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Variations in oil, protein, fatty acids and vitamin E contents of pumpkin seeds under deficit irrigation

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SUMMARY: In the present study, pumpkin (*Cucurbita Pepo* L.) was grown under water stress to determine its effects on the chemical composition of the seeds (i.e., oil, protein, fatty acids and vitamin E), in Kayseri, Turkey. Irrigation treatments were designed to supply different portions of depleted moisture within the efficient root zone of the plants (60 cm). The treatments were arranged as supplying 100% (I_{100}), 80% (I_{80}), 60% (I_{60}), 40% (I_{40}), 20% (I_{20}) and 0% (I_0) of depleted moisture through a drip irrigation system. The effects of irrigation levels on the oil content of pumpkin seeds were found to be significant ($p < 0.01$). The oil contents of irrigation treatments varied between 26% (I_0, dry) and 64% (I_{100}, full irrigation). However, the effects of deficit irrigation on protein, fatty acids and vitamin E contents were not found to be significant. The vitamin E contents varied from 41.6 – 55.3 mg/100 g; while the protein contents varied from 28.5–37.7%. Six different fatty acids (linolenic, linoleic, oleic, stearic, palmitic and myristic acid) were examined. The average concentration of palmitic, stearic, oleic and linoleic acids ranged from 10.7–12.6%, 6.4–10.4%, 39.6–48.9% and 32.4–35%, respectively. Myristic and linolenic acids were not detected in the pumpkin seeds.

KEYWORDS: *Fatty acid; Irrigation; Oil content; Pumpkin seed*

RESUMEN: ***Variaciones en los contenidos de aceite, proteínas, ácidos grasos y vitamina E de las semillas de calabaza con riego deficitario.*** En este trabajo se cultivaron calabazas (*Cucurbita Pepo* L.) en Kayseri, Turquía, con el objetivo de determinar los efectos del estrés hídrico en la composición química de las semillas (aceite, proteínas, ácidos grasos y vitamina E). Los tratamientos de irrigación se realizaron mediante el suministro de diferentes porciones de humedad dentro de la zona de la raíz eficiente de las plantas (60 cm). Los tratamientos se organizaron para suministrar 100% (I_{100}), 80% (I_{80}), 60% (I_{60}), 40% (I_{40}), 20% (I_{20}) y 0% (I_0) de humedad controlada a través del sistema de riego por goteo. Los efectos de los niveles de irrigación sobre el contenido de aceite de las semillas de calabaza fueron significativos ($p < 0.01$). El contenido de aceite en función de los tratamientos de riego varió entre el 26% (I_0, seco) y el 64% (I_{100}, riego completo). Sin embargo, los efectos del déficit de irrigación sobre los contenidos de proteínas, ácidos grasos y vitamina E no fueron significativos. Los contenidos de vitamina E variaron entre 41,6 y 55,3 mg/100 g, mientras que los contenidos de proteína variaron entre 28,5 y 37,7%. Se determinaron seis ácidos grasos (linolénico, linoleico, oleico, esteárico, palmítico y mirístico). La concentración promedio de los ácidos palmítico, esteárico, oleico y linoleico osciló entre 10,7–12,6%, 6,4–10,4%, 39,6–48,9% y 32,4–35%, respectivamente. Los ácidos mirístico y linolénico no fueron detectaron en las semillas de calabaza.

PALABRAS CLAVE: *Ácido graso; Contenido de aceite; Irrigación; Semillas de calabaza*

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1. INTRODUCTION

Fats, fatty acids and their metabolic by-products have various significant functions in the human metabolism. They usually provide an efficient source of energy, along with resistance to external factors. They constitute the basic building blocks of cell membranes (Lee, 1994). Since omega-3 and omega-6 fatty acids are not synthesized in the human body, they should be supplied externally. They are called essential fatty acids and play significant roles in the human metabolism (Erdinc *et al*., 2018).

Vegetable oils are used worldwide in human nutrition, food and industrial purposes. Despite ample sources, the world's vegetable oil production mostly comes from soybean, sunflower, rapeseed and date palms. Although pumpkin seed oil has sufficient quality attributes for human nutrition and industrial purposes, it has not been used much worldwide (Stevenson *et al*., 2007). The oil content of pumpkin seeds generally varies from 10.9–54.0% (Stevenson *et al*., 2007; Murkovic *et al*., 1996; Seymen *et al*., 2016). Pumpkin seed oil is generally used as salad dressing in Australia, Slovenia and Hungary. Although pumpkin seeds are largely used either fresh or roasted as appetizers (Murkovic *et al*., 1996; Ardabili *et al*., 2011), they have been used for medicinal purposes since ancient times.

