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DETERMINATION OF RAW AND PROCESSED BLACK OLIVE CHARACTERISTICS OF SIX CULTIVARS

Y. Ozdemir[1], A. Ozturk[1], N. A. Tangu[2], M. E. Akcay[2], U. Ozyurt[3], S. Ercisli[4*]

[1]Ataturk Central Horticultural Research Institute, Department of Food Technologies, Yalova, Turkey
[2]Ataturk Central Horticultural Research Institute, Department of Fruit Breeding, Yalova, Turkey
[3]Middle East Technical University, Department of Food Engineering, Ankara, Turkey
[4*]Ataturk University, Agricultural Faculty, Department of Horticulture, Erzurum, Turkey
Corresponding Author's email: sercisli@gmail.com

ABSTRACT

Olives can be used both for table consumption or oil production. For table consumption, curing olives is an ancient process that turns the naturally bitter raw fruit into a deliciously salty, tart snack. This study is aimed to investigate table olive characteristics of raw and processed olives from six cultivar candidates. Cultivar candidates were developed by cross breeding between Gemlik and Edinciksu cultivars. Olive from Gemlik cultivar, genitor and widespread produced table olive cultivar in Turkey, was used for comparison. Fruit and seed size with weight, water content, titratable acidity, pH, hardness, color along with oleuropein absorbance (K_{345}) value, salt, total phenolic content, antioxidant activity and sensory analysis were studied on both raw and processed olives. Results showed that the average fruit weight, titratable acidity, oleuropein absorbance value, total phenolic content and antioxidant activity of raw and processed olives of genotypes were found between 4.3-6.5 and 3.9-6.0 g, 0.27-0.38% and 0.33-0.59%, 1.08-2.52 and 0.16-0.26, 175.22-463.15 and 91.00-113.23 mg gallic acid equivalent/100 g, 2618.73-4296.23 and 130.29-147.26 µM trolox/kg, respectively. Statistically significant difference was observed between raw and processed olive for salt, total phenolic content, antioxidant activity, oleuropein absorbance and hardness. It might be caused by genetic factors, which affect skin and tissue permeability of olives during processing. Result of this study showed that there were differences on physical and chemical characteristics og genotypes and GE379 had highest total phenol content, GE397 had highest fruit weight and second highest total sensory scores and GE366 had highest sensory appearance, taste general appreciation and total scores among processed olives. GE366 and GE397 had more remarkable characteristics for production with spontaneous fermentation than other samples.

Keywords: Cross breeding, genotype, oleuropein, total phenol, antioxidant activity, sensory test.

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INTRODUCTION

Fruits are genetically very diverse groups and grown in temperate, subtropical and tropical regions and have been recognized for their human health benefits. Most of the fruits have high content of non-nutritive, nutritive, and bioactive compounds such as flavonoids, phenolics, anthocyanins, phenolic acids, and as well as nutritive compounds such as sugars, essential oils, carotenoids, vitamins, and minerals (Sahin *et al*., 2002; Ercisli *et al*., 2008; Halasz *et al*., 2010; Ercisli *et al*., 2011; Butuic-Keul *et al*., 2019; Guney *et al*., 2019)

Olive production in the world is concentrated in the Mediterranean countries including Spain, Italy, Greece, Turkey, Portugal, Tunisia and Morocco.

New olive cultivars with better table olive characteristics than standard olive cultivars have potential to increase profit in the table olive industry and to satisfy consumer expectations. One of the most important components, responsible for the nutritional benefits of olive fruit, are phenolic content, which are main secondary metabolites in olive fruits (Amiot *et al*., 1986) and can be used for selection of cultivar candidates (Bellini *et al*., 2008; León *et al*., 2008).

Crossbreeding studies have been continuing in olives and promising cultivars have been released in the last years (Arcas *et al.,* 2013). In Mediterranean countries including Turkey (Arsel and Cirik, 1994; Ozdemir *et al*., 2011), Tunisia (Trigui, 1996), Greece (Pritsa *et al*., 2003) and Italy (Bellini *et al*., 2002) crossbreeding and clonal selection studies have been conducted. These studies aimed to select genotypes characterized by early bearing, resistance to pests and to abiotic stresses, limiting alternate bearing, suitability to intensive culture and to mechanical harvesting, as well as high-quality productions, in terms of both organoleptic characteristics of fruits and oils, and high contents in substances useful for human health (Bellini *et al*., 2008; Ozdemir *et al*., 2011).

