efficiency, $$EF = \frac{\sum_{i=1}^{n}(O_i - \bar{O})^2 - \sum_{i=1}^{n}(P_i - O_i)^2}{\sum_{i=1}^{n}(O_i - \bar{O})^2} \qquad (6)$$

Coefficient of residual mass, $$CRM = \frac{\sum_{i=1}^{n}(O_i - P_i)}{n\bar{O}} \qquad (7)$$

Where; observed values; mean of the observed values; predicted values, and n number of samples.

3. Results and Discussion

3.1. Fruit yield

Fruit yield obtained from different irrigation experiments was given at Table 3a and 3b. Difference between years and fruit yield for 2 experimental years was significant with P<0.01 level based on analysis of variance (ANOVA) results. Difference between years may be attributed to the differences between rainfall amounts (121 mm for 2010, 52 mm for 2011) and temperatures (25.3 ºC for 2010, 23.8 ºC for 2011) in growing season. The highest fruit yield was obtained at reference treatment with 3-day irrigation interval. However, there was no significant difference on term of statistical analysis between

Table 3a- Evapotranspiration (ET) and fruit yield (2010)

Treatment	*ET for crop development stages (ET_a, mm)*				*Seasonal ET (mm)*	*Fruit yield (t ha^{-1})*
	Vegetative (V)	*Flowering (F)*	*Yield formation (Y)*	*Ripening (R)*		
	Days after planting (day)					
	0-21	*22-44*	*45-66*	*67-100*		
VFYR	85.2	133.8	141.0	152.2	512.2	100.4 a[1]
FYR	78.0	128.0	140.0	150.2	496.2	97.2 b
VFY	85.2	133.8	141.0	105.1	465.1	95.6 c
VFR	85.2	133.8	110.0	135.3	464.3	92.2 d
VYR	85.2	125.0	140.0	150.0	500.2	96.2 c
VF	85.2	133.8	110.0	37.0	366.0	62.8 h
VY	85.2	125.0	140.0	106.4	456.6	80.7 g
VR	85.2	125.0	65.8	93.2	369.2	48.2 k
FY	78.0	128.0	140.0	82.4	428.4	87.2 e
FR	78.0	128.0	93.4	115.7	415.1	85.4 f
YR	78.0	92.0	133.0	143.5	446.5	88.2 e
V	85.2	125.0	65.8	14.3	290.3	30.2 m
F	78.0	128.0	91.4	22.6	320.0	54.9 j
Y	78.0	92.0	133.0	44.3	347.3	61.2 i
R	78.0	92.0	90.0	109.8	369.8	45.2 l

[1], no significant difference at 0.05 level amongst mean values given in the same letters

Table 3b- Evapotranspiration (ET) and fruit yield (2011)

Treatment	*ET for crop development stages (ETa, mm)*				*Seasonal ET (mm)*	*Fruit yield (t ha^{-1})*
	Vegetative (V)	*Flowering (F)*	*Yield formation (Y)*	*Ripening (R)*		
	Days after planting (day)					
	0-22	*23-45*	*46-67*	*68-101*		
VFYR	84.9	130.4	133.8	153.4	502.5	110.7 a[1]
FYR	75.0	129.8	133.0	152.3	490.1	109.2 a
VFY	85.0	130.0	133.6	122.1	470.7	104.6 b
VFR	84.7	129.6	104.4	138.8	457.5	97.6 c
VYR	85.0	116.0	133.0	153.0	487.0	104.3 b
VF	85.0	130.0	104.0	61.4	380.4	64.2 h
VY	85.0	116.0	133.0	121.0	455.3	87.4 f
VR	85.0	116.0	72.1	85.0	358.1	40.2 k
FY	75.0	130.0	133.3	116.0	454.3	94.2 d
FR	75.0	130.0	105.1	129.0	439.1	92.1 e
YR	75.0	90.0	129.0	135.0	429.0	85.7 g
V	85.0	116.0	72.0	24.0	297.0	31.4 m
F	75.0	130.0	105.0	60.0	370.0	60.1 j
Y	75.0	90.0	129.0	106.0	400.0	63.1 i
R	75.0	90.0	67.0	74.0	306.0	36.2 l

[1], no significant difference at 0.05 level amongst mean values given in the same letters

FVYR and FYR treatments. The lowest fruit yield was observed at treatment which has irrigation only at vegetative period (V) for both experimental years.

On other treatments, yield was reduced based on water deficits of crop growth stages. While yield decrease of treatments with no irrigation was substantial at stages flowering and yield formation, yield was not considerably decreased at vegetative stages with no irrigation. Sensitivity of tomato to water stress was highest at flowering and yield formation stages.

3.2. Evapotranspiration

Both seasonal evapotranspiration and ET for different crop growth stages were given in Table 3a and 3b. The seasonal ET varied between 306 and 512.2 mm. The highest seasonal ET was found in the full irrigation treatment (VFYR) whereas the lowest seasonal ET was recorded in the V treatment, with a prolonged water deficit (79 days) after the vegetative period. Since irrigation was applied uninterruptedly in full irrigation treatment (VFYR), seasonal ET results observed in the field were congruent relative to given amount of irrigation water. On the other hand, seasonal ET observed at FYR treatment (no irrigation at vegetative stage) was quite similar to that of full irrigation treatment (VFYR). This result indicates that tomato adequately benefits from the moisture of the soil root zone at vegetative stage (Table 3a and 3b).

3.3. Sensitivity indices of the crop to water stress

The variation of sensitivity indices of the crop to water stress for crop development stages (V: Vegetative, F: Flowering, Y: Yield formation, R: Ripening) was given at Table 4. The following Equations show functions of the Jensen (1968), Minhas et al (1974), Blank (1975), Stewart et al (1977), and Rao et al (1988) models, respectively, with the sensitivity indices. Although the soil water stress sensitivity indices determined by the additive type of models were the same, the constants were different.

Table 4- Sensitivity indices of the crop to water stress in the models

Model	*Sensitivity indices of the crop to water stress for different crop growth stages*				*Constant*	*r^2*	*Std. error*
	V	*F*	*Y*	*R*			
Jensen (1968)	0.002	0.800	0.849	0.391		0.93	0.043
Minhas et al (1974)	1.547	2.306	2.431	0.543		0.93	0.053
Blank (1975)	0.006	0.628	0.479	0.562	-0.665	0.97	0.226
Stewart et al (1977)	0.006	0.628	0.479	0.562	-0.010	0.97	0.024
Rao et al (1988)	0.006	0.628	0.479	0.562		0.97	0.024

$$\frac{Ya}{Ym} = \left(\frac{ETa}{ETm}\right)_V^{0.002} \times \left(\frac{ETa}{ETm}\right)_F^{0.800} \times \left(\frac{ETa}{ETm}\right)_Y^{0.849} \times \left(\frac{ETa}{ETm}\right)_R^{0.391} \tag{8}$$

$$\frac{Ya}{Ym} = \left[1-\left(1-\frac{ETa}{ETm}\right)_V^2\right]^{1.547} \times \left[1-\left(1-\frac{ETa}{ETm}\right)_F^2\right]^{2.306} \times \left[1-\left(1-\frac{ETa}{ETm}\right)_Y^2\right]^{2.431} \times \left[1-\left(1-\frac{ETa}{ETm}\right)_R^2\right]^{0.543} \tag{9}$$

$$\frac{Ya}{Ym} = 0.006\left(\frac{ETa}{ETm}\right)_V + 0.628\left(\frac{ETa}{ETm}\right)_F + 0.479\left(\frac{ETa}{ETm}\right)_Y + 0.562\left(\frac{ETa}{ETm}\right)_R - 0.665 \tag{10}$$

$$1-\frac{Ya}{Ym} = 0.006\left(1-\frac{ETa}{ETm}\right)_V + 0.628\left(1-\frac{ETa}{ETm}\right)_F + 0.479\left(1-\frac{ETa}{ETm}\right)_Y + 0.562\left(1-\frac{ETa}{ETm}\right)_R - 0.010 \tag{11}$$

$$\frac{Ya}{Ym} = \left[1-0.006(1-\frac{ETa}{ETm}\right]_V \times \left[1-0.628(1-\frac{ETa}{ETm}\right]_F \times \left[1-0.479(1-\frac{ETa}{ETm}\right]_Y \times \left[1-0.562(1-\frac{ETa}{ETm}\right]_R \tag{12}$$

Sensitivity analysis tests were employed to determine the sensitivity indices of crop to water stress for all models. For Jensen model, the index obtained at yield formation stage was higher than those of other stages. On the other hand, sensitivity index of crop to water stress was higher at flowering stages than those of other growth stages for Minhas, Blank, Stewart and Rao models. In general, crop growth stages more sensitive to soil water stress has higher sensitivity index (Zhang et al 2002). Processing tomato is more sensitive to flowering and yield formation stages based on all model outputs, whereas flowering, yield formation and the ripening stages were the most sensitive stages based on Jensen model.

3.4. Model evaluation

Relative yield (Y_a/Y_m) obtained from field measurements and model simulations for different irrigation treatments were presented in Table 5. Table 6 summarizes statistical performance indicators associated with comparison of relative yield from field measurements and model simulations.

BIAS ranged between 0.04 and 0.10. MAD values were ranged from 0.07 to 0.10 and may be considered as very similar for each model. On the other hand, MSE was lowest in the Minhas model (0.02), followed by the Rao model with 0.04 and highest in the Jensen model (0.16). MAPE varied from 12.76 to 15.99. The lower the error measurements (BIAS, MAD, MSE, MAPE, and

Table 5- Relative fruit yield (field measurements vs model simulations)

Treatment	*Relative yield (field measurements)*	*Relative yield (model simulation)*				
		Jensen	*Minhas*	*Blank*	*Stewart*	*Rao*
VFYR	1.00	1.00	1.00	1.00	1.00	1.00
FYR	0.97	0.95	0.98	0.98	0.97	0.96
VFY	0.95	0.87	0.95	0.84	0.84	0.83
VFR	0.92	0.77	0.88	0.85	0.84	0.84
VYR	0.96	0.94	0.99	0.96	0.96	0.95
VF	0.63	0.47	0.56	0.48	0.48	0.51
VY	0.80	0.82	0.94	0.80	0.80	0.79
VR	0.48	0.41	0.40	0.50	0.50	0.56
FY	0.87	0.75	0.87	0.73	0.72	0.72
FR	0.85	0.61	0.71	0.69	0.69	0.71
YR	0.88	0.69	0.77	0.76	0.75	0.76
V	0.30	0.20	0.17	0.21	0.20	0.35
F	0.55	0.32	0.35	0.34	0.34	0.42
Y	0.61	0.44	0.53	0.39	0.39	0.47
R	0.45	0.45	0.53	0.49	0.48	0.56

Table 6- Statistics of comparison between measured and model predicted relative yields

Statistical performance indicators	*Jensen*	*Minhas*	*Blank*	*Stewart*	*Rao*
Arithmetic mean of the errors (BIAS)	0.10	0.04	0.08	0.08	0.05
Mean absolute deviation (MAD)	0.10	0.07	0.09	0.09	0.08
Mean square error (MSE)	0.16	0.02	0.10	0.11	0.04
Mean absolute percentage error (MAPE)	15.99	12.76	14.24	14.24	12.91
Root mean square error (RMSE)	0.40	0.15	0.31	0.33	0.20
Modeling efficiency (EF)	0.71	0.96	0.83	0.80	0.92
Coefficient of residual mass (CRM)	0.14	0.05	0.11	0.11	0.07

RMSE) and the higher modelling efficiency are, the better the forecasting model is. In this study, the highest modelling efficiency (EF= 0.96) between field-measured and simulated yield by the model and the lowest error parameters were achieved by Minhas model and followed by Rao model based on same statistical analyses. Both models showed relatively high modelling efficiency (>0.90). The closer the modelling efficiency is to 1, the better the consistency between the measured and predicted data, and the farther from 1, the greater the error margin in the values simulated by the model. From that point of view, modelling efficiency of Jensen model is relatively lower than those of the others (EF= 0.71). The RMSE values show how much the simulations under- or over-estimate the measurements. When considering whole experimental treatments, RMSE values most close to zero was attained by Minhas model (Table 6). When considering the values of CRM which is a tool for prediction level of the model, models predicted 5-14% lower than the field measurements. While the closest value of simulated relative yield to the field-measured relative yield was predicted by Minhas model (CRM= 5%), the lowest level of prediction was obtained by Jensen model (14%).

As seen in Table 5, the lowest difference between the model predicted and the measured relative yield in the field was found at FYR treatment which has no irrigation at vegetative development stage. The reason is that the soil water stress sensitivity index determined for vegetative stage of processing tomato for all models is much lower than the indices obtained for other crop growth stages. This result suggests that the processing tomato is not very sensitive to water stress occurred at the vegetative stage in sub-humid climates. On the other hand, the biggest differences in between relative fruit yields of field measurements and model simulations were attained at F (irrigation only at flowering stage) and Y (irrigation only at yield formation stage) treatments by Stewart and Blank models, at FR (irrigation only at flowering and ripening stages) treatment by Jensen and Minhas models, and at FY (irrigation only at flowering and yield formation stages) treatment by Rao model (Table 5). These results show that the differences in the soil water stress sensitivity indices of the multiplicative and additive type models lead different reduction levels of relative yields from model to model at different irrigation treatments.

4. Conclusions

In present study, 5 CWPMs related to relative evapotranspiration (ET) or relative ET deficit were used for the predicting relative yield or relative yield decrease of a processing tomato crop. Minhas and Rao models satisfactorily predicted relative fruit yields of processing tomato based on comparative statistical test results. Either Minhas or Rao model may be used for prediction of relative fruit yields associated with deficit irrigation under sub-humid climate conditions. One of the both models which gave the best results could be preferred by considering deficit irrigated periods. To provide better model performance, sensitivity indices of crops to water stress should be calibrated by testing in different locations. Better results may be obtained by considering different growth stages apart from four critical crop growth stages considered in this study. Besides, relative differences in sensitivity indices of processing tomato to water stress may be observed depending upon plantation date and vegetation period.

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Effects of Organic Mulching on Soil Water Potential and SPAD Values as Factors on Yield of Potatoes (*Solanum tuberosum* L.)

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ABSTRACT

In the present study, Finka and Katka potato (*Solanum tuberosum* L.) varieties were used at two localities (Leškovice and Uhříněves) and the effects of the surface mulching on chlorophyll content (SPAD values), soil water potential (SWP) and tuber yield of potato (YWP) were observed. Two mulching treatments (chopped grass and black polypropylene mulch) were used in the study. The results showed that plastic mulch can be an important factor for potato cultivation on localities with dystric cambisol soil type currently facing lack of precipitation. Organic mulching (with chopped grass) significantly influenced YWP. It favorably increased the fresh weight of tubers (over 60 mm) per plant by 81.4% in comparison with control. Additionally, it has been found that the type of mulching or varieties can be expressed by different SPAD-yield relationship. Whereas the values of SWP and SPAD values too were not significantly affected by organic mulch, they were significantly correlated with the YWP (r^2= 42.4%, $P<0.0001$ and r^2= 28.8%, $P<0.0023$). SPAD value can predict the level of tuber yield if the value was calibrated for particular potato variety. However, the highest SPAD reading value did not guarantee the highest tuber yield.

Keywords: Grass mulch; Soil water potential; SPAD value; Potato; Yield

1. Introduction

The last fifteen years have seen increasingly frequent extreme climatic events in the Czech Republic (Rožnovský 2014) and in the Central European region (Trnka et al 2016), including the occurrence of serious drought. The extent and intensity of drought also depend on water retention capacity of the landscape. Priority should be given to increase its retention capacity through mulching to the maximum (Rožnovský 2014), especially during of high rainfalls. The principle of organic mulching (or biomass thus applied) can be used to protect the soil surface (e.g. reduce runoff and erosion). Janeček (2005) reports that more than half of agricultural land in the CR is threatened by water erosion. Water erosion causes vicissitude soil degradation, which decreases the production capacity of the soil. Meteorological phenomena such as drought (Fiala et al 2016) and extreme torrential rainfall in the CR are currently a priority issue due to changing climatic conditions. Potatoes, as well as other wide-crops, are in many places in the CR associated with water erosion. Mulching may affect availability water like others physical and chemical characteristics of soil

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(Govaerts et al 2007) and enhance availability of nitrogen in the soil with increase in plant growth (Fang et al 2011). The cover of mulch influences soil moisture as well (Ramakrishna et al 2006). Mulch maintains stable soil moisture, especially in surface soil layer. The water content directly near the soil surface plays an essential role in the degradation of natural organic material by soil microbes (Hood 2001). For that reason, mulching becomes more important in moderate climate conditions. The chlorophyll (CHLO) content of a plant is a good qualitative indicator for leaf N concentration, and for several crops, it could be demonstrated that leaf N and CHLO concentration are strongly correlated (Werner et al 2005). Crop varieties differ in their genetically determined CHLO content (Minotti et al 1994; Uzik & Zofajova 2000), what should be taken into account if CHLO measurements are used for nutrient management decisions. Additionally, variation in the CHLO contents can interfere such as plant (leaf) age (Mauromicale et al 2006) or different leaf position on the plant (Busato et al 2010). One the most common portable CHLO meter is the SPAD-502 (Minolta Co., Tokyo, Japan). The SPAD meter is a simple, portable tool that measures the greenness or relative CHLO content of leaves (Busato et al 2010). It is very quick and a non-invasive method (Denuit et al 2002). Nevertheless Denuit et al (2002), some shortcomings are reported; mainly the lack of specificity caused by the environmental effect on CHLO meter values. The potato cultivar, the soil, the climate and the water status of the crop are the main factors affecting CHLO meter values (Olivier et al 1999). Correlation with CHLO content of N in the laboratory conditions for potatoes is already published Vos & Born (1993) with the result $r^2>0.95$. In field trials, Uddling et al (2007) show a lower dependence with $r^2>0.58$. This study aimed to evaluate the effect of various mulches (organic and plastic mulch) on the SPAD values, soil water potential, tuber yields of potato and other parameters that may affect growth of potatoes.

2. Material and Methods

2.1. Field experiments

They were carried out at the Experimental Station of Czech University of Life Sciences Prague in Uhříněves (50°2’0.4”N, 14°36’32”E, alt. 298 m asl) and Leškovice (49°45’46”N, 15°32’16”E, alt. 498 m asl) in years 2009-2011. Leškovice (LE) is a potato-growing region, the average of annual temperature is 6.9 °C (detailed in Table 1) and annual precipitation is 630 mm (weather station of the Potato Research Institute Havlíčkův Brod). The soil formed on paragneiss classified as dystric cambisol (IUSS Working Group WRB, 2015) was collected from the topsoil layer at experimental field of the Leškovice site (loam-sandy soils). Uhříněves (UH) is sugar beet region, the average of annual temperature is 8.4 °C and annual precipitation is 575 mm (meteorological station of the Czech Hydrometeorological Institute Prague Clementinum). At this site the texture class of this soil is a clay loam with an organic matter content of 1.74-2.12%, pH neutral with a good reserve of all essential nutrients and the type of soil is dystric cambisol (IUSS Working Group WRB, 2015) and other hydro-physical properties show the Table 2. The field experiment comprised three

Table 1- Distribution of monthly precipitation (mm) and air temperature (°C) during the years 2009-2011

Year	*Site*	*IV.*		*V.*		*VI.*		*VII.*		*VIII.*		*IX.*		*IV. – IX.*	
		mm	°C	*mm*	°C	*mm*	°C	*mm*	°C	*mm*	°C	*mm*	°C	*mm*	°C
2009	LE	4.1	13.9	70.7	13.6	108.0	14.7	92.0	18.3	66.6	18.7	17.9	15.2	359	15.7
	UH	16.0	13.6	95.3	14.7	72.0	16.1	81.9	19.5	31.8	20.0	20.2	16.1	317	16.7
2010	LE	151.0	4.3	101	11.3	74.0	16.1	155.2	18.7	173.0	16.9	90.0	10.9	744	13.0
	UH	32.0	10.1	93.1	12.6	62.0	17.9	118.0	21.6	139.6	18.6	106.0	12.4	551	15.5
2011	LE	32.0	10.4	62.6	13.7	81.0	17.4	167.0	16.9	83.2	18.5	111.0	14.6	538	15.3
	UH	20.0	11.9	46.5	15.2	95.0	18.7	166.2	17.6	85.3	19.0	33.6	15.5	447	16.3

Table 2- Soil characteristics of the experimental localities

Site	*Particle size density (determined by water pycnometer method) (g cm^{-3})*	*Saturated hydraulic conductivity (determined by Ksat device) (cm d^{-1})*	*Saturated water content (cm^{-3} cm^{-3})*	*Dry bulk density (g cm^{-3})*	*Porosity (%)*
LE	2.709	77	0.487	1.30	52
UH	2.645	1025	0.464	1.15	57

mulch treatments in a randomized block design with four replicates and plot size 7.2 m^2. Pre-sprouted seed tubers were hand-planted. Rows were apart 0.8 m, seed potatoes were placed 0.33 m apart in the rows. The plow and organic fertilization (no other nutrients were applied) were used and disease control were done according to organic agriculture (same for all treatments).

2.2. Mulching

The mulching with chopped grass (OM) and black textile mulch (PM) were compared to non-mulching control variant (C) with the mechanical cultivation (3 times up to closed crops and one before emergence). OM is material from natural meadows *Dactylis glomerata* - 40%, *Festuca pratensis* - 20%, *Lolium perenne* - 20%, *Poa pratensis* - 10%, *Alopecurus pratensis* - 10%. OM was manually spread in a 25-mm thick layer 14th day after planting (immediately after second hoeing). In plots with PM ridges were formed firstly and then covered by the black polypropylene non-woven textile and subsequently hand planted.

2.3. Soil water potential (SWP)

It was measured in all treatments (OM, PM and C) at the depth of 240 mm in 30-min intervals during period from planting to harvest with sensor Watermark 200SS-X cooperating with MicroLog SP (EMS, Brno).

2.4. Soil and plant analysis department (SPAD values)

After complete plant emergence, a SPAD was measured in the second fully expanded leaf from the apex. The readings were recorded in ten plants of each plot. The SPAD was determined by the SPAD-502 portable chlorophyll meter (Minolta Co, Tokyo, Japan). SPAD 502 meter is a small handy device, which measures light transmittance at red (650 nm, chlorophyll absorption) and near-infrared (960 nm) wavelength (Minolta 1989). The strong correlation of chlorophyll concentration and SPAD value were validated for a range of crops including potato (Vos & Born 1993). The SPAD readings were carried out in the morning, between 8:00 and 11:00 a.m. at 42, 57, 66, 76, 83 and 90 days after planting (DAP) on the same plants. The experiments carried out by Gianquinto et al (2004) showed that SPAD readings correlated to corresponding crop nitrogen content and then the highest coefficient of determination that was observed 32 days after emergence, at growing stage middle flowerings bud (about 15 days after tuber initiation). This stage is mentioned in SPAD readings. The SPAD values were correlated with YWP.

2.5. Weeds

The weight of weed biomass (WB) in all treatments were determined before harvest when the weeds were removed.

2.6. Harvest, measurement of the yield and statistical analysis

Tubers were harvested by hand. YWP (tubers > 40 mm) was determined for each plot. Data were subjected to analysis of variance using the ANOVA procedure and correlation (Pearson correlation coefficients) with SAS ver. 9.1.3. (SAS Institute Inc. 2003). Means were separated using Turkey's test at 95% level of probability only when the ANOVA F-test showed significant at 0.05 or 0.01 probability levels.

3. Results

3.1. Evaluation of soil water potential

SWP was variable (in vegetation of average from 22.4 to 119.7 kPa) due to rainfall (Table 3) and mulching materials at experimental sites. SWP developments during the year were strongly influenced by sum of rainfall and its distribution during the year. Nevertheless SWP was generally lower (respectively higher soil moisture) in the OM and PM treatments than in the C variant. Significant SWP differences in mulching treatment were observed in LE, where SWP levels were lower compared to UH (Table 3).

3.2. Evaluation of weed biomass

The type of mulch material affected the presence of weeds (Table 3). The lowest WB were found in PM (WB by 66.3% lower than C). The trend of lower WB was also at OM (lower by 11.7% comparison with C). The negative correlation between WB and YWP were not found in any of the experimental variants.

3.3. SPAD reading

Potatoes with the OM were characterized by the highest SPAD values (by 5.0%, 5.5% and 0.6% in 2009, 2010 and 2011) compared with C (Table 3). Similarly, the second-highest SPAD values were achieved with PM (on both sites in average 2009-2011). SPAD value at OM showed a medium correlation to yield of ware potatoes (r= 0.5363, P= 0.0023) and next yield components (Table 4). While at the PM was found a strong correlation (r= 0.6572, P<0.0001) between SPAD values and tuber yields (Table 5).

3.4. Yield of ware potatoes

The type of mulch material affected the YWP (Table 3). The highest YWP was found when using OM by 26.6% compared with C (28.5 t ha^{-1}). An application of OM (Figure 1) resulted in a significant increase weight of ware potatoes (fraction 55-60 mm and especially fraction over 60 mm). YWP after application of black plastic textile (PM) was similar with control. The fall of YWP at PM was recorded in UH in 2009 and 2010 (Table 3). On the contrary, during the period of tuber growth, in 2011 there was approximately 80-100 mm of precipitation more than in previous years (Table 1) and the yield of tubers was higher for PM.

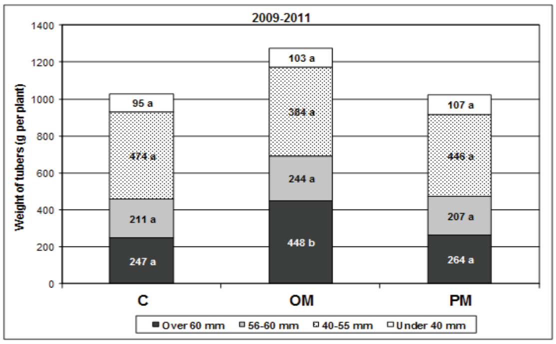

Figure 1- Weight of tubers in specific size fractions for each type of mulching (Note: same letters indicate statistically not significant differences; HSD$_{0.05}$ (over 60 mm)= 129.6, HSD$_{0.05}$ (56-60 mm)= 56.00, HSD$_{0.05}$ (40-55 mm)= 71.13, HSD$_{0.05}$ (under 40 mm)= 19.82)

4. Discussion and Conclusions

The significant differences of SWP were not observed in less rainfall locality in treatment of plastic mulching (Table 3). Link & Bower (2004) also examined the effect of plastic mulch type (permeable and non-permeable) on soil water potential too in a semi-arid ecosystem and plastic mulch had no effect on soil moisture retention (in depths 10, 30 and 60 cm in 12 experiments). However, SWP had the largest Pearson correlation coefficient (*r*) for the yield of ware potatoes (YWP) from the following factors (SWP, SPAD and WB). For control variant (C) r= -0.7188, P<0.0001, for OM r= -0.6509, P<0.0001 and for PM r= - 0.7391, P<0.0001 (Table 4, 5 and 6).

On the other hand, polypropylene (PP) textile had a positive effect on weed control in this experiment (Table 3), like similar polyethylene (PE) mulch (Ramakrishna et al 2006). We are convinced that total levels of WB were low during the main growth

Table 3- SPAD value, rainfall on vegetation, soil water potential and weight of tubers affected by different types of mulching materials (2009-2011)

Locality	*Variant of mulching*	*Rainfall on vegetation (mm)*	*Soil water potential (kPa)*	*Yield of ware potatoes (t ha^{-1})*	*Weed biomass (g m^{-2})*	*SPAD values*	*Total tuber yield (t ha^{-1})*
				2009			
LE	C	359.00	33.82	30.03	175.94	38.91	32.62
	OM		35.62	32.69	31.60	42.70	35.82
	PM		21.33	32.38	7.31	42.06	36.43
UH	C	317.00	57.04	30.34	38.33	35.63	32.52
	OM		36.50	35.49	88.99	36.18	37.51
	PM		67.24	23.67	0.35	35.71	26.54
Average of localities	C	--	45.43	30.22 AB	98.42 A	36.94	32.56 B
	OM		36.06	34.37 A	68.13 A	38.79	36.84 A
	PM		44.29	27.15 B	3.21 B	38.25	30.50 B
	HSD$_{0.05}$		NS	4.16	63.18	NS	4.02
				2010			
LE	C	744.00	36.93	43.97	229.01	37.37	48.49
	OM		37.35	52.48	252.12	39.37	57.02
	PM		29.67	34.55	58.49	38.41	40.04
UH	C	551.00	108.55	17.69	90.33	28.11	22.97
	OM		119.72	21.72	71.70	29.68	27.50
	PM		99.89	14.64	4.58	28.53	19.35
Average of localities	C	--	72.74	28.20 B	151.9 A	31.81 B	33.18 B
	OM		78.54	34.03 A	147.2 A	33.56 A	39.30 A
	PM		64.78	22.60 C	35.04 B	32.48 B	27.63 C
	HSD$_{0.05}$		NS	5.16	87.46	1.02	4.69
				2011			
LE	C	538.00	29.58	29.33	506.13	37.33	30.82
	OM		22.38	45.07	406.37	37.27	47.49
	PM		25.22	49.29	341.75	39.47	50.60
UH	C	447.00	93.19	25.43	147.41	39.74	26.37
	OM		91.43	36.09	149.76	40.14	37.00
	PM		101.66	26.39	10.61	38.41	27.54
Average of localities	C	--	61.39	26.99 B	283.80 A	38.77	28.15 B
	OM		56.91	39.68 A	256.50 A	38.99	41.19 A
	PM		63.44	35.55 A	141.80 B	38.83	36.76 A
	HSD$_{0.05}$		NS	6.28	87.40	NS	6.16
				2009-2011			
Average	C	--	59.85	28.47 B	178.10 A	35.84 A	31.30 B
	OM		57.17	36.03 A	157.30 A	37.11 A	39.11 A
	PM		57.50	28.44 B	60.02 B	36.52 A	31.63 B
	HSD$_{0.05}$		NS	3.95	50.19	1.79	3.78

Means accompanied by the same letters are not significantly different at $P \leq 0.05$

Table 4- Correlational relationship for organic mulch (with chopped grass)

OM	*WT 55-60 mm*	*WT over 60 mm*	*YWP*	*WB*	*SPAD*	*SWP*
WT 55-60 mm	1.000	0.2516	0.5636**	0.1338	0.6301***	-0.5942***
WT over 60 mm		1.000	0.6410***	0.2239	0.4763**	-0.3275
YWP			1.000	0.4974**	0.5363**	-0.6509***
WB				1.000	0.1044	-0.3662*
SPAD					1.000	-0.5538**
SWP						1.000

Weight of tubers fraction 55-60 mm (WT 55-60 mm), weight of tubers fraction over 60 mm (WT over 60 mm); significant differences of correlation test *, P<0.05; **, P<0.01; ***, P<0.001

Table 5- Correlational relationship for plastic mulch (black polypropylene textile)

PM	*WT 55-60 mm*	*WT over 60 mm*	*YWP*	*WB*	*SPAD*	*SWP*
WT 55-60 mm	1.000	0.5541**	0.8561***	0.3865*	0.5756***	-0.5864***
WT over 60 mm		1.000	0.6974***	0.5815***	0.4922**	-0.1935
YWP			1.000	0.6496***	0.6572***	-0.7391***
WB				1.000	0.2225	-0.4404*
SPAD					1.000	-0.6265***
SWP						1.000

Significant differences of correlation test *, P<0.05; **, P<0.01; ***, P<0.001

Table 6- Correlational relationship for control variant (without mulching)

Control	*WT 55-60 mm*	*WT over 60 mm*	*YWP*	*WB*	*SPAD*	*SWP*
WT 55-60 mm	1.000	0.4396*	0.5209**	0.2002	0.5677**	-0.5615**
WT over 60 mm		1.000	0.6221***	-0.1117	0.3774*	-0.1434
YWP			1.000	0.2463	0.3893*	0.7188***
WB				1.000	0.2009	-0.5243**
SPAD					1.000	-0.4691**
SWP						1.000

Significant differences of correlation test *, P<0.05; **, P<0.01; ***, P<0.001

period of tubers in all experimental variants. It was a late occurrence of weeds when the growth of the tuber and the yield of the tubers was completed. It was not about nutrient and water competition in this case. Therefore the correlation relationship between WB (determined before harvest) and YWP has been confirmed for OM and PM (Table 4 and 5). Although plastic foils are used in crop production for weed control, they have many disadvantages (Warnick et al 2006). Polyethylene foil is impermeable to precipitations and has less strength (in contrast to PP textile). Therefore, the PP textile can be used as multi-year.

Higher SPAD values, respectively higher CHLO content at mulch stands testify to their better vitality, since, as mentioned Boochs et al (1990) a loss of vitality expressed in a reduction of the CHLO content and lowers the absorption of photosynthesis active radiation (PAR). The correct use of CHLO meter can give a quick and accurate estimation of both crop nitrogen status and final YWP (Gianquinto et al 2004). We believe that increased SPAD values (Figure 2) also guarantee an increase N content in plants, respectively increase of the tuber yield. We proceed from the known truth about correlation SPAD and nutritional status respectively, N content in potato leaves (Gianquinto et al 2004). The relationship of chlorophyll content and the yield of tubers is evident here (the highest yield of tubers and the highest SPAD values for OM). Likewise, as expressed by the correlation coefficient for OM (r= 0.5363, r^2= 28.8%, P= 0.0023). An even stronger correlation was found for PM (r= 0.6572, r^2= 43.2%, P<0.0001). Gianquinto et al (2004) also showed the dependence of the SPAD levels at different sampling dates and the final yield of potato tubers in the variety Primura. This coefficient of determination ranged from r^2= 56.3% to r^2= 83.5% in their experiments.

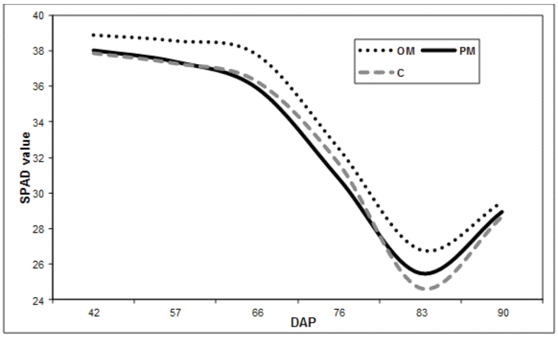

Figure 2- The evolution of SPAD values on plots with organic mulch (OM), plastic mulch (PM) and control variant (C)

OM favorably influenced the conditions during vegetation and it showed a yield increase of 7.6 t ha^{-1}. The Increase of tuber yield up to 10 t ha^{-1} was mentioned in connection with the mulching potatoes within Momirovic et al (1997).

It is difficult to achieve greater representation (proportion) of ware potatoes (tubers 55-75 mm) in organic cultivation. Sawicka et al (2007) show that comparison of the systems (ecological and conventional) and years had the strongest effect on the share of the weight of the tubers with a diameter > 6 cm as well as tuber yield of ware potatoes. The use of OM helped to significantly increase of fraction biggest tubers (over 60 mm) and tuber fraction 55-60 mm, which are popular for big consumers.

On the other hand, lower tuber yields were in accordance with the less favorable soil moisture and SWP for PM (at the term intensive growth of tubers in June and July). Due to textiles and worse rainfall penetration there was an increase SWP and yield depression, especially in the warmer and in rain poor locality as UH in 2009 and 2010 (Table 1 and 3). Therefore, we experienced higher the seasonal fluctuations in the yields of tubers at PM, on the contrary, OM stabilized the yield of tubers on the two soil-climatically different localities (LE a UH).

Based on the availability of data from the database of soil hydrophysical characteristics of the Czech Republic (Miháliková et al 2013), it is possible to derive soil hydrophysical properties from easily accessible soil properties. The diversity of precipitations and soil types at both experimental sites (LE and UH) deepens the information on the possibilities and benefits of using definite mulch material (OM or PM).

In conclusion, the expected production of organic potatoes can be based on the application of chopped grass as organic mulching. However, the highest SPAD reading value did not guarantee the highest tuber yield.

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Compost Effects on Soil Nutritional Quality and Pepper (*Capsicum annuum* L.) Yield

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ABSTRACT

There is a large amount of organic waste depending on the climatic conditions and product diversity in the agricultural areas of Turkey. These wastes can be used as a soil conditioner to reduce environmental problems and enrich soil organic matter by the transformation of organic wastes into organic manure with the process of composting. This study was performed to investigate the effects of composted greenhouse wastes (tomato residues) and animal manure on macro nutrient contents of soil and green pepper yield. Treatments were as follows: (1) Control, (2) mineral fertilizer, (3) 40 t ha^{-1} animal manure, (4) 40 t ha^{-1} animal manure + mineral fertilizer, (5) 40 t ha^{-1} tomato residuals, (6) 40 t ha^{-1} tomato residuals + mineral fertilizer, (7) 80 t ha^{-1} tomato residuals, (8) 80 t ha^{-1} tomato residuals + mineral fertilizer. Three replicates each of disturbed soil samples (two sampling) were collected (0-20 cm) from each treatment in two vegetation period and green pepper was used as a test plant in the study. In consequence of the research, significant increases were observed in crop yield and macro nutrient contents of soil. The highest increasing rate was analyzed in yield values as 305%. The most effective treatments on soil macro nutrient content and yield were determined as 40 t ha^{-1} animal manure combined with mineral fertilizer and 80 t ha^{-1} composted tomato residues combined with mineral fertilizer.

Keywords: Animal manure; Compost; Greenhouse wastes; Macro nutrients; Yield

1. Introduction

Conventional agricultural practices may include frequent and intensive tillage and the extensive use of fertilizers and pesticides. Such practices can result in a loss of organic matter (OM), leading to the degradation of cultivated soils and a decline in soil quality (Lal 2007; Peigne et al 2007; Batlle-Bayer et al 2010; Pan et al 2010). An integrated use of inorganic fertilizer with organic manure is a sustainable approach for efficient nutrient usage which enhances efficiency of the chemical fertilizers while reducing nutrient losses (Schoebitz & Vidal 2016).

Organic wastes such as animal manures, by-products of several kinds and composted residues can be used as amendments to increase soil fertility. They are important sources for enriching plant development and soil quality (Alluvione et al 2013; Li & Han 2016). Composting of the plant residuals and unusable organic wastes is an old and inexpensive method; the so-obtained compost can be used as an organic fertilizer or soil amendment

(Chang et al 2006). Applications of plant residues, cotton gin, composted urban waste, and poultry manure within high organic matter content to soils have been used to restore and maintain soil of organic matter, thus reclaiming degraded soils and supplying plant nutrients (Ros et al 2003; Walker 2003; Tejeda et al 2006).

Tomato (*Solanum lycopersicum* L.) is the most commonly grown vegetable plant in greenhouses and represented 55% of total vegetable production in Turkey. Greenhouse tomato production was determined as 458.6 t ha^{-1} in Turkey and 100 t ha^{-1} in Simav district of Kutahya in 2016 (TUIK 2017). Although production wastes or residues of tomato, their high organic matter content, are appropriate for composting, they are burned or dumped in open areas in Turkey (Kulcu 2014). Even though there are many studies about agricultural wastes, there is no sample research to solve waste disposal problems for farmers in Simav which is suitable for intensive greenhouse production. The aim of this study to evaluate these wastes as an alternative source due to the fact that composting of these wastes prevents environmental pollution and diseases and pests.

Green pepper was used as a test plant in the study. Pepper plant in our country, many arid and semi-arid areas where salinity problem commonly seen as a potential risk for croplands is one of the most important grown vegetable in greenhouses and fields. Current status of pepper plant production under greenhouses in Turkey was found as 307.2 t ha^{-1}. Total greenhouse area in Simav district was 426.1 ha and pepper production in greenhouses was 50 t ha^{-1} (TUIK 2017).

In this study, the effects of composted tomato plant residues and animal manure on macro nutrient composition of soil and pepper yield were compared.

2. Material and Methods

2.1. Site description and treatments

This study was conducted in the greenhouses of Vocational College of Simav, Dumlupinar University in Simav, Kutahya, Turkey (Figure 1). The experiment soil was classified as sandy loam with slightly alkaline reaction and some physico-chemical properties were given in Table 1. The experiment was established in a randomized block design with three replications. The plot size was 3 m x 2 m with each plot having 80 cm and 80 cm distance between and within rows, respectively with 13330 plants ha^{-1} plant population. Mono-ammonium phosphate (12% N, 61% P), potassium sulfate (50% K), ammonium nitrate (33% N), potassium nitrate (13% N, 46% K), and calcium

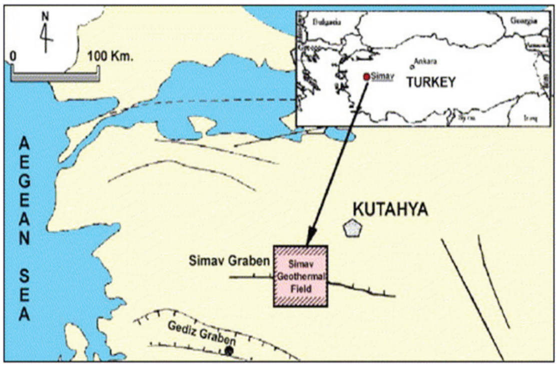

Figure 1- Location of the study area

Table 1- Some physical andchemical properties of experimental soil (Cercioglu et al 2017)

Soil texture	*Sandy loam*
Sand (%)	60.48
Silt (%)	27.64
Clay (%)	10.88
$CaCO_3$ (%)	1.80
OM (%)	1.85
Total N (%)	0.17
pH	7.71
EC (µS cm^{-1})	1467.00
P (mg kg^{-1})	104.60
K (mg kg^{-1})	206.00
Ca (mg kg^{-1})	3760.00
Mg (mg kg^{-1})	843.00
Na (mg kg^{-1})	213.30
Fe (mg kg^{-1})	7.98
Cu (mg kg^{-1})	2.15
Zn (mg kg^{-1})	3.92
Mn (mg kg^{-1})	10.10

nitrate (15.5% N, 26.5% CaO) were used as mineral fertilizers (NPK) and applied to the plots in the first hoe period after one month of planting. Animal manure (AM), and tomato residues (TR) were used as organic fertilizers and applied to the plots one day before planting.