Pumpkin seed oil is quite rich in unsaturated fatty acids and that makes it a valuable source of oil for human nutrition. Pumpkin seed oil has high oleic and linoleic unsaturated fatty acid contents and low palmitic and stearic saturated acid contents. Pumpkin seeds are also rich in vitamin E and protein (Ermiş, 2010). With these valuable quality attributes, pumpkin seeds prevent prostate enlargement, arthrolith, reduce cholesterol levels and regulate blood pressure (Stevenson *et al*., 2007). Pumpkin seeds also prevent hypertension, stimulate hyperglycemic activity and thus prevent diabetes (Stevenson *et al*., 2007; Fu *et al*., 2006; Imaeda *et al*., 1999). Pumpkin seeds are used as an important source of vitamin E (tochopherol) in Japanese diets.

Many physiological, molecular and biochemical reactions of plants are influenced by water stress. Seed fat and fatty acid compositions are largely influenced by the cultivar, soil and climate conditions and cultural practices (Nawirska-Olszanska *et al*., 2013). Parallel findings were reported in previous studies carried out with different plant species (Kaplan *et al*., 2017 for maize; Ali and Ullah, 2012 for sunflower; Kırnak *et al*., 2010, for soybean).

In Turkey, the cultivar *Cucurbita pepo* is commonly used and *Cucurbita moschata* is less frequently used for pumpkin seed production. Annually, 42181 tons of pumpkin seeds are produced from 62844.1 ha of land area in Turkey. The Kayseri province, with 15053 tons of production annually, from 31310.1 ha of land area, constitutes about 35.7% of the country's production (TUIK 2016).

Although a few studies have been conducted regarding water stress on pumpkin growth, seed yield and some seed quality properties such as 1000-seed weight and oil content, no studies have been found about the effects of water stress on the other properties of pumpkin seeds. Therefore, this study was conducted to determine the effects of different irrigation regimes on oil content, fatty acids, protein and vitamin E content of pumpkin seeds produced in the central Anatolia region of Turkey.

2. MATERIALS AND METHODS

Experiments were conducted in the experimental fields of the Agricultural Research and Implementation Center of Erciyes University for two years, in 2015 and 2016. The research site with an altitude of 1094 m is located between 34° 56' and 36° 59' east longitude and between 37° 45' and 38° 18' north latitude. Long-term and 2015–2016 climate data are respectively provided in Tables 1 and 2. As can be inferred from Table 2, the precipitation throughout the experiments was 142 mm in 2015 and 179.4 mm in 2016.

Soil samples were taken from the 0–120 cm soil profile of 3 different locations within the research site. Water samples were taken from an irrigation water supply well and analyses were performed in accordance with Ayyıldız (1990). Irrigation water has a pH of 7.60 and EC of 242 µS; thus it was classified as C1S1 (Ayyıldız, 1990). The results from the soil analysis are provided in Table 3. The stable rate of infiltration was measured as 23.3 mm/h.

Experiments were conducted in a randomized block design with 3 replications. Sowing was performed with 100-cm-row spacing and 60-cm on-row plant spacing. Since there aren't any registered varieties, the commonly-used, so-called "framed" seeds were used. Each plot had 8 rows. Two side rows and a plant row from the top and bottom sections of the plots were omitted so as to consider side-effects. About 2 m spacing was provided between the plots and 2.4 m between the blocks to prevent interactions. Based on soil analysis results, DAP fertilizer (12 kg/da N, 12 kg/da P and 10 kg/da K) was supplied through fertigation. Since the soil had sufficient K levels, K fertilization was not performed.

A drip irrigation system was used for irrigation. They system was designed in accordance with Keller and Bliesner (1990). Dripper spacing was 0.33 m, dripper discharge rate was 4 lt/h, lateral diameter was 20 mm, manifold pipe diameter was 40 mm and main pipe line diameter was 63 mm. Water flow meters were placed at the entrance of each block and applied water quantities were measured. The wetted area percentage was calculated as 66% (Keller and Bliesner, 1990).

