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Phytochemicals, Antioxidant and Chemical Properties of 32 Pomegranate Accessions Growing in Egypt

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Abstract: Arils and oils from thirty two pomegranate (*Punica granatum* L.) cultivars obtained from various sites from different regions in Egypt were evaluated for their chemical and antioxidant properties. These properties included arils juice analysis: juice content, pH, total soluble solids (TSS), titratable acidity (TA), maturity index (MI), total anthocyanin, antioxidant capacity (vitamin E and C) and ellagic acid and seed oil analysis: seed oil content % and fatty acids composition (linoleic acid , linolenic acid, arachidonic acid , lauric acid, myristic acid, palmtic acid, stearic acid, oleic acid and arachidic acid). Juice content ranged from 75.0 to 43.0, total soluble solids ranged from 20.33 to 12.27 (°Brix) and titratable acidity (TA) ranged between 2.81% and 0.30 % in pomegranate juices, respectively. The pH and vitamin C content also ranged between 4.53-2.91 and 9.48 -2.77 mg/100 ml, respectively. According to the results of HPLC, ellagic acid content of juice and peel ranged between 10.53 - 0.84 mg/100 ml and 10-50.00 mg/100 g, respectively. Total antioxidant activity measured by FRAP assay with a range of 225.17-705.50 (mmol/100 g) and 157.33-419.33 (mmol/100 ml) in peel and juice, respectively.

Key words: Pomegranate · Chemical properties · Ascorbic acid · Antioxidant activity · Juice · Anthocyanins · Peel

INTRODUCTION

Pomegranate (*Punica granatum* L.) is a tropical and subtropical fruit. One of the oldest known edible fruits and is popularly consumed as fresh fruit, beverage, as a food product and coloring agents as well as jellies [1, 2]. It is native to Southwestern Asia and spread from there to North Africa and South Europe and then to America [3]. Pomegranate locally called Rumman and one of the oldest known fruit species. The fruit was seen by ancient Egyptians as a symbol of prosperity and ambition and parts of the tree were used as treatment for tapeworm and other parasitic infections [4]. Pomegranate fruit maturity status is commonly assessed based on external (skin) color, juice color and acidity of juice [5]. Similarly, polyphenols are a main group of pomegranate phytochemicals, which act as phytochemical antioxidants with potential health related benefits. Pomegranate juice contains the highest concentration of total polyphenols in comparison to other fruit juices [6]. The red color of pomegranate juice is primarily associated and important sources of anthocyanin (cyanidin, delphinidin and pelargonidin) pigment and peel is rich in polyphenols including ellagitannins (such as punicalin, pedunculagin, punicalagin and ellagic acid) [7, 8]. These polyphenols exhibit various biological activities, such as eliminating free radicals, inhibiting oxidation and microbial growth [9]. Thus, pomegranate has become more popular because of its health-promoting phytonutritional content and high antioxidative capacity [10-12]. Therefore, production and consumption of pomegranate juice have sharply increased in recent years [13]. Recent studies in pomegranate fruits have shown that cultivar may also substantially influence the antioxidant activity and other physicochemical properties, such as skin and juice percentage, dry matter, pH, total soluble solids (TSS), total sugars, titratable acidity (TA), total phenolics and anthocyanins [14-17]. These parameters may provide important information to the consumer in terms of recognizing a more nutritional fruit [18]. Therefore, this study aims to evaluate of some pomegranate (*Punica granatum* L.) cultivars obtained from various sites from different regions in Egypt for their chemical and antioxidant properties.

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MATERIALS AND METHODS

Plant Material: Ripe fruits from 32 Pomegranate (*Punica granatum* L.) cultivars were collected from four locations (Upper Egypt, North Sinai, Ismailia and Siwa Oasis) and harvested from various sites in Egypt. Arils of fruits were hand-separated (about 100 g lots) and frozen at 20°C. Three replicates were maintained for each analysis, each replicate indicating three pomegranate fruits.

Juice Content, pH and Titratable Acidity (TA): One hundred grams of seeds were collected from each cultivar and expressed. The following data was obtained from the juice: volume of total juice from the 100 g of seeds, the pH was measured with a pH meter. After determining pH, the solution was titrated with 0.1 M NaOH to pH 8.1, monitoring with an electrode and calculated as grams of citric acid per kilogram fresh weight [19].

Total Soluble Solids Content (TSS): Total soluble solids were measured with an Atago digital refractometer PAL-1 (Tokyo, Japan). Results are reported as °Brix [19].

Maturity Index (TSS/A): Maturity index (TSS/A) up to date the following classification has been established for Spanish varieties [20, 21, 22]:

- Sweet varieties: MI = 31-98.
- Sour-sweet varieties: MI = 17-24.
- Sour varieties: MI = 5-7.