Pepper (*Capsicum annuum* L.) is grown as an annual plant and is actually herbaceous perennial that's survives and yield for several years in tropical climates (Kelley & Boyhan 2009). The material used for composting was tomato residue obtained from the greenhouses located in Simav, Kutahya. Composting was performed outdoor under a roof. The moisture content of the compost was analyzed approximately 55% by weighing the material regularly and adding water when necessary. The aeration was made by manual turning during the composting process. The composting process lasted 3 months and composting was considered complete when the C:N ratio (7.99) and temperature (35 °C) became constant. Composted tomato residues and animal manure obtained from farmers were added to the soil after composting. All doses of organic wastes were determined according to initial soil analysis results, and nutrient removal by pepper plant from soil. Some properties of these organic wastes were reported in Table 2. The treatments where: (1) Control, (2) NPK, (3) 40 t ha^{-1} AM, (4) 40 t ha^{-1} AM + NPK, (5) 40 t ha^{-1} TR (6) 40 t ha^{-1} TR + NPK, (7) 80 t ha^{-1} TR, (8) 80 t ha^{-1} TR + NPK.

Table 2- Some chemical properties of organic wastes (Cercioglu et al 2017)

Parameters	*Composted tomato residuals*	*Animal manure*
pH	8.79	8.43
EC (µS cm^{-1})	1772.00	2700.00
OM (%)	30.00	57.80
C:N	7.99.00	14.20
Total N (%)	2.18	2.35
Total P (mg kg^{-1})	1284.00	3600.00
Total K (mg kg^{-1})	5547.00	9400.00
Total Ca (mg kg^{-1})	11540.00	24200.00
Total Mg (mg kg^{-1})	2469.00	5300.00
Total Na (mg kg^{-1})	481.20	588.00
Total Fe (mg kg^{-1})	5964.00	784.00
Total Mn (mg kg^{-1})	254.00	202.00
Total Cu (mg kg^{-1})	10.60	12.30
Total Zn (mg kg^{-1})	45.50	51.60

2.2. Sampling and laboratory analyses

During the experiment, two soil samples were taken from 0-20 cm depth for each vegetation (first vegetation: November, 2014 and March, 2015; second vegetation: April, 2015 and August, 2015). The samples were air-dried and sieved through 2 mm sieve. Soil texture was determined according to Bouyoucos (1962). Soil organic matter was determined according to method of Nelson & Sommer (1982). Soil reaction (pH) in 1:1 (w/v) soil water suspension by pH meter; and electrical conductivity (EC) in the same suspension by EC meter (Kacar 1994). Calcium carbonate was measured according to Scheibler method (Schlichting & Blume 1966). Total N was analysed according to Kjeldahl method (Bremner 1965). Available P was determined by the Mo blue method in a $NaHCO_3$ extract (Olsen et al 1954). Available K, Ca, Mg and Na were determined by 1 N NH_4OAc (pH: 7) method. Samples of composted materials were collected from the bulks separately, and air-dried. The pH and EC values of composted materials were analyzed in aqueous extract, which was obtained mechanically shaking the samples for 1 hour with distilled water at a solid/water ratio of 1:10 (dry weight/volume) by using pH and EC meter, respectively (Kacar 1994). Moreover, organic matter (Nelson & Sommer 1982); total N (Bremner 1965); available P (Olsen et al 1954); available K, Ca, Na, Mg contents (Pratt 1965); and available micro nutrient contents (Lindsay & Norvell 1978) were determined.

Drip irrigation method was used according to water needs of plant in the study. Pepper plants harvested from each plot were weighed after each harvest and determined their moist weights. After these procedures, all yield values were calculated from plot area as t ha^{-1}.

2.3. Statistical analysis

Analysis of variance (ANOVA) and Duncan's tests were conducted with a $P<0.05$ significance level and 95% confidence interval using SPSS Version 25 (IBM Corp. 2017) statistical software.

3. Results and Discussion

3.1. Macro nutrient contents of soil

Soil macro nutrients (N, P, K, Ca, Na, Mg) were presented in Table 3. Total N content of soil increased in all the treatments compared to control plots. At the beginning of the experiment, total N was analyzed as 0.17%. With the application of wastes, N amounts varied between 0.11 to 0.37% during first and second vegetation periods. These values were found sufficent when compared with the limit values of soil nitrogen according to Sillanpää (1990). Compared with the control, the highest total N content of soil was obtained as 0.37% in the 40 t ha^{-1} AM + NPK and 80 t ha^{-1} TR + NPK plots, an increase of 117% in the first soil samples of first vegetation. It was closely related to the high nitrogen content of animal manure (2.35%) and composted tomato residues (2.18%) applied to the soil. Statistically, there was no significant differences among both treatments (40 t ha^{-1} AM + NPK and 80 t ha^{-1} TR + NPK) in terms of N contents of the first soil samples of first vegetation. Similar results were reported by Wang et al (2004) in plots treated with composted dairy and swine manures. According to Ayuso et al (1996), the increase may be attributed to a direct effect of organic N derived from the compost, which is slowly mineralized in soil after the composting process (Castellanos & Pratt 1981). Available P content of soil was increased by all the treatments. During all vegetation periods, soil P contents varied between 60.90 to 136.10 mg kg^{-1} with application of these materials. The highest available P content in soils was analysed as 136.10 mg kg^{-1} in the first soil samples of first vegetation under 40 t ha^{-1} AM + NPK plot. The increasing rate was 30.1% compared to the control (104.60 mg kg^{-1}). Available soil P was found in very high level (>80 mg kg^{-1}) according to Sillanpää (1990). These results were found similar to the other study about greenhouse soils (Kaplan et al 1995). Moreover, it was closely related to the high phosphorus contents of composted tomato residues (1284 mg kg^{-1}) and animal manure (3600 mg kg^{-1}) applied to the soil. The increase in the available forms of phosphorus and sulfur has explained by increase of soil organic matter content (Shang et al 2014; Siwik-Ziomek & Lemanowicz 2014). Before the applications of the materials, soil initial available K content was determined as 206 mg kg^{-1} and total K content of animal manure and composted tomato residues were 9400 mg kg^{-1} and 5547 mg kg^{-1}, respectively. Available K content of soil was significantly affected with all the treatments. The maximum K contents were measured as 503.30 mg kg^{-1}, an increase of 144% when compared to the control by the application of 40 t ha^{-1} AM + NPK in the first soil samples of first vegetation (Table 3). Similar to N content of soil, both treatments (40 t ha^{-1} AM + NPK and 80 t ha^{-1} TR + NPK) showed statistically same significance level. According to Sumner & Miller (1996), available K values were varied from sufficient level (140-370 mg kg^{-1}) to high level (370-1000 mg kg^{-1}) after material applications. An increase in the exchangeable potassium content of soil was also observed by Zhang et al (2011) after application of NPK and manure (pig, horse, cattle or sheep). Saltalı et al (2000) found that increasing rate of tobacco waste increased total N and available P, K contents. Obtained data showed that application of animal manure and plant residuals to alkaline soils improved both soil conditions and nutrient concentration of soil to increased sufficient crop production. Some authors have also found an increase in K and Mg in the soil after organic amendments (Bulluck et al 2002; Edmeades 2003). They attributed the result to the high nutrient contents of the compost and the increase of cation exchange capacity due to organic matter added. While the available Ca content of soil was found higher in second vegetation soil, the available Na and Mg content of soil was higher in first vegetation soil. Soils treated 80 t ha^{-1} TR + NPK had the highest available Ca content (4397 mg kg^{-1}) with an increase of 114% over the control. Soil available Ca values were analyzed in a high level (3500-10000 mg kg^{-1}) as compared to the limit values of Sumner & Miller (1996). As shown in Table 3, the highest Na content was determined as 149.50 mg kg^{-1} in 80 t ha^{-1} TR + NPK application in the first vegetation soil and

Table 3- Some soil chemical properties belong different applications and results of Duncan's multiple comparison tests

Treatments	*N (%)*		*P (mg kg^{-1})*		*K (mg kg^{-1})*		*Ca (mg kg^{-1})*		*Na (mg kg^{-1})*		*Mg (mg kg^{-1})*	
	I. sampling	*II. sampling*	*I. sampling*	*II. sampling*	*I. sampling*	*II. sampling*	*I. sampling*	*II. sampling*	*I. sampling*	*II. sampling*	*I. sampling*	*II. sampling*
							I. vegetation period					
(1) Control	0.17 d	0.14 f	104.60 g	100.60 h	206.00 g	181.40 h	2048 e	1943 e	65.90 g	58.90 g	427.30 g	417.60 g
(2) NPK	0.29 c	023 d	109.80 f	106.50 g	229.30 f	192.60 g	2482 d	2365 c	72.30 f	65.60 f	501.40 f	516.70 f
(3) 40 t ha^{-1} AM	0.34 b	0.29 bc	133.60 b	126.30 c	424.60 e	385.60 f	2451 d	2117 d	105.50 d	99.60 c	564.90 d	564.90 b
(4) 40 t ha^{-1} AM+NPK	0.37 a	0.30 b	136.10 a	131.30 a	503.30 a	474.80 b	2767 c	2353 c	130.10 c	88.70 d	577.20 c	530.80e
(5) 40 t ha^{-1} TR	0.28 c	0.19 e	121.00 e	118.00 f	446.30 d	430.50 d	2722 c	2409 c	131.30 c	116.40 a	553.30 e	539.00 d
(6) 40 t ha^{-1}+ TR+NPK	0.35 b	0.28 c	125.20d	121.30 e	498.50 b	463.90 c	3354 b	3068 b	144.70 b	73.20 e	593.90 b	545.50 c
(7) 80 t ha^{-1} TR	0.33 b	0.25 d	128.00 c	124.00 d	473.40c	405.00 e	3423 b	3119 b	91.60 e	74.30 e	576.30 c	537.40 d
(8) 80 t ha^{-1} TR+NPK	0.37 a	0.33 a	133.10 b	129.50 b	501.60 a	478.10 a	4397 a	3843 a	149.50 a	108.20 b	618.70 a	577.50 a
							II. vegetation period					
(1) Control	0.11 e	0.08 f	79.90 f	60.90 f	143.40 g	104.00 g	2663 f	1941 g	83.60 f	80.20 g	434.40 e	430.80 f
(2) NPK	0.21 c	0.17 c	98.50 e	95.20 e	148.40 f	107.30 f	2288 g	2058 f	91.70 d	91.00 d	443.40d	434.00 f
(3) 40 t ha^{-1} AM	0.21 c	0.17 c	122.80 b	105.30 c	316.50 e	257.10 e	2952 d	3642 c	107.60 c	101.10 b	494.10 c	471.40 e
(4) 40 t ha^{-1} AM+NPK	0.25 b	0.22 b	127.00 a	114.50 a	453.90 b	355.20 b	3965 a	4003 a	118.80 a	107.40 a	525.10 a	506.30 b
(5) 40 t ha^{-1} TR	0.15 d	0.12 e	102.70 d	93.90 e	413.60 c	262.70 d	2771 e	3098 e	86.80 e	82.70 f	496.60 c	485.00 d
(6) 40 t ha^{-1} TR+NPK	0.24 b	0.21 b	115.90 c	104.70 c	452.30 b	354.00 b	3357 c	3259 d	91.00 d	84.30 ef	514.40 b	496.30 c
(7) 80 t ha^{-1} TR	0.22 c	0.15 d	116.30 c	97.30 d	354.90 d	288.00 c	3703 b	3064 e	90.80 d	85.30 e	523.10 a	516.30 a
(8) 80 t ha^{-1} TR+NPK	0.30 a	0.25 a	122.60 b	110.90 b	464.30 a	374.40 a	3925 a	3832 b	110.70 b	98.50 c	517.20 b	505.70 b

The table presents significance levels among treatments for the measured parameters. Within columns, values followed by same letter for the treatments are not significantly different at the 0.05 probability level as 118.80 mg kg^{-1} in 40 t ha^{-1} AM + NPK application in the second vegetation soil. Available Na values were found in sufficient level (68-230 mg kg^{-1}) according to Loue' (1968). The highest Mg contents was as 618.70 mg kg^{-1} in first vegetation soils with 80 t ha^{-1} TR + NPK whereas, the highest in the second vegetation period with 40 t ha^{-1} AM + NPK (525.10 mg kg^{-1}) and 80 t ha^{-1} TR (523.10 mg kg^{-1}). Available Mg contents of soil were varied between 417.60 mg kg^{-1} (sufficient level: 160-480 mg kg^{-1}) and 618.70 mg kg^{-1} (high level: 480-1500 mg kg^{-1}) (Sumner & Miller 1996). Similarly, Agbede & Ojeniyi (2009) determined that application of poultry manure to any tillage treatment improved soil total N, available P, exchangeable K, Ca and Mg concentrations and grain yield of sorghum. Courtney & Mullen (2008) also obtained that available nutritional elements such as potassium (K), calcium (Ca), magnesium (Mg), and phosphorus (P) increased when compost was applied.

3.2. Pepper yield

All the organic wastes added to the soil significantly ($P<0.05$) enhanced pepper yield compared to the control soil. The greatest yield values were 13.10 t ha^{-1} and 15.46 t ha^{-1} in the first and second vegetation plants with the application of 40 t ha^{-1} AM + NPK. Moreover, the highest yield was reached in the plants in the soil where 40 t ha^{-1} AM + NPK was applied; and an increase of 305% over the control (7.04 t ha^{-1}) (Figure 2, Duncan's test $P<0.05$). Roe (1998) reported that compost plus fertilizer combinations have been increased efficiency. Togun & Akanbi (2003) found that the use of compost alone or compost with mineral fertilizer was better than the control. Roe et al (1997) also reported that pepper and cucumber yields were usually higher when compost was combined with mineral fertilizers. Trávník et al (1998) observed an increase in the yield from 47.6 to 83.7% for various crops fertilized with farmyard manure and NPK compared with the unfertilized crop. Moreover, Ragasits & Kismanyoky (2000) reported that simultaneous application of NPK and farmyard manure has been shown to increase wheat gluten content and quality. Vogtmann et al (1993) found that compost treatments resulted in lower vegetable yields in the first two years, but there were no different yield results after the third year. In contrast, Eghball et al (2005) showed that residue effects of manure application on crop production and soil properties can last for several years.

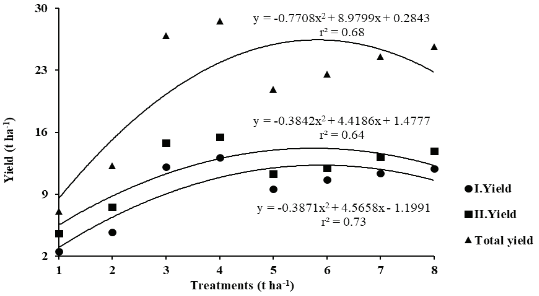

Figure 2- Pepper yield. Treatments; (1), Control; (2), NPK; (3), 40 t ha^{-1} AM; (4), 40 t ha^{-1} AM + NPK; (5), 40 t ha^{-1} TR; (6), 40 t ha^{-1} TR + NPK; (7), 80 t ha^{-1} TR; (8), 80 t ha^{-1} TR + NPK. Duncan's test ($P<0.05$)

4. Conclusions

The utilization of agricultural wastes derived from animal manure and greenhouse plants combined with or without mineral fertilizer for agronomic purposes is a potential management practices for sustainable agriculture. From the findings, it can be concluded that sandy loam soil amended with animal manure and composted tomato residues resulted in significant increase in pepper yield, total N, available P, K, Ca, Mg, and Na content of soil. The increase in yield has been mainly owing to the improvement in the nitrogen and phosphorus content of the soil. 40 t ha^{-1} AM + NPK and 80 t ha^{-1} TR + NPK applications gave the best crop yield values and improved soil nutritional quality under the conditions of this experiment. Adding different organic amendments to soil could be useful for various crops, and generally the use of plant residues could be a better option. Furthermore, the quality of used waste is an important parameter in view of agricultural management and crop productivity. Composting of greenhouse wastes promises to be an environmentally friendly alternative that converts the biodegradable wastes into a useful compost material. Composted materials could be a potential organic matter source and could also be utilized as a growing medium supplement in greenhouse vegetable productions.

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Effect of Irrigation Strategies on Yield of Drip Irrigated Sunflower Oil and Fatty Acid Composition and its Economic Returns

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ABSTRACT

A field trial was conducted to observe the effects of different irrigation strategies on the yield and the water use, oil content and marginal return of sunflower which was irrigated by means of a drip system during 2010 and 2011 under Çukurova condition of Turkey. The irrigation strategies include three irrigation intervals (A_1: 25 mm; A_2: 50 mm; A_3: 75 mm of cumulative pan evaporation) and six water levels (WL) based upon the percentages of cumulative pan evaporation (WL_1= 0.50, WL_2= 0.75, WL_3= 1.00 and WL_4= 1.25). In addition, WL_5= PRD50 and WL_6= PRD75 treatments were evaluated. They obtained water from alternative laterals 50% and 75% of the WL_3 treatment. Additionally, a non-irrigated treatment (NI) was included as control plot in the experiment. In each of the experimental years, the largest and the smallest average yields were acquired from the A_2WL_4 and NI treatments, respectively. The oil content and fatty acid composition were significantly affected by irrigation strategies. The oil content increased with the increasing amount of irrigation. Among all irrigation intervals, PRD-50 (WL_5) treatment provided the largest water use efficiency (WUE) and irrigation water use efficiency (IWUE) values in both growing seasons. In order to attain higher yields and a generated the marginal return, A_2WL_4 irrigation regime is suggested for sunflower production in the Mediterranean region. A_2WL_3 water strategy is proposed for an acceptable marginal return in case of water shortage.

Keywords: Reduced irrigation; Sunflower; Irrigation scheduling; Oil and fatty acid; Marginal return

1. Introduction

Sunflower is an important agricultural crop in most of the sunflower growing countries. In the world, 11% of crude vegetable oil production is supplied by sunflower. In Turkey, 47% of the crude vegetable oil production is supplied by the sunflower. The total production of sunflower is 1.670.716 tons in Turkey. The average yield of sunflower was 4100 kg ha^{-1} in 2016, despite changes in the regions. Turkey which has 4% ratio of sunflower production is in the first ten countries in the world (Konyalı 2017). The Mediterranean region in Turkey is defined as a semi-arid zone where most of the limited annual precipitation and uneven distribution occurs from October to May during the principal growing season. Water deficiency in this region is one of the most important factors affecting crop yield. Thus, irrigation is required during the growing season to maintain and enhance crop growth and yield. In the study conducted by Akcay & Dagdelen (2016), reported that, so as to save water and maximize the yield obtained, water saving irrigation systems should be followed; therefore, a proper irrigation scheduling is required for maximizing the yield and efficient water use. According to Asbaghy et al (2009) sunflower is not only tolerant to water stress caused by deficit irrigation (DI), but it is also high-yielding correspondingly to irrigation inputs. Numerous studies have been conducted to analyze the effect of DI in the response of crops in various areas worldwide. Recent literature revisions agree to conclude that DI is a highly convenient medium in stabilizing yields and increasing the productivity of water in water-scarce areas (Steduto et al 2012). Given the fact that irrigation water supply is scarce, DI becomes a favorable agronomic medium because water productivity is to be the goal instead of maximizing the yield per area unit. Being studied extensively in many parts of the world, deficit irrigation (DI) and partial root drying (PRD) are water-saving irrigation methods. DI is a method, by which the entire root zone is irrigated with an amount of water less than the evapotranspiration potential and the resulting minor stress has minimal effects on the yield, which consequently increases Water Use Efficiency (WUE). Finite water sources should be managed in a way that synthesizes the meeting irrigation requirements for crops and the improvement of WUE. Therefore the main goals of this research were to (a) detecting the effect of different deficit irrigation strategies on water use, seed, oil yields and oil yield-response factor of the sunflower and (b) evaluating the WUE and IWUE subject to different irrigation treatments and (c) estimating marginal return created by drip irrigated sunflower grown in the Çukurova region of Turkey.

2. Material and Methods

The field experiments of drip irrigated sunflower were conducted between April and August of two consecutive years, 2010 and 2011, at the Tarsus Location of Alata Horticultural Research Institute in Mersin, Turkey, (37º 01′ N latitude, 35º 01′ E longitude and 30 m altitude). The area is prevailed by typical Mediterranean climate and the soil of the field is identified as silty-clay-loam (SiCL) that has relatively high water-holding capacity. The available soil water in the upper 90 cm of the soil depth is 198 mm. The field capacity is 0.429 $cm^3\ cm^{-3}$ and permanent wilting point of soil is 0.207 $cm^3\ cm^{-3}$, while the mean bulk density ranged between 1.39 and 1.44 g cm^{-3}. The sunflower (cv. Oleko) seeds were planted in April 2nd, 2010 and May 5th, 2011 in rows that are 0.70 m apart. The seeding date was assigned as “0” Days After Sowing (DAS). After the crop establishment, final plant densities were calculated as 5.7 plants m^{-2} in 2010 and as 5.9 plants m^{-2} in 2011. The fertilizer treatments were performed according to soil analysis suggestions, in which equal amount of total fertilizer was provided for all treatment plots. Prior to planting on April 2nd, 2010 and on May 5th, 2011, a compose fertilizer with 15-15-15 (N, P, K) was administered at 40 N kg ha^{-1} rate; the rest of the N was administered to the experimental plots as NH_4NO_3 (33% N) at 30 kg ha^{-1} rate in May 10th, 2010 and in May 31st, 2011. In the study area, the water used was taken from channel having 7.8 pH value and 0.54 dS m^{-1} average electrical conductivity. The experiment was conducted with a split-plot design, including four replications, in which each subplot was arranged as 5 rows (8.0 m long and 3.5 m wide). The irrigation intervals were assigned as the main plot. The irrigation was applied by dripping on the main plot as soon as three different cumulative evaporation amounts, including A_1: 25 mm, A_2: 50 mm, and A_3: 75 mm, are reached. Among all irrigation intervals (A_1, A_2, and A_3), six irrigation levels were studied according to the percentages of cumulative pan evaporation (WL_1: 0.50, WL_2: 0.75, WL_3: 1.00 and WL_4: 1.25) in addition to considering WL_5 and WL_6 treatments. In WL_5 and WL_6, while one-half of the root left to dry, 50% and 75% of WL_3 was administered to the other half. Afterwards, irrigation was moved to the dry part, throughout the following irrigation. The laterals in drip irrigated plots were placed in each plant row having 70 cm distance in-between for WL_1, WL_2, WL_3 and WL_4 treatments, and in-line emitters with a discharge rate of 4.0 L h^{-1} were placed in the lateral line with 25 cm intervals (Betaplast Corp., Adana, Turkey). Two of the drip laterals were placed 20 cm away from the plant row in the WL_5: PRD-50 and WL_6: PRD-75 treatment plots. During the growing season, the system was run at a pressure of 100 kPa. As the control of these experiments, a non-irrigated treatment (NI) was also applied. A neutron probe (503 DR) was employed to measure the soil water content prior to irrigations during the growing season by using increments ranging from 30 cm to 90 cm. The neutron probes were installed on the plant row to monitor soil water throughout the growing seasons, while a Class-A pan was placed at the meteorological station next to the experimental plots. According to the physiological maturity of the plants, the yield was specified by hand harvesting three adjacent center rows with 6m sections in each plot. The seed yield and oil percentage values were noted subsequent to the harvest. The evapotranspiration (ET) value was calculated by using the water balance Equation (ET= I+R±ΔS-Dp-Rf), where ET is equal to evapotranspiration (mm), I amount of irrigation water applied (mm); ΔS change in soil water content (mm); Dp to deep percolation (mm); and Rf to runoff (Allen et al 1998). WUE and IWUE values were estimated as sunflower yield divided by seasonal ET and total seasonal irrigation water applied according to Howell (2001). The raw oil percentage was calculated by using the extraction method (Luque de Castro & García-Ayuso 1998), while the oil yield was calculated by using a function of seed yield and crude oil percentage. The fatty acid composition was analyzed by Erdemoglu et al (2003). The results obtained were presented as a relative area percentage of total fatty acid methyl esters (IOOC 2001). In order to assess the relationships between ET and the seed and oil yields attain from total ET, and the seed and oil yield data derived from the experiment, the regression analysis method was used. Within the scope of economic analysis, the irrigation cost and marginal return, in which the marginal yield was calculated as the difference between yield from irrigated treatment and non-irrigated treatment, were compared and all calculations were performed based on a unit area of 1 ha (Sezen et al 2011b). The sunflower production costs and sale prices were obtained from the Mersin Chamber of Commerce. Various expenses, including fertilizer, seed, soil cultivation, plant protection as well as land rental, labor cost for irrigation, harvesting and transportation costs constitute the production costs of sunflower. Therefore, the sum of crop production costs, yearly cost of the irrigation system, irrigation labor and water cost were evaluated in order to calculate the total cost for annual sunflower production.

All parameters were subjected to analysis of variance (ANOVA), and means were compared using Duncan test. All statistical analyses were performed using SPSS 11.5 for Windows.

3. Results and Discussion

3.1. Water use characteristics and soil water variation

The experimental area is prevailed by typical Mediterranean climate. As a result of the data analysis regarding climate, the temperatures measured during the growing seasons of 2010 and 2011 were found to be parallel to the typical long-term temperature mean of Tarsus. Table 1 summarizes the monthly climatic data compared with the long term mean climatic data for experimental area. Meteorological data were obtained from a nearby weather station. Since the rainfall distribution were unequal during the growing seasons, both experimental years (2010-2011) varied. In the first growing season (from April to July 2010), the depth of received rainfall was 112 mm, a little less than the long-term mean rainfall of 142 mm, while during the growing season of the second year, 2011, it was determined as 184 mm which was greater than that of the long-term mean rainfall as well as the first year (Table 1). The data for seasonal irrigation, crop water use (ET), WUE, IWUE and relative irrigation percentage in each treatment are given in Table 2. The first irrigation treatment was applied in May 18th, 2010 (DAS 47) and in June 7th, 2011 (DAS 33). On the other hand, the last irrigation application was performed in July 28th, 2010 (DAS 117) and on August 29th, 2011 (DAS 116). Total irrigation numbers (A_1, A_2 and A_3) were measured as 17, 8, 5 and 19, 9, 6 for 2010 and 2011, respectively. While the amounts of the applied irrigation water ranged between 199 and 603 mm based on the treatment in the 2010 growing season, non-irrigated treatment received no water. However, the irrigation water amounts ranged from 220 to 578 mm in 2011 (Table 2). Sunflower total ET varied between 268 mm for NI and 607 mm for A_2WL_4 treatment in 2010; and between 243 mm and 611 mm for NI and A_1WL_4 in 2011, respectively. Sezen et al (2011a) reported that ET values remained with the increasing amount of irrigation water and that the total ET for sunflower grown in the Eastern part of Mediterranean ranged between 269-689 mm under none-irrigated and full irrigation in 2006, and 2007.

Table 1- Historical monthly mean and growing season climatic data of the experimental area

Year	*Climatic parameters*	*Months*											
		I	*II*	*III*	*IV*	*V*	*VI*	*VII*	*VIII*	*IX*	*X*	*XI*	*XII*
2010	T_{mean} (°C)	10.9	11.8	14.5	18.5	21.1	24.4	27.2	29.1	26.9	21.4	18.1	14.5
	T_{max} (°C)	14.6	16.7	21.0	24.3	26.9	29.9	32.1	35.1	33.3	28.6	37.2	21.0
	T_{min} (°C)	7.2	7.3	8.8	11.4	15.7	19.2	22.7	23.1	21.3	15.6	10.5	8.8
	P (mm)	108.0	105.0	19.0	37.0	71.0	4.0	0.0	5.0	15.1	33.5	0.2	251.5
	E (mm)	34.5	43.5	83.4	118.2	152.9	175.5	202.0	222.0	184.8	113.6	78.9	83.4
	RH (%)	74.2	70.4	65.5	74.0	74.2	75.2	76.6	72.2	67.5	64.7	49.0	65.5
2011	T_{mean} (°C)	9.8	10.7	13.5	16.3	20.3	24.1	27.2	28.0	25.9	20.0	12.1	13.5
	T_{max} (°C)	15.3	17.3	19.8	22.0	26.2	28.6	31.0	33.4	32.1	27.6	18.7	19.8
	T_{min} (°C)	5.0	5.7	7.9	11.2	15.0	17.8	19.3	21.7	18.7	11.1	6.4	7.9
	P (mm)	71.0	36.0	77.0	89.0	70.0	25.0	0.0	0.0	8.0	19.4	27.0	183.9
	E (mm)	34.8	40.5	77.5	85.8	121.6	135.3	164.0	182.5	147.5	102.6	64.5	77.5
	RH (%)	68.0	67.0	70.2	69.1	70.4	75.2	77.1	70.8	66.8	52.0	60.2	70.2
Long term	T_{mean} (°C)	8.9	9.8	13.5	17.5	22.0	26.0	28.5	29.0	26.3	22.0	15.2	13.5
	T_{max} (°C)	19.5	21.0	26.2	31.9	35.7	37.3	37.6	39.2	38.2	36.0	28.7	26.2
	T_{min} (°C)	-1.9	-1.7	2.8	6.2	11.3	16.2	20.5	21.0	16.1	11.3	4.5	2.8
	P (mm)	111.8	79.0	55.4	55.1	45.7	18.0	12.3	12.3	17.8	37.1	87.9	55.4
	E (mm)	45.2	55.6	90.2	118.0	167.9	222.1	240.1	229.9	181.7	130.2	72.2	90.2
	RH (%)	70.9	71.4	66.5	67.7	67.1	68.2	73.2	72.4	66.3	62.2	65.3	66.5

T_{mean}, mean air temperature; T_{max}, maximum air temperature; T_{min}, minimum air temperature; P, rainfall; RH, relative humidity; E, evaporation

The course of soil water storage during the 2010 and 2011 growing seasons of sunflower for each irrigation frequency (A_1, A_2 and A_3) are shown in Figure 1a-f, respectively. In the A_1 and A_2 irrigation frequencies, soil water contents of treatment plots remained fairly high as compared to A_3 irrigation treatments. Soil water remained higher in the WL3 and WL_4 plots (in A_1 and A_2 irrigation intervals) than in the deficit treatments (WL_1, WL_2, WL_5 and WL_6) considered. Available soil water in WL_3 and WL_4 treatment plots remained above 50% throughout the growing season except the WL_3 irrigation interval. The soil water storage within the 90 cm depth gradually decreased towards the end of the season in heavy stress treatment (NI) in both experimental years and resulted in soil water contents below wilting point towards the end of the growing season of sunflower.

3.2. Growth stages for sunflower

The time spent on harvesting and on the various phenological growth stages (Steduto et al 2012)

Table 2- Amount of irrigation water, ET, seed yield, relative yield, oil content, oil yield, WUE and IWUE and fatty acid composition (palmitic, stearic, oleic, linoleic) values for different treatments in 2010-2011 periods

Years	Treatments	Irrigation (mm)	ET (mm)	Seed yield (kg ha^{-1})**	1000 Seed weights (g)**	Oil content (%)**	Oil yield (kg ha^{-1})	WUE (kg m^{-3})	IWUE (kg m^{-3})	Fatty acids (%)			
										Palmitic (C16:0)*	Stearic (C18:0)**	Oleic (C18:1)**	Linoleic (C18:2)**
2010	A_1WL_1	225	354	2908 cd	75.50 bc	34.40 ef	1000	0.82	1.29	4.03	2.50 abc	87.73 a	3.23 f
	A_1WL_2	338	471	3295 bcd	82.20 abc	36.30 cdef	1196	0.70	0.97	4.43	2.30 bcde	87.33 abc	3.50 ef
	A_1WL_3	450	542	3793 b	83.10 abc	42.80 abc	1623	0.70	0.84	5.67	1.87 hijk	85.83 hi	4.83 b
	A_1WL_4	563	597	4075 ab	89.10 a	43.50 ab	1777	0.68	0.72	6.07	1.73 jk	85.50 ij	4.97 ab
	A_1WL_5	225	359	3650 bc	84.00 abc	38.80 bcdef	1420	1.02	1.62	5.47	2.20 cdefg	86.50 defg	4.53 c
	A_1WL_6	338	460	3345 bcd	83.12 abc	42.10 abcd	1408	0.73	0.99	5.30	2.00 efghij	86.30 fgh	4.37 cd
	A_2WL_1	212	339	2703 d	75.00 bc	35.30 def	954	0.80	1.28	4.67	2.37 abcd	87.07 bcd	3.33 f
	A_2WL_2	318	446	3558 bc	83.00 abc	37.50 bcdef	1334	0.80	1.12	4.97	2.33 abcd	86.87 cdef	3.73 e
	A_2WL_3	424	516	4030 ab	84.20 abc	44.00 ab	1777	0.78	0.95	6.07	1.80 ijk	84.57 k	5.00 ab
	A_2WL_4	530	607	4758 a	85.30 ab	48.30 a	2298	0.78	0.90	6.63	1.60 k	84.13 k	5.17 a
	A_2WL_5	212	349	3993 ab	80.40 abc	40.70 bcdef	1625	1.14	1.88	5.47	2.17 defgh	86.07 ghi	4.53 c
	A_2WL_6	318	436	3813 b	83.70 abc	43.20 ab	1647	0.87	1.20	5.90	1.90 ghijk	85.20 j	4.90 ab
	A_3WL_1	199	360	2675 d	72.60 c	34.10 f	912	0.74	1.34	3.90	2.63 a	87.73 a	2.57 g
	A_2WL_2	298	425	2943 cd	74.20 bc	35.50 def	1045	0.69	0.99	4.00	2.57 ab	87.53 ab	2.73 g
	A_3WL_3	397	514	3740 bc	80.70 abc	40.80 bcde	1530	0.73	0.94	5.17	1.97 fghij	86.37 efgh	4.83 b
	A_3WL_4	603	603	4080 ab	82.20 abc	42.60 abc	1734	0.68	0.68	5.50	1.83 ijk	86.20 gh	4.87 ab
	A_3WL_5	199	356	3633 bc	84.80 abc	36.40 cdef	1319	1.02	1.83	4.27	2.23 cdef	86.50 defg	4.20 d
	A_3WL_6	298	426	3440 bcd	82.70 abc	39.50 bcdef	1355	0.81	1.15	4.80	2.07 defghi	86.93 bcde	4.37 cd
	NI	0	268	1670	65.10	33.60	563	0.62	-	3.73	2.90	88.50	2.20
2011	A_1WL_1	236	396	2855 i	78.50 cdef	39.70 ij	1138	0.72	1.21	3.93 hi	1.90 cd	88.67 a	4.00 gh
	A_1WL_2	348	452	3335 h	82.00 bcd	40.80 ghi	1366	0.74	0.96	4.67 efg	1.90 cd	87.67 cdef	4.43 ef
	A_1WL_3	461	513	3940 de	84.00 bc	43.70 de	1726	0.77	0.85	5.23 bcd	1.60 fg	86.13 i	5.03 abc
	A_1WL_4	578	611	4560 b	90.50 a	46.60 b	2116	0.75	0.79	5.47 bc	1.53 gh	85.93 ij	5.17 ab
	A_1WL_5	236	385	3695 efg	69.80 h	41.80 fgh	1550	0.96	1.57	5.10 cde	1.63 efg	86.97 fgh	4.63 cdef
	A_1WL_6	348	443	4025 cd	73.4 fgh	42.60 efg	1721	0.91	1.16	4.90 def	1.77 de	86.40 ghi	4.87 bcd
	A_2WL_1	221	320	3430 gh	75.10 efgh	39.30 ij	1351	1.07	1.55	4.73 ef	1.77 de	88.20 abc	4.33 fg
	A_2WL_2	328	416	3700 efg	76.50 defg	40.10 hi	1487	0.89	1.13	4.90 def	1.70 ef	87.87 bcd	4.47 def
	A_2WL_3	436	491	4118 cd	78.30 cdef	46.80 b	1934	0.84	0.94	5.63 ab	1.53 gh	85.33 jk	5.17 ab
	A_2WL_4	546	595	4890 a	80.20 bcde	51.10 a	2499	0.82	0.90	5.90 a	1.40 h	85.03 k	5.40 a
	A_2WL_5	221	327	4115 cd	69.40 h	43.70 def	1798	1.26	1.86	5.10 cde	1.63 efg	87.10 efg	4.83 bcde
	A_2WL_6	328	400	4273 c	71.00 gh	45.60 bc	1953	1.07	1.30	5.33 bcd	1.57 fg	86.20 i	5.03 abc
	A_3WL_1	220	342	2375 j	70.60 gh	37.10 k	885	0.69	1.08	3.43 j	2.23 a	88.57 ab	2.60 i
	A_2WL_2	327	423	2805 i	72.10 gh	38.20 jk	1076	0.66	0.86	3.63 ij	2.13 ab	88.20 abc	2.93 i
	A_3WL_3	436	495	3635 fg	83.30 bc	42.60 efg	1554	0.73	0.83	4.87 def	1.70 ef	86.43 ghi	4.77 bcde
	A_3WL_4	546	605	4080 cd	84.10 bc	44.40 cd	1817	0.67	0.75	5.10 cde	1.67 ef	86.33 hi	4.93 bc
	A_3WL_5	220	329	3560 fgh	85.20 ab	40.20 hi	1436	1.08	1.62	4.27 gh	1.90 cd	87.80 cde	3.67 h
	A_3WL_6	327	400	3743 ef	86.60 ab	40.80 ghi	1531	0.94	1.14	4.53 fg	2.00 bc	87.43 def	4.63 cdef
	NI	0	243	1700	63.50	36.10	614	0.70	-	3.30	2.40	89.20	1.70

Datas are emitted as a mean ± SD. *, $P<0.05$ and **, $P<0.01$

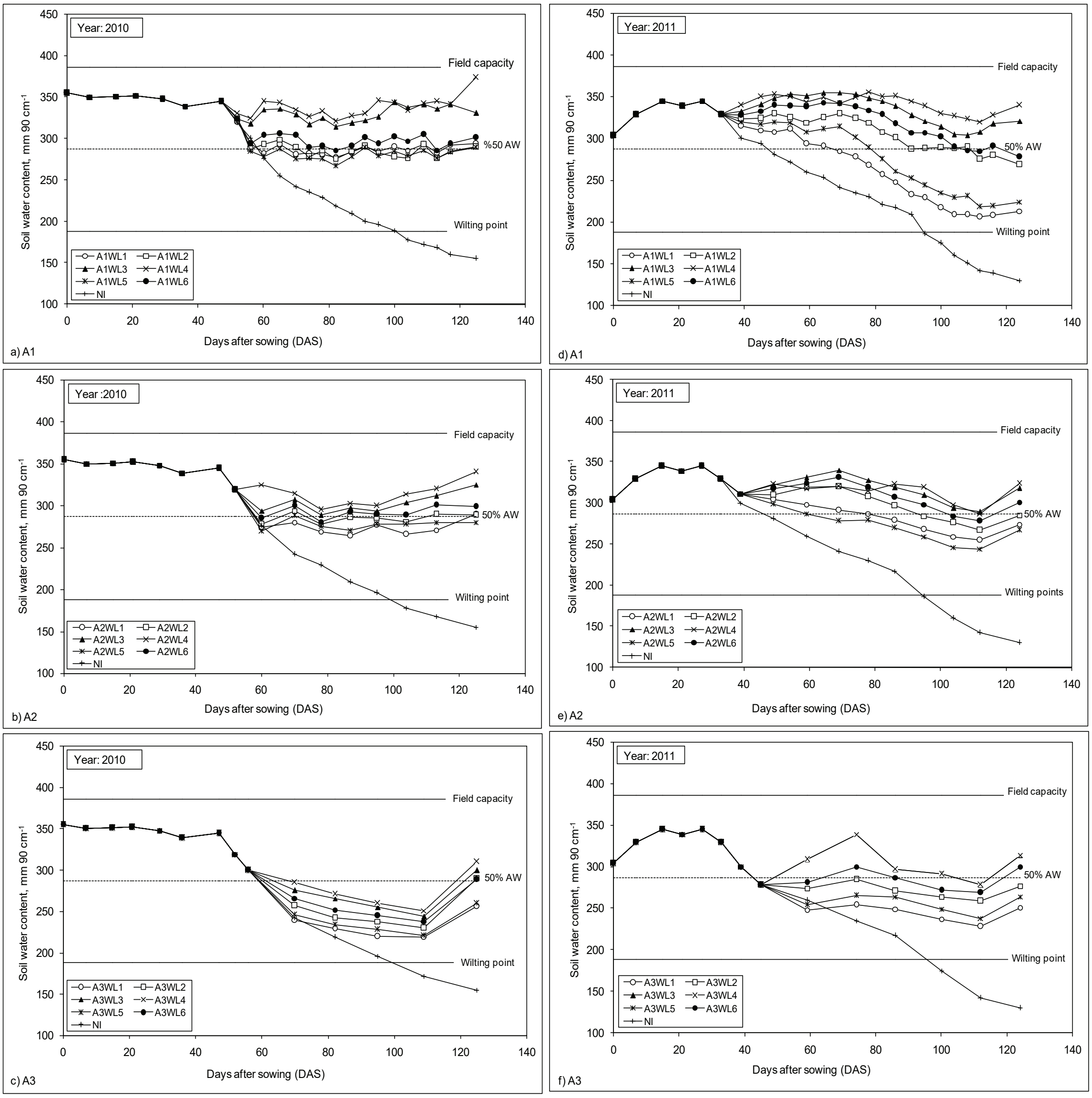

Figure 1- Course of soil water storage during the 2010 (a, b, c) and 2011 (d, e, f) sunflower growing season in all treatments

were noted as number of days after sowing (DAS). The total length of the growing season for sunflower was calculated as 125 days in 2010, while it was 124 days in 2011. Sezen et al (2011a) have reported growth stages of sunflower varying from 129 to 132 days in drip irrigation and varying from 121 to 132 days in sprinkler irrigation conditions in east Mediterranean part of Turkey.