TABLE 1. Long-term climate data for the Kayseri province (1950-2016)

Climate Data	Jan	Feb	Mar	Apr	May	Jun	Jul	Aus	Sep	Oct	Nov	Dec
T_{avr} (°C)	−1.7	0.1	4.7	10.6	15.0	19.0	22.2	22.0	17.3	11.8	5.5	0.6
T_{max} (°C)	4.1	6.1	11.4	17.7	22.5	26.8	30.6	30.7	26.5	20.4	13.0	6.4
T_{min} (°C)	−6.9	−5.2	−1.4	3.1	6.8	9.7	11.9	11.4	7.3	3.5	−0.9	−4.5
U_2 ($m \cdot s^{-1}$)	1.1	1.3	1.6	1.7	1.4	1.3	1.3	1.2	1.1	1.0	1.0	1.1
Precipitation (mm)	35.2	36.5	41.8	52.1	51.8	39.5	10.5	8.8	15.0	28.0	32.4	37.4
RH_{avr} (%)	76.6	74.0	68.3	62.3	61.2	55.8	49.5	49.2	54.1	63.6	71.7	77.1
RH_{max} (%)	96.4	96.6	96.7	96.3	95.3	93.0	87.0	87.1	94.3	97.0	97.1	97.4
RH_{min} (%)	38.8	34.4	22.5	18.6	19.0	19.4	17.2	16.9	17.4	20.3	28.2	36.8
Sunshine Duration (hour)	3.0	4.0	4.8	6.2	8.3	10.4	11.9	11.4	9.1	6.7	4.8	3.0
Solar Radiation (MJ m^{-2}.day)	7.0	9.8	13.1	16.1	19.0	21.8	22.7	20.6	17.0	11.6	8.0	6.1
ETo (mm/ay)	24	31	62	92	119	137	158	147	107	70	40	26

U_2: Wind speed at 2 m height, RH: Relative Humidity, ETo: Reference Evapotranspiration.

TABLE 2. Climate data for experimental years (2015–2016)

Climate Data (2015)	May	June	July	August	
T_{avr} (°C)	15.89	18.15	22.15	26.69	
T_{max} (°C)	22.55	24.33	30.06	32.67	
T_{min} (°C)	9.34	11.98	14.24	20.71	
Wind Speed (m/s)	1.75	1.46	1.69	1.87	
Precipitation (mm)	25.6	114.8	0.4	1.2	
RH_{max} (%)	77.62	85.72	70.56	60.26	
RH_{min} (%)	31.79	39.71	23.52	22.82	
Climate Data (2016)	**April**	**May**	**June**	**July**	**August**
T_{avr} (°C)	14.02	14.83	20.41	23.33	25.38
T_{max} (°C)	20.4	26.7	34.6	37	34.8
T_{min} (°C)	4.5	4.4	7.5	10.8	14.5
Wind Speed (m/s)	1.57	1.88	1.75	1.81	1.81
Precipitation (mm)	0	151.8	25.6	2	0
RH_{max} (%)	65.2	80	78.2	66.1	62.4
RH_{min} (%)	25.5	34.4	30.8	21.1	19.99

TABLE 3. Soil characteristics of the research site

	Soil depth			
Soil characteristics	**0–30 cm**	**30–60 cm**	**60–90 cm**	**90–120 cm**
Texture	Loamy	Loamy	Clay-Loam	Loamy
EC, dS/m	0.22	0.173	0.258	0.191
pH	8.13	8.17	8.14	8.23
Field Capacity, Pw_{FC} (%)	23	26	26	25
Permanent Wilting Point, Pw_{PWP} (%)	10.73	11.38	9.3	9.37
Bulk Density, g/cm^3	1.27	1.24	1.22	1.28
Organic Matter, %	1.25	1.05	0.69	0.73
Lime, %	2.54	5.83	3.15	6.2
N, kg/ha	21.5	10.5	4.00	4.00
P_2O_5 , kg/ha	20.5	11.5	6.00	2.00
K_2SO_4 , kg/ha	271.6	376.4	310.1	310.1

Soil moisture levels within the 120 cm soil profile were monitored on every other day with a neuron meter (CPN 503DR Hydroprobe) and irrigation was initiated when 35–40% of available water within the root zone was depleted. Plant efficient root depth was taken as 60 cm (Allen *et al*., 1998). Irrigation intervals varied during the growing season because of irrigation scheduling based on soil moisture depletion. In total, irrigation was carried out 12 times in 2015 and 10 times in 2016.