The present study was aimed to determine characteristics of raw and processed (spontaneous fermentation was used for processing) black olives of six cultivar candidates and comparing them with those of olives from Gemlik cultivar, one of the widespread standard table olive cultivars in Turkey. Determined olive characteristics were used for final selection steps of these olive cultivars candidates.

MATERIALS AND METHODS

In this study, olives from six cultivar candidates as well as Gemlik cultivar were evaluated. These cultivar candidates were developed by cross-breeding of Gemlik and Edincik su cultivars. These trees were planted at 1.5 m x 3 m within and between rows on field in olive genotype observation orchard of Ataturk Central Horticultural Research Institute (Yalova/Turkey) in 2001. Maturity index of olives were followed according to Berenguer *et al*. (2006) and olives were randomly handpicked in 2013-2014 and 2014-2015 years. Code of olives and their maturity index were given in Table 1.

Method of table olive production: Olives were processed to table olive according to method of spontaneous fermentation in brine (Anonymous, 2014; Ozdemir *et al*., 2011). Olives were kept in 10% brine, also plates and stones were put as force source on olive fruits (3.5 kg/m^2) to accelerate olive debittering and increase dry matter of olives. Salt concentration were controlled in brine at 3 days intervals and adjusted to 7%. Olives were fermented in brine at ~16 °C until pH fall to 5.0 approximately in 6 months.

Physical and Chemical Analysis: An official method according to TS 774 (1992) has been used to determine fruit weight and flesh to seed ratio. The average weight of fruit and flesh to seed ratio was determined using 100 fruits. Fruit and seed size were measured using digital compass (Mitutoyo, Japan). Color values of olive skin were measured with a color meter (Konica Minolta, Japan). Maturity index was determined after Hassan *et al*. (2011). Salt contents were determined after the method described by Garcia *et al*. (1991). Texture hardness of olive was measured with fruit hardness tester (W.O.W FRH-5, Japan). A conventional oven (75±2 °C) was used to determine water content of fruits (Esti *et al*., 1998). Folin-Ciocalteu method was used for total phenol determination (Thaipong *et al*., 2006) and antioxidant activity was determined by DPPH method (Usenik *et al*., 2008). Absorbance value of oleuropein was determined by spectrophotometric method at 345 nm according to Mastorakis *et al*. (2004). pH, titratable acidityand sodium chloride content were determined according to official method (TS 774, 1992).

Sensory Analysis: Sensory profile of processed olives were determined by the panel of ten trained judges with 9 point scale (Aponte *et al.* (2010). Three sessions as 1 hour (4-5 samples/session) were conducted to complete the analysis. Olive sample preparation, serving and tasting were arranged following Galán-Soldevilla and Ruiz Pérez-Cacho (2010). The appearance and color attributes were assessed by the whole panel on the complete sample before carrying out the tasting. Firstly odor, after that ease of separation from seed, taste, bitterness and finally, general appreciation attributes were evaluated (Galán-Soldevilla *et al*., 2013).

Statistical analysis: The experiment was laid out under completely randomized design with four replications. Data related to physical and chemical characteristics of raw and processed olives were subjected to analysis of variance (ANOVA) and means were separated by LSD test ($p \leq 0.05$). SPSS 16.0 for Windows (SPSS Inc., Chicago, USA) was used for raw data analysis. There were no differences between 2013-2014 and 2014-2015 years thus the data of both years were pooled.

RESULTS AND DISCUSSION

Consumers prefer high fruit weight and flesh to seed ratio; moreover, price of olive increases in parallel to these values (Son, 2004). Therefore, these values are required to be as high as possible for a new developed cultivar. Fruit and seed size with fruit weight and flesh to seed ratio of raw olive are shown in Table 2. No significant differences were observed among the physical characters of olives except fruit weight. In this study, fruit weight and flesh to seed ratio was observed higher than reported by Rallo *et al*. (2008) and Arji and Bahmanipour (2014) but similar Medina *et al*. (2010) for evaluated olive cultivars and cultivar candidates. The differences could be due to different materials used (cultivars, genotypes, accessions), growing conditions, soil properties etc.