3.3. Seed yield

Sunflower seed yields and yield components at the 1% level were remarkably influenced by interaction of irrigation intervals (A) and irrigation levels (WL). Based on Duncan test (Table 2), A_2WL_4 treatment was placed in the first group ($P<0.01$) in the first experimental year. The minimum yield was found in the non-irrigated treatment as 1670 kg ha^{-1} in 2010 and 1700 kg ha^{-1} in 2011. Even though PRD-50 treatments (WL_5) of A_1, A_2, and A_3 intervals received about 50% less irrigation of the water that was administered to the WL_3 plots of A_1, A_2 and A_3 irrigation intervals, the reduction in seed yields of WL_5 at A_1, A_2, and A_3 treatments were only 3.8, 0.9 and 2.9% in 2010 and 6.2, 0.1, and 2.1% in 2011, respectively, as compared to WL_3 in both years (Table 2). It was found that seed yield decreased significantly when the amount of irrigation was lowered. Based on Duncan test (Table 2), A_2WL_4 treatment was situated in the first group ($P<0.01$) while A_1WL_4 treatment was fallen into the second group in 2011.

3.4. Water use efficiency (WUE) and irrigation water use efficiency (IWUE)

It was observed that irrigation regimes affected WUE and IWUE values considerably (Table 2). WUE values varied between 0.62 kg m^{-3} in NI treatment to 1.14 kg m^{-3} in the A_2WL_5 for the first experimental year, while it varied between 0.66 kg m^{-3} in A_3WL_2 and 1.26 kg m^{-3} in the A_2WL_5 for the second year. The highest WUE values were measured in A_2WL_5 for both growing years. In the study carried out by Sezen et al (2011a), similar results were attained for sunflower grown under water stress conditions. Depending on the treatment applied, IWUE values varied between 0.68 and 1.88 kg m^{-3} in 2010 and varied between 0.75 and 1.86 kg m^{-3} in 2011. For both experimental years, the highest IWUE values were observed in A_2WL_5. In addition, Sezen et al (2011b) reported that the irrigation strategies in sprinkler and drip systems affected the IWUE values significantly. Akcay & Dagdelen (2016) reported that the WUE and IWUE values were affected by the irrigation intervals and levels. WUE varied from 0.70 kg m^{-3} to 1.21 kg m^{-3} among treatments in both years.

3.5. ET-seed yield and ET - oil yield relationships on sunflower

Regression analysis showed that there was a linear relationship between seed and oil yield with total ET at the 0.05 level of significance for both years (Figure 2a-b). It was demonstrated that the oil content is strongly influenced by the figure of ET throughout the growing season.

3.6. Seed weight

According to the analysis of variance administered to the results obtained in 2010 and 2011; sunflower seed weight at the 1% level were statistically

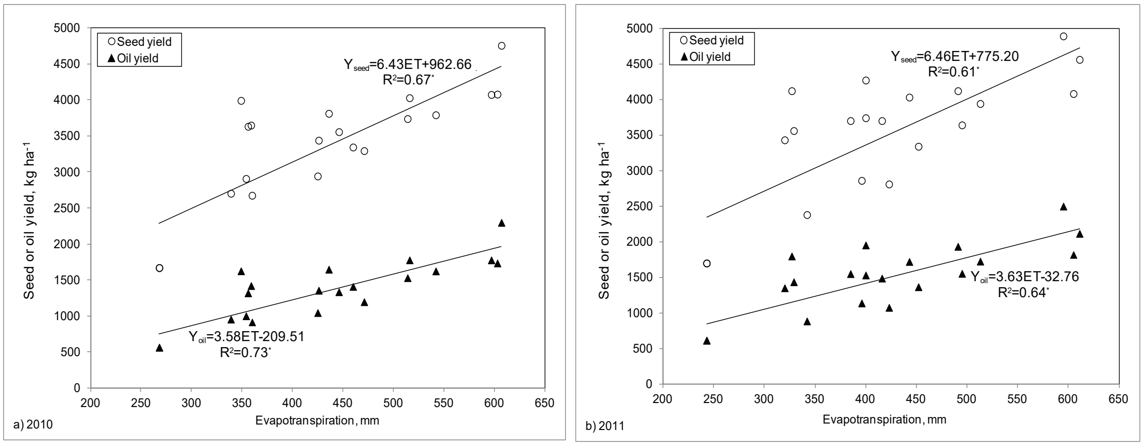

Figure 2 a, b- Interaction between evapotranspiration (ET)-seed or oil yield in 2010(a) and 2011(b)

influenced by interaction of irrigation intervals (A) and irrigation levels (WL). As for 1000 seed weights, it varied from 65.0 to 89.2 g in 2010 while the values were between 63.6 and 90.6 g in 2011. The lowest and the highest values in 1000 seed weights were obtained at NI and A_1WL_4 in 2010 and 2011, respectively (Table 2). Langeroodi et al (2014) reported that 1000 seed weight of the sunflower seeds varied between 54.3 and 68.0 g according to the treatments.

3.7. Oil yield, oil percentage and fatty acid compounds

The oil yield and percentage was significantly influenced (P<0.01) by the irrigation intervals and irrigation levels in both years. The oil content increased with the increasing amounts of irrigation in the treatments, in which the greatest oil contents were attained from the A_2WL_4 (48.33 and 55.10%) treatment in both years (Table 2). NI yielded the least oil content (33.70 and 36.10%). Asbagh et al (2009) reported that the oil content of the sunflower seeds varied between 34.3% and 39.1% in non-irrigated conditions and between 38.5 and 42.7% in irrigated conditions in different varieties. In this study, the results which demonstrated the oil percentage correspond to those of Asbagh et al (2009) and Sezen et al (2011a), who suggested that the oil percentage increased with the increased use of irrigation water. Results showed that although stress did not affect the oil percentage, it reduced the oil yield via severe reduction in grain yield. Both the irrigation interval and irrigation levels had a significant effect on the sunflower oil yield in both growing seasons (Table 2), hence it reveals the increase in oil yield with the increasing amount of irrigation water. The maximum average oil yield was obtained in A_2WL_4, followed by A_2WL_3, and the minimum oil yield was obtained from the NI treatment. According to Essiari et al (2014), the fatty acid composition is an important parameter in identifying the quality of the oils. The major fatty acids were oleic acid (84.13- 89.20%), linoleic acid (1.70-5.17%) palmitic acid (3.30-6.07%) and stearic acid (1.40-2.90%) in sunflower oils (Table 2). It was determined that there is a negative correlation between oleic acid and linoleic acid. Baldini et al (2002) suggested that there is a positive correlation between the amount of oleic acid and ET during the vegetative period of the plant. However, Asbagh et al (2009) observed an increase in the amount of linoleic and palmitic acid and a decrease in the amount of stearic acid and oleic acid with irrigation. It was found that there are significant linear relationships between the oleic acid and linoleic acid contents compared to evapotranspiration for 2010 and 2011 growing seasons (Figure 3a-b). The water stress significantly affects the content of unsaturated

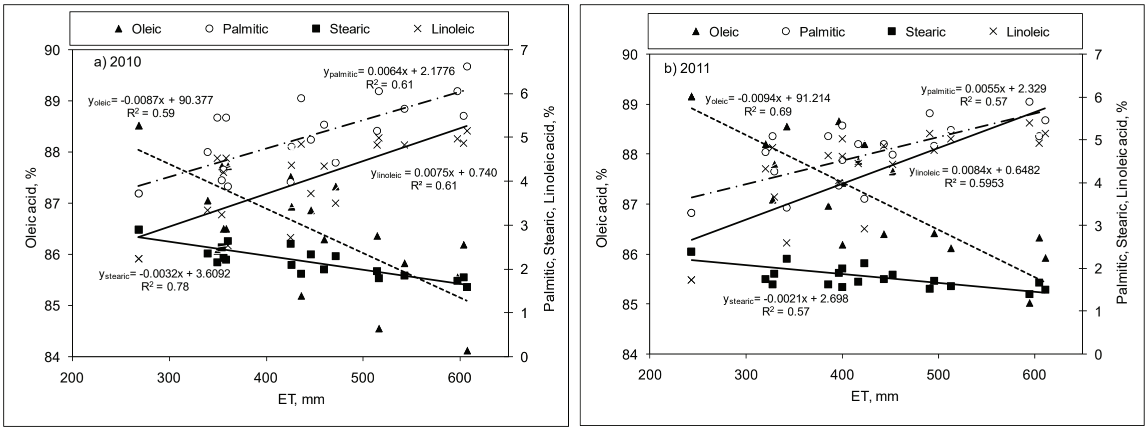

Figure 3 a, b- The interaction between ET and oleic, palmitic, linoleic and stearic acid contents for all treatments in 2010(a)-2011(b)

(oleic and linoleic acid) fatty acids. As ET increased oleic acid content decreased, while linoleic acid concentration increased in 2010 and 2011 growing seasons. In their study, Sezen et al (2011a) acquired similar results for sunflower grown under water stress conditions in Eastern Mediterranean part of Turkey.

3.8. Economical evaluation

In this study, the economical evaluation was performed by analyzing the results based on the means of the operation, investment and production costs for both years and the results are shown in Table 3. Within the scope of economical evaluation, the maximum return of 581 US$ ha^{-1} was obtained from the A_2WL_4 treatment. It was observed that lower irrigation levels caused a decrease in marginal return per irrigation interval. In the context of marginal return, a significant difference was found between the irrigation intervals and irrigation levels. The marginal return was increased in all irrigation intervals as the water supply increased. In the economic analyses, the marginal return and irrigation costs were compared and for the evaluations the marginal yield was calculated, which is the difference between yield from irrigated treatment and non-irrigated treatment. The results indicated that A_1WL_3 and A_1WL_4 in A_1, A_2WL_2, A_2WL_3, A_2WL_4 and A_2WL_5 in A_2, and A_3WL_3 and A_3WL_4 in A_3 were the economical treatments since they generated higher income over the irrigation cost. Among them A_2WL_4 was the most economical treatment and recommended. It was found that A_2WL_3 treatment is a plausible alternative in areas where access to irrigation water is expensive or less than demanded.

4. Conclusions

In this study, the effects of different irrigation strategies on the seed and oil yield, water use, WUE and IWUE in Çukurova region of Turkey throughout the sunflower growing seasons of 2010 and 2011 were analyzed in terms of amount and frequency. A_2WL_4 treatment presented the highest yield as 4758 and 4890 kg ha^{-1} respectively for both years. The oil contents were considerably affected depending on different irrigation intervals and levels. Furthermore, the results demonstrated that the WUE and IWUE values diminished depending on the increase in irrigation intervals. Lower WUE and IWUE were achieved by the means of the same irrigation level in A_1 and A_3 irrigation intervals compared to A_2 interval. It was determined that there is a significant linear relationship between sunflower seed and oil yield, and total ET, the oil content of sunflower as well as total ET in both of the experimental years. In the context of economic evaluation, the marginal return from the A_2WL_4 treatment under drip irrigation was determined to be logical in areas having no water scarcity. Under water scarcity conditions, however, it was determined that A_2WL_3 treatment may generate an acceptable marginal return. It has been thought that the results will help to adopt deficit irrigation method, by which net financial returns are enhanced.

In conclusion, it is suggested to apply A_2WL_4 treatment (cumulative pan evaporation: 50±5 mm, WL_4= 1.25) in drip irrigated sunflower production for the purpose of achieving a higher yield in Çukurova region of Turkey. For two consecutive experimental years, the total amount of irrigation water for suggested treatment (A_2WL_4) was found to be 530 mm and 546 mm, respectively, while the total amount of seasonal water use for A_2WL_4 was found to be 607 mm and 595 mm, respectively. The number of irrigation treatments varied from 8-9 in 2010, and 8-10 days in A_2 treatments.

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Table 3- The economical evaluation on sunflower for all treatments based on average of two years data

Treatments	*(1) Irrigation water (mm)*	*(2) Irrigation water (m^3 ha^{-1})*	*(3) Irrigation duration for the irrigation season (h)*	*(4) Labor cost for irrigation (\$ h^{-1})*	*(5) Total cost for irrigation labor (\$) (3x4)*	*(6) Water price (\$ m^{-3})*	*(7) Water cost (\$ ha^{-1}) (2x6)*	*(8) Irrigation system cost (\$ ha^{-1})*
A_1WL_1	231	2310	10.1	1.7	16.6	0.1	231	4446
A_1WL_2	343	3430	15.0	1.7	24.8	0.1	343	4446
A_1WL_3	456	4560	20.0	1.7	33.0	0.1	456	4446
A_1WL_4	571	5710	25.0	1.7	41.3	0.1	571	4446
A_1WL_5	231	2310	10.1	1.7	16.6	0.1	231	6983
A_1WL_6	343	3430	15.0	1.7	24.8	0.1	343	6983
A_2WL_1	217	2170	9.5	1.7	15.7	0.1	217	4446
A_2WL_2	323	3230	14.2	1.7	23.4	0.1	323	4446
A_2WL_3	430	4300	18.9	1.7	31.2	0.1	430	4446
A_2WL_4	538	5380	23.6	1.7	39.0	0.1	538	4446
A_2WL_5	217	2170	9.5	1.7	15.7	0.1	217	6983
A_2WL_6	323	3230	14.2	1.7	23.4	0.1	323	6983
A_3WL_1	210	2100	9.2	1.7	15.1	0.1	210	4446
A_2WL_2	313	3130	13.7	1.7	22.6	0.1	313	4446
A_3WL_3	417	4170	18.3	1.7	30.2	0.1	417	4446
A_3WL_4	575	5750	25.2	1.7	41.6	0.1	575	4446
A_3WL_5	210	2100	9.2	1.7	15.1	0.1	210	6983
A_3WL_6	313	3130	13.7	1.7	22.6	0.1	313	6983
NI	0	0	0.0	1.7	0.0	0.1	0	0

Treatments	*(9) Irrigation system cost for 1 ha (\$ h^{-1}) (8/6 years)*	*(10) Yearly cost for the irrigation system (\$ ha^{-1}) (5+7+9)*	*(11) Yield (kg ha^{-1})*	*(12) Marginal yield (kg ha^{-1})*	*(13) Sunflower sales price (\$ kg^{-1})*	*(14) Marginal return (\$ ha^{-1} year^{-1}) (12x13)*	*(15) Return (\$ ha^{-1} year^{-1}) (14-10)*
A_1WL_1	741	989	2882	1197	0.6	724	-265
A_1WL_2	741	1109	3315	1630	0.6	986	-123
A_1WL_3	741	1230	3867	2182	0.6	1320	90
A_1WL_4	741	1353	4318	2633	0.6	1593	239
A_1WL_5	1164	1411	3673	1988	0.6	1202	-209
A_1WL_6	1164	1532	3685	2000	0.6	1210	-322
A_2WL_1	741	974	3067	1382	0.6	836	-138
A_2WL_2	741	1087	3629	1944	0.6	1176	89
A_2WL_3	741	1202	4074	2389	0.6	1445	243
A_2WL_4	741	1318	4824	3139	0.6	1899	581
A_2WL_5	1164	1397	4054	2369	0.6	1433	37
A_2WL_6	1164	1510	4043	2358	0.6	1427	-84
A_3WL_1	741	966	2525	840	0.6	508	-458
A_2WL_2	741	1077	2874	1189	0.6	719	-357
A_3WL_3	741	1188	3688	2003	0.6	1212	23
A_3WL_4	741	1358	4080	2395	0.6	1449	91
A_3WL_5	1164	1389	3597	1912	0.6	1156	-233
A_3WL_6	1164	1499	3592	1907	0.6	1153	-346
NI	0	0	1685	0	0.6	0	0

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Effects of Leaf Surface Energy on Pesticidal Performance

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ABSTRACT

Surface energy is widely used in the industry to predict behavior of spray droplets on solid surfaces. The targets of pesticide applications which are used extensively in agricultural production are mainly plant leaf surfaces. Digitization of leaf surfaces to estimate the spread and adhesion of a pesticide application is an important approach in providing descriptive information. In this regards, from intensive agricultural products *Triticum aestivum* L., *Citrus sinensis, Fragaria ananassa, Vitis vinifera* L., *Cucumis sativus, Capsicum annuum* L. culture plants, *Elymus repens* and *Sinapis arvensis* from weeds were used to determine surface energy. The leaf surface energies were determined by evaluating the contact angles of the drips while obtained from surface tension and its components from known liquids pure water, diiodomethane and formamide liquids on the surface of the leaves according to five different methods. Wu and Equation of State methods have been found to give more accurate results than other methods. *Elymus repens* and *Triticum aestivum* L. plants among the statistically three significant grouped leaves were reduce the spreading and sticking of droplets applied on the leaves by providing a more spherical droplet formation. The *Fragaria ananassa* leaves have encouraged the higher surface energy that they have the spread of the drips on the leaf surface.

Keywords: Contact angle; Diiodomethane; Leaf; Spray; Surface tension; Wu

1. Introduction

It is predicted that, in spite of the increasing population, the consumption of projections of pesticide use will increase in order to obtain sufficient crops from declining agricultural areas. Researches related to pesticide application have mostly focused on pesticide formulations and the physical properties of liquids and leaves. As a result of the researches, it has been found out that various factors have an effect on the success of applications (spray), these are; epicuticular wax, epidermal cell structure, wax crystals, amount of wax, shape, composition (Wohlfahrt et al 2006; Puente & Baur 2011; Massinon & Lebeau 2012), chemical functional groups on the leaf surface, leaf roughness, leaf hairs, general shape of epidermal cells, cuticle folds, hairs (trichomes) (Wagner et al 2003), nanostructure of wax crystals (Khayet & Ferna´ndez 2012), enhanced wetting in some plant species with open trichome pattern caused by capillary action (Holloway 1970), lattice arrangement and structure of molecules (Brewer et al 1991; Wang et al 2014).

It is known that the application of liquid to different leaf surfaces results in different contact angles and spreading levels on the surfaces. This happens due to different surface energies produced by the molecular structure in the chemical structure of each leaf. The estimation of a study related to water adhesion is an easy and valuable tool that can be used to quantify the adhesion degree of a particular plant surface (Fernández et al 2014). Various different models have been developed in order to determine the surface energy of an object. Five of the commonly used methods were used in the experiments. The general information on these is briefly as follows.

The surface energy of a solid object is not a quantity that can be measured directly. It is determining the droplet contact angles applied to the solid surface as a function of the surface tension based on the evaluation of Young's equation (Equation 1) according to different models. It is based on the spread of the droplet surface tension on the target according to the size of the surface energy of the solid.

$$\gamma_2 = \gamma_{12} + \gamma_1 \cos\theta \tag{1}$$

It is generally based on the prediction that the surface energy of a solid γ_2 is equal to the sum of the surface tension of the liquid γ_1 and the interfacial tension θ which is derived from the cosine of the horizontal component of the droplet contact angle on the solid surface γ_{12}. Contact angle (θ) is the quantitative measurement of the wetting rate of a solid surface by a liquid. The other approaches foreseen in line with this basic principle are as follows;

According to Zisman, the surface energy of the solid matter is determined by the droplets' contact angles formed on the surface of the applied liquids. Here, the values corresponding to the cosine of the contact angle data (cos θ) are drawn in the form of surface tension of the liquids, and with a contact angle of 0°, the highest surface tension value for the liquid which will completely wet the solid is obtained by being arranged for $\cos\theta = 1$ ($\theta = 0^{o}$). This is considered equal to the surface energy of the solid (Zisman 1964).

In the Equation of State method, in order to calculate the surface free energy with the contact angle, the second unknown variable must be determined (Moy & Neumann 1987; Li & Neumann 1992).

$$\gamma_{12} = \gamma_1 + \gamma_2 - 2\sqrt{\gamma_1.\gamma_2}.e^{-\beta(\gamma_1-\gamma_2)^2} \tag{2}$$

$$\cos\theta = -1 + 2\sqrt{\frac{\gamma_2}{\gamma_1}}.e^{-\beta(\gamma_1-\gamma_2)^2} \tag{3}$$

The polar and dispersive parts of the surface tension are not taken into consideration. Experimentally, the value of 0.0001247 $(m^2\,mJ^{-1})^2$ is determined for the constant β (Hansen 2004; Krüss 2017).

OWRK and Fowkes models are based on the assumption of the sum of interfacial interactions dependent on the polar (^{p}) and dispersive (^{d}) properties of the solid measured with the measurement liquid (Fowkes 1964; Owens & Wendt 1969; Kaelble 1970; Rabel 1971).

$$\gamma_{12} = \gamma_1 + \gamma_2 - 2(\sqrt{{\gamma_1}^d.{\gamma_2}^d} + \sqrt{{\gamma_1}^p.{\gamma_2}^p}) \tag{4}$$

Since ${\gamma_2}^p$ and ${\gamma_2}^d$ are not known in the Equations 4 and 5, two liquids, one of which is polar and the other is dispersive, should be used for the solution. Water, formamide polar liquids, and diiodomethane are the most commonly used dispersive liquids.

Wu's method involves the approach that the use of the harmonic mean of the polar and dispersive components in determining the surface energy yields more reliable results compared to the geometric mean used in OWRK & Fowkes methods.

$$\gamma_{12} = \gamma_1 + \gamma_2 - 4\left(\frac{{\gamma_1}^d.{\gamma_2}^d}{{\gamma_1}^d + {\gamma_2}^d} + \frac{{\gamma_1}^p.{\gamma_2}^p}{{\gamma_1}^p + {\gamma_2}^p}\right) \tag{5}$$

Wu's method is mostly used for free surface energy calculations for objects with low surface free energy (up to 30-40 mJ m^{-2}) (Wu 1973).

In acid-based methods, the polar component is divided into acid and base components, and the Equation is written as:

$$\gamma_{12} = \gamma_1 + \gamma_2 - 2(\sqrt{{\gamma_1}^d.{\gamma_2}^d} + \sqrt{{\gamma_1}^+.{\gamma_2}^-} + \sqrt{{\gamma_1}^-.{\gamma_2}^+}) \tag{6}$$

In order to solve the Equation, at least three liquids with known properties are required. One of them is a fluid with dispersive properties (e.g., diiodomethane), the other is a bipolar fluid with polar properties (e.g., water, formamide) (Krüss 2017).

In the study, it is aimed to determine the reactions of some of the conventional products used in intensive farming against the spray applied in the agricultural struggle, as meaningful quantitative relations. Therefore, alteration of the droplets applied to the leaf surfaces based on the leaf types will be determined in terms of energy. On the other hand, the possibility of developing alternative approaches for the preparation of effective pesticide tank mixes arranged based on plant species has been examined by comparing the obtained data with the surface tensions of the application liquid.

2. Material and Methods

In order to determine the impacts of surface energy on pesticide applications, plants that are commonly produced and frequently exposed to pesticide applications were used in the experiments. *Triticum aestivum* L., *Citrus sinensis, Fragaria ananassa, Vitis vinifera* L., *Cucumis sativus, Capsicum annuum* L. cultivated plants *Elymus repens* and *Sinapis arvensis* weeds obtained from Cukurova University research (production) farms were placed in a pot to maintain the living features, and the leaves were cut and the necessary measurements were made in due course.

Pure water, 99% pure formamide and diiodomethane in the Middle East Technical University Central Laboratories are used in the contact angle, surface tension, interfacial tension and surface energy measurements that provide information on the surface properties affecting the liquid such as spread, wettability, absorption, surface tension, etc. In order not to affect the properties of other liquids, operations were carried out by drawing each liquid into three different syringes. "Sessile drop technique", which is the most commonly used technique on flat surfaces for contact angle measurements, was used. Since the shape and contact angle of droplets on solid surfaces are dependent on the liquid's effect on the surface tension, liquid surface tensions should be known in order to determine the surface energies of objects. For this, the optical contact angle and surface tension measurement device produced by KSV firm Attention Theta (Goniometer) was used, which analyzes the droplet shape based on time by saving the images of the droplet.

The leaves to be measured were separated from the plants from the cross sectional areas by about 1 cm^2 by means of scissors, and placed on the sample stage of the device by carefully sticking them onto the glass slide using double sided tape to prevent any deformations on the surface. The sample stage of the device can be easily adjusted forward-backward, up-down and right-left. The syringes, into which liquid was drawn, were attached to the Goniometer, a hanging droplet was formed at the tip of the needle, and was contacted with a leaf surface. Meanwhile, falling of droplet can also be monitored on the screen. Then, the right and left contact angles were started to be measured in real time with the software's detection of the droplet wall. At least five drops of the same liquid were measured and the angles of both sides of the contacting droplets were determined. Asymmetric measurements (when the difference between the angles of both sides were higher than five degrees) were removed from the data. The surface tension given in Table 1, and the contact angles and standard deviation values given in Table 2 and the average contact angles and standard errors obtained on the leaf surfaces that given in Table 3 are obtained in real time by opening the saved measurement images to analyze further through the use of statistical function.

The contact angles on the leaf surfaces, and the surface tension data of the liquid used were calculated in the Goniometer software, in accordance with the guidelines for surface energy calculation methods. The surface energy values of the plant leaves were obtained as in Table 4.

The statistical analyzes were carried out with version 18 of the SPSS package program. Differences between the plant species were evaluated according to Tukey multiple comparison test for 5% level of significance.

Table 1- Liquid surface tension parameters

Heavy phase	γ^{tot} *(mN m^{-1})*	γ^{d} *(mN m^{-1})*	γ^{p} *(mN m^{-1})*	γ^{+} *(mN m^{-1})*	γ^{-} *(mN m^{-1})*
Water	72.8	21.8	51.0	25.5	25.5
Diiodomethane	50.8	50.8	0.0	0.0	0.0
Formamide	58.0	39.0	19.0	2.3	39.6

Table 2- The contact angles and standard deviations of the liquids formed on the surfaces of the leaves

Method	*Crop*	*Water*	*SD*	*Diiodomethane*	*SD*	*Formamide*	*SD*
Contact angle (θ°)	*Citrus sinensis*	1 00.2^{b*}	5.9	62.2^{a}	7.6	69.2^{ab}	5.6
	Triticum aestivum L.	126.4^{c}	6.1	93.2^{b}	6.9	144.8^{c}	2.5
	Fragaria ananassa	85.8^{b}	6.3	56.2^{a}	11.7	59.8^{a}	6.3
	Vitis vinifera L.	109.0^{c}	2.5	67.8^{a}	6.8	90.6^{b}	10.2
	Cucumis sativus	94.6^{b}	8.4	58.4^{a}	10.6	56.6^{a}	6.0
	Capsicum annuum L.	87.0^{b}	11.7	53.8^{a}	6.1	80.9^{b}	7.9
	Elymus repens	150.2^{c}	8.0	97.8^{b}	8.5	138.2^{c}	3.2
	Sinapis arvensis	98.2^{b}	5.2	61.0^{a}	2.6	74.0^{ab}	1.2

*, the difference between the same letters is insignificant at the $P<0.05$ level

Table 3- The contact angles and standard errors that form on the leaf surfaces

Crop	*Mean* (θ°)	*SE*
Citrus sinensis	77.2^{ab}	4.6
Triticum aestivum L.	121.5^{c}	5.9
Fragaria ananassa	67.3^{a}	4.1
Vitis vinifera L.	89.1^{b}	4.8
Cucumis sativus	69.9^{ab}	5.1
Capsicum annuum L.	71.6^{ab}	4.2
Elymus repens	128.8^{c}	6.2
Sinapis arvensis	78.5^{ab}	4.2

*, the difference between the same letters is insignificant at the $P<0.05$ level

3. Results and Discussion

3.1. Contact angle measurements

The surface energy of an object depends on its ability to meet the requirements of the particular model selected for the analysis of the liquids that are deemed appropriate for use. In order to obtain more consistent results and make comparisons, the most preferred ones in the literature were selected from among the liquids meeting the specified conditions. The liquids used in the experiments and some of their properties are given in Table 1. It can be observed that water and formamide liquids have both dispersive and polar properties, and also charge values. It can be understood that the diiodomethan liquid is of a dispersive nature.

When the aim is to characterize the objects through the components of surface energy, conversion of contact angle data to surface energy values is utilized. The contact angles obtained from the leaf surfaces are given in Table 2 and Table 3. *Fragaria ananassa* when the lowest contact angle was obtained from the leaves, *Cucumis Sativus, Capsicum annuum* L, *Citrus sinensis ve Sinapis arvensis* the leaf surfaces formed higher contact angles. While the leaves *Triticum aestivum* L. and *Elymus repens* formed the surfaces that produced the highest contact angles, the leaves *Vitis vinifera* L. produced contact angles at intermediate values.

When the contact angles are examined based on the liquids, in Table 1, inversely proportional contact angles were obtained from the liquids with high dispersive components. While the diiodomethane liquid formed the smallest contact angle on the leaf surfaces, greater contact values proportional to the dispersive value were obtained from the formamide liquid. It has been determined that the

Table 4- Liquid surface energy parameters

Crop	*Method*	γ^{tot} *(mN m^{-1})*	γ^{d} *(mN m^{-1})*	γ^{p} *(mN m^{-1})*	γ^{+}	γ^{-}
Citrus sinensis	Acid-Base	27.721	27.745	-0.024	1.088	-0.011
	Equation of State	29.006	29.006			
	OWRK/Fowkes	30.039	29.312	0.727		
	Wu	33.017	30.190	2.827		
	Zisman	35.303				
Triticum aestivum L.	Acid-Base	-7.312	12.227	-19.539	-3.300	2.960
	Equation of State	8.426	8.426			
	OWRK/Fowkes	8.584	8.177	0.408		
	Wu	11.550	16.414	-4.864		
	Zisman	-12.114				
Elymus repens	Acid-Base	9.146	8.564	0.583	-1.581	-0.184
	Equation of State	5.980	5.980			
	OWRK/Fowkes	9.320	7.433	1.887		
	Wu	8.420	13.115	-4.695		
	Zisman	4.137				
Fragaria ananassa	Acid-Base	28.331	24.502	3.829	2.345	0.816
	Equation of State	33.340	33.340			
	OWRK/Fowkes	32.908	27.154	5.754		
	Wu	37.372	28.138	9.235		
	Zisman	24.326				
Vitis vinifera L.	Acid-Base	19.932	21.289	-1.357	-0.579	1.173
	Equation of State	20.888	20.888			
	OWRK/Fowkes	19.990	19.886	0.105		
	Wu	23.407	23.537	-0.130		
	Zisman	21.612				
Cucumis sativus	Acid-Base	18.399	23.432	-5.033	2.957	-0.851
	Equation of State	31.531	31.531			
	OWRK/Fowkes	31.158	28.019	3.139		
	Wu	35.405	28.821	6.584		
	Zisman	32.160				
Sinapis arvensis	Acid-Base	27.967	27.667	0.300	0.115	1.306
	Equation of State	27.866	27.866			
	OWRK/Fowkes	27.855	26.932	0.923		
	Wu	31.275	28.559	2.715		
	Zisman	30.500				
Capsicum annuum L.	Acid-Base	21.274	30.701	-9.427	-1.145	4.116
	Equation of State	30.540	30.540			
	OWRK/Fowkes	29.550	26.182	3.369		
	Wu	33.598	27.505	6.092		
	Zisman	18.298				

water which has a high polar character formed the highest contact angle on the leaf surfaces. In Table 2 and Table 3, the standard deviation and standard error values of the contact angles are also given. It can be understood that the results between the data from the standard error values showing the standard deviation obtained from the same leaf surface and the variation (alteration) in the distribution of the average are close. The small standard deviation values in the contact angles also indicate the flatness and homogeneity of the surfaces.

3.2. Surface energy measurements

Characterization of plants through a parameter that is a combination of physicochemical properties of the leaves, will provide guiding suggestions for pesticide applications based on a condition that is created accordingly. In order to obtain an effective pesticide performance, it is required that the producers recognize the plant to which they will apply pesticide, and develop appropriate spray alternatives.

The size of the surface energy the molecular gravity that the leaf has while the droplet spreads on it in comparison with the surface tension the molecular gravity that liquid possesses is the determinant. The surface energy values obtained according to different models from the same leaf surfaces are shown in Table 4. These methods include single parameter calculations as well as surface energy values determined by two and three-component processes. Thus, different results have appeared due to different approaches in determining the leaf surface energies. In this context, more than one quantity is obtained instead of a single value describing the leaf surface. In Table 4, the fact that some results have negative values show that leaf surface tensions are less than liquid surface tensions. Also, the leaves forming low contact angles in the second level based on the Acid-Base model formed *Cucumis sativus* 18.399 mN m^{-1} surface energy and the mean contact angle of 69.9^0. While it was expected that a lower contact would be formed with *Citrus sinensis* 27.721 mN m^{-1}surface energy, it has been specified that higher contact angles such as 77.2^0 were formed. Namely, as a result, *Cucumis sativus* with lower surface energy spread the droplet further on its surface when compared with *Citrus sinensis*. Since this contradicts the definition of surface energy, it should be accepted as an erroneous approach.

In order to avoid obtaining incorrect results from data analysis, it is necessary to select the correct model and make the evaluations accordingly. In this context, when the results should be directly proportional for the conversion of contact angles to surface energy, with the exception of models the Wu's and Equation of state methods, have produced antiparallel results with the contact angles. It is understood that other methods outside these two models produce erroneous results. It has been reported in the literature that both methods give accurate results for low surface energy measurements (Wu 1973; Moy & Neumann 1987).

In the statistical analysis for Table 3 and Table 4 carried out, *Fragaria ananassa* formed the first independent group as the leaves with highest surface energy from among the types of plants separated into three different groups based on their surface energies and contact angles. *Cucumis Sativus, Capsicum annuum* L. *and Sinapis arvensis* leaves with lower surface energies, produced results that can be evaluated statistically as both the first and second group. Statistically, *Vitis vinifera* L. leaves were ranked as the second independent group with the surface energies they possessed. *Elymus repens* and *Triticum aestivum* L. were the third group as the leaves with the lowest surface energies.

4. Conclusions

In Table 4, it can be observed that specified surface energy values differ based on the selected method and the fluid used. In order to make an evaluation regarding which of the numeric value(s) describe the plants correctly, the results produced by the methods should be examined based on the surface energy definitions. Determining the impact rates of the changes in spray behavior applied to an object based on the model that produces the best results will make the results more sensible. Otherwise, the analyzes to be carried out will lead to incorrect evaluations. On the other hand, it would be more appropriate to refer to a surface energy value range rather than a single constant value to make precise judgments based on the surface energies. The use of surface energy values with a certain numerical range instead of constant quantitative data for defining objects will be a more correct approach because of the definition of surface energy, which is not a definite case. Accordingly, the surface energies of the leaves and the application liquid surface tension value should be compared, since the application liquid with low surface tension can help droplets to more easily attach, spread and adhere to the leaf surfaces for leaves with low surface energy, it should be taken into consideration as an important factor for its use. Since the plant leaves with high surface energy will spread to the surface further, the use of liquids with higher surface tension will produce better results in preventing the leakage of the application fluid.

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Comparisons of Physical and Chemical Characteristics of Eggs Obtained using Hens Reared in Deep Litter and Free-Range Systems

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ABSTRACT

This study was designed to compare the physical and chemical qualities of eggs obtained using hens reared in free-range and deep-litter systems. A total of 300 Lohmann Brown hens (150 for each housing system) were used. In the free-range system, 4 m^2 grazing area was allocated for each hen. The hens were taken into layer house at 16 weeks of age. Hen egg production reached up to 50%, produced eggs were randomly sampled once every 4 weeks until 52 weeks of age and physical characteristics of eggs from each housing system were measured. Results revealed that there was no significant difference in egg shell color, egg weight, breaking strength, shell thickness, shape index, specific gravity, yolk color, albumen index, haugh unit, meat and blood spots of eggs from two housing systems. However, there was a significant difference in yolk index. It was determined that housing systems effected the chemical content of the egg and the eggs obtained from free-range system were significantly richer in essential amino acids, vitamin D$_3$ and biotin.

Keywords: Hens; Free-range system; Deep-litter systems; Egg quality

1. Introduction

One of the biggest challenges of human beings in today's world is to maintain healthy diet, which is strongly associated with life quality. A key component of maintaining the healthy diet is to consume adequate and balanced amount of animal- and plant-based products. One of the excellent source of animal-based proteins is egg. This is because egg proteins are highly digestible (98%), high in biological value (94%), and rich in essential amino acids (Altan 2015).

After the consideration of animal rights in egg production systems has started to gain importance, different alternative housing systems with a consciousness of food safety were developed in order to produce high quality eggs. This goes along with consumers' demands that they prefer to consume eggs produced using such systems (Anderson 2009).

The genotype of the hens (Hanusová et al 2015) and the housing systems used in production (Doley et al 2010; Nistor et al 2014; Angelovicova et al 2014; Yang et al 2014; Nistor et al 2015) significantly affect the egg quality. It has been observed that different hen genotypes used in production react differently to different housing systems (Leyendecker et al 2001).

However, in various studies, findings support the fact that eggs produced from hens reared in free-range system are higher in quality (Krawczyk & Calik 2006; Senčic et al 2006; Krawczyk & Gornowicz 2010; Yang et al 2014; Nistor et al 2014), compared to other systems. This is mainly attributed to the fact that hens reared in free-range system are exposed to direct sunlight, have plenty of space for movement, and they can access to green grass and different food resources in open space.

Since alternative production systems are important in terms of both variation and animal welfare in egg production, it is required to study and put forward the effects of these production systems on productivity and quality of eggs in detail. Although various studies exist on this manner, they are not adequate to piece together in order to discover the best system for the production of eggs with high quality. In this study, it is aimed to determine the physical and chemical qualities of eggs obtained from hens reared in deep litter and free-range systems and pointing out the differences between both housing systems.

2. Material and Methods

In the study, a total of 300 hens consisting of 150 Lohmann Brown hybrids in each of deep-litter and free-range system groups were used. The study was carried out with a research henhouse in the city of Ordu, which is located in the Black Sea Coast of Turkey. In the free-range system, out of the henhouse, a green area of 4 m^2 was allocated for each hen. Five hens were placed in each meter square into henhouse. Chicks were transferred following hatchery to an environmentally controlled growth house and were exposed to 10 h daily lighting until the age of 16 weeks. The hens were placed in henhouse with windows at 16 weeks of age. After the 18 weeks of ages, the lighting period was increased for 1 hour periodically in every week until the daily lighting reached 16 h. Once it reached 16 h, no more increase was made in the daily lighting and it was stabilized at 16 h. Water and feed were provided add libitum in both rearing systems throughout the experiment. The hens were fed with 1. period layer diets according to NRC (1994).

2.1. The physical quality characteristics of the egg

The physical characteristics were determined in eggs once every 4 weeks from onwards 50% production age of hens, and totally 160 eggs were used in each group. For this purpose, the eggs were brought into the laboratory and kept at room temperature for 24 hours. Afterwards, the following characteristics were determined.

2.1.1. Egg weight

It was determined by weighting with a scale at 0.01 g sensitivity.

2.1.2. Shape index

It was determined by using a digital caliper to measure width and height of the egg.

2.1.3. Specific gravity (g cm^{-3})

It was calculated with Equation 1.

Specific gravity (g cm^{-3}): Weight in air (g) / Weight in air (g) - Weight in pure water (g) (1)

2.1.4. Shell breaking strength

It was determined by using a shell breaking strength measurement tool in (kg cm^{-2}).

2.1.5. Shell color

It was determined by using the shell color scale developed by Hy-Line Company.

2.1.6. Eggshell thickness

It was measured in mm by using a micrometer.

2.1.7. Albumen index

It was calculated with Equation 2.

Albumen index: Height of albumen (mm) / Average of length and width of albumen (mm) * 100 (2)

2.1.8. Yolk index

It was calculated with Equation 3.

Yolk Index: Egg yolk height (mm) / Egg yolk diameter (mm) * 100 (3)

2.1.9. Haugh unit

It was calculated with Equation 4.

Haugh unit: $100 \text{ Log } (H + 7.57 - 1.7\ G^{0.37})$ (Haugh 1937). (4)

Where; H, albumen height (mm); G, egg weight (g)

2.1.10. Meat and blood spots

Eggs having meat and blood spots were counted and expressed as %.

2.1.11. Yolk color

It was determined by Roche color scale with 15 yellow color shades.