Different irrigation levels were experimented in this study. Experimental irrigation levels were created through supplying different portions of depleted moisture within the efficient root zone of the plants. The treatments were arranged as supplying 100% (I_{100}), 80% (I_{80}), 60% (I_{60}), 40% (I_{40}), 20% (I_{20}) and 0% (I_0) of depleted moisture.

Sowing was performed on 5 May, 2015 and 29 April, 2016 and harvesting was done on 24 August, 2015 and 10 August, 2016. Irrigation programs were initiated on 19 June, 2015 in the first year and on 13 June, 2016 in the second year. Chemicals were not applied since there were not any pests or diseases.

2.1. Chemical analyses performed on pumpkin seeds

2.1.1. Seed vitamin E contents

The vitamin E concentration of pumpkin seeds was determined with the aid of a liquid chromatography system (AOAC, 2000). The sample preparation process was composed of two basic stages. The first stage was the saponification stage of a certain quantity of sample with potassium hydroxide in a water-ethanol ambient. In the second stage, the non-saponified fraction was extracted with hexane. The resultant extract was evaporated in a nitrogen ambient and analyzed in the liquid chromatography system. The seeds were ground and the oil was extracted. The resultant extract was then subjected to vitamin E analysis. About 0.1 g sample was weighed on a precise balance (±0.0001 g) and placed into screwed-cap test tubes. The oil samples were supplemented with 0.2 g ascorbic acid as antioxidant. The samples were then supplemented with 5.5 mL saponification solution (45% water, 55% ethanol, 11% KOH (w/v)). The residual air over the samples was removed with nitrogen gas. The test tubes were screwed-tight, and placed in a heated ultrasonic water bath for 30 minutes at 80 °C for saponification. The samples were cooled down instantly and passed to the liquid-liquid extraction phase. The samples were initially supplemented with 1.5 mL distilled water and then with 3 mL n-hexane and vortexed for 3 minutes to obtain a homogeneous mixture. The Tubes were centrifuged at 2000 rpm for 10 minutes to accelerate phase separation. The upper hexane phase of the centrifuged samples was taken into 30 mL vials. The same extraction process was repeated and the resultant hexane gas was added to the previous portion. The hexane phase in 30 mL vials was then evaporated through nitrogen flow. The residue left over the bottom of the vial was dissolved in 0.5 ml methanol and sample preparation was finalized. HPLC analyses were performed in an Agilent 1100 series LC system. Separation was performed in an analytic column filled with 250 × 4.6 mm size 5 µm diameter particles. Chromatograms were recorded with the aid of measurements made at 325 nm wave length with an Agilent 1100 Diode Series Detector. Vitamin E concentrations were determined from the peaks of the chromatograms.

2.1.2. Seed oil analysis

The oil analysis of pumpkin seeds was performed with the aid of the Soxhlet method (AACC, 2000). Seeds were ground and dried before the analyses and oil was extracted from the seeds with petroleum ether. The solvent evaporated and the remaining portion was dried to constant weight at 100 °C.

2.1.3. Seed protein analysis

The automatic nitrogen determination device (Leco, ABD) operating in accordance with Dumas principles was used for protein analyses. For this purpose, about 0.2 mg sample were placed in the pre-heated Leco FP 528 nitrogen analyzer at 850 °C and nitrogen contents were directly read from the device.

2.1.4. Fatty acid composition of seeds

Ground and dried seed samples were subjected to fatty acid composition analyses in a Gas Chromatographer (Tulukcu *et al*., 2012). Initially, previously extracted and frozen samples (−24 °C) were thawed and 100 mg sample were placed in 15 ml centrifuge tubes. For the methylation process, the samples were supplemented initially with 100 µL 2 N KOH and then with 3 mL hexane. The samples were vortexed for 15 seconds to obtain a homogeneous mixture and centrifuged at 5000 rpm for 5 minutes. Following the centrifuge, 1.5 mL upper phase were transferred into vials and made ready for fatty acid composition analyses in the gas chromatographer (Agilent 6890, Ariz., ABD) equipped with an Atom Ionizing Detector equipped with an ID HP-88 column (100 m long and 0.25 mm in diameter). The injection temperature was 250 °C; oven temperature was 103 °C. Temperature was increased 1 minute later from 103 °C to 170 °C with a 6.5 °C increase per minute, and then increased from 170 °C to 215 °C with a 2.75 °C increase per minute. Finally, the temperature was set at 230 °C for 5 minutes. Helium was used as the carrier gas at a flow rate of 2 mL/min. The split ratio was 1/50.