Color values, hardness, pH and titratable acidity of raw olive samples are presented in Table 3. The hardness of a material is measured as its surface resistance to penetration against an indenter. It is also an important table olive quality criterion (Castro-Garcia *et al*., 2009). In present study, hardness, pH and titratable acidity of olive samples were observed between 470-510 g, 5.32-5.63 and 0.22-0.38% oleic acid, respectively. Olives has four surface colors; green, turning color, natural black and ripe olives (BOE, 2001). In this research table olives were categorized in natural black olive.

Water and total phenol content, oleuropein absorbance value and antioxidant activity of raw olives are given in Table 4. Oleuropein absorbance value is an indicator of olive bitterness. Low oleuropein absorbance value of olives is required for olives since, it provide easiness and short time for debittering of olives (Boskou *et al*., 2015, Ozdemir *et al*., 2014). On the contrary, antioxidant activity and total phenol content of olives are required at high values because of their role on prevention against certain diseases especially cancer (Dimitrios, 2006, Kris-Etherton *et al.,* 2002). Antioxidant activity was in accordance with the results of Uylaser *et al*. (2000). However, oleuropein absorbance values and total phenolic content were higher than the results from the previous studies (Kumral *et al.,* 2009; Sánchez Gómez *et al.,* 2006). This disparity may be due to use of different material.

The criteria for choosing an appropriate new cultivar for table olives can be listed as proper shape with good size, high flesh/stone ratio, ease in releasing the seed and texture (Sánchez Gómez *et al.,* 2006). Fruit and seed size, fruit weight and flesh to seed ratio of processed olives were given in Table 5. Flesh to seed ratio requirement is at least 5 for new table olive cultivar candidates according to breeding researchers (Varol *et al*., 2009). In this research flesh to seed ratio of all table olives had higher value than 5.

Glossy black color and high flesh hardness of table olives were a few of the required criteria (Boskou *et al*., 2015, Varol *et al*., 2009). Color values, hardness, pH and titratable acidity of processed olives were given in Table 6. When bitterness removed (fruits fermented), they are oxidized by exposure to air and this process improves the skin color (Fernandez Diez, 1991). In this research, olives did not expose to air particularly in a processing step, instead air only contacted to olive during processing steps. Black color values of spontaneous fermented olive without oxidation step were less than oxidized table olive (Boskou *et al.,* 2015).

Water, salt and total phenol content, oleuropein absorbance value and antioxidant activity of processed olives were shown in Table 7. Oleuropein absorbance of the olives decreased during fermentation, which is related with diffusion to brine (Ozdemir *et al*., 2014). Oleuropein absorbance value and total phenolic content of the samples were in accordance with the previous studies (Ben Othman *et al*., 2008; Morello *et al.,* 2004*;* Uylaser *et al*., 2000). Titratable acidity content of the processed table olives were lower than Poiana and Romeo (2006); moreover, water content were higher than Kumral *et al.* (2009).

Debittering steps were used in table olive processes to loss bitter taste and astringency due to phenolic compounds (Ben Othman *et al*., 2008). Oleuropein was important phenolic component and it is responsible from this bitter taste (Panagou, 2006). Oleuropein absorbance value gives information about the content of oleuropein in olive samples (Kumral *et al*., 2009). In this research after processing, 85.18-90.69% loss was detected in oleuropein absorbance value.

With processing, olive fruit pulp loss of phenolic compounds; nevertheless, table olives still remain as an important source of phenolic compounds (Boskou *et al*., 2006). In this research after processing olives, total phenol content and antioxidant loss were determined between 44.11-78.26% and 94.63-96.97%, respectively. GE379 and Gemlik showed higher total phenol content after processing. Despite using the same production method, statistically significant difference was determined in salt content of processed olive samples, which changed between 3.83-4.43%. This difference was thought to be associated with the olive skin permeability and tissue hardness properties of olives, which depend on genetic factors of olive cultivar candidates.