2.2. Chemical quality characteristics

The chemical analyses of the eggs were done in Food Institute Laboratory of Marmara Research Center. For this purpose, a total of 120 eggs, with 60 eggs from each group were used. The tested eggs were obtained from hens which are 52 weeks old. The chemical analyses targeted in this study were total energy (Atwater method), A, E, B_1, B_2, B_6, folic acid, niacin, B_5, B_7, K_2, D_3, B_{12} vitamins (HPLC-FLD method), alanine, aspartic acid, glutamic acid, serine, glycine, histidine, arginine, threonine, proline, tyrosine, valine, methionine, leucine, isoleucine, phenylalanine and lysine amino acids (UFLC-UV method), omega-3 and omega-6 oil acids (IUPAC IID 19 method), selenium and cholesterol analyses (Chromatography method) were done.

2.3. Statistical analysis

For all traits taken into account in the study, the control of normal distribution was done by using Kolmogorov-Smirnov test. The effects of group, time (linear, quadratic and cubic), and interaction on the internal and external egg quality characteristics were analyzed with MIXED procedure of the SAS software. While the time effects were significant for all mentioned characteristics, the age*group interaction effects were found to be statistically insignificant. For this reason, the time effect was removed from the model, and only the groups were compared for all traits. T-test was used in the evaluation of the traits which fulfill the assumptions. Nonparametric data were analyzed by Mann-Whitney test. For the data expressed as rates and %, angle transformation was carried out. Data were analyzed with Minitab 16 software (Anonymous 2010).

3. Results and Discussion

Research findings relating external quality characteristics have been presented in Table 1, and those concerning internal quality characteristics are presented in Table 2. It was found that there is not a difference between housing systems in terms of the researched characteristics of egg shell color, weight, breaking strength, shell thickness, shape index, specific gravity, yolk color, albumen index, haugh unit, meat and blood spot proportion ($P>0.05$). On the other hand, it was found that there is a significant difference in terms of yolk index between the systems ($P<0.05$). The yolk index of eggs obtained from hens reared in free-range system was found to be higher than those reared in deep litter system.

Chemical analysis results of the eggs produced through free-range and deep litter systems are given in Table 3. Differences occurred in the food

Table 1- External quality characteristics of eggs

Groups		*Shell color*	*Egg weight (g)*	*Breaking strength (kg cm⁻²)*	*Shell thickness (mm)*	*Shape index*	*Specific gravity (kg cm⁻³)*
	n	*Median*	$\overline{X} \pm S_{\overline{x}}$	$\overline{X} \pm S_{\overline{x}}$	$\overline{X} \pm S_{\overline{x}}$	$\overline{X} \pm S_{\overline{x}}$	$\overline{X} \pm S_{\overline{x}}$
Free-Range	160	90	61.907±0.549	2.858±0.095	0.377±0.002	78.792±0.344	1.085±0.0005
Deep-Litter	160	90	61.358±0.655	2.813±0.108	0.378±0.003	78.425±0.326	1.086±0.0006
P		0.337	0.522	0.754	0.722	0.457	0.330

Table 2- Internal quality characteristics of eggs

Groups		*Yolk color*	*Albumen index*	*Haugh unit*	*Yolk index*	*Meat-blood spot range (%)*
	n	*Median*	$\overline{X} \pm S_{\overline{x}}$	$\overline{X} \pm S_{\overline{x}}$	$\overline{X} \pm S_{\overline{x}}$	-
Free-Range	160	13	11.313±0.225	91.056±0.720	49.074±0.255	47.570±3.680
Deep-Litter	160	13	10.696±0.278	89.092±0.992	48.124±0.338	40.770±4.330
P		0.898	0.083	0.101	0.023*	0.234

material composition of the eggs produced. These differences can be attributed to the fact that 1) hens reared in free-range system are more dynamic compared to those reared in deep litter system, 2) hens reared in free-range system are exposed to direct sunlight and 3) hens reared in free-range system have access to reach other food sources including green grass.

Table 3- Findings related to chemical analysis of eggs

Chemical analysis	*Unit*	*Free-range systems*	*Deep-litter systems*	*P value*
Gross energy	Kcal 100 g^{-1}	133.00±3.840	130.00±3.750	0.606
Moisture	g 100 g^{-1}	76.73±2.220	78.23±2.260	0.660
Ash	g 100 g^{-1}	0.88±0.0250	0.86±0.024	0.603
Crude protein	g 100 g^{-1}	12.19±0.352	11.50±0.332	0.227
Carbohydrate	g 100 g^{-1}	1.53±0.044	0.09±0.002	0.000
Lipid	g 100 g^{-1}	8.67±0.250	9.32±0.269	0.152
Cholesterol	mg 100 g^{-1}	457.16±13.200	503.69±14.500	0.077
L-Alanine	mg 100 g^{-1}	450±13.000	396±11.400	0.036
Glycine	mg 100 g^{-1}	499±14.400	464±13.400	0.150
L-Valine	mg 100 g^{-1}	900±26.000	750±21.700	0.011
L-Leucine	mg 100 g^{-1}	1142±33.000	932±26.900	0.008
L-Isoleucine	mg 100 g^{-1}	733±21.200	593±17.100	0.007
L-Threonine	mg 100 g^{-1}	620±17.900	812±23.400	0.003
L-Serine	mg 100 g^{-1}	771±22.300	1006±29.000	0.003
L-Proline	mg 100 g^{-1}	590±17.000	458±13.200	0.004
L-Arginine	mg 100 g^{-1}	176±5.080	524±15.100	0.000
L-Aspartic acid	mg 100 g^{-1}	289±8.340	861±24.900	0.000
L-Methionine	mg 100 g^{-1}	510±14.700	462±13.300	0.073
L-Glutamic acid	mg 100 g^{-1}	782±22.600	1529±44.100	0.000
L-Phenylalanine	mg 100 g^{-1}	780±22.500	634±18.300	0.007
L-Lysine	mg 100 g^{-1}	683±19.700	1223±35.300	0.000
L-Histidine	mg 100 g^{-1}	182±5.250	293±8.460	0.000
L-Tyrosine	mg 100 g^{-1}	543±15.700	491±14.200	0.070
Se (Selenium)	mg 100 g^{-1}	0.281±0.008	0.406±0.012	0.001
Vitamin B_5 (Pantothenic acid)	mg 100 g^{-1}	2.53±0.073	2.52±0.072	0.927
Vitamin B_7 (Biotin)	µg 100 g^{-1}	27±0.780	3.60±0.100	0.000
Omega-6 fatty acids	g 100 g^{-1}	2.38±0.069	2.29±0.066	0.396
Omega-3 fatty acids	g 100 g^{-1}	0.14±0.004	0.13±0.003	0.144
Vitamin K_2	mg 100 g^{-1}	9.69±0.28	11.16±0.320	0.026
Vitamin D_3 (Cholecalciferol)	µg 100 g^{-1}	2.20±0.063	0.93±0.0260	0.000
VitaminA (Retinol,beta carotene)	µg 100 g^{-1}	81.46±2.35	133.5±3.850	0.000
VitaminB_{12} (Cyanocobalamin)	µg 100 g^{-1}	0.75±0.022	0.78±0.023	0.391
Vitamin E (Alfa tocoferol)	mg 100 g^{-1}	2.10±0.061	5.51±0.160	0.000
Vitamin B_1 (Thiamin)	mg 100 g^{-1}	0.074±0.002	0.060±0.002	0.007
Vitamin B_2 (Riboflavin)	mg 100 g^{-1}	0.29±0.008	0.26±0.007	0.056
Vitamin B_6	mg 100 g^{-1}	0.100±0.003	0.070±0.002	0.001
Folic acid	µg 100 g^{-1}	28±0.810	31±0.900	0.068
Niacin	mg 100 g^{-1}	0.067±0.002	0.067±0.003	1.000

Breaking strength was not affected by the housing systems. While this finding shows similarity to those reported by Angelovičová et al (2014) and Clerici et al (2006), it contradicts with findings reported by Torges & Matthes (1975), Krawczyk & Calik (2006), Hidalgo et al (2008), Krowczyk & Gornowicz (2010). These differences may be due to genotype and breeding conditions.

In this study, it has been determined that the housing systems do not affect egg weight. However, Doley et al (2010) have found that egg weight is higher in deep-litter system, and Pavlovski et al (1992) have pointed out that egg weights differ in cage, deep-litter and free-range systems. Lewko & Gornowicz (2011) have found that egg weight is higher cage than litter and free-range systems. In another study, eggs in free-range system have been found to be heavier than those in cage system (Senčic & Butko 2006), whereas Torges & Matthes (1975), Pavlovski et al (2004), Clerici et al (2006), Samiullah et al (2014) and Wegner (1982) have reported that eggs produced in free-range system are lighter that those produced in cage system. These discrepancies in different studies could arise from the fact that the free-range system have not reached a standard structure like the other systems, which result in egg production with different quality parameters.

In the present study, both housing systems did not affect egg shell thickness. This finding disagrees with the reports made by Pavlovski et al (2001), Senčic et al (2006), Angelovičová et al (2014), Yang et al (2014), and Krowczyk & Gornowicz (2010). On the other hand, Samiullah et al (2014) that have reported Shell thickness of eggs obtained from hens reared through cage system are higher than those obtained from hens reared in free-range system. In the present study, similar results were obtained in terms of specific gravity. It is thought that, this is caused by the fact that the egg shell thicknesses in both systems were similar. Ozcelik (2002) reported that there is an important relationship between egg weight and specific gravity, shell weight, shell thickness, specific gravity and shell weight and shell thicknesses in quail eggs.

There is not a difference between the two housing systems in terms of egg shape index. This was an expected result because the egg shape is determined in the magnum section of the egg canal and genotype is more effective on this than environmental factors. This finding agree with those reported by Lewko & Gornowicz (2011). However, Pavlovski et al (2004), Senčic et al (2006), Sekeroglu et al (2010) have reported that housing systems are effective on shape index.

No difference was found between the housing systems in terms of yolk color. However, this finding contradicts with Torges & Matthes (1975), Torges et al (1976), Pavlovski et al (2001), Senčic et al (2006), Senčic & Butko (2006), Lewko & Gornowicz (2011), Galis et al (2012) who reported that housing systems are effective on yolk color. This might be attributed to the fact that adequate amount of color pigments (Xanthophylls and Canthaxanthin) were included in the feed used for both systems in the present study. In case that the feed given to hens is poor in these substances, it is an expected outcome that the yolks of the eggs fed with additional green grass is yellower.

In the present study, the housing systems showed similarity in terms of albumen index and haugh unit. This agrees with the findings of Senčic et al (2006) who reported that eggs produced using hens reared in cage and free-range systems show similar characteristics in terms of albumen index and haugh unit. In a similar study, Samiullah et al (2004) have reported that albumen height and haugh unit are higher in cage systems compared to free-range system. In a study comparing eggs produced through cage, aviary and free-range systems, the best albumen quality was reached in eggs produced with free-range systems Pavlovski et al (2001). Pavlovski et al (2004) stated that haugh unit is lower in eggs produced through deep litter system when eggs produced through cage and free-range systems are compared. Dikmen et al (2017) have reported that eggs in the free-range system were better quality than eggs from convencional-cage and enriched-cage systems. Lewko & Gornowicz (2011) stated that was not differences in terms of haugh unit among litter, cage and free-range housing systems.

Eggs produced through free-range systems have a higher value in terms of yolk index compared to those produced through deep litter system. This might be due to the lower moisture leve of eggs produced through free-range system. No literature has been found on the effects of housing systems on yolk index. In their study comparing eggs produced through cage and free-range systems, Senčic et al (2006) reported that there is no difference in terms of yolk index.

It was found that the housing systems did not have an effect on meat and blood spots. However, it was observed that the proportions of meat and blood spots were rather high in both systems. Both genetic factors and environmental factors are effective on meat and blood spots. Hence, pointing out that heredity and other environmental factors are effective on the formation of meat and blood spots. Lerner et al (1951) have reported that the degree of heredity is approximately 0.5. In the literature, adequate information has not been found on the effects of housing systems on meat and blood spots.

Antioxidants such as vitamin A and vitamin E have been found to be lower in free-range hens because they find more opportunities for movement.

It was observed that the eggs produced by hens reared in free-range systems contain a higher level of vitamin D_3. This is attributed to the fact that hens are exposed to direct sunlight in this system. This vitamin is known to be effective especially on bone development and human psychology.

It was determined that eggs produced by hens reared in free-range system had a higher value in terms of vitamin B_7 (Biotin). In addition to taking part in oil, protein and carbohydrate metabolisms as a coenzyme, Biotin also plays important role on bone marrow and nerve tissues, hair and nails.

It was observed that there was no significant difference between the free-range and deep litter system in terms of cholesterol. Similar result was also observed by Torges et al (1976) who pointed out that there is no difference between eggs produced through free-range, deep litter and cage systems in terms of total cholesterol. Nistor et al (2014) have reported that the protein ratio is 10.35%, 9.97%; dry matter content is 23.37%, 22.96% and the oil ratio in yolks is higher in eggs produced through conventional cage system than free-range eggs, respectively. Radu-Rusu et al (2014) have reported that the total oil amount is 11.40 g 100 g^{-1}, 10.78 g 100 g^{-1}, cholesterol amount is 211 mg 60 g^{-1}, 202 mg 60 g^{-1}, total energy amount is 0.36 MJ egg^{-1} and 0.35 MJ egg^{-1} for eggs produced through cage and free-range systems, respectively. Galis et al (2012) have reported in a study comparing eggs produced through organic, free-range, aviary and cage systems that the protein and water ratio is highest in free range system eggs, ash ratio is lowest in cage system eggs and highest in organic system eggs.

It has been determined in this study that valine, leucine, isoleucine, methionine and phenylalanine are rich in the free-system and lysine and threonine are rich in the deep-litter system which are essential for humans. Other amino acids contents were similar in free-range and deep-litter systems. Küçükyılmaz et al (2012) have reported that eggs produced in the organic system were poor in yolk omega-3 content when compared to eggs laid by hens reared in the conventional system.

4. Conclusions

In this present study, it was determined that the free-range and deep litter systems are effective on egg quality characteristics. The aminoacid compositions of eggs are significantly affected by the housing systems; more importantly, eggs produced through free-range system were observed to be richer in essential amino acids, compared to deep litter systems. In general, since eggs produced through free-range system had more Vitamin D_3, Vitamin 7 (Biotin) and less total cholesterol, they were determined to be higher in quality compared to deep litter system eggs. Thus, it has been concluded that preferring eggs produced trough free-range systems would be important for human health, especially in regions where humans expose to less sunlight.

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Growth and Mortality Rates of *Cornu aspersum*: Organic Snail Culture System, Black Sea Region

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ABSTRACT

The study was aimed to examine a snail organic culture system and describe the cultivation properties of *Cornu aspersum.* The environmental parameters of the culture system and their effects on the growth and mortality rates of *C. aspersum* were determined between November 2014-October 2015. Snails were fed *Spinacia sp*. (spinach), *Urtica sp.* (nettles), Brassica *oleracea sp* (cabbage) and formulated diet. The feeding and growth rates increased with increasing temperature. Shell height growth rate was the highest in spring while the live weight growth rate was the highest in summer. Mortality rate of the baby snail was higher between November 2014 and May 2015 due to stress conditions such as handling and varying temperatures during their first stages of life. High mortality observed in adults could be associated with the spawning activity of the matured snails that caused physiological exhaustion. The result showed that the best culture cycle for *C. aspersum* was from spring to autumn in Black Sea region and in order to prevent post-reproductive mortality, snails reached to marketable size should be harvested.

Keyword: Land snail; Ecological culture; Extensive production

1. Introduction

Cornu aspersum (synonym: *Helix aspersa*), dominating the world market, are quite abundant in Turkey because of the topographical structure, favorable weather and environmental conditions of the country. Snail exporting is important, since the contribution to the maintenance of economic growth for Turkey (TUIK 2017). However, the snail production is only based on the gathering individuals from wild populations. On the other hand, overharvesting, human impacts and climate change have been affecting the natural snail population and it decreased in the last decade (FAO/Fishstatj 2018) in Turkey. Therefore, snail farming is necessary to prevent overexploitation and ensure sustainable production for the country.

Appropriate farming system for any given species provides optimal living conditions, supports growth and survival, minimizes risks and optimizes production. Humidity, temperature and feeding are vital factors to control growth and survival for heliciculture. Snails aestivate if the temperature is >30 °C and hibernate if the temperature is <5 °C (Cobbinah et al 2008). Growth of snails is mainly determined by genetic factors, although it is influenced by many other parameters such as stocking density (Dupont-Nivet et al 2000), environmental conditions (Garcia et al 2006), the management and sexual maturation. Reproduction activity could affect the life history of snails. High reproductive activity drains somatic energy reserves (e.g. carbohydrate and lipid) and limits the energy available for biochemical systems. Thus, the cost of reproduction compromises immune function, decreases protection against stress and reduces adult survivorship (Harshman & Zera 2007). In addition, offspring's mortality is strongly affected by adverse environmental conditions. Adverse environmental conditions cause to increase maintenance costs under adverse environmental conditions and affects survival during early development stage of the gastropoda (Diederich & Pechenik 2013).

Generally, three snail culture systems are described; intensive (indoor), semi-intensive (indoor/outdoor-mixed) and extensive (outdoor) cultures (Cobbinah et al 2008). Extensive system involves the breeding site which has a protection from the wind, a sprinkler system to keep the substrate moist and troughs made from wood or building blocks covered with plastic netting to hinder predators, requiring minimal financial input (Bryant 1994). Extensive snail farming can be certified as ecological and organic if the soil's conditions and the management are appropriate to the principles of IFOAM (1998). The key principle relates to the integration of wildlife, habitats and farming is the principle of ecology (Toader-Williams & Golubkina 2009). Begg (2009) also detailed organic principles for snail farming.

C. aspersum, frequently used in snail farming since their high reproductive capacity, can adapt to every climatic and farming condition (Avagnina 2012). It is clear that the future of *C. aspersum* farming has an interesting lucrative potential in Turkey. In this study, we investigated the culture of one-month-old snails in an extensive system according to Begg's organic principles (2009) in Black sea region. It was aimed to test workability and profitability of the system for the Black sea region, to describe the properties of *C. aspersum* farming and reveal practices of heliciculture.

2. Material and Methods

One-month-old snails with a mean shell height of 6.68±0.06 mm and a mean weight of 0.11±0.00 g were used in an extensive culture area located at the Scientific Research Center (SUBITAM) of Sinop University between November 2014-October 2015.

2.1. Study organism

Cornu aspersum (synonym: *Helix aspersa*), a hermaphroditic species, is a terrestrial pulmonate gastropod mollusc. The formation of a thickening and recurving lip at the edge of shell aperture indicates maturation (shell heigh>27 mm) (Daguzan 1982; Begg 2003). The reproductive behaviour involves that snails gets in touch with their tentacles, solidified dart comes out, the genital vebt pushed out and copulation happens (Avagnina, 2012).

2.2. Establishment of snail culture area

Extensive snail culture system was examined because of its suitability for the Black Sea regional climate. The culture system was maintained according to Begg's organic heliciculture principles (2009) which includes that a) crops are planted without the use of synthetic fertilizers, b) no chemicals are used in the snail fields, c) crops are planted densely to help prevent weed growth, d) finished crops are ploughed back into the ground as 'green manure' crops, e) physical controls are maintained for unwanted weeds and pests, f) ecological benefits of natural sunlight, organic soil.

110 square meters' area was fenced with a galvanized iron as the perimeter fence with a depth of 35-40 cm to the bottom and supported by iron posts. The sprinkler irrigation system, connected to water tanks filled with tap water, was established to provide water equitably to the production area. The culture area was divided into three pieces as parallels (B1, B2 and B3) with mesh fences of 9 m length and 1 m width and included 80 cm pathways between experimental fields (Figure 1). The downward

facing flaps and 4 pieces electric copper wires with a diameter of 0.80 mm (12 watts cm^2) were integrated to the system to prevent snails from climbing up the sides of the fencing. 8 slight slope wooden boards (50 cm x 50 cm) were placed in each field for feeding and establishing shelter area for snails. The culture area was covered with greenhouse nylon until May 2015 due to prevent harsh winter conditions. The culture area was covered with a bird protection mesh after removing greenhouse nylon. The roof pitch of the system was oriented according to prevailing wind directions.

Soil in the experimental culture area was analyzed prior to the study. Sandy humus soil was added for improving water holding capacity and organic matter content of the culture area since the soil texture was mostly clayey. pH, organic matter and moisture contents of the soil then were determined.

After excavating soil, three preferred plant types were planted as a row of *Spinacia sp*. (spinach), *urtica sp.* (nettles) and *Brassica oleracea sp* (cabbage) to each experimental field in order to determine the most preferred plants. Plants development was monitored and unwanted plants were removed from the experimental areas.

Artificial diet was prepared according to the organic feed rules (Blair 2008). The formulation and biochemical composition of the diet were given in Table 1. Feed was supplied every other day on the wooden boards during sunset time (between 5 and 8 pm).

Table 1- Ingredient and biochemical composition of diet

Diet formulation		*Biochemical composition of diet (based dry-weight, %)*	
Soybean flour	16	Protein	33.33
Corn gluten	11	Lipid	3.85
Wheat flour	28	Carbohydrate	22.26
Canola oil	3	Ash	40.56
Dicalciu phosphate	5	Moisture	3.85
Pectin	1		
Limestone	33		
Sodium chloride	0.5		
Vitamin premix	1.5		
Mineral premix	1		

2.3. Environmental parameters

Temperature and humidity were measured every morning (9:00 am) and sunset time from inside and outside of the culture area by TFA 5013 (electronic equipment). Natural ventilation was supplied by opening door and window of the greenhouse when temperature increased inside the culture area. Supplemental irrigation was also supplied as needed before sunset time when moisture was deficient.

2.4. Growth and mortality rate

300 snails were placed in one square meter of unit (separated with wooden block) in each field (B1, B2 and B3). Then the unit was enlarged by one square meter for every month with increasing growth. One hundred snails was randomly sampled from each field (B1, B2, B3) for the biometric parameters in every week until June, then sampling were carried out every two weeks period. Live weight (total weight of snail) was measured by weighing live animals to the nearest 0.001 g and shell height was measured to the nearest 0.1 mm with a caliper (Figure 1).

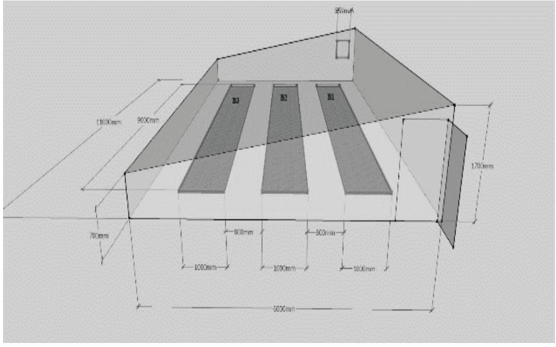

Figure 1- Schematic drawing of the organic snail culture system

Weekly shell height and live weight were calculated on a monthly base to allow for a clear pattern in the growth rates. Monthly shell height growth rate (SHGR %) and live weight growth rate (LWGR %) were calculated from the following formulate;

$$\text{SHGR (\%)} = [(L_2 - L_1) / L_1] \times 100 \qquad (1)$$

LWGR (%)= $[(W_2-W_1) / W_1] \times 100$ (2)

Where; L_1 and L_2 are the mean shell height and W_1 and W_2 are the mean live weight in a month.

The mortality rate was not determined before May due to fragile shells. The first mortality rate was determined in May 2015 by subtracting snails in May from the starting number of snails. After May, empty snail shells were counted and removed to determine mortality rate and followed monthly.

Mortality rate (%)= $100 (N_t - N_0)$ (3)

Where; N_t is the number of dead snails removed after t time and N_0 is the number of snail at the beginning of the experiment.

2.5. Statistical analyses

Data were analyzed for significant differences in means using ANOVA's, with significance levels set at P<0.05 and the normality of the variation of data was verified using the software program MINITAB 16 software. The variability of shell height and live weight were analyzed as the coefficient of variation (CV) in Microsoft Office Excel. A correlation matrix analysis was used to determine the relationships between the environmental and growth parameters.

3. Results and Discussion

3.1. Environmental factors

Monthly day and night temperature of the culture area (A), temperature of natural and culture environment (B), monthly day and night humidity of the culture area (C) and humidity of natural and cultured environment (D) were shown in Figure 2. Day and night temperature differences was the highest (7.72 ºC) in April (P<0.05). There was no difference in humidity between day and night inside the culture system when greenhouse nylon was covered (P>0.05) (Figure 3). However, there was significant difference in humidity between day and night after the greenhouse nylon was removed on May 2015 (P<0.05). pH, organic matter and moisture values of the soil were 7.00, 30.67% and 24%, respectively.

It is reported that if soil structure is not suitable for snail culture, soil should be improved in order to ensure for healthy development of snails (Begg 2003). In the present study, organic matter in the soil increased by adding humus soil and water permeability increased by adding sandy soil. After soil improvement, the soil structure was suitable for snail and plant breeding. Lucas & Davis (1961) declared that if soil pH is around 7.2, it indicates that the soil is rich in calcium. Calcium rich soil is desirable property because it supports shell growth in snail culture.

The observation on developing plants showed that cabbages grew fast in the culture area. The spinach did not grow enough because of the shade of the enlarging cabbage leaves. The leaves of cabbage hardened quickly due to the high growth of the cabbage. However, this situation did not have an adverse effect on the snails feeding since cabbage mostly consumed by snails. Snails started to consume more formulated feed after cabbage leaves. In addition, enlarging cabbage leaves also obstructed to effectively use sprinkler irrigation system. On the other hand, the culture area covered greenhouse nylon was also regulated the circulation of humidity with preventing humidity lose when temperature was higher at the outside.

3.2. Growth rate

Matured snails were not within a certain size range because they were collected from nature, therefore no standard growth was achieved from the obtained offspring. Many studies revealed that cultured mature snails should be used for optimal and regular growth (Murphy 2001; Cobbinah et al 2008; Begg 2009). Monthly shell height growth was significantly different (P<0.05). SHGR was the highest (8.58%) in May while LWGR was the highest (4.11%) in June (Figure 3).

Temperature varied between 11-12 °C in the culture area until February and there was no significant growth rate in these time intervals. It was

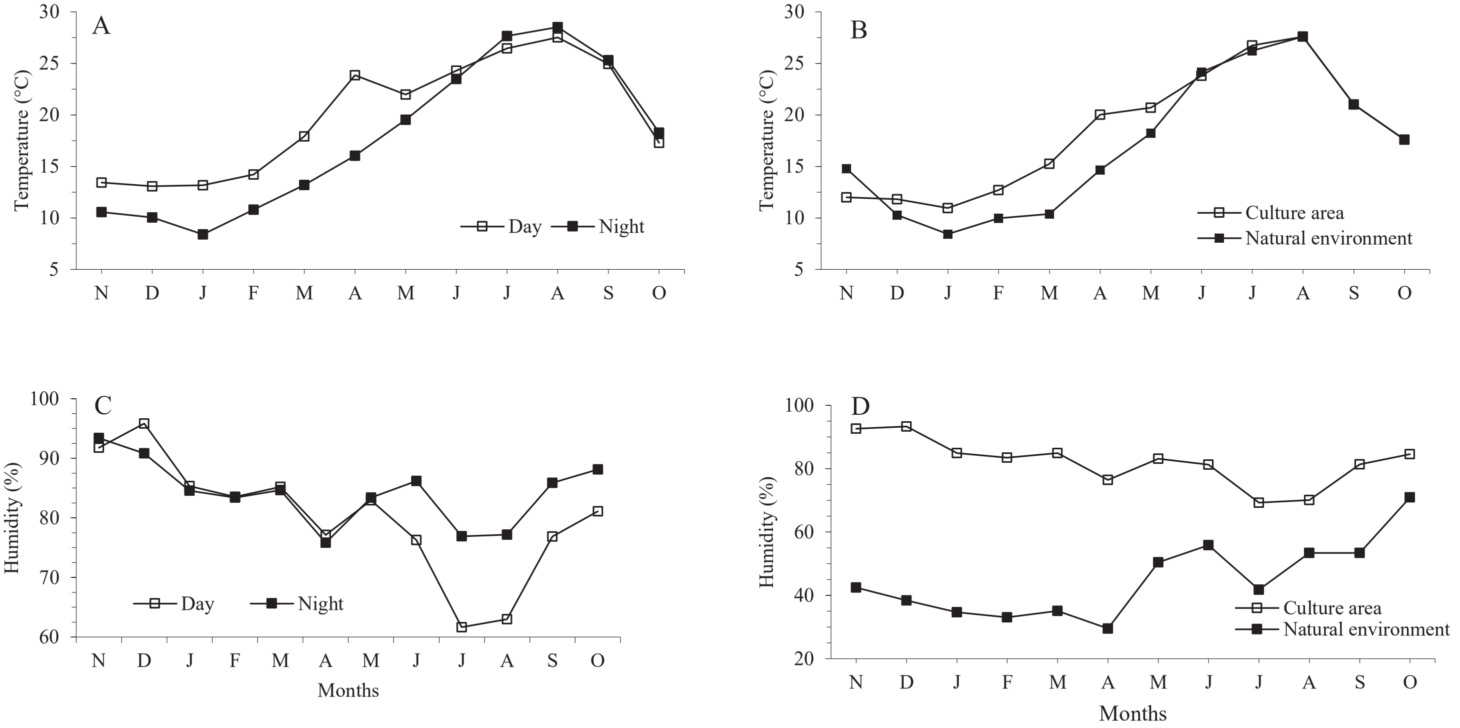

Figure 2- Monthly day and night temperature of the culture area (A), temperature of natural and culture environment (B), monthly day and night humidity of the culture area (C) and humidity of natural and cultured environment (D) during experiment

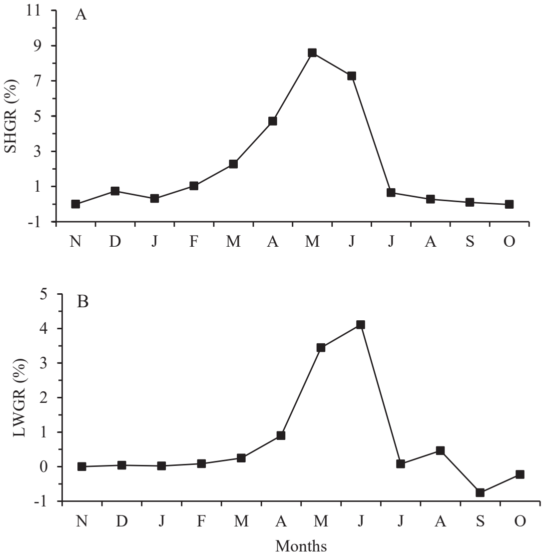

Figure 3- Monthly shell height growth rate (SHGR %) (A) and live weight growth rate (LWGR %) (B) of snails during experiment.

demonstrated that snails were not active below 12 °C and decreased or stopped feeding. After February, the growth rate increased with the increasing temperature above 12 °C. In April, the feeding rate of snails was increased with the increasing temperature about 19 °C. Furthermore, the highest growth was in May although the temperature difference was significant in April and May. It could be said that snails with the 13.91±0.28 mm shell height had better tolerance to environmental stress caused by the difference between night and day. Many studies showed that continuous growth was noticed in spring which demonstrated that most terrestrial snails show a faster growth rate during spring in nature (Staikou et al 1988; Hatziioannou et al 1989). Length growth of land snails' ceases with lip formation and reaches sexual maturation (Choat & Schiel 1980; Koene & Ter Maat 2004). In the present study, snails with 31.58±0.31 mm shell height growth rate started to decrease after May and reproductive behavior was observed. Daguzan (1982) reported that *H. aspersa* reached maturity with shell heights reaching over 27 mm and marketable size of this species is between 25 and 32 mm (Lazaridou-Dimitriadou et al 1998). In the present study, 25% of snails reached shell heights of over 27 mm within seven months, while 90% of them reached to the same size in June within eight months. Approximately all of hatched snails reached marketable size in nine months (July 2015). Ligaszewski et al (2007) declared that *Helix pomatia* needed two year farming cycle from hatching to maturity in an unheated greenhouse farming system in Poland. In Greece, *C. aspersum* reached marketable size varied from 2.5-5 months indoor farming system (Lazaridou-Dimitriadou et al 1998). The life cycle of *C. aspersum* was highly affected by the climatic conditions of the region (Chevallier 1977) and farming system. In Australia, one third of the hatched snails reached marketable size before they were 12 months old at an extensive system. Lazaridou-Dimitriadou et al (1998) reported that snails reached marketable size in 4-5 months under intensive farming conditions, depending origin, instead of 18 months which is needed in nature.

3.3. Mortality rate

Our study showed that offsprings experienced high stress such as variation in weather conditions and handling stress in the first stages of their life-cycles between November and May (Figure 4).

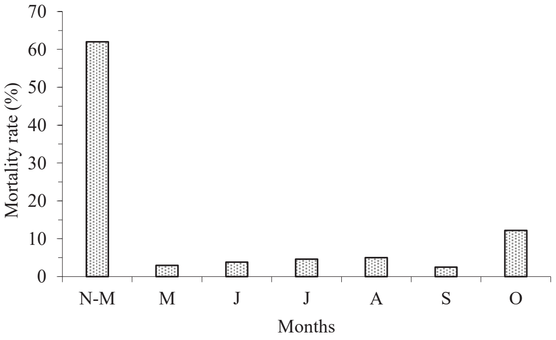

Figure 4- Mortality rate (%) between November 2014 and October 2015 (N-M: Mortality from November to May)

In November the juvenile snails (6.68 mm shell height) were taken from the hatchery (temperature range 18-20 °C; humidity range 84-87%) and placed to the covered culture area where they were exposed to relatively cold weather and temperature difference. In addition, they were under handling stress caused by the weekly measurement procedure of this sensitive stages. Snails could be damaged during the measurement and were not able to repair their shells since they had low feeding or no feeding rate for generating enough energy for repairing their shells under inappropriate conditions like low temperature. Hence, high mortality was observed between November and May. Many studies revealed that animals has lower ability to tolerate stress during their first stages of life (Zippay & Hofmann 2010; Gheoca 2013; Diederich & Pechenik 2013).

Snails showed reproductive behaviors after reaching sexual maturity in May and continued throughout summer and autumn. This showed that snails were constantly mating and producing eggs throughout the summer when suitable environmental was provided. However, high mortality rates were recorded in August, September and October. Negative live weight growth rate was also observed after August due to death of mature snails (Figure 4). Thus, spawning activity of matured snails could affect immune system and caused to post-reproductive mortality. Many studies showed similar results that mortality rate was higher in animals showing higher reproductive efforts because of reducing body maintenance and immune capacity, and hence it might cause physiological exhaustion (Baur & Baur 2000; Barker 2001; Carvalho et al 2008). In addition, farming system types, snail species and rearing density also effect snails' mortality rate. Dupont-Nivent (2000) reported quite high mortality of C. *aspersum* as 21% in indoor system in France. Ogogo et al (2011) found 1.4% cumulative mortality of *Archactina* spp. in survey of snail farming (farming systems: concrete trench, wire quaze fence, wooden cage) in Nigeria.

4. Conclusions

Additional studies should be carried out to further investigate which plants should be grown together without competition with the others for snail farming at the Black sea region.

More controllable system should be established to minimize the difference between night and day temperatures in the winter months, and therefore, low mortality and increased growth rate could be obtained.

The largest and the same sized mature snails should be selected as much as possible for breeding stock from the initial collected wild snails to obtain uniform growth of offspring and for successful breeding.

The most suitable environmental conditions for snail culture were provided during the Spring-Summer-Autumn seasons in the Black Sea region. The production activities might be more efficient with the addition of shading and irrigation at these seasons to reduce the effect of hot and dry air in summer. However, irrigation should be carried out according to temperature value of the environment to prevent snails from thermal stress.

After snails reached the market size which is the same size at maturation, breeding stock should be separated and the rest should be immediately harvested. Otherwise, mature snails will continue to reproduce which causes mortality by physiological exhaustion.

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Tractor Lifetime Assessment Analysis

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ABSTRACT

In this paper, two different approaches in analyzing the tractor lifetime assessment are presented. The first one is based on reliability theory and the other one is based on the relevant experience that was implemented in the ASABE standards. In this way, the dependence of tractor reliability and lifetime on working conditions is presented through two models verified in the paper. Tractors from two different producers were analyzed. Experimental data were collected during the tractor working engagement at the fields of Agricultural Corporation Belgrade (ACB). Analyzing the obtained data it is possible to find the mismanagement in the tractor usage. Removing them it is possible to extend the period of tractor utilization. In this way the overall organization of tractor-machinery system on a farm can significantly be improved.

Keywords: Standards; Tractor; Reliability; Remaining lifetime

1. Introduction

Tractors are one of the most used power units on the agricultural farms. Apart from agriculture, they are used as basic or drive machines in the mining and construction engineering systems. Tractor working environment varies significantly from one place to another so it is very difficult to estimate its influence on the tractor overall lifetime. Calculation of operational life of complex machines, despite designer's effort, is performed using some probability prediction model, which is based on assessment made by experienced designers and analogies with existing machines and experiences gained during their operation, including corrections related to differences of installed equipment. Anyhow, exact calculation of the operational life during design is not possible, hence it is about aspired operational life (Polovina et al 2010). During systems operation, based on the working and maintenance parameters it is possible to accurately define reliability and remaining capability of technical system. It is also possible to define the critical condition when the system does not fulfil its functionality. In Ebramhimipour & Suzuki (2006), the effectiveness was defined as overall indicator which contains efficiency, reliability and availability. In Miodragovic et al (2012), the effectiveness was defined as total indicator of reliability, maintainability and functionality. This is justified concerning the fact that the availability contains reliability and, thus, these two cannot be analyzed separately. Effectiveness, as a parameter, is very suitable for the analysis of technical systems such as tractors. There are new concepts that use money and costs parameters specially the maintenance cost parameters. According to Plessis (2007), there are three models of analyzing the costs and equipment service lifetime relation, with the aim of determining the moment of replacement of earthmoving equipment. These methods include the replacement that is primarily done on intuition, age-based replacement and replacement after performing an economic analysis. A machine must be replaced when a supposed frequency of breakdowns becomes so high that the machine is not reliable any longer. Finally, a machine must be replaced when the costs of repair begin to increase the average unit costs of accumulation beyond the minimum ones. For example, equipment manufacturer - Komatsu, has developed a model for determination of time frame for replacement of mining equipment; for the agricultural machines a model for assessment of remaining lifetime on the base of ASABE standard was developed. In any case, complete overview of the tractor lifetime is required as optimization process that synthesizes the cost and reliability (Previati et al 2011). Agricultural systems demand detailed planning and control of relevant biological, technical, technological and other processes (Mileusnic et al 2010). Among others, machinery statistics represents a crucial information that influences the agricultural technique management. The adequate data basis of this kind is an initial point for the appropriate decision-making. Miodragovic et al (2012) established the model for effectiveness determination according to fuzzy sets theory utilization. There by the fuzzy sets were used to analyze reliability, maintainability and functionality performances (partial indicators of effectiveness) as well as and for their integration into effectiveness. On the basis of data acquired on various Serbian farms (Tomantschger et al 2011), the frequency distribution and probability density function of the engine lifetime (up to the overhaul is done) has been obtained. An original mathematical model, which includes the differential Equation with adequate conditions, has been developed for this purpose. It is clear that all the models have, as their base, the reliability i.e. only with the reliable machines the high performance, low working and maintenance costs can be expected. Dalmiş et al (2017) and Ekinci & Çarman (2017) dealt with the tractor efficiency problem. Dalmiş et al (2017) analyzed the effects of materials fatigue on the exploitation parameters of three point hitch tractor system, using the method of finite elements. They also analyzed the effects of some drive tires properties on the improvement of tractive efficiency.

The aim of this paper is the use the experimental data from the field for the working productivity and the lifetime assessment analysis. The idea was to use exploitation data of the two tractor models for showing the methods for lifetime assessment analysis; the first one based on the basic model known from the reliability theory, and the other one based on the ASABE standards specially developed for these purposes. In this way, the verification of the models is done between themselves where the first one is strictly theoretical and the other one is specialized for the situation.

2. Material and Methods

Reliability engineering is a sub-discipline within system sciences. Reliability within the time-depending systems is defined as a time function $R(t)$ and can have the value between 0 and 1 or between 0 and 100%. Reliability can be also given as the number of successfully finished tasks and the total number of the system tasks ratio. In the case where for every moment of time, a system has all the tasks finished successfully, the reliability is 1 i.e. 100%. In the other case, when $R(t_1)= 0$, it can be said that the time t_1 is the end of lifetime. Essentially, for one system can be said that it is at the end of its lifetime when the failure rate (λ) begins to increase rapidly.