The total analysis duration was about 40 minutes and at the end of this period, the device software was used to determine the detention times of the fatty acids. Comparisons were made with standard retention times to identify each fatty acid. Results were expressed as % of total fatty acid composition.

2.2. Statistical analysis

Data were subjected to variance analysis using SAS (SAS Inst., 1999) statistical software. The LSD multiple range test was employed to compare the treatment means as a complement of the ANOVA procedure.

3. RESULTS

Irrigation water quantities applied to pumpkin seed plants in I_0, I_{20}, I_{40}, I_{60}, I_{80} and I_{100} treatments were measured as 24, 70.8, 117.9, 164.6, 211.4 and 258.4 mm, respectively, in 2015 and as 13, 104.4, 195.8, 287.3, 378.8 and 470.2 mm, respectively, in 2016.

The vitamin E contents, fatty acids, oil and protein contents of different irrigation treatments are provided in Table 4.

While different irrigation treatments did not have significant effects on the vitamin E contents of the pumpkin seeds in 2015, the effects of irrigation treatments on vitamin E contents were found to be significant in 2016 (Table 5). Vitamin E contents varied from 41.6–55.3 mg/100g in 2015 and from 45.0–55.3 mg/100g in 2016. The lowest vitamin E content was obtained from I_{40} treatments in 2015 and 2016 and the greatest vitamin E content was obtained from I_{20} treatment in 2015 and from I_{60} treatment in 2016.

Linoleic, linolenic, oleic, stearic, palmitic and meristic acids were investigated in pumpkin seeds and linolenic and meristic acids were not encountered in the seeds. Although linolenic and meristic acids are among the main components of pumpkin seed, these acids are included in trace levels and some researchers could not determine linolenic or meristic acid (Potočnik *et al*., 2018; Meru *et al*., 2018; Lazos, 1986; Rezig *et al*., 2012). Irrigation treatments did not result in significant changes in the fatty acid compositions of the pumpkin seeds in either year (Table 5). Linoleic acid contents varied from 33.0–34.35 in 2015 and from 32.4–35.0% in 2016. Oleic acid was the dominant fatty acid in both years. Oleic acid contents varied from 45.3–48.9%

Table 4. Effects of different irrigation treatments on fatty acids, oil, protein and vitamin E contents of pumpkin seeds

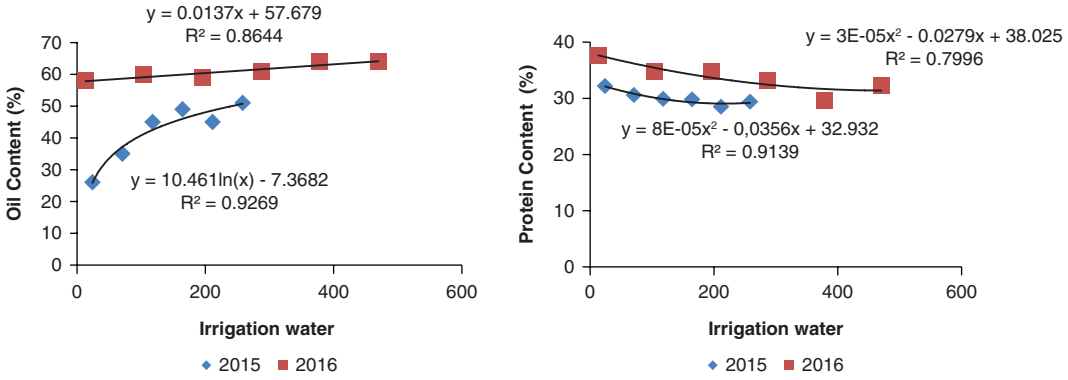
Year	Treatments	Vitamin E (mg/100 g)	Linoleic acid (%)	Oleic acid (%)	Palmitic acid (%)	Stearic acid (%)	Oil content (%)	Protein content (%)
2015	I_0	51.9±3.83	33.8±2.15	45.3±2.58	10.9±0.25	7.6±1.19	26±1.98c	32.2±0.19
	I_{20}	55.3±9.01	34.3±0.68	47.3±0.70	10.7±0.51	7.7±0.44	35±5.94b	30.6±2.30
	I_{40}	41.6±1.65	34.1±1.25	45.5±2.69	12.4±1.23	8.5±0.58	45±2.77a	29.9±3.98
	I_{60}	46.0±4.40	33.0±0.75	48.9±0.31	11.3±0.39	6.9±0.14	49±2.01a	29.8±2.43
	I_{80}	46.1±4.17	34.2±0.50	48.4±1.61	11.1±0.14	6.4±1.06	45±5.80a	28.5±2.09
	I_{100}	46.8±5.66	33.9±1.96	48.2±1.84	11.3±0.19	6.7±0.20	51±0.31a	29.4±1.18
2016	I_0	47.4±8.64bc	32.4±3.98	39.6±4.16	11.5±0.94	10.4±0.80	58±3.22b	37.7±1.53a
	I_{20}	50.4±6.87abc	34.4±3.16	43.9±3.07	10.8±0.70	10.3±0.29	60±2.54ab	34.7±1.51ab
	I_{40}	45.0±7.27c	34.0±1.31	43.4±0.62	11.7±1.00	10.4±0.28	59±4.73b	34.8±0.59ab
	I_{60}	53.9±7.64ab	33.4±4.03	44.5±3.81	12.4±1.04	9.2±0.99	61±4.60ab	33.2±3.19bc
	I_{80}	48.2±11.49bc	35.0±2.95	43.1±3.19	12.6±1.22	9.1±0.80	64±3.94a	29.61.61±c
	I_{100}	55.3±2.26a	34.5±0.80	43.8±0.77	11.8±0.73	9.6±0.86	64±2.89a	32.3±4.41bc