Fermentation improved flavor and texture characteristics (Sánchez Gómez *et al*., 2006) and color as a consequence of the different anthocyanin polymerization (Romero *et al*., 2004). Result of sensory evaluation of processed olives was shown in Table 8. In order to produce spontaneous fermented black table olives, fruits should be ripe (not over ripe). It means at the end of the season olive fruit retain an excellent color after processing but their texture is not hard enough to attract consumer (Sánchez Gómez *et al*., 2006). Further more, in this research olives were harvested between 5,1-5,6 maturation index and processed with spontaneous fermentation. After that color, appearance, taste and general appreciation scores were determined higher than 5.0 by sensory evaluation test.

As a result of the sensory evaluation of olive cultivars, appearances as well as taste and general appreciation were affected sensory criteria. In other words, GE366 had statistically highest scores for his sensory attributes and total score. It was followed by GE379 and GE397. GE409, GE076 and GE320 had lower total score than Gemlik cultivar, which was used for comparison. Previous studies indicated compositional and sensory differences of horticultural crops (Zia-Ul-Haq *et al*., 2014; Gecer *et al*., 2020).

Table 1. Olives and their maturity index (average of 2013-2014 and 2014-2015 years)

Cultivar codes	Maturity Index
GE-076	5.6
GE-320	5.1
GE-366	5.7
GE-397	5.2
GE-409	5.3
GE-493	5.6
Gemlik	5.4

Table 2. Fruit and seed size, fruit weight and flesh to seed ratio of raw olives (average of 2013-2014 and 2014-2015 years)

Sample	Fruit length (cm)	Fruit width (cm)	Seed length (cm)	Seed width (cm)	Fruit weight (g)	Flesh to seed ratio
GE076	2.2	1.9	1.4^{NS}	0.8	4.3c	5.5
GE320	2.5	2.1	1.6	0.9	5.9b	5.7
GE366	2.2	2.0	1.4	0.9	5.4b	5.6
GE379	2.3	2.0	1.4	0.8	4.8c	5.5
GE397	2.6	2.1	1.8	1.0	6.5a	5.8
GE409	2.5	2.2	1.6	1.0	6.2a	6.0
Gemlik	2.5	1.8	1.4	0.7	4.4c	5.3
$LSD_{0.05}$	NS	NS	NS	NS	NS	NS

Different letters in the same column refers to a statistically significant difference at 5% level of probability. NS: Non significant

Table 3. Color values, hardness, pH and titratable acidity of raw olive (average of 2013-2014 and 2014-2015 years).

Sample	Color values			Hardness (g)	pH	Titratable acidity (% oleic acid)
	L	a	b			
GE076	25.04^{NS}	5.51^{NS}	-0.14^{NS}	500^{NS}	5.51^{NS}	0.30b
GE320	28.78	5.46	0.80	490	5.46	0.22d
GE366	22.19	5.43	0.66	510	5.43	0.37a
GE379	26.48	5.32	0.50	470	5.32	0.34b
GE397	27.05	5.48	0.42	480	5.48	0.27c
GE409	26.87	5.63	0.75	510	5.63	0.32b
Gemlik	28.22	5.52	0.43	490	5.52	0.38a

Different letters in the same column refers to a statistically significant difference at 5% level of probability. NS: Non significant

Table 4. Water, total phenol, oleuropein absorbance value and antioxidant activity of raw olives (average of 2013-2014 and 2014-2015 years)

Sample	Water (%)	Oleuropein absorbance value (K_{345})	Total phenolics (gallic acid mg/ 100g)	Antioxidant activity (µM trolox/ kg)
GE076	71.85^{NS}	2.52a	463.15a	4296.23a
GE320	71.56	1.39c	316.84b	3261.35b
GE366	72.03	1.85bc	221.53c	3680.66b
GE379	70.31	2.04b	202.61cd	3304.52b
GE397	71.14	1.26c	175.22d	2618.73c
GE409	70.10	1.12cd	219.53c	3142.16bc
Gemlik	72.27	1.08d	320.17b	4268.53a

Different letters in the same column refers to a statistically significant difference at 5% level of probability. NS: Non significant

Table 5. Fruit and seed size, fruit weight and flesh to seed ratio of processed olive (average of 2013-2014 and 2014-2015 years).