Failure rate can be defined based on the failure function *f*(*t*) as:

$$\lambda(t)=\frac{f(t)}{R(t)};f(t)=\frac{dF(t)}{dt} \tag{1}$$

Where; Failure probability (unreliability) represented by the following Equation.

$$F(t)=1-R(t) \tag{2}$$

Failure rate is the frequency of fails of a technical system or element. Failure rate is often called as the rapidity of failure. Relation between failure rate and lifetime can be presented in form of diagram (Figure 1). With the technical systems that are not fully worked out in the sense of construction and functionality (produced in pieces such as plants etc.) the above mention relation can be described via so called bathtub curve. This curve is characterized by three periods where during the first period system is in the stage of working itself out. During the second period, which is the most time consuming period, systems enters the stationary state. In the third period systems has failures and reaches it lifetime period. For the technical systems that are produced in the large series (tractors, etc.) there is no need for the first period (interrupted line on Figure 1) i.e. these systems are starting their usage in Normal life regime. In this regime the failure rate is low and continues up to end of lifetime where failure rate increases rapidly. If the systems is fully checked during time in sense of recording the periods of failure and based on the probability theory, functions *R*(*t*) and *λ*(*t*) can be determined (3-4). In general, these functions can be used for assessment of remaining lifetime. When solving the problem in the domain of failure theory two-parameter Weibull distribution showed as the most suitable (Weibull 1951). In this distribution, the reliability function and failure rate can be determined by the following equations, respectively

$$R(t)=e^{-\left(\frac{t}{\eta}\right)^{\beta}} \tag{3}$$

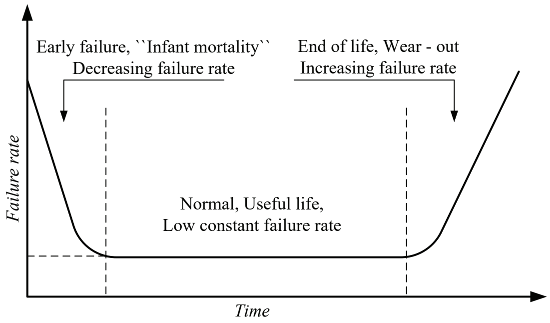

Figure 1- The bathtub curve, hypothetical relation failure rate versus time

$$\lambda(t)=\frac{\beta}{\eta}\cdot\left(\frac{t}{\eta}\right)^{\beta-1} \tag{4}$$

Where; *β* is shape parameter and *η* is scale parameter.

The mean time to failure (*MTTF*) was determined with below Equation.

$$MTTF=\eta\cdot\Gamma\left(1+\frac{1}{\beta}\right) \tag{5}$$

Where; Gamma function (*Γ*) is represented by the following Equations.

$$\Gamma(p)=\int_0^{\infty}t^{p-1}e^{-t}dt; \tag{6}$$

$$p=1+\frac{1}{\beta} \tag{7}$$

Weibull function is used due to its parametric shape and the possibilities of the other distribution laws. For the needs of lifetime management, it is very important to precisely define the moment *t* when system should be withdrawn from engagement (from work). In Figure 1, this is the moment when failure rate begins to increase rapidly (III period). It is the time period when *R*(*t*) function falls down on a certain low point. These two approach are theoretical. In real conditions, technical systems usually have a possibility for some kind of reparation which then complicates these graphical presentations.

2.1. Assessment model of remaining lifetime

Calculation of remaining value (RV_n) as a percentage of the list price for farm equipment at the end of *n* years of age and after *h* average hours of use per year using the following equation and the coefficients which depends on the power of the engine on the tractor is shown in Table 1.

Table 1- Remaining value coefficients

Equipment type *Farm tractors*	C_1	C_2	C_3
TSmall < 60 kW	0.9810	0.0930	0.0058
Medium 60-120 kW	0.9420	0.1000	0.0008
Large > 112 kW	0.9760	0.1190	0.0019

$$RV_n = 100\left[C_1 - C_2\left(n^{0.5}\right) - C_3\left(h^{0.5}\right)\right]^2 \tag{8}$$

To include the inflation effects, the list price of farm equipment should be multiply by $(1+i)^n$ where *i* is the average annual inflation rate, *n* is the age of the machine. Machine effectiveness can be defined as a ratio between machine productivity in the real, field conditions and maximum theoretical productivity (ASABE 2009). The effectiveness in the field conditions includes improper usage of machine in sense of its working width, time losses caused by operator itself and field characteristics (Table 2). Expenditures are necessary to keep a machine operable due to wear, part failures, accidents and natural deterioration. The costs for repairing a machine are highly variable. Good management may keep costs low. Indices of repair and maintenance costs are shown in ASABE (2009). The size of the machine, as reflected by its list price and the amount of use are factors affecting the costs. Both the use and costs are expressed in an accumulated mode to reduce variability. In times of rapid inflation, the list price must be increased to reflect inflation effects. Accumulated repair and maintenance costs at a typical speed can be determined with the following relationships using the repair and maintenance factors *RF*1 and *RF*2 (ASABE 2009) and the accumulated use of the machine (ASABE 2006).

$$C_{rm} = (RF1)\cdot P\cdot\left[\frac{h}{1000}\right]^{(RF2)} \tag{9}$$

Where; C_{rm} is accumulated repair and maintenance cost; *RF*1 and *RF*2 are repair factors. *P* is machine price. During rapid inflation, the original list price must be multiplied by $(1+i)^n$, *h* is the accumulated hours of machine use.

For the model, two time-digressive amortisation methods were used. The first method is the digital digressive amortization. The lowering of the amortisation quotas that are in the arithmetic sequence, can have a value like the amortisation from the last period of usage.

$$a_k = \frac{V_a}{1+2+...+n}\cdot\left[n-(k-1)\right] \tag{10}$$

Where; a_k is amortisation level in the given periods of lifetime; V_a is the based amortization, *n* is usage period as years, and *k* is current period.

The second method is the geometrical digressive amortization when amortisation quotes are decreasing as elements of the geometrical sequence.

Table 2- Field efficiency, field speed, and repair and maintenance cost parameters

Machine	*Field efficiency (%)*		*Field speed*		*Estimated life (h)*	*Total life R & M cost*	*Repair factors*	
	Range	*Typical*	*Range*	*Typical*		*(%)*	*RF1*	*RF2*
ASAE D497.6								
Tractor 4x2	-	-	-	-	12000	100	0.007	2.000
Tractor 4x4	-	-	-	-	16000	80	0.003	2.000

$$a_k = a_n \cdot q^{(n-k)} \tag{11}$$

$$a_n = V_a \cdot \frac{q-1}{q^n - 1} \tag{12}$$

Where; a_n is amortisation at the end of lifetime period and q is geometrical sequence ratio.

3. Results and Discussion

In this paper, as an illustrative example of agriculture machinery evaluation for lifetime assessment, the comparative analyses of two tractors, Fendt Vario 920 (A-type tractor) and John Deere 8520 (B- type tractor), are contain. Based on their engagement the following data about the time of failure are obtained. Experimental data collected from "ACB" have also taken into account the time of the specific intervention on the every tractor form. In all four cases, the number of collected data was $n<30$, so, for the calculation of cumulative distribution function $F(t)$, Median rank (MR) also known as Bernard's approximation, was used (Table 3). Concerning the fact that these method are well known, only the reliability and failure rate functions are presented, as well as the mean time to failure values from 13 to 16 Equations, for tractors A1, A2, B1 and B2, respectively. The calculations used well-known tools: median rank, probability plotting paper, last square method and Kolmogorov-Smirnov test.

Table 3- Time to failures for observed tractors, hours

A1 *FV-920; F28*	*A2* *FV-920; F30*	*B1* *JD 8520; Inv. no. 36*	*B2* *JD 8520; Inv. no. 37*
6374	4766	4682	4168
6583	4896	6682	6610
6840	7367	8980	7631
8145	7685	9790	9122
8365	8139	10493	9614
8636	9359	10646	9630
9332	10380	10862	9695
10765	14796	10865	10168
11349		11051	10434
12276		11717	11180
13476		11981	11376
13670		12210	11689
13745		12470	11936
15369		12563	12385
16096		12675	12634
		12743	12785
		12757	
		12836	
		13200	
		13721	
		14023	

Kolmogorov-Smirnov test (K-S) is the most common method for testing of hypothesis of established distribution law. K-S test compare empirical cumulative distribution function and theoretical function $F(t)$= 1- $R(t)$, on the base of their distance D_n. Necessity is that the distance is less than the critical value $D_{n\alpha}$, and we can conclude that the data is a good fit with the specified

$$R(t) = e^{-\left(\frac{t}{11941.94}\right)^{3.55}}; \lambda(t) = 2.97 \cdot 10^{-4} \cdot \left(\frac{t}{11941.94}\right)^{2.55}; MTTF = 10750.55 \text{ hours} \tag{13}$$

$$R(t) = e^{-\left(\frac{t}{9523.81}\right)^{2.78}}; \lambda(t) = 2.92 \cdot 10^{-4} \cdot \left(\frac{t}{9523.81}\right)^{1.78}; MTTF = 8209.39 \text{ hours} \tag{14}$$

$$R(t) = e^{-\left(\frac{t}{12273.77}\right)^{4.00}}; \lambda(t) = 3.26 \cdot 10^{-4} \cdot \left(\frac{t}{12273.77}\right)^{3.00}; MTTF = 11124.70 \text{ hours} \tag{15}$$

$$R(t) = e^{-\left(\frac{t}{11213.06}\right)^{3.80}}; \lambda(t) = 3.39 \cdot 10^{-4} \cdot \left(\frac{t}{11213.06}\right)^{2.80}; MTTF = 10134.96 \text{ hours} \tag{16}$$

distribution law. Critical value can be found in the Kolmogorov-Smirnov table. The largest distance in presented case study is for tractor B1 and for data i= 6: D_n= 0.15976. For the given example (tractor B1) that contains n= 22 data, according to K-S test for goodness of fit, the acceptable difference between empirical and theoretical value is: $D_{n;\alpha}$= $D_{22;0.05}$= 0.285 (O'Connor & Kleyner 2012). Usually calculated with level of significance α= 0.05. Since D_n= 0.160<0.285= $D_{10;0.05}$, we conclude that the data is a good fit with the Weibull distribution. K-S test shows that the actual model is satisfactory accuracy. The simplest parameter for comparing two tractors is mean time to failure (*MTTF*). Concerning this parameter the B1 tractor is the best having the mean time to failure of 11124.70 hours. The worst results were obtained for A2 tractor with the 8209.39 hours. Based on the reliability function ($R(t)$) and Figure 2, the same conclusions, about the most reliable tractor can be obtained. For example, if 12000 hours are considered to be a reference time, it can be seen that for tractor B1 there is 40% probability that it is working properly while with the A2 tractor this probability is 15%. If it is said that probability that a systems works properly is 20% then tractor B1 will achieve this value after 13800 hours, tractor A1 after 13650 hours, tractor B2 after 12700 hours and tractor A2 after 11300 hours. Based on the failure rate function ($\lambda(t)$) and Figure 3, it can be concluded that A- type tractor is less susceptible to failure rate increment. In any case it can be seen that failure rate starts to grow in the 12000-15000 hours interval, when λ>0.0005. Based on the average values of the above parameters it can be generally concluded that B- type tractor is slightly better compared to A- type tractor. Table 4 gives the results obtained for A- type tractor, using the digital digressive amortisation model (according to ASABE standard). Purchasing tractor price was 162000 € and the price after the 10th year of usage is 52024 €. Inflation rate was adopted on the basis of average inflation rate in EU of 1.6%. The prevailing interest rate was 4.4% while real interest rate was 2.8%. From Table 4 can be seen that, as far for the tractor unit the accumulated costs are starting to grow after the seventh year of usage. For the same tractor geometrical digressive amortisation was used, with the same input parameters. It can be seen that with the given amortisation rate unit accumulation costs are starting to increase in the tenth year of tractor usage. The same trend in the results, according to the both criteria, was observed for the B- type tractor (Tables 3 and 5). Purchasing price of B- type tractor was 155900 € and after the ten years of usage the value was 50065 €. If results of applied ASABE standards and theory of reliability are compared it can be seen that in the case of digital digressive amortisation method they are almost identical. With this model 11200 hours are the point where one should think about replacing the tractor and purchasing the new one. In this moment the unit accumulation costs are starting to increase. Theoretical model states that for the B- type tractor reliability starts to decrease at 11200 hours from 37% up to 50% and in the case of A- type tractor the reliability falls to 21% even down to 45% meaning that there is a higher probability that tractor is out of function than the probability that operates well. In addition, failure rate is just starting to increase at the value of 12000-15000 hours meaning that right time for tractor replacement should be planned at the interval of 11200 hours. All this indicates that when the time of tractor replacing is in question these methods are compatible. However, the geometrical digressive amortisation method states that the time when unit accumulation costs start to increase is after 16000 hours of tractor usage. The reliability of A- type tractor is between 2 and 6% while it is 6% for B- type tractor. For sure this is the time when tractor should be replaced. If not the productivity of tractor it will significantly decrease. Results of the theoretical model show that the B-type tractor is a better solution than A- type concerning the reliability during the period of usage. According to the validation results, all the tractors are still in use. B- type tractor has 16096 working hours and the procedure for its replacement has started. A- type tractor has 14023 working hours and it is still in usage. Hypothesis, on the beginning of the research, was that the lifetime of the equipment depends on technical and economical parameters. The first one can be expressed via availability and reliability and the other one can be expressed via economic indicators. Regarding the results tendencies, these two approaches are in accordance. This paper proves it and the case study has served as a proof of the hypothesis on the beginning. Similar results were obtained by Tomantschger et al (2011) applying the different models.

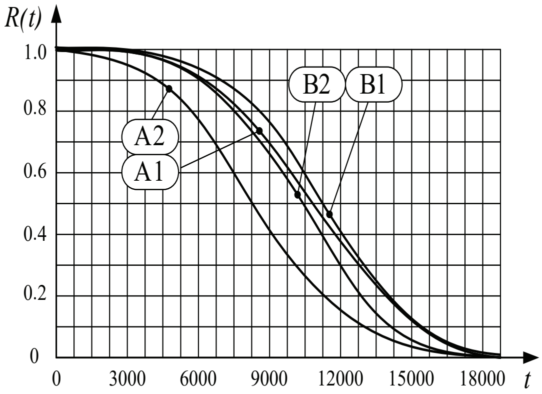

Figure 2- Reliability function

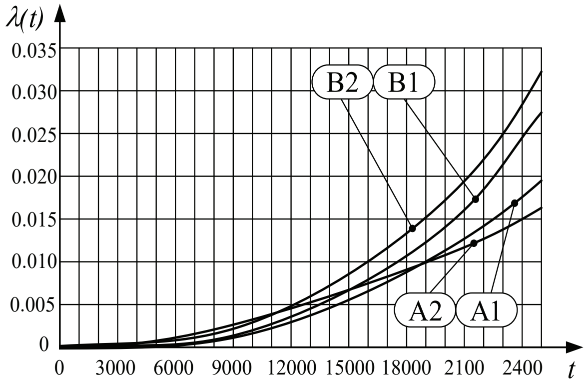

Figure 3- Failure rate function

Table 4- Digital digressive amortization model for A type tractor

End of year	*Rest of value (€)*	*T_o (€)*	*Amortization (€)*	*Interest (€)*	*Acum. amortization (€)*	*Acum. interest (€)*	*Acum. T_o (€)*	*Acum. T_{total} (€)*	*Acum. use (h)*	*Unit acum. cost (€ h^{-1})*
1	164805	1264	25063	4615	25063	4615	1264	30942	1600	19.34
2	142249	3873	22556	3983	47619	8598	5137	61354	3200	19.17
3	122199	6606	20050	3422	67669	12020	11743	91432	4800	19.05
4	104655	9468	17544	2930	85213	14950	21211	121374	6400	18.95
5	89617	12462	15038	2509	100251	17459	33673	151383	8000	18.93
6	77086	15592	12531	2158	112782	19617	49265	181664	9600	18.92
7	67061	18863	10025	1878	122807	21495	68128	212430	11200	18.96
8	59542	22280	7519	1667	130326	23162	90408	243896	12800	19.05
9	54529	25851	5013	1527	135339	24689	116259	276287	14400	19.18
10	52024	29560	2506	1457	137845	26146	145819	309810	16000	19.36

Table 5- Geometrical digressive amortization model for B type tractor

End of year	*Rest of value (€)*	*T_o (€)*	*Amortization (€)*	*Interest (€)*	*Acum. amortization (€)*	*Acum. interest (€)*	*Acum. T_o (€)*	*Acum. T_{total} (€)*	*Acum. use (h)*	*Unit Acum. cost (€ h^{-1})*
1	116328	1216	66391	3257	66391	3257	1216	70864	1600	44.29
2	83132	3728	33196	2328	99587	5585	4944	110116	3200	34.41
3	66534	6357	16598	1863	116185	7448	11301	134934	4800	28.11
4	58235	9112	8299	1631	124484	9079	20413	153976	6400	24.06
5	54086	11992	4149	1514	128633	10593	32405	171631	8000	21.45
6	52011	15005	2075	1456	130708	12049	47410	190167	9600	19.80
7	50974	18153	1037	1427	131745	13476	65563	210784	11200	18.82
8	50455	21441	519	1413	132264	14889	87004	234157	12800	18.29
9	50196	24872	259	1406	132523	16295	111876	260694	14400	18.10
10	50065	28452	130	1402	132653	17607	140328	290678	16000	18.16

4. Conclusions

Technical systems lifetime assessment is very complex and responsible task. Practically, it is not possible to precisely define the lifetime assessment parameters but only to give their estimation regarding the precisely defined technical system working conditions.

In this paper, two models for analyzing the tractor lifetime assessment are presented. Models are presented on the theoretical level but are developed thorough the case study. The first one is theoretical and it is based on reliability theory and the other one is special and is based on the ASABE standards. This standard uses empirical data and data about the working conditions, working regimes to give the estimated period of tractor usage. Results show that reliability theory confirms the results and their tendencies obtained by the ASABE standards. In this way, both models are verified. This is of a great practical contribution since one of the models is of a practical and the other of the empirical nature. Conclusions can be summarized in the next two statements: reliability and availability are decreasing through the time while the maintenance expenses are increasing. For each and every machine and working environment, the moment when the reliability as intense decrease and expenses intense increase, can be determined based on the proposed model. Results show that these two moments are very close to each other.

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Morphological Characterization of Bottle Gourd (*Lagenaria siceraria* (Molina) Standl.) Germplasm and Formation of a Core Collection

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ABSTRACT

Bottle gourd [*Lagenaria siceraria* (Molina) Standl.] genotypes were collected from different parts of Turkey and a core collection (CC) was established based on morphological characteristics. The collection was grown for morphological characterization and seed multiplication. Seeds were produced from 322 genotypes (Entire Population- EP) representing diversity of entire country by selfing and the accessions were characterized for 25 quantitative and 21 qualitative morphological characteristics. Important variation was observed in fruit size, cotyledon dimensions, leaf dimension, flower size, fruit length (8.7-155.7 cm), fruit shape (flat-elongate), seed weight (3.8 g/100 seeds -38 g/100 seeds) among accessions as previously reported. UPGMA cluster, and principal component analyses cluster of quantitative and qualitative characteristics of EP were assayed. Results of the assays together with geographic origin of the genotypes were analyzed to establish a core population (CC) consisting of 100 genotypes. High correlation between the CC and the EP (r= 0.974 for normalized Mantel Statistic Z) demonstrated that CC represents the most of total genetic variation with minimum redundancy in bottle gourd germplasm of Turkey. The CC will enable efficient and cost-effective management and utilization of bottle gourd germplasm.

Keywords: Core collection; Genetic resources; Qualitative; Quantitative; Turkey

1. Introduction

Bottle gourd, calabash [*Lagenaria siceraria* (Molina) Standl.] belongs to the *Cucurbitaceae* family, is generally accepted as “the white-flowered gourd” due to its white petals. *L. siceraria,* an annual, monoecious and vigorously climbing species, is one of the earliest domesticated plant species for human utilization (Decker-Walters et al 2004). Bottle gourd is extensively distributed, and ocean currents are believed to have mediated migration of *L. siceraria* from Africa to America, and consequently the species is divided into two subspecies: Asian bottle gourd [*L. s.* subsp. *asiatica* (Kobyakova) Heiser] and American/African bottle gourd (*L. s.* subsp. *siceraria*) (Kistler et al 2014). In Europe, bottle gourd has been a known species in northern Italy since the Iron Age (Schlumbaum & Vandorpe 2012). Whether bottle gourd originated from Africa or Asia is the subject of studies all over the world including Turkey; no historical finding about bottle gourd has been reported, however, recent molecular analysis suggested that Turkish bottle gourds are mix of African and Asian bottle gourds (Gürcan et al 2015).

The mature dried fruits of bottle gourds are used by people throughout the world as containers, kitchen utensils, musical instruments, for artistic purposes or in some coastal regions, fishing net holders (Decker-Walters et al 2004). Young leaves, shoots, seeds, fresh tendrils, shoot and leaves have been also utilized for cooking and some therapeutic aims (Loukou et al 2007). Additionally, *L. siceraria* has a potential to be used rootstock for watermelon since it is likely to have resistance against different biotic and abiotic stress factors including Fusarium wilt (Yetisir et al 2003), salinity and water-logging of soil (Yetisir & Uygur 2009).

Collection, characterization and presentation of genetic resources, and their use by breeders and producers provide a clear path to achieve sustainable agriculture (Given 1987). The conserving plant germplasm has been remarkably progressed in the last decades. In many cases the increasing number of germplasm collections has become to limit their accessibility and utilization in plant breeding and research. Difficulties related to utilization of large germplasm in research purposes led the researchers to establish "core collections" (CC) which does not replace the existing collection, but it is off limits size and chosen to represent the genetic diversity of a large collection (Frankel 1984). Several recent studies on collection and evaluation of bottle gourd genetic resources have been conducted based on traits in USA (Decker-Walters et al 2004), Kenya (Morimoto et al 2005), Serbia (Mladenović et al 2012) and Turkey (Yetisir et al 2008). In the previous study (Yetisir et al 2008) bottle gourds were collected in just Mediterranean region of the country. The study demonstrated in high morphological variation and indicated importance of germplasm collection over the entire country. Here a) we collected and studied morphological (quantitative and qualitative traits) characterization of the bottle gourds in the all regions of the Turkey where bottle gourds grown and b) additionally, a CC consisting of 100 genotypes were established the first time. This study is the most comprehensive on the collection and morphological characterization of Turkish bottle gourd accessions.

2. Material and Methods

2.1. Collection of bottle gourds and morphological characterization

Bottle gourd genotypes used in this study were collected under two projects (TOVAG 3216 and TOVAG 111O117) financially supported by TUBİTAK and introduced from different countries. Additionally, bottle gourd accessions obtained from international gene banks were investigated in this study. Available seeds (maximum up to 24) of the collected 418 genotypes were planted on March 10th 2012 in multi-pots filled with a mixture of perlite and peat (1:2) in a greenhouse at the Alata Horticultural Research Station in Erdemli, Mersin. The experimental area receives annual average rainfall of 500-600 mm and the soil characteristics are sandy-loam with a low level of lime. Ten seedlings for each accession with 2-3 true leaves were planted into the field with 3 x 0.5 m spacing.

Following the soil analysis report, 50 kg N ha^{-1}, 100 kg P_2O_5 ha^{-1} and 50 kg K_2O ha^{-1} were added to soil during soil tillage as base fertilizer. Water was applied by drip irrigation and an additional 50 kg N ha^{-1} and 50 kg K_2O ha^{-1} fertilizers were provided by fertigation methods during the growing period. All measurements and observations were made on three representative plants for each genotype. Each accession was characterized for 25 quantitative (Table 2) and 21 qualitative traits (Table 3) according to the descriptor list published by the Biodiversity International for *Cucurbitaceae* (2007).

2.2. Data analyzing and selection of accessions for core collection

After obtaining characteristic values for the successfully grown bottle gourds, the phylogenetic relationships among bottle gourds were analyzed by unweighted pair group method arithmetic average (UPGMA) and by principal component analysis (PCA). A morphological distance matrix based on all traits was generated by applying the Euclidean coefficient, which was subsequently used for the preparation of a UPGMA dendrogram with the Numerical Taxonomy Multivariate Analysis System (NTSYS-pc) version 2.1 software package (Exeter Software, Setauket, NY, USA) (Rohlf 2000). For the PCA, the data from the entire population (EP) were analyzed using XLSTAT Statistical Analysis Software together with Pearson correlation coefficient (Kovach Computing Service 2013) based on the 25 quantitative and 21 qualitative variables. The UPGMA dendrogram, PCA, and geographic origin of the gourds were used to select establishing a CC representing genetic variation of the entire bottle gourds. All exotic accessions and the Turkish accessions with the most extreme traits were included in the CC. Source and accession numbers of CC genotypes can be seen at Figure 2. Correlation between the EP and CC were tested by producing and comparing the distance matrices using Mantel test of NTSYS-pc software, which is performed by Pearson product-moment correlation coefficient "r".

3. Results and Discussion

Of the total 418 accessions including 380 Turkish accessions and 38 exotic accessions, seed of 96 accessions did not germinate and the remaining accessions germinated and produced healthy intact plants. A total of 322 produced seedlings and were grown in the experiment field successfully. Of these 322 accessions, 292 were Turkish and the remaining 30 accessions were exotic. Seeds produced from these 322 accessions were deposited in D8 Seed Bank, Ankara, Turkey, (http://www.d8seedbank.org/ContactUs.aspx) and available for breeders and researchers upon request. Twenty-five quantitative and 21 qualitative characteristics were investigated for these 322 accessions resulting in a significant variation among the Turkish and exotic accessions (Table 1. and 2.). The fold difference between minimum and maximum values for the quantitative traits ranged from 1.6 (blooming time for male and female flower) to 15 (fruit length) folds. Interestingly, fruit dimension varied from 8.7 cm to 155.7 cm in length while fruit diameters ranged from 4.6 cm to 27.7 cm. Seed size also showed notable variation with about 10 fold differences between minimum and maximum value of 100 seed weight. A remarkable range of variation was also identified in seedling, leaf and other fruit characteristics: hypocotyl length (1.4-9.9 cm), leaf length (8.3-24.6 cm), male flower width (2.6-13.1 cm), female flower length (3.8-11.8 cm), petiole length (5.5-30.5 cm), and fruit shell thickness (2.8-16.3 mm).

With respect to qualitative characteristics, plant growth habit (all prostrated), corolla color (all white), sex type (all monecious) and tendril shape and branching (all branched and coiled) did not show differences. However, the remaining traits exhibited notable variation and were separated into several different classes (Table 2). Nine observed traits distributed in three classes while seven observed characteristics separated into four classes. The highest-class number was identified in fruit shape with nine classes (Round, elongated, pyriform, cylindrical, dumbbell, elliptical, flattened, curved, and crooked neck) followed by five classes in leaf shape (oval, round, kidney, heart, and slightly lobed) and pattern of secondary color of fruit skin (Absent, speckled, spotted, streaked, and bisectional). Prior studies showed that Turkey is not the genetic origin of *L. siceraria*, and the bottle gourds could have been introduced from both Africa and Asia through multiple gateways (Gürcan et al 2015). However, present study confirmed that Turkish bottle gourd germplasm has still a significant variation. At present study, the most obvious phenotypical difference was found in fruit shape and volume. Significant variability of seed and fruit characteristics of bottle gourd accessions from different part of Kenya

Table 1- Minimum, maximum and mean values for 25 quantitative variables of the entire and core collection of bottle gourd genotypes in the Turkish germplasm collection

Characteristics	*Minimum*		*Maximum*		*Mean±SE*	
	EP	*CC*	*EP*	*CC*	*EP*	*CC*
Hypocotyl diameter (mm)	1.5	1.7	4.4	4.4	2.8±0.4	2.9±0.4
Hypocotyl length (cm)	1.4	2.1	9.9	8.2	4.2±1.3	4.2±1.2
Cotyledon length (cm)	2.3	4.8	7.6	7.5	5.7±0.9	5.8±0.8
Cotyledon width (cm)	1.5	2.4	4.3	4.0	3.13±0.4	3.16±0.4
Leaf length (cm)	8.3	8.3	24.6	20.1	13.8±2.1	14.2±2.3
Leaf width (cm)	8.9	8.9	26.1	26.0	16.4±3.0	16.9±3.8
Petiole length (cm)	5.5	6.2	30.5	29.8	14.4±3.7	14.7±3.8
Petiole diameter (mm)	2.5	2.5	11.0	10.0	6.0±1.3	6.1±1.3
Main stem diameter (mm)	3.9	3.9	15.3	14.4	10.0±1.6	9.8±1.7
Internode length (cm)	8.4	8.1	25.9	25.9	13.2±2.4	13.5±2.8
Male flower length (cm)	3.1	3.1	5.9	5.3	4.3±0.5	4.2±0.6
Male flower width (cm)	2.6	3.8	13.1	12.4	5.5±0.9	5.5±1.0
Ovarium diameter (mm)	4.0	4.0	14.6	14.6	9.2±1.8	9.3±1.8
Ovarium length (mm)	1.6	1.6	8.3	6.7	3.3±0.8	3.2±0.8
Female flower length (cm)	3.8	3.8	11.8	8.9	5.9±1.0	5.9±1.4
Female flower width (cm)	3.0	3.2	7.6	7.0	5.0±0.7	4.9±0.7
Male flower blooming time (DAS)	51.0	51.0	80.0	80.0	63.0±5.9	63.4±6.2
Female flower blooming time (DAS)	51.0	51.0	86.0	86.0	66.0±6.6	65.7±7.1
Fruit length (cm)	8.7	8.7	155.7	130.5	54.5±18.8	53.9±1.7
Fruit diameter (cm)	4.6	2.3	27.7	53.0	16.3±3.7	8.6±4.9
Fruit shell thickness (mm)	2.8	2.8	16.3	16.3	6.8±2.0	6.8±2.3
Seed length (cm)	0.7	0.7	2.5	2.5	1.8±0.2	1.8±0.2
Seed width (cm)	0.4	0.4	1.2	1.1	0.8±0.1	0.8±0.1
Seed thickness (mm)	2.3	2.0	4.4	4.0	3.3±0.4	3.3±0.4
100 seed weight (g)	3.8	3.8	37.3	34.3	21.4±5.3	21.3±5.6

EP, entire accessions; CC, core collection

(Morimoto et al 2005) and Serbia (Mladenovic et al 2012) were recorded. More recently, Mashilo et al (2017) reported that presence or absence of fruit neck, fruit shape, degree of neck bending and fruit neck length positively correlated the phenotypic variation of South African bottle gourd landraces. In the family of *Cucurbitaceae*, the significant range of seed and fruit phenotypic variation has been also indicated in *C. pepo* (Paris 2001), *L. siceraria* (Morimoto et al 2005) and *C. maxima* (Balkaya et al 2009).

We aimed to form a CC representing genetic variation of the entire bottle gourds we possess. In order to determine most appropriate composition in the CC, initially we included the accessions with extreme values such as extremely large and small fruits. The dendrogram and PCA of the EP were used to select the most diverse bottle gourds in Turkish germplasm paying attention their distribution on the dendrogram and PCA and excluding closely positioned genotypes. Additionally, we paid attention to include samples from different regions of the country to represent geographical diversity of country. At first, our selection resulted in total 116 accessions. After that, UPGMA dendrogram and PCA plots were constructed several more times to reduce the number of accessions in the CC. Finally, we terminated selection reaching the

Table 2- Distribution of 21 qualitative variables of *L. siceraria* from Turkish germplasm

Characteristics	*Distribution (%)*								
Cotyledon position	Open	Closed	Intermediate						
	50	39	11						
Leaf shape	Oval	Round	Kidney	Heart	Slightly lobed				
	2	15	3	79	1				
Leaf size	Small	Big	Intermediate						
	11	26	63						
Secondary color on leaf	Absent	Light green	Silvering						
	98	1.5	0.5						
Leaf margin	Smooth	Dented							
	11	89							
Leaf lobe	Absent	Shallow	Intermediate	Deep					
	16	77	6	1					
Leaf pubescence, ventral surface	Absent	Low	Intermediate	High					
	1.5	59	37	2.5					
Leaf pubescence, dorsal surface	Absent	Low	Intermediate	High					
	0.5	19	57	23.5					
Leaf blistering	Absent	Present							
	66	34							
Flower size	Small	Big	Intermediate						
	15	21	64						
Female flower position	Main stem	Lateral branch	Both						
	0.5	95	4.5						
Peduncle intersectional shape	Round	Slightly angular	Sharply angular						
	11	42	47						
Fruit ribs	Absent	Superficial	Intermediate	Deep					
	79	1.5	4	0.5					
Fruit stem end shape	Depressed	Flattened	Rounded	Pointed					
	19	31	45	5					
Fruit blossom end shape	Depressed	Flattened	Rounded	Pointed					
	50	23	13	14					
Fruit skin texture	Smooth	Finely wrinkled	Netted						
	34	64	2						
Time of maturity	Early	Late	Intermediate						
	48	42	10						
Predominant fruit skin color	Green	Yellow	Milky-Brown						
	87	11	2						
Secondary color on fruit skin	White	Cream	Yellow	Other					
	1	84	13	2					
Pattern of secondary fruit color	Absent	Speckled	Spotted	Streaked	Bisectional				
	11	57	21	8	3				
Fruit shape	R.	E.	P.	Cy.	D.	E.	F	C.	Cr. N.
	1.5	6	18	1	8	0.5	0.5	8	57

R, Round; E, Elongated; P, Pyriform; Cy, Cylindrical; D, Dumbbell; E, Elliptical; F, Flattened; C, Curved; Cr. N, Crooked Neck

Table 3- Principal components for 25 quantitative variables of all bottle gourd accessions

Variable	*Entire population*					*Core collection*				
	PCA1	*PCA2*	*PCA 3*	*PCA4*	*PCA5*	*PCA1*	*PCA2*	*PCA3*	*PCA4*	*PCA 5*
Eigenvalue	5.11	2.93	2.66	1.94	1.72	5.57	2.97	2.45	2.15	1.68
Variability (%)	20.42	11.74	10.63	7.75	6.87	22.28	11.89	9.82	8.62	6.71
Cumulative %	20.42	32.16	42.79	50.54	57.41	22.28	34.17	43.99	52.61	59.32
	Contribution of the variables (%)					*Contribution of the variables (%)*				
Hypocotyl diameter (mm)	0.67	0.36	4.96	0.11	1.63	0.28	2.72	1.42	4.36	2.91
Hypocotyl length (cm)	0.02	0.29	1.38	10.09	4.96	0.49	0.71	0.98	0.12	24.13
Cotyledon length (cm)	1.98	0.41	11.34	0.14	14.55	2.57	2.12	5.71	1.12	10.22
Cotyledon width (cm)	2.48	1.31	9.06	0.70	14.88	1.73	5.93	5.28	0.53	16.44
Leaf length (cm)	8.44	0.00	7.40	0.18	7.66	10.86	0.01	0.26	5.37	1.97
Leaf width (cm)	10.43	0.01	6.43	0.56	6.82	11.07	0.11	0.00	6.34	4.20
Petiole length (cm)	7.72	0.01	7.62	0.27	4.44	9.69	0.14	0.02	5.35	2.85
Petiole diameter (mm)	5.42	1.23	5.88	0.18	4.86	8.37	2.66	0.03	6.69	1.38
Main stem diameter (mm)	0.22	4.66	0.00	2.07	3.60	0.08	6.13	9.16	0.09	3.06
Internodium length (cm)	0.68	3.08	0.78	8.13	5.44	0.37	4.85	9.55	2.34	0.13
Male flower length (cm)	6.71	1.43	1.42	1.23	3.85	8.64	0.60	0.40	0.72	0.79
Male flower width (cm)	6.29	0.89	1.07	1.03	2.90	6.42	0.98	0.29	2.92	2.07
Ovarium diameter (mm)	2.29	6.15	3.66	0.00	1.18	2.28	2.62	1.34	2.57	8.76
Ovarium length (mm)	3.90	16.91	0.50	0.04	3.60	3.36	15.97	0.55	5.26	0.26
Female flower length (cm)	6.50	14.25	0.07	0.39	5.71	5.40	14.43	0.43	2.81	0.14
Female flower width (cm)	7.12	0.70	1.91	0.46	8.39	8.49	1.95	0.20	0.20	0.08
Male flower blooming time (DAS)	2.10	0.12	5.67	31.11	0.00	0.47	0.06	26.73	3.10	8.43
Female flower blooming time (DAS)	2.37	0.00	4.91	30.68	0.02	0.81	0.02	25.94	1.94	8.02
Fruit length (cm)	1.17	13.76	3.50	0.21	0.71	0.88	6.32	0.36	6.76	2.63
Fruit diameter (cm)	3.12	12.53	0.00	0.03	0.38	1.08	1.94	0.02	8.84	0.07
Fruit shell thickness (mm)	3.92	1.08	0.00	0.09	0.01	3.81	2.47	0.11	1.50	0.94
Seed length (cm)	4.21	4.11	8.43	3.04	0.75	2.34	5.86	1.04	11.60	0.29
Seed width (cm)	3.63	8.49	2.38	2.25	1.14	4.52	10.17	1.56	3.22	0.07
Seed thickness (mm)	2.61	2.10	5.01	3.68	1.07	1.10	2.42	4.36	8.56	0.06
100 seed weight (g)	6.01	6.10	6.61	3.33	1.48	4.88	8.83	4.23	7.70	0.10

number 100. Correlation between the EP and CC were confirmed by the Mantel test in which "r" could be in the range of -1 to +1. When "r" is close to -1 this indicates strong negative correlation between the EP and +1 indicates strong positive correlation. We found "r"= 0.974 (= normalized Mantel Statistic Z) indicating that there is very strong positive correlation between the EP and CC. The UPGMA dendrogram of CC revealed their genetic relationships (Figure 2). The dendrogram separated genotypes into two main clusters without any correlation with geographic origin and traits. The upper cluster on the dendrogram contained 50 accessions located closely. The lower cluster consists of the other highly diverse 50 genotypes. In cluster analysis, it was shown that grouping was not closely related with collection sites and phenotypically similar genotypes did not take place in the same group.

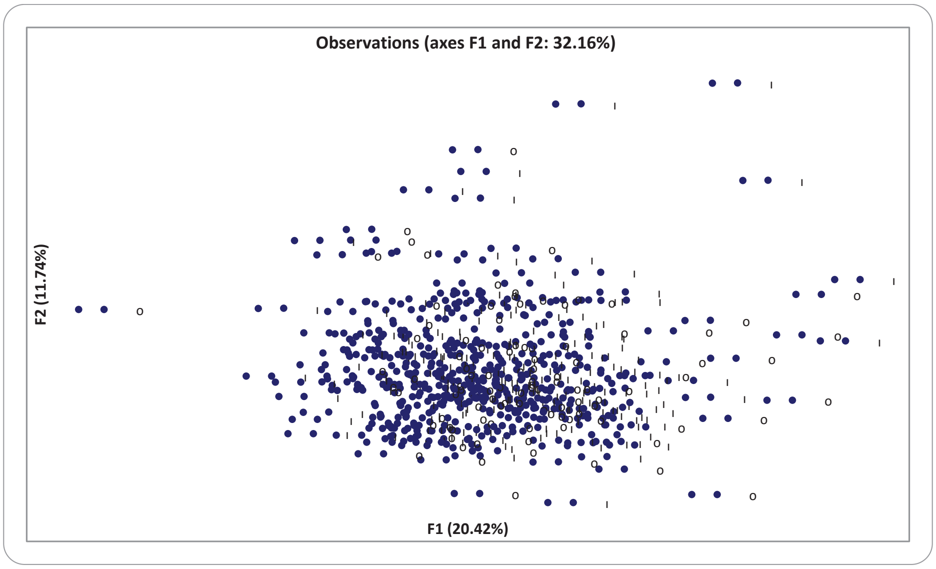

Figure 1- Principal component analysis all genotypes including the core collection collected from Turkey. The first principal component accounted for 20.42% of the variability and the second explained an additional 11.74%. 'o' represent the accessions of core collection and 'ı' represents remaining

Twenty-five quantitative and 21 qualitative traits of all bottle gourd germplasm were analyzed to establish principal components. The Figure 1. depicts the plot of the first two components of EP including CC. For the 25 qualitative traits, the first five principal components for EP and CC explained a total variation of 57.4% and 59.3%, respectively, and the percentage variability of the first four principal components is greater than seven from qualitative scoring (Table 3). The first principal component (PC1), predominantly of leaf attributes (leaf length and leaf width) and some flower traits (male flower length, male flower width and female flower width) explained 20.4% and 22.3% of the observed variation in the EP and core accessions, respectively. Considering the 21 qualitative variables, the first five principal components for the EP explained a total variation of 34.7%. PC1, predominantly of leaf attributes [leaf pubescence lower (23.8% of PC1 variation) and leaf pubescence upper (21.7% of PC1 variation)] and blossom-end fruit shape (11.4% of PC1 variation) explained 8.2% of the total variation. The results of PC analsis for 21 qualitative traits were not presented since qualitative characters were found to be less informative comparing quantitative characters. We have not observed distinct groupings based on geographical origin, plant growth traits nor fruit shape. Neither principle component analysis revealed any groping based on geographical origin.