In the table each value represents the mean ± standard deviation of the tree replicates of the analyses

Table 5. Level of significance (P) from the variance analyses of chemical components of pumpkin seeds

Source of Variations	D.F.	Vitamin E (mg/100 g)	Linoleic acid (%)	Oleic acid (%)	Palmitic acid (%)	Stearic acid (%)	Oil (%)	Protein (%)
2015								
Irrigation Level	5	0.122ns	0.828ns	0.187ns	0.096ns	0.062ns	0.0001**	0.242ns
2016								
Irrigation Level	5	0.024*	0.932ns	0.952ns	0.190ns	0.070ns	0.020*	0.0185*

ns: Non significant *: Significant at 5% level and **: significant at 1% level.



In the figureeach value represents the mean ± standard deviation of the tree replicates of the analyses

FIGURE 1. Relationships between irrigation water quantities and oil and protein contents of pumpkin seeds

in 2015 and from 39.6–44.5% in 2016. Palmitic acid contents varied from 10.7–12.4% in 2015 and from 10.8–12.6% in 216. The stearic acid contents of the pumpkin seeds varied from 6.4–8.5% in 2015 and from 9.1–10.4% in 2016. Fatty acids exhibited similar variations in both years.

Irrigation treatments had significant effects on the oil contents of the pumpkin seeds in both years (Table 5). Oil contents varied from 26–51% in 2015 and from 58–64% in 2016. In 2015, the lowest oil content was obtained from the I_0 treatment and the greatest oil content was obtained from the I_{100} treatment. There were three statistical groups in 2015 and the I_{100}, I_{80}, I_{60} and I_{40} treatments were placed in the same statistical group. In 2016, the lowest oil content was obtained from I_0 and the greatest oil content was obtained from the I_{100} treatment. There were two statistical groups in 2016 and the I_{20}, I_{60}, I_{80} and I_{100} treatments were placed in the same statistical group. The oil contents in the seeds increased with increasing irrigation water quantities.

While irrigation treatments did not have significant effects on the protein contents of the pumpkin seeds in 2015, the effects of irrigation treatment on protein contents were found to be significant in 2016 (Table 5). In both years, the greatest protein content was obtained from the I_0 treatment and the lowest oil content was obtained from the I_{80} treatment. Protein contents decreased with increasing irrigation treatments.

The relationships between the applied irrigation water quantities and the oil and protein contents in the pumpkin seeds are presented in Figure 1. Irrigation water quantities significantly correlated with oil and protein contents. There was a significant logarithmic relationship between irrigation water quantities and oil contents in 2015 ($R^2 = 0.93$) and a linear relationship was observed in 2016 ($R^2 = 0.86$). On the other hand, there were polynomial relationships between irrigation water quantities and protein contents ($R^2 = 0.91$, $R^2 = 0.80$, respectively) in both years.