Sample	Fruit width (cm)	Fruit length (cm)	Seed width (cm)	Seed length (cm)	Fruit weight (g)	Flesh to seed ratio
GE076	1.4c	1.8d	0.8^{NS}	1.4^{NS}	3.9e	5.3^{NS}
GE320	1.5c	2.4ab	0.9	1.6	5.4b	5.7
GE366	1.6bc	2.1c	0.9	1.4	5.0bc	5.9
GE379	1.7ab	2.4ab	0.8	1.4	4.7cd	5.5
GE397	1.9a	2.6a	0.9	1.8	6.0a	5.8
GE409	1.7ab	2.4b	1.0	1.6	5.3b	6.0
Gemlik	1.8ab	2.4b	0.8	1.4	4.5d	5.4

Different letters in the same column refers to a statistically significant difference at 5% level of probability. NS: Non significant

Table 6. Color values, hardness, pH and titratable acidity of processed olive (average of 2013-2014 and 2014-2015 years).

Sample	Color values			Hardness (g)	pH	Titratable acidity (% oleic acid)
	L	A	b			
GE076	30.42^{NS}	8.61b	7.01b	260c	4.60^{NS}	0.49b
GE320	34.68	11.69a	10.51a	290bc	4.65	0.59a
GE366	26.16	5.21d	2.99d	330ab	4.79	0.33c
GE379	25.78	7.83c	2.85d	290bc	4.82	0.47b
GE397	29.18	9.16ab	4.11c	350a	4.83	0.36c
GE409	29.15	11.53a	6.31b	340ab	4.72	0.48b
Gemlik	27.81	7.66c	2.92d	380a	4.55	0.49b

Different letters in the same column refers to a statistically significant difference at 5% level of probability. NS: Non significant

Table 7. Water, salt and total phenol, oleuropein absorbance value and antioxidant activity of processed olives (average of 2013-2014 and 2014-2015 years)

Sample	Water (%)	Oleuropein absorbance value (K_{345})	Salt (%)	Total phenolics (gallic acid mg/ 100g)	Antioxidant activity (µM trolox/ kg)
GE076	64.74e	0.26a	3.83d	100.68b	130.29^{NS}
GE320	69.03a	0.18cd	4.17b	97.61b	140.60
GE366	63.88e	0.19bc	4.07bc	91.05c	141.07
GE379	68.06ab	0.19ab	4.43a	113.23a	138.68
GE397	65.10de	0.18b	3.90cd	91.00c	140.55
GE409	67.26bc	0.16d	4.07bc	96.09bc	147.26
Gemlik	66.36cd	0.16d	4.20b	111.16a	140.56

Different letters in the same column refers to a statistically significant difference at 5% level of probability. NS: Non significant

Table 8. Sensory evaluation scores of processed olives (0-9) (average of 2013-2014 and 2014-2015 years)

Sample	Appearance	Color	Odor	Ease of separation from seed	Taste	Bitterness	General appreciation	Total Score
GE076	5.7c	5.5^{NS}	5.8^{NS}	6.5^{NS}	6.0ab	0.3^{NS}	6.5b	36.2^{NS}
GE320	5.5c	5.0	6.0	7.7	4.8b	0.5	5.3c	33.2
GE366	7.2a	7.5	5.9	7.0	7.2a	0.3	7.2a	43.7
GE379	6.7b	7.7	6.2	6.8	7.3a	0.4	6.5b	40.5
GE397	7.0b	6.8	5.9	7.0	6.5ab	0.3	7.0a	40.8
GE409	6.5b	6.5	6.1	6.0	4.8b	0.3	5.0c	33.2
Gemlik	7.5a	7.3	6.2	6.3	5.5ab	0.4	6.2b	38.3

Different letters in the same column refers to a statistically significant difference at 5% level of probability. NS: Non significant.

Conclusion: According to results significant differences were observed for titratable acidity, oleuropein absorbance value, total phenol content and antioxidant activity of raw olive. Also fruit width, length and weight, color values, hardness, titratable acidity, oleuropein absorbance value, water, salt and total phenol content, appearance, taste and general appreciation sensory scores of processed olives were significantly different. GE076 (463.15 mg gallic acid equivalent/100 g) among raw olives but GE379 (113.23 mg gallic acid equivalent /100 g) among processed olives had high total phenol content. GE366 was found to have the highest total sensory scores and the GE320 lowest. GE397 was found to have highest fruit weight with good flesh to seed ratio and second highest total sensory scores.

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