In this study, phenotypic characteristics have been used for characterization and evaluation of Turkish bottle gourd germplasm with exotic accessions resulted in establishment of a CC representing genetic diversity of the EP. The CC concept was introduced by Frankel (1984) and further the concept was developed by Brown (1989) and van Hintum (1999). Creating a CC requires definition of materials, division of the entire group into sub-groups by stepwise and finally selecting of the entries from each group to establish a core group that represents the diversity of EP as well as possible (van Hintum 1999). It is interesting that this approach does not include geographical distribution although studies show that significance of geographical information as a criterion for

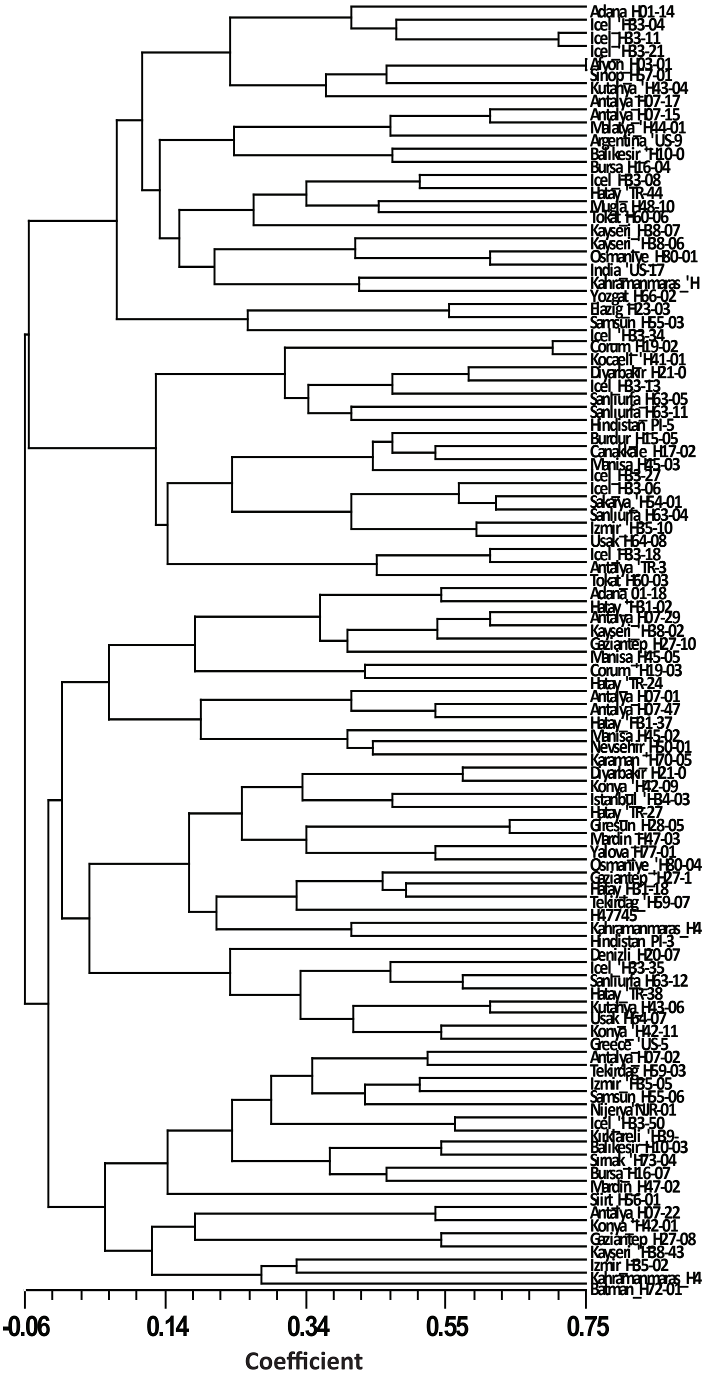

Figure 2- UPGMA depiction of core collection including 100 accessions. Name of the accessions shows source region followed by accessions number

establishment of CC (Rodrigues et al 2004; De Cristo-araújo et al 2015). We established a CC consisting of 100 accessions following generally the procedure for the selection of a core collection described by van Hintum (1999) and furthermore we included geographical distribution as a criteria during selection. The significant positive correlation (Mantel's "r"= "r"= 0.974) between EP and CC could be shown as successful accomplishment of CC of bottle gourd. Moreover, the high correlation between EP and CC was supported by PCA.

4. Conclusions

In the present study, Turkish bottle gourd accessions collected from all over Turkey and derived from USA and India were morphologically characterized and significant genetic diversity was determined between accessions despite the knowledge that Turkey is not the genetic origin of bottle gourds. Bottle gourd genotypes were characterized for 25 quantitative and 21 qualitative traits. It was concluded that quantitative traits were more discriminative than qualitative traits. Quantitative characteristics of cotyledon, leaf, flower, fruit and seed were found to be more discriminative than others traits. One hundred bottle gourd accessions were chosen based on the morphological characteristics investigated in this study to create a core collection representing the majority of the genetic variation of all genotypes within the entire Turkish bottle gourd germplasm collection.

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Abbreviations and Symbols					
cm	centimeter	kg	kilogram	N	Nitrogen
ha^{-1}	Per Hectare	m	meter	P_2O_5	Phosphorus pentoxide
K_2O	Potassium oxide	mm	millimeter	Subsp	Subspecies

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Investigation of the Temporal Variation of Water Quality in Ziyaret Pond Basin

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ABSTRACT

Urbanization, rapid population growth, intensive land use and land cover changes have negative impact on water quality. In this research, Amasya Ziyaret Pond's water basin flows' various physical and chemical specifications for 2014 water year (from 01 October 2013 to 29 September 2014) were studied. Temperature, pH, ammonium, nitrite, nitrate, potassium, sulphate, phosphorus, conductivity (EC) specifications of the basin pond flows were identified with the analysis of the water samples collected. Basin flows were estimated with the MIKE 11 NAM hydrological model. The variations between the instant flows values and parameters identified periodically in the research basin and the effect of basic management applications such as fertilization are identified. Eutrophication risk caused by phosphorus, nitrate, nitrite was identified.

Keywords: Surface water quality; Water pollution; Physical and chemical properties of waters; Ziyaret Pond

1. Introduction

Numerous issues such as industrialisation, population increase, rapid urbanization, improper land usage, climate change, excessive input use in agricultural production are central for the contemporary environmental problems, with detrimental effects on the sustainability of water resources.

A region's water quality and its water catchment basin's topography can be defined by evaluating the region's hydrogeology and hydrology together with the climate factors and human effects (Carpenter et al 1998; Jarvie et al 1998). In rural basins, surface waters are most susceptible to contamination since various pollutants such as fertilizers, pesticides, etc. are easily transported to streams, reservoirs and seas. Increasing intensity of agricultural activities and energy production based on water resources are fuelling the need for water (Graveline et al 2014; Bellin et al 2016). Intensive agricultural activities have also led to an increase in the use of pesticides and fertilizers and deterioration of surface water quality. While the applied nitrogenous fertilizers are directly transported, the phosphorous and potassium fertilizers can be carried away from the source together with the sediment. Rivers have the highest responsibility in hosting and transporting the pollutants (Ahmed et al 2011).

Many human-induced effects are affecting pollution in water resources negatively. The solution of the problem may be possible by monitoring the seasonal variation of the pollution in rural basins for sustainable planning and the appropriate management of land and water resources. Several studies have been carried out in different regions for this purpose. In a study carried out, monitoring the water quality in Cinarli county of Hafik province in Sivas, monthly water samples were taken from six stations during 2011-2013, the river found to be exposed to pollutants and reported to be at risk (Mutlu et al 2016). As a result of a study conducted in China in four different basins and with five water quality parameters, the water quality was significantly different in settlement and non-settlement areas. Water quality showed positive correlation with forestlands while presenting negative relation with impermeable and agricultural areas (Chen et al 2016).

In this research carried out in Amasya Ziyaret basin pond between 2013-2014, some chemical and physical properties of the basin flows were determined by the analysis of the instant water samples. The annual flow hydrograph of basin is combined with the measured water analysis values to determine the annual total chemical transport amount for the basin. The effects of basic soil management practices such as fertilization on basin are discussed. Water quality of the Amasya Ziyaret Pond, which was built for irrigation purposes, is tried to be determined according to water pollution control regulation (WPCR) and the possible negative effects of pollution elements on the irrigation area are investigated.

2. Material and Methods

2.1. Research location

Research was carried out in the Ziyaret Pond basin, located 4 km from the centre of Amasya province in Central Black Sea Region. Ziyaret Pond Basin is 28 km^2 and its water source is Degirmendere. The amount of organic matter content in the basin soil is medium and pH is neutral or slightly alkaline. The soil contains moderately lime and the $CaCO_3$ content increases towards the sub soil surfaces (KHGM 1991). As the basin is located in the inner part of the Central Black Sea region, transition from the temperate climate of the coastal area to the harsh climate of the inner part is dominant. The average annual precipitation is 443 mm (DMI 2016). The catchment basin consists of agriculture, pasture and forest areas. Orchards, forage and field crops cultivating are carried out on the agricultural lands. The fertilization trends in farmland of the basin have been determined as a result of land surveys and interviews with farmers. Nitrogen, phosphorus and potassium composite fertilizers are applied to fruit trees in autumn, while potassium nitrate and calcium nitrate based fertilizers are applied in spring. Wheat, winter barley, rye and triticale are planted October, applying phosphor and first half nitrogen fertilizers, and second half of the nitrogen fertilizer in spring at tillering stage, in the basin.

2.2. Water analysis

Within the 2014 water year, 18 water samples with 250 mL sterile plastic containers were taken from a permanent measuring point established in the Degirmendere stream reaching the pond. Water samples were delivered to the laboratory promptly and analysed within 3 hours from the collection.

pH, electrical conductivity (EC), ammonium, nitrite, nitrate, potassium, phosphorus and sulphate analyses were carried out for the water samples in the laboratory. EC measurements were realized by conductivity meter and pH measurements were performed with pH meter in the laboratory. Ammonium, nitrite, nitrate, potassium, phosphorus and sulphate contents were determined with suitable methods and test kits using DR5000 benchtop spectrometer (Hach-Lange Germany) (APHA 1998).

2.3. Determination of annual transportation

Basin flow data are needed to determine the annual total of various chemicals transported from the basin area. Since daily flows in the Basin were not measured, basin flows were estimated using the MIKE 11 NAM hydrological simulation model.

MIKE11 NAM is a rainfall-runoff model developed by Danish Hydraulic Institute (DHI 2009). The model is deterministic, lumped and conceptual rainfall-runoff model that calculate overland flow, interflow and base flow.

In the study, the precipitation-flow section of the model was used and MIKE 11 NAM data entries were realised in 3 groups: installation parameters (parameters related to watershed area and soil properties), model parameters (time constants and drift for surface flow, threshold values for subsurface flow and base flow), meteorological data (daily precipitation and daily potential evaporation). The installation and parameters required by the model are obtained from soil and topographic maps and field studies, while the precipitation and evaporation data are obtained from Amasya Meteorological Station data.

3. Results and Discussion

3.1. Some measured physical and chemical values of the Basin stream

The highest water temperature of the basin flows was measured as 22.4 °C in August and the lowest 10 °C in January. It was observed that the temperature of the water decreased from October to January, and increased from January to August. From August onwards, the water temperature had the tendency to autumn decrease. The solubility of salts in water is generally directly proportional with the increase in temperature (Temponeras et al 2000). Therefore, it is envisaged that the solubility of the chemicals transported from the basin to the Ziyaret Pond between April and October will be higher and more effective. When the temperature values are compared with Water Pollution and Control Regulation (Anonymous 1988), the water reaching the pond was in first class water quality in terms of temperature.

The pH values of basin flows were measured throughout the water year. The pH value of the water feeding the pond did not change very much during the autumn and winter months (8.42-8.56) remaining low. However, the pH value (8.88-8.97) tended to increase in the spring months when the flows increased, reaching the highest pH value (9.78) in August. This rise in water pH measured in summer can be attributed to increased air circulation in the water and the photosynthesis of phytoplankton communities consuming CO_2 present in the water. In winter, the declining trend of pH is due to the decrease in the flow and oxygen content, as well as the structure of the materials carried by the rain waters feeding the basin. The results of the study showing that the pH value is the lowest in November (6.79) and the highest in July (8.44) conducted by Alvarel-Rogel et al (2006) in Menor Lagoon in Spain, overlaps with the findings of the study we carried out in the basin. According to WPCR (2008), when evaluated in terms of pH value, the pond water quality was 2nd class in December and February, 3rd class in October, November, January, March, April, May and September while it was 4th class in June, July and August. The measured EC values of the water samples ranged from 692 µS cm^{-1} to 1221 µS cm^{-1}. The highest conductivity value was observed as 1221 µS cm^{-1} in October and 692 µS cm^{-1} at the lowest in July. Conductivity values were higher in autumn and winter compared to spring and summer months. Conductivity varies with concentration, mobility of existing ions present and water temperature (Garrison 1998). Higher stream flows in spring and summer months compared to autumn and winter months led to a decrease in EC values.

Ammonium was not detected in December, January and February at the Ziyaret Pond stream. The highest ammonium value was 0.284 mg L^{-1} in May. The ammonium values of the water feeding the pond increased from March, reaching the highest value (0.293 mg L^{-1}) in May and began to fall again. Ammonium values showed a rapid increase in the spring months (0.01-0.293 mg L^{-1}) and reached the highest value at the annual level. In winter months ammonium was not detected in the water, while in autumn months ammonium values observed to be close to each other (0.015-0.049 mg L^{-1}). Lopes et al (2006) reported similar findings in a study they conducted in Portugal, Ria de Aveiro lagoon observing the highest NH_4^+ values during the summer months and lowest in the winter months. The increase in ammonium values from spring can be attributed to several causes such as; a) increased temperatures creating higher microbial activity causing increased organic material decomposition resulting the appearance of the ammonium, b) conversion of nitrate to ammonium by deoxidation event, and c) mixing of organic fertilizers used in agricultural land during the periods of rain and irrigation, to the stream by the surface and subsurface flows. The ammonium content of the Degirmendere stream feeding Ziyaret basin was 1st class according to the WPCR (2008). In a study carried out by Odabasi & Buyukates (2009) in Saricay river between 8th July and 6th August, the NH_4^+ values observed between 0.04-3.68 mg L^{-1}. The NH_4^+ values of the Ziyaret Pond was found as 0.016 and 0.023 mg L^{-1} in July and August, respectively, which are lower than the NH_4^+ values obtained in the study conducted in Saricay river. Research carried out in the Lagoa de Araruama lake in Rio de Jenerio, Brazil, suggests that the ammonium content was in transition towards a eutrophic level (Souza et al 2003). Although the Ziyaret Pond ammonium content does not present a eutrophic hazard yet, it will be beneficial to consider the possibility of a transition towards eutrophic levels during rainy seasons, given that the study was conducted in an arid season.

The highest nitrite concentration was 0.73 mg L^{-1} in June and the lowest was 0.085 mg L^{-1} in March. The amount of nitrite feeding the pond started to increase (0.053 mg L^{-1}) from March and a sharp decline was seen after June. Seasonally, the lowest nitrite concentration is in winter while the highest values are reached in spring and summer months. The basin stream's water is classified as 4th class when the nitrite concentration is compared with the limit values of WPCR (2008). In a study carried out at Damsa Dam Lake, Mert et al (2010) identified the nitrite level within 3rd class waters, according to WPCR (2008). It can be argued that Damsa Dam lake has a better water quality in terms of nitrite level. Nitrite does not accumulate in the environment and is readily converted to nitrate since it is an intermediate product (Boyd 1990). However, in waters which nitrification is not sufficient, observing higher levels of nitrite is also possible (Secer 1997). Ziyaret Basin stream's nitrite concentration started to increase from the spring months. Increased leaching with increasing rainfall in the spring, the saturation of the soil and the decrease in O_2 concentration in the water constitute anaerobic conditions. Consequently, nitrite concentration in the environment increases because it could not be sufficiently oxidized to nitrate. With the decrease in precipitation and temperatures, the nitrite concentration of the Ziyaret basin level has decreased.

The highest nitrate value of the Ziyaret Pond stream was found as 22.6 mg L^{-1} in February and the lowest nitrate concentration was 8.15 mg L^{-1} in August. Seasonally, nitrate concentration in the waters was partially increased in the spring, autumn and winter periods and reached its lowest level in summer. The fluctuations in nitrate concentration in the spring can be attributed to two factors. The first one is related with the nitrate fertilizers used in agricultural fields mixing into stream. The second one is the decrease in the O_2 concentration of the water caused by the increase in precipitation and temperature, which further causes nitrite, insufficiently increasing to nitrate, transforming to nitrogen oxide under reducing conditions changing the nitrate concentration in the basin flows. Alvarez-Rogel et al (2006) reported an increase in nitrate concentration with the effect of agricultural areas around the lagoon, in their study of the Mar Menol lagoon in Spain. Ziyaret Pond stream's nitrate values varied between 2nd and 4th class water quality according to WPCR (2008). Protective measures are required to reduce the risk of nitrate-induced eutrophication in the pond.

The highest and lowest potassium value in the basin stream was found to be 26.6 mg L^{-1} in June and 0.22 mg L^{-1} in July, respectively. Potassium content was on the increase in spring months.

Despite the highest potassium value measured in June, it showed a sharp decline in July.

It was observed that although the concentration of potassium in the water has risen from July, the potassium content of the water falls with the beginning of autumn. The highest determination of potassium value in summer is considered to be due to the application of commercial fertilizers with potassium content to agricultural areas in summer, especially to fruit gardens, mixing with water and reached to the pond.

The highest and lowest phosphorus value of basin stream is found as 0.916 mg L^{-1} in September and 0.186 mg L^{-1} in June, respectively. Phosphorus content showed a significant increase while it was lowest during summer. When the phosphorus values identified in the Ziyaret Pond Basin were compared with the WPCR (2008) values, the lowest and highest water quality were within the range of 3rd class and 4th class (0.916 mg L^{-1}), when the lowest (0.186 mg L^{-1}) and highest (0.916 mg L^{-1}) values were taken into consideration, respectively. Souza et al (2003) observed in a study realised in Brazil that most of the changes in the inorganic phosphorus amount were due to human activities. Similarly, a significant part of the changes in phosphorus content reaching to Ziyaret Pond are due to commercial fertilizers applied by people to agricultural lands. In the autumn months, the amount of phosphorus started to increase as a result of the phosphorus-containing commercial fertilizers mixing into the pond feeding water. The amount of phosphorus in the soil is reduced when the plants begin to absorb the phosphorus from the soil into the plants by spring. Thus, the amount of phosphorus reaching the pond through surface runoff and leaching was reduced. Furthermore, the decrease in the amount of adsorbed phosphorus transported to the pond with the sediment, due to low surface flows during the arid summer months is also effective in this reduction. Thus, it is observed that the amount of phosphorus in Degirmendere has started to fall rapidly from the last months of spring.

The highest and lowest sulphate values were observed as 25.2 mg L^{-1} in September and as 11.9 mg L^{-1} in January, respectively. The sulphate concentration in the water continued to decline from October to January. The SO_4^{-2} amount in the water feeding the pond began to increase from winter to spring. The solubility of gypsum ($CaSO_4.2H_2O$) in the soil increases with increasing temperatures. Gypsum with increased solubility, leached with the spring rainfall mixing to the water feeding the pond in the form of SO_4^{-2} caused this increase. According to WPCR (2008), the sulphate content of the Ziyaret Pond Basin is in the 1st class water quality. Sen & Golbasi (2014) in the study they carried out at Hazar Lake-Kurk Stream during the seven-month period when the Stream was in flow, found the average SO_4^{-2} value as 20.375 mg L^{-1}. The SO_4^{-2} concentration of Ziyaret Pond was found to be 16.958 mg L^{-1} in 12 months period, which is lower than the study conducted in Kurk Stream.

3.2. Seasonal and yearly total transport values to the pond with the basin stream

In the scope of the study, some physical and chemical properties of surface water were determined with water samples taken from basin stream in 18 different periods in 2014 water year. The amount of flows in the basin must be known in order to be able to determine the annual total chemical transport reaching the pond. Basin flow amount should be known in order to identify the annual chemical transport reaching the basin.

Mike 11 Nam Hydrological model is used in the study to identify the daily flow amount of the basin. NAM is prepared with 9 parameters, representing surface zone, subsurface zone and ground water storage. Description of the model parameters for Degirmendere stream and their effects is presented in Table 1. The application of MIKE 11 model for rainfall runoff estimation was realized in two stages in the study. The first stage, in which the calibration process was realized to determine optimum values of the model parameters. In the second stage the streamflow was simulated the streamflow using the estimated model parameter during the calibration process. In the calibration procedure, several model parameters were adjusted by way of applying trial and error method to obtain optimum values. There were not daily flow records in the basin to optimize the model parameters and check the validity of the model estimation. In order to test the model success, the basin stream flows measured on five different dates; on 05th and 29th October 2013, 09th and 23rd November 2013 and 23rd September 2014, were compared with the same day model prediction results for the same dates. Stream flows observed and simulated on five different occasions were presented as 0.00658 $m^3 s^{-1}$ - 0.00022 $m^3 s^{-1}$; 0.00694 $m^3 s^{-1}$ - 0.00108 $m^3 s^{-1}$; 0.00704 $m^3 s^{-1}$ - 0.00542 $m^3 s^{-1}$; 0.00602 $m^3 s^{-1}$ - 0.00458 $m^3 s^{-1}$; 0.00463 $m^3 s^{-1}$ - 0.07599 $m^3 s^{-1}$ respectively. Observed and simulated flows were compared statistically by, two non-parametric approaches, called Kolmogorov-Smirnov and Shapiro Wilk, were applied to all of the observed and predicted data, for normality analysis. Their results showed that the full data set was not normally distributed. Therefore, the Mann-Whitney test was used to assess whether or not the two data sets (observed flows and predicted flows time series) obtain from the same population. The method implied that the median of them is not statistically different each other.

The flow hydrograph of 2014 water year predicted according to the annual simulation result realised with Mike 11 Nam model is given in Figure 1. According to the model estimates, daily basin flows showed a decrease in the autumn months, which was around 50 L s^{-1} in winter months, and increasing with the melting snow and precipitation received in spring, its highest estimated flow value of 383.4 L s^{-1}, was on 30.04.2014. There are no periods without flow in the basin.

In order to calculate the total amount of ammonium, nitrite, nitrate, potassium, phosphorus and sulphate reached Ziyaret Pond annually, analysis results determined on the water samples' collection dates, are interpolated for the period between two measurement dates and converted to daily value comprising 365 days. To calculate the total daily chemical transportation, daily estimated water analysis results are multiplied by the flow amount

Table 1- Different parameters of the NAM model

NAM parameter description	*Effect*	*Parameter value*
Area of catchment, km^2	Volume of water yield	28
Maximum water content in surface storage (U_{max}), mm	Overland flow, infiltration, evapotranspiration, interflow	10
Maximum water content in root zone storage (L_{max}), mm	Overland flow, infiltration, evapotranspiration, base flow	100
Overland flow runoff coefficient (C_{QOF}), dimensionless	Volume of overland flow and infiltration	0.5
Time constant for interflow(C_{KIF}), hours	Drainage of surface storage as interflow	1000
Root zone threshold value for overland flow (T_{OF}), dimensionless	Soil moisture demand that must be satisfied for overland flow to occur	0.5
Root zone threshold value for interflow (T_{IF}), dimensionless	Soil moisture demand that must be satisfied for interflow to occur	0.5
Groundwater recharge threshold (TG), dimensionless	Soil moisture demand that must be satisfied for groundwater recharge to occur	0
Time constant for routing overland flow (C_{K1}), hours	Routing overland flow along catchment slopes and channels	10
Time constant for routing inter flow (C_{K2}), hours	Routing interflow along catchment slopes	10
Timing constant for base flow (C_{KBF}), hours	Routing recharge through linear groundwater recharge	2000

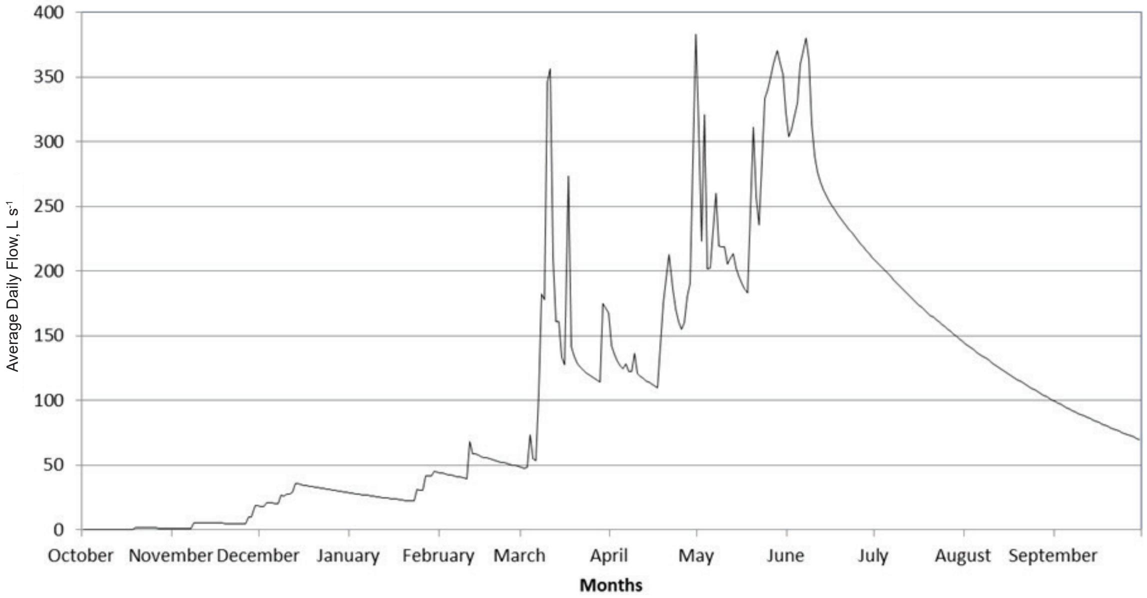

Figure 1- Ziyaret Pond basin 2014 water year hydrograph

and time. Total annual transport is determined by adding calculated daily values (Table 2).

Total ammonium, nitrite and nitrate nitrogen forms reaching the pond varied seasonally. Ammonium is transported from high to low in the spring, summer, autumn and winter. Nitrite has been transported more during the spring and summer seasons, respectively, and has been transported in

Table 2- Monthly and seasonal transportation values for amonium, nitrit, nitrate, potassium, phosporus, sulphate (kg)

Parameters	*Autumn*				*Winter*			
	September	*October*	*November*	*Total*	*December*	*January*	*February*	*Total*
Ammonium (NH_4^+)	4.02	0.03	0.24	4.29	0.03	0.00	0.00	0.03
Nitrite (NO_2^-)	19.59	0.20	1.46	21.25	7.73	5.28	7.98	20.99
Nitrate (NO_3^-)	2556.75	28.29	255.85	2840.89	1557.96	1589.45	2718.53	5865.93
Potassium (K^+)	2243.09	9.19	23.06	2275.35	71.40	56.49	103.74	231.63
Phosphorus (P_2O_5)	101.47	0.73	6.32	108.52	35.70	35.55	56.44	127.69
Sulphate (SO_4^{-2})	4924.66	29.23	226.83	5180.72	1006.46	921.79	1612.51	3540.76
Parameters	*Spring*				*Summer*			
	March	*April*	*May*	*Total*	*June*	*July*	*August*	*Total*
Ammonium (NH_4^+)	9.93	52.43	339.39	401.75	180.95	7.42	7.13	195.50
Nitrite (NO_2^-)	37.83	186.82	429.91	654.56	597.76	15.04	16.68	629.48
Nitrate (NO_3^-)	6835.01	9339.58	22001.55	38176.14	15782.58	4875.15	2944.08	23601.81
Potassium (K^+)	914.53	3888.96	10139.78	14943.27	21137.52	98.05	1936.31	23171.89
Phosphorus (P_2O_5)	187.26	259.04	560.73	1007.02	478.05	219.46	151.58	849.09
Sulphate (SO_4^{-2})	6495.51	11320.54	17507.47	35323.52	17526.01	7182.84	6123.84	30832.68

close quantities in the autumn and winter months. Nitrate transport was mostly in spring and summer, followed by winter and autumn. Nitrate has been the most transported among the nitrogen forms followed by nitrite nitrogen. Nitrogen transport in the form of ammonium was less. It is estimated that the low transport soil in the form of ammonium is created with the effect of fixing ammonium nitrogen. The potassium transported to the pond with the surface waters occurred most in summer followed by spring months. While, increased surface flows with the snow melting during the spring months, are effective with the transportation of potassium, more potassium was transported in the summer months, when the flows were relatively low, due to more intensive use of potassium fertilizers applied to agricultural areas in the spring. Phosphorus transportation was observed during spring, summer, winter and autumn months, from high to low, respectively. Existing phosphorus transport is at a level creating eutrophication risk in the pond and precautions are required.

4. Conclusions

The water temperature values of Degirmendere have changed in parallel with the air temperature, however did not exceed quality water upper limit of 25 °C. The pH values were strongly alkaline in October, November, December, January, February, March, April and May while very strongly alkaline in June, July, August and September. According to the "Water Pollution Regulation", the pH value of the water was 3rd class in October, November, December, January, February, March, April and May, while it was 4th class in June, July, August and September.

Particularly SO_4^{-2} has been transported (74877.68 kg $year^{-1}$) to Ziyaret Pond from its basin. NO_2^- 1326.28 kg $year^{-1}$, NO_3^- 70484.78 kg $year^{-1}$ and NH_4^+ 601.57 kg $year^{-1}$ reached the pond. The annual transport values of K^+ and P_2O_5 have been identified as 40622.14 kg $year^{-1}$, 2092.31 kg $year^{-1}$, respectively.

Degirmendere has 3rd class water quality according to K^+, NH_4^+ ve SO_4^{-2}, and 4th class water quality according to NO_2^- content.

According to the results of this research, it can be said that there is a risk of pollution and eutrophication for the pond in terms of NO_2^- content, the phosphorus content carries potential risk, and there is no significant pollution risk in terms of other chemical parameters discussed. In order to prevent water pollution in the pond and to sustain the ecological balance, it is recommended to take a holistic approach for the pollutants and pollution problem in the basin and at non-basin level. Sewerage network should be built in places where there is none in the basin. As the pond and its surroundings are used as a picnic area especially in the summer months, the amount of solid waste around the pond is increasing. For this reason, the solid wastes that accumulate around the pond should be gathered regularly as much as possible and people gathering for recreational purposes such as picnic should be informed about this. The use of fertilizers and pesticides in the basin should be taken under control. The amount and quality of the fertilizers used in agricultural areas should be determined according to the nutrient value of the soil, the plant species to be grown, while preventing excessive use of fertilizers.

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The Impact of Soil Conditioners on Some Chemical Properties of Soil and Grain Yield of Corn (*Zea Mays* L.)

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ABSTRACT

The goal of this study was to explore the effects of various soil conditioners on selected properties (pH; organic matter, OM; electrical conductivity, EC; cation exchange capacity, CEC) of a *Xerofluvent* soil and corn yield. During the experiment, four amendments were applied in an experimental set of plots: tobacco waste compost (TWC), poultry manure (PM), bio-humus (BH), and chemical fertilizer (NPK). Soils were treated with TWC at the rate 50 t ha^{-1}, PM at the rate 4 t ha^{-1}, BH at the rate 10 t ha^{-1} and NPK at the rate 300 kg ha^{-1}, respectively. All organic conditioners were increased soil pH, OM, CEC, EC and corn yield when compared with the control soil and these parameters have been changed from 5.7% to 333%. The most effective soil conditioners were determined as tobacco waste compost, bio-humus, and poultry manure. The findings of current study suggest that 50 t ha^{-1} TWC should be added to soil as a priority for improving properties of a *Typic Xerofluvent* soil and crop yield.

Keywords: Bio-humus; Poultry manure; Soil chemical properties; Tobacco waste; Yield

1. Introduction

Nowadays, the use of organic treatments for soil nutrient improvement is getting important for sustainable productivity and soil nutrient management. The decline of organic matter in soil, as a consequence of intensive soil cultivation practices, has been identified as one of the most important threats to soil quality (Lal 2007; Batlle-Bayer et al 2010). Use of organic manures alongside chemical fertilizers often lead to increased soil organic matter, soil structure, water holding capacity and improved nutrient cycling and helps to maintain soil nutrient composition, cation exchange capacity and biological activities (Saha et al 2008). While chemical fertilizers are important input to enhance crop productivity, over reliance on chemical fertilizers is associated with decline in some soil properties and crop yields over time (Hepperly et al 2009). Hence, integrated use of chemical fertilizers with organic manures is a sustainable approach for efficient nutrient usage which enhances efficiency of the chemical fertilizers while reducing nutrient losses (Schoebitz & Vidal 2016). Compost amendments could contribute significantly to the improvement of the soil organic carbon content in the long term (Barral et al 2009) and hence to the chemical (nutrients), physical (structure and moisture retention) and biological (soil life) quality of the soil (Herencia et al 2011; Odlare et al 2011; Ozores-Hampton et al 2011). Plant residuals can be used as a nutrient supplement in the agricultural fields. The kinetics of plant residual decomposition in soil and their carbon and nitrogen mineralization are largely influenced by the quality of the plant materials, i.e. by their origin and composition (Heal et al 1997). Animal manures may contribute to improving the physical and biological properties of soil (Li & Han 2016) and are important source of Ca, Mg, S, and micronutrients; they contain only low and highly amounts of N, P, and K (Odedina et al 2011). Among the different sources of organic manure which have been used in plant production, poultry manure is found to be the most concentrated in terms of nutritional status (Obire & Akinde 2006). The efficiency of compost usage in agriculture mostly depends on the quality of the compost which is closely related to its stability and maturity. Some physico-chemical properties (pH, temperature, C:N ratio, cation exchange capacity, total organic C, NH_4^+, phenols, humic-like substances) have been used to control compost quality. Nevertheless, it is difficult to add these parameters across a wide range of composts prepared from different organic wastes (Kayıkçıoğlu & Okur 2011). Tobacco is an important agricultural plant in the Aegean region of Turkey and 60.5% of Turkey's total tobacco production is grown in this region according to 2016 data (TUIK 2017). Tobacco plant residues from the primary production and cigarette manufacture are classified as an agro-industrial waste. This waste contains high amounts of organic matter and nicotine and is known to be a toxic and hazardous compound if the nicotine content exceeds 500 mg kg^{-1} dry weight (Wang et al 2004; Piotrowska-Cyplik et al 2009). In this investigation, tobacco waste compost (TWC), poultry manure (PM), bio-humus (BH), and chemical fertilizer (NPK) at different ratios was applied to soil and the influences of these conditioners on selected chemical properties of a sandy loam soil and corn yield were compared.

2. Material and Methods

2.1. Site features and properties of conditioners

The experiment was laid out at the Agricultural Research Farm of Ege University in Menemen, Izmir, Turkey (38°58′35.51″-38° 58′36.03″N; 27°03′84.56″-27°03′89.81″E). The aim of this study to compare the role of different materials on soil chemical properties and corn yield. The soil at the study site is characterized by sandy loam texture with slightly alkaline reaction and classified as a *Typic Xerofluevent* (Soil Survey Staff 2006). The general properties of the soil are shown in Table 1. Experimental treatments were as follows: (1) Control soil, C (No treatment); (2) Poultry manure, PM at 4 t ha^{-1} plus NPK fertilizer at 300 kg ha^{-1}; (3) Bio-humus, BH at 10 t ha^{-1} plus NPK fertilizer at 300 kg ha^{-1}; (4) NPK fertilizer at 300 kg ha^{-1}; (5) Tobacco waste compost, TWC at 50 t ha^{-1}. Since tobacco waste compost had very rich organic matter and nutrient content, it was applied to the soil (at a higher dose than other materials) without NPK. Moreover, some researchers reported that tobacco waste compost had positive responses to soil properties and yield without any mineral fertilizer (Okur et al 2008; Cercioglu et al 2012). The plot sizes were 5 m x 3 m and replicated four times according to a randomized block design. The test plant grown on the study field was corn (*Zea Mays* L.), planted during April-May-June and harvested in October of each growing season. The organic treatments [tobacco waste compost (TWC), poultry manure (PM) and bio-humus (BH)] were applied to the soil only once at the beginning of the experiment (first year). Some properties of these materials are given in Table 2 and Table 3. Tobacco wastes obtained from Izmir Kemalpaşa Socotab Factory were added to soil after composting process. Composting of tobacco waste was performed outdoor under a roof. The moisture content of the compost was analyzed approximately 55% by weighing the material regularly and adding water when necessary. Aeration was made by manual turning during the composting. After 3 months, when the temperature of the compost decreased to the ambient level, composting was completed. Both of bio-humus (composted plant residues) and poultry manure were gathered from organic manure industry. All rates of treatments were determined according to initial soil analysis results, uptake of nutrients by plant, and recommendations from producers of organic manure. Moreover, some additional chemical fertilizers (ammonium sulfate, triple superphosphate, and ammonium nitrate) were applied, and drip irrigation method was used in the study.

2.2. Soil sampling and analytical determinations

Two soil samples (0-20 cm) were collected (planting and harvest period) each growing season from the center of each plot. To determine the initial physical and chemical properties of the soil, soil samples were air-dried and passed through a 2-mm sieve prior to analysis. Particle-size distribution was determined according to Bouyoucos (1962) and porosity was determined according to Danielson & Sutherland (1986). Organic matter concentration (Nelson & Sommers 1982), pH (Jackson 1967), electrical conductivity (Rhoades 1996) and cation exchange capacity (Rhoades 1982a) were determined. Calcium carbonate was measured by the Scheibler method (Tüzüner 1990). Total N was analyzed by the Kjeldahl method and available K, Ca, Mg and Na were determined by the 1 N NH_4OAc (pH: 7) method. Ca, K and Na were determined by flame emission spectrometry and Mg was determined by flame atomic absorption spectrometry (AAS) (Kacar 1995). Available P was determined by the Mo blue method in a $NaHCO_3$ extract (Olsen & Sommers 1982). Available Fe, Cu, Zn and Mn were obtained with 40 mL DTPA+$CaCl_2$+TEA extract method and were found by atomic absorption spectrometry (Lindsay & Norvell 1978). Grain yield measurements were performed by 10 crops from each plots and the values were calculated by measuring total weight, corncob weight, and grain weight in t ha^{-1}.

Table 1- Some soil properties for the Ege University Menemen Agricultural Research Farm study site (sandy loam)

Depth (cm)	*Sand (%)*	*Silt (%)*	*Clay (%)*	*pH*	*EC (dS m^{-1})*	*OM (%)*	*$CaCO_3$ (%)*	*Total N (%)*
0-30 cm	55.28	36.00	8.72	7.78	0.72	1.11	4.70	0.07

Table 2- Main chemical quality characteristics of tobacco waste compost (TWC), poultry manure (PM), and bio-humus (BH)

Material	*pH*	*EC (dS m^{-1})*	*OM (%)*	*C:N*	*$CaCO_3$ (%)*
TWC	9.18	49.50	33.60	22.40	7.06
PM	8.60	54.50	44.90	25.80	12.00
BH	7.88	9.20	46.50	29.30	26.00

Table 3- Nutrient status of tobacco waste compost (TWC), poultry manure (PM), and bio-humus (BH)

Material	*%*					*ppm*				
	N	*P*	*K*	*Ca*	*Mg*	*Na*	*Fe*	*Cu*	*Mn*	*Zn*
TWC	0.87	0.27	1.94	7.44	0.63	794.80	14500.00	119.00	442.00	124.00
PM	1.01	0.34	2.19	9.44	1.20	5663.00	2200.00	72.20	536.30	648.60
BH	0.92	0.20	0.69	11.76	0.92	993.50	12400.00	34.80	433.80	86.14

2.3. Statistical analysis

Analysis of variance (ANOVA) and Duncan's tests were performed with a $P \leq 0.05$ significance level and 95% confidence interval using the statistical package, SPSS Statistics 25.

3. Results and Discussion

3.1. Soil chemical properties

Soil pH values were significantly affected by all the treatments and varied between 7.49 and 7.92 ($P \leq 0.05$, Figure 1a). Soil under TWC treatment had significantly higher pH values than the other treatments. The highest pH (7.92) was observed with an increase of 3.6% over the control in the first growing season. There were no significant differences between the PM and BH treatments in the first and third growing season ($P \leq 0.05$). The study conducted by Mabuhay et al (2006) agreed with these results; they found that soil pH increased when organic and chemical fertilizers were applied to agricultural lands. Nastri et al (2009) observed very slight soil pH response to addition of either organic or inorganic fertilizers. Giannakis et al (2014) reported that compost application increased soil pH from 7.80 to 8.10 and 8.20 in the 50 and 100 t ha^{-1} application rates, respectively, at the 0-15 cm soil layer. Soil electrical conductivity (EC) was significantly different among all the treatments and varied between 0.45 and 1.95 dS m^{-1} ($P \leq 0.05$, Figure 1b). PM and TWC treatments showed same significant effect on soil EC values in the second growing season; TWC and NPK treatments also showed same significant effect in the third growing season ($P \leq 0.05$). Release or solubilization of ions during compost incorporation may have resulted in the increased values of EC observed at the beginning of the study. Applying composted tobacco waste and poultry manure to soil raised the EC due to high level EC values of these materials (TWC: 49.50 dSm^{-1}; PM: 54.50 dSm^{-1}). Several researchers reported that addition of organic manure and compost to the soils significantly increased electrical conductivity (Candemir & Gulser 2011; Morugan-Coronado et al 2011; Cercioglu et al 2012). Addition of TWC, BH and PM were increased significantly soil organic matter (OM) at each growing season (Figure 2a). According to the results, OM values were significantly ($P \leq 0.05$) greater (124%) in the TWC treatment when compared with the control treatment in 2009. Third growing season showed the greatest OM values (2.51 and 2.45%) by the PM and TWC treatments among all the growing seasons. The soil OM content was increased because of high OM content of organic materials (see Table 2). The increase in the levels of soil OM was expected, since, organic sources have the ability of increasing soil OM content (Ojeniyi 2000). Cercioglu et al (2012) reported that addition of composted tobacco waste on a loamy soil also increased soil organic matter content. Nevertheless, some studies showed that OM increases were temporary, since the organic material is rapidly mineralized by soil microorganisms (Mechri et al 2007; Di Serio et al 2008). Cation exchange capacity (CEC) is defined as the measure of the total capacity of a soil to hold exchangeable cations and indicates the negative charge present per unit mass of soil (Peverill et al 1999). With application of all the treatments, CEC values varied between 114.1 and 196.6 cmol (+) kg^{-1} (Figure 2b). The greatest CEC value was obtained as 196.6 cmol (+) kg^{-1} in the second growing season by increasing the rate 31% in the BH treatment. The TWC treatment also showed greater CEC values (124 and 145.6 cmol (+) kg^{-1}) in the first and third growing seasons. The increase in CEC of soil, as the result of organic material addition, has been reported by several researchers (Qian et al 2004; Jien & Wang 2013).