4. DISCUSSION

Pumpkin seed production has increasing popularity compared to cereals because of profitability, easy storage and marketing. Rich oil, fatty acids, protein and vitamin E contents increase the significance of pumpkin seed cultivation even more.

Pumpkin seeds are quite rich in vitamin E. Rezig *et al*., (2012) reported the vitamin E contents of pumpkin seeds (*Cucurbita maxima*) as 41.9 mg/100 g. Stevenson *et al*., (2007) worked with pumpkin seeds of 12 different cultivars and reported vitamin E contents between 45.4 and 70.9 mg/100 g. In another study carried out in Iran, the average vitamin E content in pumpkin seeds (*Cucurbita pepo* Subsp. pepo Var) was reported as 88.8 mg/100g (Ardabili *et al*., 2007). The present findings comply with those earlier ones. Vitamin E is a natural antioxidant. The vitamin E contents in pumpkin seed oil largely depend on the fatty acid contents and compositions of the seeds. Vitamin E contents are also greatly influenced by post-harvest conditions (Nakic *et al*., 2006). Environmental conditions affect the fatty acids, amino acids, minerals and vitamins of different pumpkin genotypes (Erdinç *et al*., 2018). Because the vitamin E content of pumpkin seed depends on water stress and other factors, we may not find regular effects of water stress on vitamin E.

Pumpkin seeds were mostly composed of linoleic, oleic, palmitic and stearic acids. Pumpkin seeds generally had high unsaturated fatty acid (Linoleic+Oleic) contents. Fatty acid contents varied from 79.1–82.65 in 2015 and from 72–78.3% in 2016. Stevenson *et al*., (2007) reported the unsaturated fatty acid contents of pumpkin seeds as between 73.1–80.5%. Meru *et al*., (2018) reported the unsaturated fatty acid contents as between 78.6–86.1%. Oleic acid was the dominant fatty acid in the present study and it was followed by linoleic acid. The present findings were in good agreement with the findings of Nakic *et al*., (2006). The lowest acids were identified as stearic (6.4–10.4%) and palmitic acids (10.7–12.6%). The present findings

comply with the results of earlier studies (Lazos 1986; Sekerci *et al*., 2017; Nawirska-Olszanska *et al*., 2013; Seymen *et al*., 2016).

Pumpkin seed oils are mostly unrefined. Therefore, they are not used as cooking oil, but mostly used as salad dressing because of the color, strong aroma and fatty acid composition. Linoleic acid usually reduces the heat balance of the oil, thus it not suitable to be used as cooking oil (Nederal *et al*., 2012; Potacnik *et al*., 2018).

The Present oil contents (26–64%) comply with the findings of earlier studies carried out with cucurbita species. Meru *et al*., (2018) reported the oil contents of pumpkin seeds as between 29.33 and 48.41%. Lazos (1986) reported the average oil content of pumpkin seeds as 45.4% and Ardabili *et al*., (2011) as 41.6%. In another study, the oil contents of pumpkin seeds were reported as between 10.9 and 30.9% (Stevenson *et al*., 2007). Differences in oil contents of the seeds largely depend on cultivars, agricultural practices and water management regimes.

The present protein contents also comply with the findings of previous studies. Glew *et al*., (2006) reported the average protein content of pumpkin seeds as 51.4%, Lazos (1986) as 32% and Ardabili *et al*., (2011) as 25.4%. Al-Khalifa (1996) reported the protein contents as 26.5% for Cucurbita pepo and 24% for *Cucurbita moschata*. The Present protein contents varied from 28.5–37.7%. These values are lower than the values reported by Glew *et al.,* (2006), but well comply with the findings of other studies. Differences in protein contents are mostly attributed to differences in cultivars, growing conditions and climate parameters.

5. CONCLUSIONS

The pumpkin seeds of this experiment have high vitamin E content, which makes them desirable for human health. Vitamin E content was not changed with irrigation water amounts. Although water stress decreased the oil's components, fatty acid content was not significantly changed. Oil content was improved by increasing irrigation amounts and the highest oil content was acquired by applying full irrigation. Oleic, linoleic, palmitic and stearic acids were the main components of the pumpkin seeds. Irrigation also improved pumpkin seed protein content. We concluded that pumpkin seed properties are beneficial for human health and their production could be hastened by proper irrigation management strategies.

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