3.2. Corn yield

Grain yield values significantly varied between 6.62 and 24.36 t ha^{-1} by all the treatments ($P \leq 0.05$, Figure 3). Poultry manure treatment had the greatest yield (24.36 t ha^{-1}) value in the first harvest with an increase of 42.3% over the control. Since the second and third growing season had an extreme dryness, yield results decreased during this time of the study. The addition of PM and TWC to soils significantly increased grain yield ($P \leq 0.05$). Positive

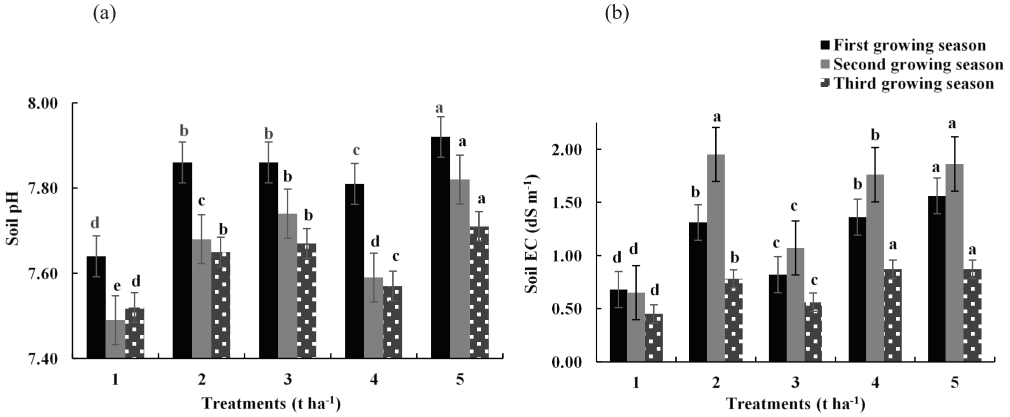

Figure 1- Changes in (a), soil pH; (b), soil EC (dS m^{-1}). Treatments 1, control soil; 2, poultry manure (4 t ha^{-1}) +NPK (300 kg ha^{-1}); 3, bio-humus (10 t ha^{-1})+NPK (300 kg ha^{-1}); 4, NPK (300 kg ha^{-1}); 5, tobacco waste compost (50 t ha^{-1}). The error bars represent the mean±SE of four replicates (n= 4). The letters presents significance levels among treatments for each growing season according to Duncan's test ($P \leq 0.05$)

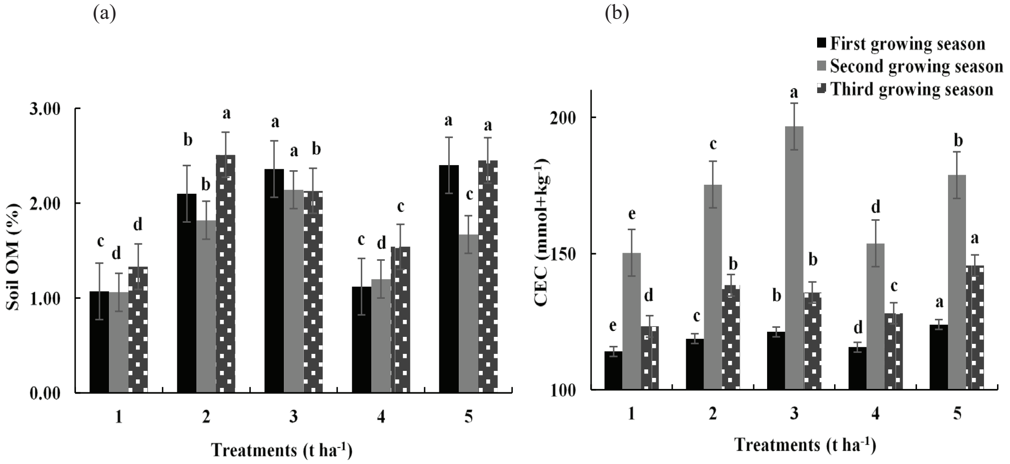

Figure 2- Changes in (a), soil OM (%); (b), soil CEC (cmol+kg^{-1}). Treatments 1, Control soil; 2, Poultry manure (4 t ha^{-1})+NPK (300 kg ha^{-1}); 3, Bio-humus (10 t ha^{-1})+NPK (300 kg ha^{-1}); 4, NPK (300 kg ha^{-1}); 5, tobacco waste compost (50 t ha^{-1}). The error bars represent the mean ± SE of four replicates (n= 4). The letters presents significance levels among treatments for each growing season according to Duncan's test ($P \leq 0.05$)

yield responses in various plants to the addition of composted tobacco waste have been mentioned in several studies (Jakubus & Czekala 2002; Cercioglu et al 2012; Cercioglu 2017). Ojeniyi & Adeniyan (1999) reported that poultry manure can effectively improve soil fertility, yield and nutrient composition of plant. Similarly, Garg & Bahla (2008) found that higher grain yield with increased poultry manure could be because of balanced nutrients supply throughout the growth and development stages of plant. Studies conducted by Ayoola & Makinde (2009), obtained greater grain yield in poultry manure treatments and lower in chemical fertilizer and control treatments. Poultry manure and chemical fertilizer combinations can improve the efficiency of nutrients uptake and availability to plant (Warren et al 2006). Similar yield results found in the current study has also occurred in other studies.

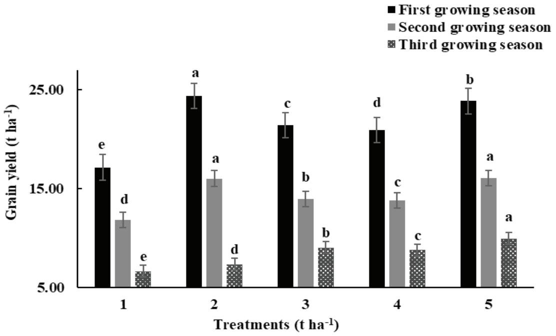

Figure 3- Changes in grain yield (t ha^{-1}). Treatments 1, Control soil; 2, Poultry manure (4 t ha^{-1})+NPK (300 kg ha^{-1}); 3, Bio-humus (10 t ha^{-1})+NPK (300 kg ha^{-1}); 4, NPK (300 kg ha^{-1}); 5, tobacco waste compost (50 t ha^{-1}). The error bars represent the mean±SE of four replicates (n= 4). The letters presents significance levels among treatments for each growing season according to Duncan's test (P≤0.05)

4. Conclusions

Generally, organic materials added to the soil significantly (P≤0.05) enhanced soil pH, electrical conductivity, organic matter content, and cation exchange capacity compared to the control soil. Improvement of the soil chemical properties is important for plant yield especially for Izmir where its plant and fruit production are prominent among agricultural industries of Turkey. The benefited methods and results from this study have demonstrated that soil chemical properties are changed significantly by addition of different organic treatments. Similarly, grain yield of corn was changed by applying these amendments. However, occasionally using these materials may cause some problems. For instance, high salinity of poultry manure is the most important factor limiting the use of it. It is therefore recommended to apply to the soil after analyzing the salt content of poultry manure. Additionally, due to the fact that experiment soil has high sand content, provides permeable structure and the applications do not cause a soil pollution problem. Nevertheless, they might cause pollution of groundwater. Hence, these materials should be added to the soil (tobacco waste compost, poultry manure and bio-humus) for improving soil properties of a *Typic Xerofluvent* soil.

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Biochemical Characterization of Fig (*Ficus carica* L.) Seeds

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ABSTRACT

Fig fruit has been a typical component of the health-promoting Mediterranean diet since centuries. One of the fig fruit's parts responsible for health effects is fig seeds that do not come to mind. Firstly, the proximate compositions of Sarilop fig seed cultivars were determined in this study. It was seen that the fig seeds were rich in oil and carbohydrate contents. Then, the mineral contents of fig seeds were analyzed by UV-vis spectrophotometer and an atomic absorption spectrometer. The major minerals in fig seeds were found as Ca, K and P. Moreover, the fatty acid compositions of the seeds were evaluated by gas chromatography. The fig seed oil had greater amount of the unsaturated fatty acids than saturated fatty acids. The chemical compositions of fig seeds are presented for the first time in this study.

Keywords: Fatty acids; *Ficus carica* L.; Fig seeds; Mineral content

1. Introduction

Fig is a fruit, which belongs to the Moraceae family and has more than 750 varieties (Guvenc et al 2009). Among them, *Ficus carica* L. is an edible fruit species. Figs can be consumed as raw, dried, canned, or in other preserved forms (Mawa et al 2013). Commercial producers of figs are in countries such as Turkey, Egypt, Morocco, Spain, Greece, California, Italy, Brazil, and other countries with hot dry summers and mild winters. Turkey leads in the fig production with 30% of the world fig production (Nakilcioğlu & Hişil 2013; Mehta et al 2014). It is an important constituent of the Mediterranean diet due to its energy, minerals, dietary fiber, amino acids and antioxidants compounds such as phenolic compounds and vitamins contents (Oliveira et al 2010; Bucić-Kojić et al 2011; Amessis-Ouchemoukh et al 2017). The fruits, root and leaves of fig trees are known for quite a number of therapeutic properties such as their use in traditional medicine for the treatment of cardiovascular, respiratory (sore throats, coughs, and bronchial problems) and gastrointestinal (colic, indigestion, loss of appetite, and diarrhea) disorders and as anti-inflammatory and antispasmodic remedy (Mawa et al 2013; Amessis-Ouchemoukh et al 2017).

One of the attractive parts of whole figs effecting both their nutritional values and health effects is their seeds. The interior portion of fig is a white, inner ring containing a seed mass bound with jelly-like flesh (Badgujar et al 2014). Fig seeds vary greatly in size such as large, medium, small, or minute and their number is from 30 to 1600 per fruit (Lansky et al 2008; Badgujar et al 2014). For example, the mean of seed number per a fruit is 420 and 1000 seed weights are 1.28 g for 'Sarilop' fig cultivars (Çalişkan et al 2012). In a fig, there are numerous edible seeds and they are usually hollow, unless pollinated. The characteristic nutty taste of dried figs is also provided from their seeds. Nowadays, the fig seeds may be used as edible oil and lubricant source (Badgujar et al 2014).

Fig is a fruit that is consumed together with many seeds found in its jelly-like flesh, with or without its peel. For this reason, it is considered that the fig seeds contribute considerably to the nutritional composition of fig. The aim of the present study is to determine the chemical properties and nutritional composition of fig seeds obtained from different fig cultivars produced in Turkey. This study may contribute to improvement and enhancement of the information contained in the literature about chemical and nutritional properties of figs. Moreover, this study is very important in terms of the fact that the chemical composition of fig seed is the first to be enlightened.

2. Material and Methods

2.1. Materials

2.1.1. Samples

In this study, Sarilop fresh figs grown in three different locations (Incirliova, Germencik, Nazilli) were bought from local markets in Turkey. They were chopped into a water bath and their seeds were extracted. After the extracted seeds were washed several times with water, they were left to dry at room temperature for one week (Engin et al 2011). The obtained fig seeds were stored at 4 ºC until analysis.

2.2. Methods

2.2.1. Proximate composition analyses

The chemical compositions (moisture, ash, oil and protein) of fig seeds were determined by the official methods reported by the Association of Official Analytical Chemists (AOAC 2000). The carbohydrate contents of the samples were calculated by subtracting their moisture, ash, protein and fat contents from 100. The proximate compositions were expressed as a percentage.

2.2.2. Mineral analyses

2.5 g of the fig seeds were weighed in the porcelain crucible and incinerated at 550 C in a muffle oven during 8-10 h. The ashes that are white-grayish in color were slightly moistened and dissolved with three parts of 2 mL of 6 N hydrochloric acid. Then, they were filtered into a 25 mL volumetric flask. After the filter was cleaned three times with 3 mL of deionized water that was also added to the volumetric flask, it was filled with deionized water until level (López et al 2008). Mineral analyses were applied by using the obtained solution. The Mg, Zn, Fe, Cu, Na, K, Ca and Mn measurements were performed with a Model contrAA 700 high-resolution continuum source atomic absorption spectrometer (Analytik Jena, Jena, Germany), with a flame atomize. The amounts of P in the samples were determined by UV-vis spectrophotometer (Optizen Pop, Mecasys Co., Ltd., Korea) according to AOAC method 965.17-1966 (AOAC 1996). The mineral contents were expressed as mg of mineral per kg for samples.

2.2.3. Fatty acids analysis

The oil of fig seeds was extracted at room temperature with n-hexane, and then, was methylated directly according to IUPAC method 2.201 (Paquot & Hautfenne 1987). The compositional analysis of fatty acid methyl esters was performed by using a gas chromatograph (Agilent 7820A) equipped with a flame ionization detector and a capillary column (Agilent DB-23, 30 m x 0.25 mm I.D, 0.25 μm film thicknes) according to the modified method of Yıldız-Turp & Serdaroğlu (2008). The carrier gas is H_2, which was used at a flow rate of 1 mL min^{-1}. Sample was injected (1 μL) with a split mode (ratio 50:1). Injector temperature and detector temperature were adjusted at 225 ºC and 250 ºC, respectively. Column oven temperature increased from 100 ºC (hold 4 min) to 240 ºC at a rate of 3 ºC min^{-1}, and kept at 240 ºC for 15 min. Fatty acids composition of samples was identified with retention times of individual fatty acid obtained from commercial standard (Restek, Food industry FAME mix-37 components, cat.# 35077) and the defined each peak was expressed as a percentage of the total area of all peaks.

2.2.4. Statistical analysis

All assays were performed in triplicate. Data analyses were carried out using SPSS software v 20.0 and Duncan multiple range test at $P<0.05$ probability level.

3. Results and Discussion

3.1. Proximate composition of fig seeds

The moisture contents of fig seeds were from 5.54 to 5.64% while the ash contents of them were 2.99% (Table 1). There were not found significant differences in the moisture and ash contents of fig seeds obtained from different fig varieties ($P<0.05$). Morton (1987) and Kim et al (1992) reported moisture content of the fresh figs as 77.5-88.70% and ash content of the fresh figs as 0.44-0.85%. Compared to whole fig, the lower moisture contents in the fig seeds are expected due to the fact that the jelly-like flesh found in the whole figs is higher amount than the fig seeds. The reason why fig seeds have higher ash content is also that jelly-like meat is not found in fig seeds. The fact that fig seeds have low moisture and high ash contents is a very positive feature. Because, low moisture content increases the shelf life of product and high ash content also increases the possibility of existed more minerals having positive health effects in the structure of fig seeds.

The oil and protein contents of the fig seeds ranged from 23.06 to 23.67% and from 14.74 to 15.07, respectively (Table 1), these values are higher compared to data reported by Morton (1987) and Kim et al (1992). They reported the oil and protein contents of fresh figs as 0.14-0.31% and 0.70-1.3%, respectively. This is evidence that the fig seeds contribute significantly to the protein and oil contents of the figs. Besides, it was determined that the fig seeds in Germencik location had the highest contents of oil and protein and the fig seeds in Nazilli location had the lowest contents of oil and protein ($P<0.05$). Generally, the presence of high amounts of oil and protein in fig seeds is an indication that they may be beneficial to health because they are rich in fatty acids and contain essential amino acids.

Fig seeds were found to be good sources of carbohydrate. The obtained results demonstrated that the carbohydrate contents of fig seeds were 52.62-53.66% (Table 1), which was significantly higher than the carbohydrate contents (17.1-20.3 g 100 g^{-1} fresh fig) of figs (Morton 1987; Guvenc et al 2009). The significant differences in carbohydrate contents of the analyzed samples determined ($P<0.05$). Although the fig seeds in Nazilli location had the highest amount of carbohydrate content, the fig seeds in Germencik location had the lowest amounts of carbohydrate contents.

3.2. Mineral content of fig seeds

The mineral levels of fig seeds were analyzed by UV-vis spectrophotometer (for only P analysis) and an atomic absorption spectrometer with a flame atomize. The performance characteristics of the HR-CSFAAS method were given in Table 2.

Table 1- Some chemical properties of fig (*Ficus carica* L.) seeds

Location	*Moisture content (%)**	*Ash content (%)**	*Oil content (%)*	*Protein content (%)*	*Carbohydrate content (%)*
Incirliova	5.59±0.07	2.99±0.01	23.37±0.15ab	14.92±0.12ab	53.13±0.19ab
Germencik	5.64±0.06	2.99±0.01	23.67±0.14a	15.07±0.07a	52.62±0.11b
Nazilli	5.54±0.04	2.99±0.01	23.06±0.10b	14.74±0.14b	53.66±0.21a

Data are expressed as mean ± standard deviation in triplicate (n= 3). Different letters (a, b, c, d, and so on) in the same column show significant differences ($P<0.05$). *, no statistically significant differences ($P>0.05$)

Table 2- Performance characteristics of the HR-CS-FAAS method

Minerals	*LOD (mg L^{-1})*	*LOQ (mg L^{-1})*	*RSD (%)*	*Calibration range (mg L^{-1})*	*r*	*Regression equation*	*R (%)*
Mg	0.47	1.56	6.50	0.2-1.5	0.9050	y= 0.1559549+0.7635861 x	98.50
Mn	0.09	0.31	2.50	0.2-1.5	0.9961	y= 0.0085974+0.2259809 x	89.20
Zn	0.19	0.63	9.50	0.2-1.5	0.9840	y= 0.0819164+0.2810132 x	96.80
Fe	0.13	0.43	6.80	0.5-3.8	0.9970	y= 0.0094184+0.0671127 x	95.60
Ca	1.14	3.80	2.10	1.0-4.0	0.9488	y= 0.0059524+0.0582419 x	87.40
Cu	0.31	1.05	0.80	0.5-4.0	0.9561	y= -0.0004395+0.3672192 x	94.00
Na	0.49	1.62	0.90	1.0-2.5	0.9635	y= 0.3499030+0.4490676 x	98.80
K	0.33	1.10	1.60	0.5-4.0	0.9924	y= 0.0330507+0.2026023 x	94.50

LOD, limit of detection; LOQ, limit of quantification; RSD, relative standard deviation; r, correlation coefficient; R, recovery in coffee samples

There was a significant difference ($P<0.05$) among samples in terms of mineral contents (Mg, Zn, Fe, Cu, Na, K and P) in fig seeds, except Ca and Mn (Table 3). The results statistically expressed that Ca, K and P were the major minerals and Cu and Mn were also existed in trace amount in the analyzed samples ($P<0.05$). Aljane et al (2007) also found K and Ca as major minerals of fresh figs among the analyzed minerals (K, Ca, Mg, Na, Zn). In the study of Morton (1987), Ca, P, Fe, Na and K contents of fresh figs were investigated and major minerals of fresh figs were determined as K, Ca and P, which was similar to the results of this study. Mineral contents of fig seeds varied according to location characteristics; the fig seeds in Germencik location in terms of Mg, Zn and Cu contents, the fig seeds in Nazilli location in terms of Fe, Na, K and P contents come forward ($P<0.05$). Minerals have key roles in the body to do necessary functions for healthy and lengthy life which is from building strong bones to transmitting nerve impulses (Gharibzahedi & Jafari 2017). It is very important to determine the high-mineral foods because a balanced diet is aimed at providing almost all of the minerals needed for the body.

3.3. Fatty acid composition of fig seeds

In the oil of fig seeds, 14 fatty acids were identified (Table 4). Statistically significant differences were generally found between individual fatty acid contents of samples except heptadecanoic, α-linolenic and lignoceric acids ($P<0.05$). The total saturated (SFA) and unsaturated fatty acid (MUFA and PUFA) percentages of fig seeds were in range 7.10-10.98% and 89.02-92.9%, respectively. Similar to the study of Morton (1987), the major fatty acids of fig seeds were quantified as palmitic, oleic, linoleic and γ-linolenic acids. Arachidic acid was detected in smaller amount compare to other fatty

Table 3- Mineral contents (mg kg^{-1}) of fig (*Ficus carica* L.) seeds

Location	*Mg*	*Mn**	*Zn*	*Fe*	*Ca**	*Cu*	*Na*	*K*	*P*
Incirliova	49.82± 0.01b	7.90± 0.19	20.77± 0.23b	52.31± 0.11b	187.80± 3.71	5.29± 0.03ab	70.24± 0.41ab	169.78± 0.07b	178.36± 2.83c
Germencik	53.66± 0.01a	8.03± 0.10	22.56± 0.43a	50.26± 0.15c	185.67± 3.54	5.34± 0.01a	69.45± 0.28b	166.73± 0.03c	181.78± 2.02ab
Nazilli	46.36± 0.04c	7.77± 0.28	18.26± 0.37c	58.11± 0.12a	190.42± 5.66	5.25± 0.03b	70.82± 0.19a	172.83± 0.06a	186.21± 1.41a

Data are expressed as mean ± standard deviation in triplicate (n= 3). Different letters (a, b, c, d, and so on) in the same column show significant differences ($P<0.05$). *, no statistically significant differences ($P>0.05$)

acids. The concentrations of fatty acid components in the fig seeds was observed to be generally lower compared to those of the fresh whole figs analyzed by Guvenc et al (2009). It was noteworthy that the amounts of linoleic and γ-linolenic acids in the fig seeds were higher than linoleic and γ-linolenic acid contents (1.46%) of the fresh figs. Also, the fig seeds in Germencik location came out as the sample with the highest PUFA and the lowest SFA.

It could have been said that the fig seed was a better source of linoleic and γ-linolenic acids. The fig seed oil is healthy oil which is rich in nutrients. It can be considered a commercial source, due to the fact that it contains w-3 (α-linolenic and cis-5, 8, 11, 14, 17-eicosapentaenoic acid) and w-6 (linoleic acid) fatty acids. These fatty acids provide important contributions to human health and are known as essential fatty acids. It is also important that the amount of unsaturated fatty acids in fig seed oil is higher than that of saturated fatty acids.

As a result of this study, it was determined that the biochemical characteristics of fig seeds obtained from Sarilop fresh figs which are grown at different locations were different from each other. It is thought that this difference is caused by the difference of location (soil structure, irrigation etc.) where the figs are grown, although the fig seeds obtained from the same fig cultivar are examined. The fig seeds of the Germencik location were identified as sample having the highest fat, protein, some minerals (Mg, Zn and Cu) and PUFA contents in this study.

4. Conclusions

This is the first study to investigate the chemical properties and nutritional compositions of fig seeds. Fig is a fruit which considered as a functional food due to its unique physicochemical properties. The contribution rate of fig seeds to the nutritional characteristics of figs was determined for the first time by this study. It has been discovered that fig seeds

Table 4- Fatty acid compositions (%) of fig (*Ficus carica* L.) seeds

Fatty acids	*Common name*	*Location*		
		Incirliova	*Germencik*	*Nazilli*
C16:0	Palmitic acid	7.40±0.380a	3.58±0.250b	7.00±0.130a
C16:1	Palmitoleic acid	0.06±0.003ab	0.06±0.003a	0.05±0.001b
C17:0	Heptadecanoic acid*	0.06±0.003	0.06±0.002	0.06±0.001
C18:0	Stearic acid	3.38±0.020ab	3.73±0.180a	2.97±0.330b
C18:1n9c	Oleic acid	16.82±0.070b	17.79±0.030a	16.97±0.050b
C18:2n6c	Linoleic acid	32.81±0.750b	37.95±0.780a	31.80±0.470b
C20:0	Arachidic acid	0.02±0.003ab	0.03±0.001a	0.02±0.001b
C18:3n6	γ-linolenic acid	37.87±0.160b	41.80±0.960a	39.09±0.140b
C20:1	cis-11-Eicosenoic acid	0.26±0.010a	0.18±0.010b	0.23±0.030ab
C18:3n3	α-Linolenic acid*	0.30±0.020	0.25±0.020	0.30±0.030
C21:0	Heneicosanoic acid	0.04±0.002b	0.08±0.005a	0.04±0.005b
C20:3n6	cis-8,11,14-Eicosatrienoic acid	0.09±0.004a	0.04±0.005b	0.09±0.001a
C24:0	Lignoceric acid*	0.03±0.004	0.04±0.001	0.03±0.001
C20:5n3	cis-5,8,11,14,17-Eicosapentaenoic acid	0.39±0.040a	0.24±0.030b	0.11±0.010c
∑ SFAs		10.98±0.260a	7.10±0.140b	10.24±0.320a
∑ MUFAs*		17.22±0.150	17.05±0.300	17.48±0.320
∑ PUFAs		71.80±0.110c	75.85±0.150a	72.28±0.002b

Data are expressed as mean ± standard deviation in triplicate (n= 3). Different letters (a, b, c, d, and so on) in the same line show significant differences (P<0.05). *, no statistically significant differences (P>0.05). ∑ SFAs, total saturated fatty acids; ∑ MUFAs, total monounsaturated fatty acids; ∑ PUFAs, total polyunsaturated fatty acids contribute significantly to the chemical properties of figs by their rich nutritional content. It is obvious that fig seeds have significant support on the health benefits of figs. Considering the findings obtained in this study, the use of fig seeds as a pharmaceutical supplement can be possible and the commercialization of the fig seeds can come to the fore.

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Effects of Additive Intercropping on Mineral Uptake of Onion and Fenugreek at Different Densities

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ABSTRACT

Increasing world population demands for food, limited natural resources have created the risk of food security for current and future generations. Intercropping is regarded as an effective strategy in sustainable agriculture. Therefore, current study was carried out to evaluate the nutrient uptake of onion and fenugreek intercropping system during two growing season of (2015-2016 and 2016-2017) at a research field in Kerman, Iran. Two factorial experiments based on completely random blocks design with three replications were carried out. The treatments in this study were including: onion densities as the first factor (30, 25 and 20 plants m^{-2}) and fenugreek densities as the second factor at three levels of 25, 15 and 12 plants m^{-2} with sole cropping of two species at these densities. Results showed that in the both of years, mineral concentration and uptake (N, P and K) of onion leaf and bulb and fenugreek in intercropping increased in comparison with mono cropping for two species. In both years, the highest and lowest nutrient content of onion was observed in the densities of 30 and 20 plant m^{-2}. In general, it can be said that intercropping systems have some positive effects on nutrient content and uptake of onion.

Keywords: Fenugreek; Intercropping; Nutrient content; Onion; Plant nutrient uptake

1. Introduction

Onion (*Allium cepa* L.) is an important economic vegetable crop which unique flavor is appreciated by people in every part of the world. Also, it is one of the most prominent commercial vegetable crop grown in Iran and thought to be originated in central Asia (Anthon & Barrett 2003). Having high mineral and organic contents, onions are essential for human health (Raj & Yadav 2005). It has low branched roots, so needs to supply adequate minerals. Intercropping onion with legumes have some advantages as follows: improved nutrient availability, soil structure, hormonal effects and reduced pest and disease incidence through rhyzodeposition. Fenugreek (*Trigonella foenugraecum* L.) is an annual crop from Leguminosae, being used as condiment and dried leaves to prepare spice. The vegetative parts are also rich in vitamins A, B and iron. In addition to fixing nitrogen, forming a cover on the soil surface, it hinders weeds growth and diminishes soil erosion (Basch et al 2003).

Intercropping is a crop management system which involves two or more economic species grown together for at least a part of their respective productive cycle and planted sufficiently close to each other, so that inter-specific competition can occur (Ouma & Jeruto 2010). The main advantage of intercropping is the more effective usage the available resources, reduced the nutrients leaching, keeping the soil fertility and enhanced productivity as compared the sole crop cultivation (Lithorgidis et al 2011). A significant reason for intercropping is the improvement and maintenance of soil fertility and increasing the productivity per unit of land (Pozdíšek et al 2011). Intercropping enhances soil fertility via biological nitrogen fixation of legumes, improves the soil protection through more ground cover than sole cropping, and provides better lodging resistance for crops vulnerable to lodging than when grown in monoculture (Ouma & Jeruto 2010). A legume, grown in associated to other crop, especially with a cereal, have normally more productivity in cropping systems (Kamran Khan et al 2015). Thus modern farming system intercropping implies a key strategy for sustainable agriculture (Jackson et al 2007). Intercropping is a way to introduce more biodiversity into agro-ecosystems and the results obtained from intercropping studies indicate that increased crop diversity could increase the number of ecosystem services provided (Lithourgidis et al 2011). Enhancing productivity via more effective use of available resources (light, water, fertilizer, etc.) is possible through intercropping which leads to diminished weed pressure and plant health protection (Hauggaard-Nielsen et al 2003; Banik et al 2006).

Inorganic nutrients including nitrogen (N), phosphorus (P) and potassium (K) are fundamental macronutrient for growth and development of all living organisms. Being absorbed in large amounts, nitrogen contributes significantly to raise the onion production (Myaka et al 2006). Phosphorus plays a vital role in cellular energy transfer, respiration, photosynthesis and root development (Alam et al 2016). In soils which are contain moderately low P, onion growth and yield can increase by applied P. The beneficial effect of potassium can be found in different traits of agricultural products, such as photosynthesis, translocation of photosynthesis, regulation of plant, activation of plant catalysts and resistance against pests and disease (Malavolta 2006). There are many studies demonstrating the superiority of intercropping legumes include intercropping wheat with soybean (Lithourgidis et al 2011), wheat with fenugreek (Wasaya et al 2013) and maize with soybean (Owusu & Sadick 2016). Also intercropping of legume crop (pigeon pea) in maize added up about 60 kg N ha^{-1} and improves the fertility status of soil (Myaka et al 2006). Therefore, the objective of this study was to evaluate the intercropping system effect and density on plant nutrient concentration and uptake of onion and fenugreek.

2. Material and Methods

Two experiments were conducted during two successive growing seasons (2015-2016 and 2016-2017) in Agricultural Research Center; Kerman, Iran. (31˚7′ N, 57˚14′ E and 1749 MASL). Some properties of soil were shown in Table 1. A factorial set of treatments was arranged within completely randomized blocks design (RCBD) with three replications. Intercropping pattern was additive series. The first factor was three onion distance (8, 10 and 12 cm) for densities (300000, 250000 and 200000 plant ha^{-1}), and the second factor comprised of fenugreek distance (10, 15 and 20 cm) for plant populations of 250000, 150000 and 120000 plant ha^{-1} in both intercropping and mono cropping system. Fertilization was done according to soil analysis results and nutritive requirement of species. Any insect, pest and disease infections were not seen in the experimental field. In the present study, onion seed cv. 'Rossen' were planted in nursery, onions forty-five day old onion seedlings and fenugreek seeds were planted in the right side and fenugreek seed was sown in the opposite side of rows, by hand in December 2015 and September 2016 in intercropping treatment. Each single crop in the mono treatments was cultivated in the same site of lateral. All other agronomic operations except those under study were kept normal and uniform for all treatments. The fenugreek was harvested when plants reached the peak of vegetative growth, with marketable 2 and 4 month after planting before flowering. Onion bulbs were harvested 4 month after transplanting (when 80% of onion leaves had fallen). At maturity, five plants from each plot were collected randomly to determine parameters. The plant sample (leaf and bulb) of onion and (shoot) of fenugreek were collected from each plot and were dried for 48 hrs in hot air oven at 65±5 °C. Finally ground samples were passed through 0.5 mm mesh sieve and were used for chemical determination of nitrogen, phosphorus and potassium. The concentrations of potassium and phosphorus were determined by nitric perchloric and nitric acid digestion methods. Phosphorus was measured by the vanadate- molybdate method using a spectrophotometer HITACHI U-1100 at wavelengths of 430 nm and nitrogen was measured by the spectrophotometer HITACHI U-1100 at wavelengths of 650 nm that involved changing the form of organic nitrogen to the ammonium ($NH4^+$) form with concentrated sulfuricacid and then measuring the amount of ammonium production (Baethgen & Alley 1989), and K was determined using a flame photometer (Model 405G) methods described by (Cottenie 1980).

Nutrient uptake= Nutrients contents (mg) × Dry weight (g)

Statistical analysis: Analyses of variance were applied to the data using SAS (Statistical Analysis System) version 9.1. And Excel software was used to draw figures. Then, means were compared by Duncan's multiple range test.

3. Results and Discussions

A: Onion, According to the results, the year effect on nutrient concentration and uptake was significant ($P<0.01$) (Table 2). In the first year, leaf and bulb concentrations (N, P, K) of nutrient in onion were higher than the second year (Table 3). In the second year, nutrient uptakes were higher than the first. Average temperature and rainfall data (Table 4) clearly show that in the second year, high temperatures at the early planting of onion affected on root growth and morphology of plants, and resulted in higher nutrient uptake by onions. Soil temperature has long been thought as a prominent ecological factor which determines a variety of structural and functional characteristics in managed and natural ecosystems. In some ecosystems, the root zone temperature is the most important factor in determining net primary productivity (Risser 1985). Soil thermal regime has a dramatic effect on root growth and morphology. (Camilia 2001) discovered that the mineral status depends on some environmental factors including temperature, rainfall, parent material, plant species, and fertilization practice. The exact mechanism for temperature-induced changes in nutrient uptake capacity is not clearly understood. Root-zone temperature might affect nutrient absorption capacity by changing fluidity of the fatty acids in root plasmalemma. The most notable are the direct effects of soil temperature on biogeochemical processes that regulate N and water availability in the soil (Parsons et al 1994).

Nutrients concentration (N, P, and K): The effects of onion and fenugreek densities on N, P and K concentration were significant ($P \leq 0.01$) and ($P \leq 0.05$). Nutrients concentration in leaf was

Table 1- Some physicochemical properties of soil before sowing (depth: 0-30 cm) in 2015-2016 and 2016-2017 seasons

Years	*Texture*	*Sand*	*Silt*	*Clay*	*Organic carbon (%)*	*pH*	*EC (dSm^{-1})*	*Total N (%)*	*P ($mg\ kg^{-1}$)*	*K ($mg\ kg^{-1}$)*
2015-2016	loam	45.72	28.88	25.40	0.41	7.60	1.30	0.02	8.40	175
2016-2017	loam	46.20	28.20	25.60	0.23	7.90	1.26	0.03	9.40	200

Table 2- Analysis of variance of intercropping onion and fenugreek on mineral concentration and uptake by plant

Source of variation	*df*	*Concentration*						*Uptake*		
		NLeaf	*NBulb*	*PLeaf*	*PBulb*	*KLeaf*	*KBulb*	*N*	*P*	*K*
Year (y)	1	1.200**	3.900**	0.060**	0.920**	5.12*	5.03**	52093.17**	1133.09**	131659.00*
Error	4	0.040	0.130	0.017	0.002	0.55	1.25	276.50	7.11	530.90
Onion (A)	2	0.660**	0.110 ns	0.017*	0.019*	1.22*	4.85**	820.65**	48.49**	4855.20**
Fenugreek(B)	3	0.450**	0.820**	0.110**	0.037**	8.74**	5.51**	454.60*	10.32*	2058.60*
A×B	6	0.070 ns	0. 070 ns	0.003 ns	0.009 ns	0.96 ns	0.39 ns	310.85 ns	7.94 ns	598.70 ns
Y×A	2	0.011 ns	0.006 ns	0.001 ns	0.006 ns	0.40 ns	0.11 ns	11.06 ns	4.77 ns	247.80 ns
Y×B	3	0.010 ns	0.160 ns	0.006 ns	0.029**	1.25*	0.03 ns	244.49 ns	4.33 ns	1474.40*
Y×A×B	6	0.010 ns	0.060 ns	0.003	0.010 ns	0.26 ns	0.20 ns	440.43 ns	9.36 ns	264.00 ns
Error	44	0.060	0.080	0.004	0.005	0.43	0.92	229.36	4.90	387.30
Cv (%)		5.770	8.730	9.480	15.390	9.22	25.69	19.43	17.60	18.30

*, **, significant at $P \leq 0.05$, $P \leq 0.01$ respectively; ns, not significant

Table 3-The effect of years on mineral concentration and total mineral uptake of onion

Seasons	*Concentration (mg g^{-1} DW)*						*Uptake (mg DW plant^{-1})*			*Uptake (kg ha^{-1})*		
	N Leaf	*N Bulb*	*P Leaf*	*P Bulb*	*K Leaf*	*K Bulb*	*N*	*P*	*K*	*N*	*P*	K
Y_1 (year)	4.50 a	3.52 a	0.70 b	0.60 a	7.37 a	4.01 a	51.04 b	8.62 b	64.61 b	15.32 b	2.64 b	20.22 b
Y_2 (year)	4.24 b	3.05 b	0.76 a	0.38 b	6.84 b	3.48 b	104.83 a	16.55 a	150.10 a	28.34 a	4.48 a	40.57 a

Means with similar letter in each column are not significantly ($P<0.05$) different by Duncan Multiple Range test

Table 4- Average temperature and rainfall during growth seasons

		2015-2016		*2016- 2017*	
Date	*Temperature (°C)*	*Rainfall (mm)*	*Date*	*Temperature (°C)*	*Rainfall (mm)*
December	13.5	59.8	September	29.2	0.0
January	13.7	0.2	October	22.4	0.0
February	19.6	4.0	November	16.0	8.3
March	24.8	7.6	December	15.0	0.0
April	29.7	0.8	January	13.3	148.1

decreased by increasing onion density (Table 2). N, P and K maximum and minimum content in leaf were observed at densities of 20 and 30 plants m^{-2}, respectively (Table 5). No significant difference observed in bulb N concentration among onion densities (Table 2). While, the highest and the lowest P and K concentration of leaf and bulb were recorded in densities of 20 and 30 onions per m^{2}, respectively (Table 5). Also, N, P and K concentration of bulb and leaf were increased by intercropping onion with fenugreek as this trait was more at various densities of fenugreek compared to onion mono cropping. However, the effect of various densities of fenugreek on N, P and K concentration of bulb and leaf were statistically similar (Table 5). Also the interaction effect of year and fenugreek density had a significant effect on the P concentration of bulb and K concentration of leaf (Table 2). The lowest P concentration of leaf and bulb were obtained in density of 0 fenugreek (sole cropping of onion), while P concentration of bulb was increased by intercropping onion with fenugreek as this trait was more at various densities of fenugreek compared to onion mono cropping in the first year. However, the effect of various densities of fenugreek on P concentration of bulb was statistically similar in the second year (Figure 1). The highest concentration of K (8.24 mg g^{-1} DW) was obtained under intercropping with 12 plants m^{-2} fenugreek in the first year, and the lowest concentration of K (5.78 mg g^{-1} DW) was observed in (sole cropping) of onion in the second year (Figure 2).

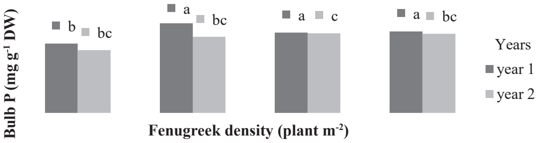

Figure 1- The interaction effect of year and fenugreek on P concentration of onion bulb

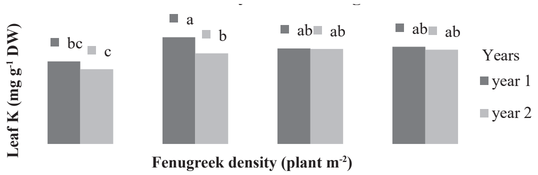

Figure 2- The interaction effect of year and fenugreek densities on K concentration of onion leaf

Nutrient Uptake: The effects of onion and fenugreek densities on nutrient uptake of onion were significant ($P \leq 0.01$) and ($P \leq 0.05$), respectively (Table 2). Increasing onion densities, nutrient uptake by plant was decreased. The maximum and the minimum uptake of nutrient were observed at densities of 20 and 30 plants m^{-2}, respectively (Table 5). Also, nutrient uptake was increased by intercropping onion with fenugreek as this trait was more at various densities of fenugreek compared to onion mono cropping. However, the effect of various densities of fenugreek on nutrient uptake of onion was statistically similar. Also the interaction effect of year and fenugreek density on the K uptake was significant (Table 2), the highest (172.16 mg plant^{-1}) was obtained under intercropping with 12 plants m^{-2} fenugreek in the second year, and the lowest K uptake was related to densities of 0 (sole cropping) onion and was statistically similar in the first year (Figure 3). The results show uptake ha^{-1} values (Table 6); high densities of two crops confirms to the highest uptake ha^{-1} (26.14, 4.67 and 38.02 kg ha^{-1}) for N, P and K recorded in combination treatment

Table 5- Effect of intercropping of onion and fenugreek on the mineral content and total mineral uptake of onion

Density (plants m^{-2})	*Concentration (mg g^{-1} DW)*						*Uptake (mg DW Plant^{-1})*		
	N Leaf	*N Bulb*	*P Leaf*	*P Bulb*	*K Leaf*	*K Bulb*	*N*	*P*	*K*
0nion									
30	4.27 b	3.24 a	0.70 b	0.46 b	6.85 b	3.26 b	72.03 b	11.17 c	92.98 c
25	4.27 b	3.25 a	0.73 ab	0.50 ab	7.20 ab	3.83 a	78.06 ab	12.56 b	107.72 b
20	4.56 a	3.36 a	0.75 a	0.52 a	7.27 a	4.14 a	83.72 a	14.01 a	121.42 a
Fenugreek									
0	4.16 b	2.97 b	0.61 b	0.44 b	6.08 b	2.92 b	71.01 b	11.78 b	99.03 b
25	4.38 a	3.38 a	0.75 a	0.49 b	7.38 a	4.10 a	77.87 ab	12.24 ab	108.15 ab
15	4.50 a	3.33 a	0.77 a	0.48 b	7.35 a	3.91 a	80.10 ab	12.78 ab	112.02 ab
12	3.38 a	3.45 a	0.78 a	0.55 a	7.61 a	4.03 a	82.75 a	13.54 a	120.29 a

Means with similar letter in each column are not significantly ($P<0.05$) different by Duncan Multiple Range test of 30: 25 plants m^{-2} onion and fenugreek at mean of two years, while the lowest uptake ha^{-1} (14.99, 2.63 and 22.11 kg) for N, P and K was found in sole cropping treatment with 20 plants m^{-2} (onion) density for mean of years. Soil nutrient which were absorbed by plant is a crucial resources for plant metabolism, especially to build cell structure and plant grows and development. Nutrient absorption in plant roots, therefore will be similar with the nutrient in plant tissue and it correlates positively with organic compound which was resulted from plant metabolism (Alam et al 2016). Onion is more vulnerable to nutrient shortages than most crop plants because of their shallow and unbranched root system; therefore, they cannot maintain adequate nutrients uptakeing such as phosphorous and potassium which are known to diffuse slowly through the soil solution (Brewester 1994). Different nutrients (N, P and K) uptaking by leaves and bulbs of onion influenced by intercropped fenugreek. In intercropping system, root interactions could raise the root extent and microbial activity in rhizosphere (Zhang et al 2013). Inter- specific interaction in rhizosphere compositely affects nutrient availability and uptaking in intercropping system (Hauggard-Nielsen 2003; Li et al 2010). Intercropping onion with fenugreek increased the N, P and P content in leaf and bulb and total uptake N, P and K. This was probably due to higher nitrogen fixation by the bacteria causing more utilization of all the nutrients by crop thus lead to more N, P and K content in leaf and bulb. Studies have presented advantages of intercropping with legumes including tomato with common bean (Abd El-Gaid et al 2014), Barely with fenugreek (Kamran Khan et al 2015), and wheat with fenugreek (Wasaya et al 2013). Nutrient uptake was significantly influenced by cropping systems where onion plants were more nutrient uptake in plots that had onion intercropped with fenugreek at wider spacing compared with other treatment. This could have been contributed by the fixed nitrogen which improved the plant nutrient uptake over other treatments. Although intercropping at narrower spacing also had fixed nitrogen, but the plant was strong completion which led to increase nutrient uptake (Table 5). Also, low densities of onion plant leads to reduction in competition between onion and fenugreek plants for resources and enhances soil fertility via atmosphere nitrogen fixation into the soil (150 tons year^{-1}). Legumes increases soil conservation through more cover on ground than sole cropping (Oroka & Omoregie 2007). Results indicated that maize and soybean intercropping caused an increase in phosphorus (5.25 mg kg^{-1}) (Owusu & Sadick 2016). (Zhang et al 2009) studied the effects of maize- peanut and stated that intercropping could increase soil bacteria in the both of maize and peanut root zones. Also, improved uptake of N, P and K nutrients might be attributed to higher dry matter accumulation with density decrease leading to higher uptake of N, P and K. Similar results of increased uptake of N, P and K due to wider row spacing were reported by (Thavaprakaash & Velayudham 2007) in baby corn. The increase in the onion density enhanced the uptake of plants ha^{-1} (leading to an increase in uptake). In general, the more N uptake, the more crop tendency to absorb more P and K. This is similar to the findings of (Rathika 2013) in maize. (Gahoonia et al 2006) stated that plant nutrients such as (K) and (P) mostly remain fixed in soils and their bioavailability to plant roots is diffusion- limited. The increased uptake of nutrients is possibly due to higher photosynthetic rate and nitrate reductase activity led to better uptake of N by the crop. Generally, when the uptake of N is more, the crop would have a tendency to absorb more P and K. This is in line, these findings are in accord with those of (Kumar Dubey et al 2012).

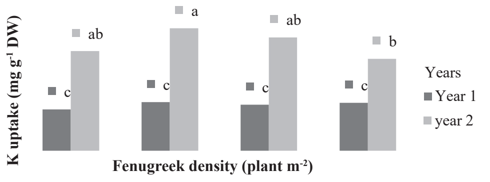

Figure 3- The interaction effect of year and fenugreek on K uptake of onion

B. Fenugreek, The result revealed that the effect of onion and fenugreek density were significant

Table 6- The effect of intercropping onion with fenugreek on the N, P and K uptake at two years

Treatments (Onion: Fenugreek)	*Uptake (kg ha^{-1})*		
	Mean of two years N	*Mean of two years P*	*Mean of two years K*
Density 30	18.91 cd	3.05 cd	24.22 c
Density 30: 25	26.14 a	4.67 a	38.02 a
Density 30: 15	25.77 a	4.00 abc	35.19 a
Density 30: 12	25.20 ab	4.13 ab	31.81 ab
Density 25	18.80 cd	2.97 cd	23.88 c
Density 25: 25	24.70 ab	3.73 abc	32.22 ab
Density 25: 15	24.49 ab	3.86 abc	35.96 a
Density 25: 12	20.58 bc	3.33 bcd	31.35 ab
Density 20	14.99 d	2.63 d	22.11 c
Density 20: 25	21.73abc	3.66 abc	31.97 ab
Density 20: 15	22.51 abc	3.69 abc	32.14 ab
Density 20: 12	18.19 cd	2.95 cd	25.84 bc

Means with similar letter in each column are not significantly (P<0.05) different by Duncan Multiple Range test

on mineral content and total mineral uptake of fenugreek ($P \leq 0.01$) in intercropping of onion and fenugreek. Fenugreek means comparisons (Table 7) revealed that the highest uptake plant ha^{-1} was related to 25 plants m^{-2} density and there was no significant difference in content and uptake by plant among fenugreek treatments. In intercropping system, the maximum mineral content of fenugreek was obtained under mono cropping (sole fenugreek), while no significant difference was observed in intercropping treatments. In contrast, the minimum total mineral uptake of fenugreek was obtained under mono cropping (sole fenugreek), while there was no significant difference in uptake among intercropping treatments (Table 7). Also, effects of onion and fenugreek densities interactions were significant on P and K uptake of fenugreek ($P \leq 0.05$). The highest fenugreek P and K uptake were observed

Table 7- The effect of intercropping of onion and fenugreek on the mineral content and total mineral uptake of fenugreek

Density (plants m^{-2})	*Concentration (mg g^{-1} DW)*			*Uptake (mg DW Plant^{-1})*		
	N	*P*	*K*	*N*	*P*	*K*
Fenugreek						
25	4.69 a	0.79 a	7.71 a	18.10 a	3.04 a	29.75 a
15	4.81 a	0.77 a	7.93 a	15.65 a	2.58 a	26.15 a
12	4.79 a	0.78 a	7.96 a	19.50 a	3.24 a	32.87 a
Onion						
0	5.15 a	0.84 a	8.43 a	12.90 b	2.12 b	21.20 b
30	4.56 b	0.77 b	7.63 b	18.94 a	3.10 a	31.32 a
25	4.63 b	0.75 b	7.71 b	19.91 a	3.35 a	34.01 a
20	4.72 b	0.76 b	7.70 b	19.31 a	3.25 a	31.83 a

Means with similar letter in each column are not significantly (P<0.05) different by Duncan Multiple Range test in intercropping with 15:25 (fenugreek: onion) densities, while the lowest fenugreek P and K uptake were recorded mono cropping (sole onion) (Figure 4 and 5). Legumes are distinguished to fix atmospheric nitrogen into the soil, thus enriching soil fertility, and help to satisfy the N needs of cereals. Fenugreek being leguminous crops did not compete with onion for N for its growth and development besides, fixing of atmospheric N in soil. Also, onion and fenugreek have different root depths and architectures as well as different requirements for soil N, P and K. Fenugreek roots act as a source of nitrogen for plant as a result enriches the soil with nitrogen. Therefore, increase in the nutrient concentration of fenugreek was associated with increase in uptake of these nutrients in fenugreek. This can again be supported by synergistic uptake mechanism of N, P and K and their assimilation which is manifested in the form of increase in yield along with higher nutritional demand for plant growth. These results are in close conformity with findings of (Tuncturk et al 2011).

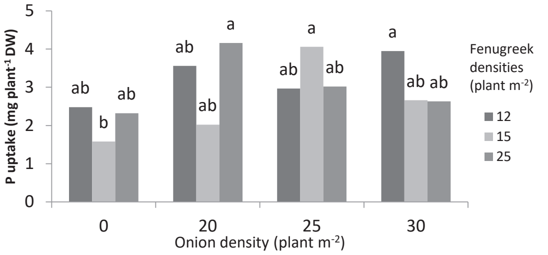

Figure 4- The interaction effect of onion and fenugreek on P uptake of fenugreek

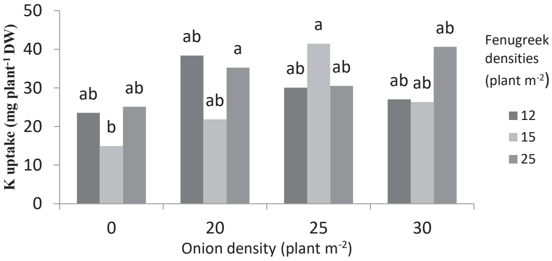

Figure 5- The interaction effect of onion and fenugreek on K uptake of fenugreek

4. Conclusions

This study was conducted in order to evaluate the effects of densities of onion and fenugreek in mono cropping and intercropping systems. According to the results, indicated that the mineral content and total mineral uptake by onion was increased when onion was intercropped with fenugreek. Therefore, the onion-fenugreek intercrop would be suitable to small-scale farmers who do not have adequate resources for purchase of chemical nutrients. The system would also be very ideal for organic vegetable production, in which chemical application is not desirable.

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Effects of *Tobacco Etch Virus* (TEV) on the Yield and Quality of Karaisali Pepper Populations

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ABSTRACT

The Eastern Mediterranean Region has a significant potential for capia pepper production in Turkey. In the region, a population named as 'Karaisali pepper' with special characteristics, is grown. The population is well-adapted to the region and grown for processing. Karaisali pepper is usually preferred for red pepper production due to its high dry matter content. Survey studies conducted on Karaisali pepper in 2014 and 2015 demonstrated that the *Tobacco etch virus* (TEV) was the most common and destructive virus affecting Karaisali pepper. In the present study, three pure lines derived from Karaisali pepper were used to assess their susceptibilities against TEV. The experiment included mechanically inoculated infected plants and healthy pepper plants in the control group. The pepper plants were inoculated with the TEV using mechanical inoculation method during the four-leaf stage. The plants were observed periodically after mechanical inoculation. Each repetition was analyzed based on the total yield, pepper paste yield, fruit size, soluble solid content, fruit color and market value. The result indicated that, in average, TEV reduced Karaisali pepper yield by 77.5% and pepper paste yield by 33.6%. Furthermore, the average fruit length (37%), fruit diameter (21.4%), fruit wall thickness (14.2%), fruit volume (60.2%) and first quality fruit ratio were also decreased. The results confirm the threats of TEV in pepper production.

Keywords: *Tobacco etch virus*; Pepper; Yield; Quality

1. Introduction

Pepper is cultivated on 805.166 acres in Turkey and total production of pepper were 2.608.172 tons; 1.107.713 tons of it were capia pepper production (TSI 2017). The Karaisali pepper is a local population, widely cultivated as a main and second crop in Adana province, with a high dry matter content, and it is used for pepper paste and grilled pepper production in the industry. Different viruses significantly affect the growth and development of different plant species. For that reason, Friess & Maillet (1997) for chickweed, Johnson & Main (1983) for tobacco, Taiwo & Akinjogunla (2006) for cowpea, Nascimento et al (2006) for peanut and Al-Saleh et al (2007) for peanut carried out comprehensive studies. Different viruses in pepper cause considerable amount and quality losses. Pazarlar et al (2013) studied changes in some growth (plant leaf number and area, plant biomass, plant height, root length, and plant stem diameter) and physiological (photosynthetic pigments, relative water content (RWC) and proline content) parameters of pepper (*Capsicum annuum* L.) varieties as they were affected by *Tobacco mosaic virus* (TMV) infection. Infected plants showed various degrees of stunting, necrosis on stems, leaves and fruits, mosaic symptoms on leaves, deformations, defoliation of leaves, and reduction in fruit size. TEV is among the major viruses that infect pepper plants cultivated in open fields and cause infections (Buzkan et al 2012; Fidan & Keleş Öztürk 2013). TEV is transmitted in a non-persistent manner by aphids and its most important vector is *Myzus persicae*. TEV is observed as single infection on pepper or as mixed infections with *Potato Y virus* (PVY). Its incidence could be as high as 100% at harvest time (Padgett et al 1987). Yield reduction due to TEV can reach up to 70% (Koenning & McClure 1981). Murphy & Morawo (2017) evaluated TEV strains HAT, Mex21, and N for their pathogenicity and effects on growth of 'Calwonder' pepper. Effects on plant growth parameters closely reflected disease symptoms induced by each TEV strain. HAT-infected Calwonder plants did not differ from the healthy control for plant height, internode lengths, and above ground fresh weight of shoots. In the present study, a greenhouse experiment was constructed to investigate response of pepper plants against the TEV agent, which has been identified in Karaisali pepper populations and noticed that it has an impact on the crop. In this experiment, three prominent pure Karaisali pepper lines were inoculated with TEV using mechanical inoculation method and the yield and quality losses in Karaisali pepper population due to this virus were determined.

2. Material and Methods

K7, K25, K34 pure pepper line seeds obtained from the Alata Horticultural Research Institute in Mersin were planted. These specific lines were selected because of their overall promising horticultural attributes. The plants were grown in a greenhouse having full control and drip irrigation systems, and some chemical fertilizers and pesticides were applied when necessary. In pomological analyzes scales, rulers, refractometer, calipers, 2 and 5 liter beakers were used for measurements.

2.1. Cultivation of Karaisali pepper population lines

The line reaction experiment was carried out in greenhouses in the Alata Horticultural Research Institute in 2015 and 2016 years. The experiment was conducted using a randomized blocks design, with a control for each line, five repetitions and 25 plants were grown in each repetition. The experiment included virus-infected pepper plants and healthy pepper plants in the control group. The seeds were planted in viols located in a climate room during the first week of March due to climate requirements and the developed seedlings were transferred into the fully-controlled greenhouses in the middle of April when they were at the stage of 3-4 leaf and routine applications were applied.

2.2. Inoculation of virus isolate by mechanical inoculation method

During the surveys carried out in 2014-2015, TEV, which was the most destructive virus species in Karaisali pepper, local population cultivated widely in Karaisali district of Adana province, was isolated from a single virus infected pepper plant and inoculated on test pepper plants for reproducing the virus. Leaf samples obtained from pepper plants, which were determined to be infected by TEV using double antibody sandwich enzyme-linked immunosorbent assay (DAS-ELISA) test, were crushed in a porcelain container with 0,02 M potassium phosphate buffer (pH 7.0) (0.02 M KH_2PO_4, 0.02 M Na_2HPO_4) containing 0.1% 2-mercaptoethanol at the rate of 1:5 (w/v). The obtained inoculum was filtered through 2 layers of cheesecloth. The leaves of the pepper lines were dusted with carborumdum powder then the inoculum was rubbed on to the leaves using a sponge (Çelik et al 2010). After inoculation, the plant leaves were washed with tap water to remove carborundum powder and plant residues. The virus symptoms were observed 10-15 days after inoculation and confirmed by the DAS-ELISA (Clark & Adams 1977).

2.3. Fruit yield

At this stage, pepper fruits harvested from the control and virus-infected plots were classified based on the fruit size and the total fruit weight in each plot was determined. Pepper paste was produced from these Karaisali pepper lines, and the paste yield was determined.

2.4. Fruit quality

When the pepper fruits in plots reached the harvest maturity, the following fruit quality properties were examined (IPGRI 1995). For fruit quality examination, pepper fruits collected at the second harvest were used. For every repetition in healthy and infected plots, 25 pepper fruits representing each line were selected. A total of 375 healthy and 375 infected pepper fruits were examined for 3 lines, 125 pepper fruits each line. The effect of TEV on average fruit diameter (cm), average fruit length (cm), average fruit volume (mL), fruit wall thickness (mm), fruit color, fruit total dry matter production (%), fresh and dry weight of all green parts (kg) and market value (1st quality: 16-18 cm, smooth, shiny, with the color specific to the species; 2nd quality: small fruits, deformed, cracked, with distorted color) of the fruits were determined. The results of the experiment were analyzed with JMP statistics software LSD test and at 5% significance level. In the experiment related to responses of pepper lines to TEV, the yield and quality losses caused by the virus in peppers were determined by the following formula:

The % effect of the reaction test= (Application/control) × 100

3. Results and Discussion

3.1. Analysis of fruit and paste yields

The yield was recorded during five harvesting periods. In 2016, however, three harvesting periods were made due to low fruit yield in the infected plots and five harvests were made in healthy plots. The average yield values, loss ratios and analysis results for the healthy and infected line plots are presented in Table 1. The yield loss ratios caused by TEV infection were determined for K7 (59.9% and 95.1%), K25 (48.2% and 86.3%) and K34 (13.4% and 82.5%) lines in 2015 and 2016, respectively. Nutter et al (1989) also conducted field experiments in northeast Georgia to quantify the effect of TEV epidemics on yield of pepper. They found that early season infection reduced yield 74% in 1986 and 73% in 1987. The disease severity varied between 2015 and 2016 in the line reaction experiments. In both years, the experiment was conducted in mid-April. There were differences in yield between two consecutive years, probably due to impact of climatic changes that occurred in the immediate aftermath of inoculation, probably affecting the propagation and spread of the virus in the plant. In the healthy and infected plots in both years, the decrease in the yield was parallel and correlated. In the present study, K34 line was the least affected line by TEV infection based on average yield. Ramkat et al (2006) was conducted a study in a greenhouse to determine the effect of mechanical inoculation of *tomato spotted wilt tospovirus* (TSWV) on the severity of the disease on tomato varieties ‘Cal J’, ‘Marglobe’, ‘Money maker’, ‘Roma’ and ‘Riogrande’ and its impact on yield. Mechanical inoculation reduced total yield of ‘Cal J’, ‘Riogrande’, ‘Money maker’, ‘Marglobe’ and ‘Roma’ by 60, 55.3, 45.1, 40.3 and 27%, respectively. The effects of TSWV on yield and quality of tomato fruits were also studied in Samsun Province of Turkey in 2004. It was found that TSWV caused 42.1% and 95.5% reduction in yield and marketable value of tomato, respectively, (Sevik & Arlı-Sokmen 2012). Furthermore, pepper paste was made using each repetition of healthy and infected plots of three pepper lines. Statistical analysis regarding the paste weight is presented in Table 1. Loss ratios demonstrated that K34 line exhibited the highest loss with 41.2%, followed by K25 line with 25.7% and K7 line with 19.1% loss in 2015. In 2016, the line with the highest loss was K25 with 39.6%, followed by K7 line with 36.7%. The K34 line was the least reactive line with a 26%

loss. Pepper paste was produced from pepper fruits and the dry matter content was measured with a refractometer. It was determined that the TEV did not decrease the dry matter content in the pepper paste (Table 2).

3.2. Fruit quality

3.2.1. Dry matter production in fruit

Pepper fruits were crushed and total dry matter content of fruit was examined with a digital refractometer. Examination of the water-soluble dry matter (WSDM) content of pepper fruits in the control and infected plant lines demonstrated that the difference between the applications was not statistically significant at 5% level (Table 2). When infected pepper lines were compared to healthy plots in 2015, it was found that WSDM value was 0.1 brix-0.6 brix high, while in 2016, there was a 0.4-2.6 brix decrease in the WSDM value in all infected plant lines.

3.2.2. Fruit weight

Average fruit weight and percentage loss ratios for the lines were determined. Fruit weight loss was the highest in K34 line (48.5%) in 2015, followed by K7 (46.1%) and K25 (45.7%) lines. In 2016, the K7 line exhibited the highest loss with 56.4%, followed by K25 with 53.1% loss and K34 with 42% loss (Table 3). Padgett et al (1990) also reported that, TEV affected pepper yield by reducing the average weight of the fruit and the number of fruit. Ramkat et al (2006) observed that, TSWV inoculation on tomatoes caused reduction of varying magnitudes in total fruit weight when compared to their health controls.

3.2.3. Fruit diameter

Table 3 demonstrates that the highest loss in fruit diameter among the lines in 2015 was observed in K34 line with 22.2%, followed by K7 line with 20% loss and K25 line with 12.1% loss, the highest loss in fruit diameter in 2016 was observed in K34 line with 20.5% loss, followed by K25 with 17.9% and K7 with 17.2% (Table 3).

3.2.4. Fruit length

Based on the average fruit length, the difference between applications in 2015 and 2016 was significant at 5% for all lines. While K25 line exhibited the highest loss in 2015 with 41.9%, K7 exhibited 35.1% and K34 exhibited 30.1% loss. In 2016, the K25 line exhibited the highest loss with 32.1%, followed by K7 with 24.8% and K34 with 23.9% loss (Table 4). Fruit length of Karaisali pepper line is an important property in determining the market value. The minimum fruit length loss was recorded with K34 line in both experiment years.

3.2.5. Fruit volume

It was found that the difference in fruit volume between healthy control and infected K7, K25 and K34 line plants was significant at 5% level for 2015 and 2016 (Table 4). In the 2015 reaction experiment, the highest loss in fruit volume among the applications was observed in K25 line with 49.7%, followed by K34 with 49% and K7 with 45.6% loss. In 2016, the least fruit volume loss was observed in K34 line with 53.3% among the K7, K25 and K34 lines. Fruit volume is important since it demonstrates that the fruit is fully grown in shape and size. When the line reaches its unique fruit volume, the dry matter/water ratio would be good, and the food quality increases. Because, this affects the balance of the secondary compounds in the fruit as well. This ensures a good taste. Furthermore, the volume and length of the fruit is important, facilitating the use of these types of peppers in industrial pepper paste and grilled pepper production.

3.2.6. Fruit wall thickness

In both years, the difference between applications in K7, K25 and K34 lines was not significant at 5% significance level (Table 5). However, it was determined that there were losses in fruit wall thickness of all lines at varying ratios. The highest loss was found in K7 line with 16.6%, followed by K25 with 11.8% and K34 with 10.1% in 2015. In 2016, the highest loss was found in K34 with 13.2%, followed by K7 with 11.8% and K25 9%.

Table 1- Average fruit and paste yields and loss ratio of Karaisali pepper lines

Application	*Yield kg 125 plant $^{-1}$*						*Pepper paste yield kg 25 fruit $^{-1}$*					
	K7		*K25*		*K34*		*K7*		*K25*		*K34*	
	2015	*2016*	*2015*	*2016*	*2015*	*2016*	*2015*	*2016*	*2015*	*2016*	*2015*	*2016*
Healthy (Control)	284.5^{ab}	106.9^{b}	298.5^{a}	80.8^{c}	255.0^{ab}	149.5^{a}	0.204^{cd}	0.271^{b}	0.374^{b}	0.317^{a}	0.675^{a}	0.322^{a}
Virus infected	114.0^{d}	5.3^{d}	154.7^{cd}	11.0^{d}	220.8^{bc}	26.2^{d}	0.165^{d}	0.070^{d}	0.278^{c}	0.082^{d}	0.397^{b}	0.166^{c}
Average	199.3^{a}	56.1^{b}	226.6^{a}	45.9^{b}	237.9^{a}	87.9^{a}	0.185^{c}	0.171^{b}	0.326^{b}	0.200^{b}	0.536^{a}	0.244^{a}
Loss ratio (%)	59.9	95.1	48.2	86.3	13.4	82.5	19.1	36.7	25.7	39.6	41.2	26.0
2015 year			2016 year				2015 year			2016 year		
LSD 5% application: 7.64			LSD 5% application: 1.67				LSD 5% application: 0.0544			LSD 5% application: 0.0249		
LSD 5% line: 9.35			LSD 5% line: 2.04				LSD 5% line: 0.0666			LSD 5% line: 0.0306		
LSD 5% application × line: 13.23			LSD 5% application × line: 2.05				LSD 5% application × line: 0.0942			LSD 5% application × line: 0.0432		

Table 2- Content of water-soluble dry matter (WSDM) for Karaisali pepper fruit and paste (brix)

Application	*Fruit WSDM (water-soluble dry matter)*						*Pepper paste WSDM (water-soluble dry matter)*					
	K7		*K25*		*K34*		*K7*		*K25*		*K34*	
	2015	*2016*	*2015*	*2016*	*2015*	*2016*	*2015*	*2016*	*2015*	*2016*	*2015*	*2016*
Healthy (Control)	6.72^{c}	6.68^{b}	8.06^{ab}	8.10^{a}	7.12^{c}	7.78^{a}	51.64^{a}	46.46^{ab}	47.72^{b}	47.76^{a}	45.86^{b}	38.22^{b}
Virus infected	7.28^{bc}	6.74^{b}	8.42^{a}	7.72^{a}	7.24^{c}	6.20^{b}	47.02^{b}	50.93^{a}	53.48^{a}	47.18^{a}	46.54^{b}	43.32^{ab}
Average	7.00^{b}	6.71^{b}	8.24^{a}	7.91^{a}	7.18^{b}	6.99^{b}	49.33^{a}	48.69^{a}	50.45^{a}	47.47^{a}	46.20^{b}	40.77^{b}
2015 year			2016 year				2015 year			2016 year		
LSD 5% application: n.s			LSD 5% application: 0.506				LSD 5% application: n.s			LSD 5% application: n.s		
LSD 5% line: 0.58			LSD 5% line: 0.620				LSD 5% line: 2.59			LSD 5% line: 6.01		
LSD 5% application × line: n.s			LSD 5% application × line: 0.620				LSD 5% application × line: 3.66			LSD 5% application × line: n.s		

n.s, nonsignificant

Table 3- Average fruit weight, fruit diameter and % loss ratio of Karaisali pepper lines

Application	*Fruit weight (g)*						*Fruit diameter (cm)*					
	K7		*K25*		*K34*		*K7*		*K25*		*K34*	
	2015	*2016*	*2015*	*2016*	*2015*	*2016*	*2015*	*2016*	*2015*	*2016*	*2015*	*2016*
Healthy (Control)	1525.6^{c}	1120.62^{c}	2150.0^{b}	1348.54^{b}	2609.6^{a}	1739.1^{a}	3.48^{b}	2.86^{b}	3.33^{b}	2.82^{b}	4.48^{a}	3.92^{a}
Virus infected	822.4^{e}	488.58^{d}	1167.4^{d}	632.58^{d}	1344.4^{cd}	1008.14^{c}	2.80^{c}	2.40^{c}	2.90^{c}	2.25^{c}	3.54^{b}	3.14^{b}
Average	1174.0^{c}	804.59^{c}	1658.7^{b}	990.56^{b}	1977.0^{a}	1373.61^{a}	3.14^{b}	2.63^{b}	3.11^{b}	2.54^{b}	4.01^{a}	3.53^{a}
Loss ratio (%)	46.1	56.4	45.7	53.1	48.5	42.0	20	17.2	12.1	17.9	22.2	20.5
2015 year				2016 year			2015 year			2016 year		
LSD 5% application: 137.59				LSD 5% application: 121.71			LSD 5% application: 0.24			LSD 5% application: 0.22		
LSD 5% line: 168.51				LSD 5% line: 149.06			LSD 5% line: 0.29			LSD 5% line: 0.27		
LSD 5% application × line: 238.31				LSD 5% application × line: n.s			LSD 5% application × line: n.s			LSD 5% application × line: n.s		

n.s, nonsignificant

Table 4- Average fruit length, fruit volume and % loss ratio of Karaisali pepper lines

Application	*Fruit length (cm)*						*Fruit volume (mL)*					
	K7		*K25*		*K34*		*K7*		*K25*		*K34*	
	2015	*2016*	*2015*	*2016*	*2015*	*2016*	*2015*	*2016*	*2015*	*2016*	*2015*	*2016*
Healthy (Control)	15.44^{b}	13.34^{b}	19.78^{a}	15.90^{a}	15.29^{b}	14.20^{b}	1692^{b}	774^{b}	2458^{a}	1108^{a}	2668^{a}	1335^{a}
Virus infected	10.04^{c}	9.96^{c}	11.46^{c}	10.76^{c}	10.72^{c}	10.81^{c}	920^{d}	196^{d}	1236^{cd}	346^{cd}	1362^{bc}	624^{bc}
Average	12.74^{b}	11.65^{b}	15.62^{a}	13.33^{a}	13.00^{b}	12.50^{ab}	1306^{b}	485^{c}	1847^{a}	727^{b}	2015^{a}	980^{a}
Loss ratio (%)	35.1	24.8	41.9	32.1	30.1	23.9	45.6	74.7	49.7	68.8	49.0	53.3
2015 year				2016 year			2015 year			2016 year		
LSD 5% application: 1.18				LSD 5% application: 0.97			LSD 5% application: 223.72			LSD 5% application: 174.12		
LSD 5% line: 1.44				LSD 5% line: 1.19			LSD 5% line: 274.00			LSD 5% line: 213.26		
LSD 5% application × line: 2.04				LSD 5% application × line: n.s			LSD 5% application × line: n.s			LSD 5% application × line: n.s		

n.s, nonsiginificant

3.2.7. Fruit color

Fruit color was examined based on hue value (fruit color tone), a value (red-green) and L value (light-dark color) and statistical analyzes were conducted (Tables 5 and 6). Examination of the fruit color hue value demonstrated that there was no significant difference between healthy and infected plots in K7, K25 and K34 lines for 2015 at 5% significance level. However, in 2016, a significant difference was found between the healthy and infected plots at 5% significance level in K7 and K34 lines, however the difference was not significant in K25 line, similar to 2015 findings. The disease affected the K7 and K34 lines to acquire their true color. Based on 2015 data, the difference between the plots in all lines was not statistically significant based on fruit color a value (red-green) at 5% significance level. The difference between the values for K7 and K34 lines was statistically significant in 2016 (Table 6). The difference between healthy and infected plots of all lines based on the fruit color L value (light-dark color) was not significant at 5% significance level (Table 6).

3.2.8. Fresh and dry weight of green parts

Initially, the fresh weight of the green parts of the pepper plants removed after the final harvest was determined, and statistical analysis was conducted (Table 7). It was determined that for 2015, the maximum loss in fresh weight was recorded in K25 line (51.6%), followed by K7 (48.4%) and K34 (37.5%) lines for 2015. Based on green parts fresh weight, highest loss was recorded in K34 line (46.5%) in 2016, followed by K7 line (46.3%) (Table 7). The lowest loss was obtained by K25 line (25.1%). Low green parts weight reflects lower photosynthesis, which in turn reduces the production of assimilative materials, reducing the yield and quality. In a study conducted by Murphy & Bowen (2006), the correlation between root length and fresh weight was investigated on the isolates obtained from bell pepper plants mixed infected with CMV and PepMoV for each virus. For this purpose, they conducted three applications (only CMV, only PepMoV, and CMV+PepMov) and inoculated the viruses they isolated to pepper cultivars and used the ELISA method for the diagnosis of viruses. In each experiment, they measured root length development and surface fresh weight. They found that these values were significantly lower for CMV+PepMoV mixed infection when compared to species infected with a single virus. On the other hand, Murphy & Morawo (2017) reported that above ground fresh weight of TEV-strain Mex21 infected pepper plants was significantly less than for the healthy control. Al-Saleh et al (2007) was evaluated selected peanut cultivars for reaction to TSWV in field plots in 2001, 2002, and 2003. They reported that across all three times of inoculation, significant reductions in fresh weight were observed in all cultivars except Okrun when inoculated at 5 days post-planting. Dry Weight of Green Parts: The green parts fresh weight was obtained for all lines and they were placed in brown paper bags and dried up to 10% humidity in an incubator at 55-60 ºC and dry weigh data were obtained, and statistical analysis was conducted (Table 7). In 2015, the highest loss was recorded in K34 line (46.4%), followed by K7 (45.9%) and K25 (42%). The highest loss in 2016 was recorded in K7 line (51.5%), followed by K34 (43.7%).

3.2.9. Market value

Market value of the fruits was determined as follows: 1st quality: 16-18 cm, smooth, shiny, with the color specific to the species; 2nd quality: small fruits, deformed, cracked, with distorted color. The first and second quality counts and ratios for healthy plots for both years are presented in Table 8, and for infected lines; the same values are presented in Table 9. In general, when 2015 and 2016 figures are examined respectively, it was determined that the highest ratio of first quality pepper in healthy plots was observed in K25 (96.8%, 69.6%) line, while the same ratio decreased to 19.2% and 16% in the infected plots. Ramkat et al (2006) was found that TSWV caused a severe decrease in the marketable yield of the tomato fruits that was significant across the varieties. A 90% reduction in marketable yield was observed in variety ‘Cal J’ followed by ‘Marglobe’, ‘Riogrande’ and ‘Money maker’, with 62, 41 and 40%.

Table 5- Average fruit wall thickness, fruit color (Hue) and % loss ratio of Karaisali pepper lines

Application	*Fruit wall thickness (mm)*						*Fruit color (Hue value)*					
	K7		*K25*		*K34*		*K7*		*K25*		*K34*	
	2015	*2016*	*2015*	*2016*	*2015*	*2016*	*2015*	*2016*	*2015*	*2016*	*2015*	*2016*
Healthy (Control)	3.80^{a}	2.46^{bc}	4.15^{a}	3.23^{a}	3.96^{a}	3.41^{a}	27.63^{ab}	40.65^{a}	29.63^{a}	38.52^{a}	26.65^{b}	33.40^{ab}
Virus infected	3.17^{b}	2.17^{c}	3.67^{ab}	2.94^{ab}	3.56^{ab}	2.96^{ab}	26.04^{b}	20.90^{cd}	29.02^{a}	28.69^{bc}	27.83^{ab}	13.52^{d}
Average	3.49^{a}	2.31^{b}	3.91^{a}	3.08^{a}	3.76^{a}	3.18^{a}	26.84^{b}	30.77^{a}	29.33^{a}	33.60^{a}	27.24^{b}	23.46^{b}
Loss ratio (%)	16.6	11.8	11.8	9.0	10.1	13.2						
2015 year				2016 year			2015 year			2016 year		
LSD 5% application: 0.34				LSD 5% application: 0.31			LSD 5% application: N.S			LSD 5% application: 5.47		
LSD 5% line: N.I				LSD 5% line: 0.37			LSD 5% line: 1.60			LSD 5% line: 6.70		
LSD 5% application × line: n.s				LSD 5% application × line: n.s			LSD 5% application × line: n.s			LSD 5% application × line: n.s		

n.s, nonsiginificant

Table 6- Average fruit color "L" and "a" and % loss ratio of Karaisali pepper lines

Application	*Fruit Color (L value)*						*Fruit Color (a value)*					
	K7		*K25*		*K34*		*K7*		*K25*		*K34*	
	2015	*2016*	*2015*	*2016*	*2015*	*2016*	*2015*	*2016*	*2015*	*2016*	*2015*	*2016*
Healthy (Control)	34.59^{a}	39.39^{b}	33.11^{ab}	41.41^{ab}	31.05^{b}	40.11^{ab}	30.17^{b}	33.82^{a}	25.50^{c}	33.04^{a}	25.34^{c}	26.36^{ab}
Virus infected	34.96^{a}	42.76^{ab}	33.02^{ab}	43.62^{a}	31.11^{b}	41.17^{ab}	33.14^{a}	16.98^{cd}	26.52^{c}	24.39^{bc}	25.65^{c}	9.85^{d}
Average	34.78^{a}	41.07^{a}	33.07^{a}	42.51^{a}	31.07^{b}	40.64^{a}	31.66^{a}	25.40^{a}	26.00^{b}	28.72^{a}	25.50^{b}	18.11^{b}
2015 year				2016 year			2015 year			2016 year		
LSD 5% application: n.s				LSD 5% application: 2.16			LSD 5% application: n.s			LSD 5% application: 4.76		
LSD 5% line: 1.82				LSD 5% line: N.I			LSD 5% line: 2.02			LSD 5% line: 5.84		
LSD 5% application × line: n.s				LSD 5% application × line: n.s			LSD 5% application × line: n.s			LSD 5% application × line: n.s		

n.s, nonsignificant

Table 7- Average fresh and dry weights of green parts and % loss ratio of Karaisali pepper lines

Application	*Fresh weight of green parts (kg)*						*Dry weight of green parts (kg)*					
	K7		*K25*		*K34*		*K7*		*K25*		*K34*	
	2015	*2016*	*2015*	*2016*	*2015*	*2016*	*2015*	*2016*	*2015*	*2016*	*2015*	*2016*
Healthy (Control)	1.52^{a}	0.97^{a}	1.35^{ab}	0.60^{b}	0.72^{c}	0.66^{b}	0.39^{a}	0.27^{a}	0.31^{a}	0.17^{bc}	0.18^{b}	0.17^{b}
Virus infected	0.79^{bc}	0.52^{bc}	0.65^{c}	0.45^{bc}	0.45^{c}	0.35^{c}	0.21^{b}	$0.13b^{cd}$	0.18^{ab}	0.11^{cd}	0.10^{ab}	0.10^{d}
Average	1.15^{a}	0.75^{a}	1.00^{a}	0.53^{b}	0.59^{b}	0.51^{b}	0.30^{a}	0.20^{a}	0.25^{ab}	0.14^{b}	0.141^{b}	0.13^{b}
Loss ratio (%)	48.4	46.3	51.6	25.1	37.5	46.5	45.9	51.5	42.0	33.7	46.4	43.7
2015 year				2016 year			2015 year			2016 year		
LSD 5% application: 0.287				LSD 5% application: 0.123			LSD 5% application: n.s			LSD 5% application: 0.04		
LSD 5% line: 0.351				LSD 5% line: 0.151			LSD 5% line: 0.116			LSD 5% line: 0.04		
LSD 5% application × line: n.s				LSD 5% application × line: n.s			LSD 5% application × line: n.s			LSD 5% application × line: n.s		

n.s, nonsignificant

Table 8- Market value of healthy plots of Karaisali pepper lines

Lines	*2015 Year*					*2016 Year*			
	Pepper number of repetition	*1. Quality number of peppers*	*2. Quality number of peppers*	*1. Quality ratio %*	*2. Quality ratio %*	*1. Quality number of peppers*	*2. Quality number of peppers*	*1. Quality ratio %*	*2. Quality ratio %*
K7	125	55	70	44.0	56.0	36	89	31.2	71.2
K25	125	121	4	96.8	3.2	87	38	69.6	30.4
K34	125	46	79	36.8	63.2	55	70	44.0	56.0

Table 9- Market value of infected plots of Karaisali pepper lines

Lines	*2015 Year*					*2016 Year*			
	Pepper number of repetition	*1. Quality number of peppers*	*2. Quality number of peppers*	*1. Quality ratio %*	*2 .Quality ratio %*	*1. Quality number of peppers*	*2. Quality number of peppers*	*1. Quality ratio %*	*2. Quality ratio %*
K7	125	2	123	1.6	98.4	4	121	3.2	96.8
K25	125	24	101	19.2	80.8	20	105	16.0	84.0
K34	125	8	117	6.4	93.6	9	116	7.2	92.8

4. Conclusions

The present study involving yield and quality analyses demonstrated that the K34 line among the 3 good pepper lines obtained from the selective breeding of Karaisalı pepper populations by Alata Horticultural Research Institute Directorate was the less affected line by TEV when compared to other lines scrutinized in the study. The breeding institution applied to Seed Registration and Certification Center for the registration of the K34 line, which demonstrated high yield and quality properties and the lowest impact by the TEV agent and the seed was finally registered as the "Hayriye" variety. This study has shown the magnitude of yield and quality losses due to varying rates of TEV infection in Karaisali pepper lines grown under controlled conditions. To mitigate the damage caused by TEV, the cultural measures such as ways to prevent virus infection can be undertaken. However, the use of resistant cultivars could be the ultimate solution. Since significant differences among the pepper lines for the loss and damage were also recovered, our results suggest that new pepper cultivars exhibiting overall Karaisalı pepper attributes and minimizing the TEV damage could be developed.

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Lawrence K C, Funk D B & Windham W R (2001). Dielectric moisture sensor for cereal grains and soybeans. *Transactions of the ASAE* 44(6): 1691-1696

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Conference Proceedings (Full papers)

Yağcıoğlu A, Değirmencioğlu A & Cağatay F (1999). Drying characteristics of laurel leaves under different drying conditions. In: *Proceedings of the 7th International Congress on Agricultural Mechanization and Energy*, 26-27 May, Adana, pp. 565-569

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