Seed Oil Extraction: Seed oil content was determined by extraction method described by AOAC [23] No.963.15 by using the petroleum ether (40-60°C) as a solvent.

Determination of Total Amount of Anthocyanins: Spectrophotometric analyses were performed under the following conditions: 10 ml of juice were filtered and diluted with 90 ml of a EtOH/HCl mixture previously prepared mixing 79.7 ml of anhydrous ethyl alcohol with 20.3 ml of HCl (37%). The absorbance has been measured at 535 nm, by an UV-Vis Hitachi U-2000 spectrophotometer, using 1 cm cells. The calibration curve has been obtained by measuring absorbance of standard solutions of pure cyanidin-3-glucoside chloride (99.5% purity grade tested by HPLC, Extrasynthese, Genay, France) [24].

Determinations of Vitamin C: Vitamin C was determined by HPLC. A volume of 50 ml of each Pomegranate juice was homogenized with 40 ml of an extraction solution (30 g/1 meta-phosphoric acid + 80 g/1 % acetic acid). The resulting mixture was filtered under suction and adjusted up to 100 ml with distilled water. Samples were filtered through a 0.45 µm membrane filter and duplicates of 20µm for each extract were analyzed by HPLC. Results are expressed as milligrams of ascorbic acid per 100 ml juice. Separation of ascorbic acid was performed by HPLC using a Hypersil BDS C8 (5 µm) stainless steel column (250 mm x 4.6 mm) (Thermo Electron, United Kingdom). The flow rate was fixed at 1.5 ml/min. A UV- vis detector was set at 245 nm; chromatographic data and UV-vis spectra were collected, stored and integrated using chromostar light software. The calibration curve was built with one concentration level an ascorbic acid standard solution (100 mg/ml in a solution 30 g/1 meta-phosphoric acid + 80 g/1 % acetic acid) [25].

Determination of Total Antioxidant Activity and Ellagic Acid

HPLC Analysis: The chromatographic analysis was carried out on HPLC system Hewlett Packard, HP 1090 liquid chromatograph). Analysis of ellagic acid was performed according to the method of Gil *et al.* [26]. Before injection, each juice was centrifuged in an eppendorf tube (4 min at 5000 rpm) and the centrifuged supernatant was allowed to pass through a 0.45 lm PTFE filter. Injection volume was 50 ll. An RP C18 Nucleosil 100 (12.5 cm _ 5.0 mm _ 5.0 lm) column was used for the separation of sample components. Mobile phase consisted of solvent A (2.5%, v/v, solution of acetic acid in water) and solvent B (2.5%, v/v, solution of acetic acid in methanol) at different ratios, the gradient profile was 100% A at 0-5 min, 90% A at 15 min, 50% A at 45 min and 100% A at 55 min. Flow rate was 1.0 ml/min. Chromatograms were recorded at 510 nm. Each compound was quantified by comparing its peak area against the standard curve obtained specifically for the reference solutions containing that compound. To obtain the standard curves, five different concentrations of ellagic acid (0.0012-0.01 mg/100 ll). Antioxidant capacity (vitamin E and vitamin C) of PJs were determined by method as recommended by Lee *et al.* [27] as the most trustable methods specifically for PJs. The method based on the evaluation of, the free radical scavenging effect of plant extracts was assessed by the decolouration solution of DPPH radical of all cultivars juice according to Lee *et al.* [27] using standard concentration as vitamin E (0.1g/100 ml methanol) and as vitamin C (0.1g/100 ml methanol).

Determination of Total Fatty Acid: Separation of fatty acids was accomplished according to A.O.A.C.[28] using Gas Liquid Chromatography, Trace GC Ultra, Thermo Scientific ,fatty acids of standards and samples were converted to methyl esters using ethereal solution of diazomethane.

Statistical Analysis: Data were analyzed statistically by one way ANOVA, using analysis of variance and differences among the means were determined for significance at P>0.05 using Least Significant Difference test and the system programmed SAS.

RESULTS AND DISCUSSION

Chemical analysis for 32 Pomegranate (*Punica granatum*) cultivars were collected from four locations (Upper Egypt, North Sinai, Ismailia and Siwa Oasis) and divided to two groups:

- Arils juice analysis: juice content, pH, total soluble solids (TSS), acidity, maturity index (MI), total anthocyanin, antioxidant capacity (vitamin E and vitamin C) and ellagic acid.
- Seed oil analysis: Seed oil content % and fatty acids composition saturated and unsaturated fatty acids (linoleic acid, linolenic acid, arachidonic acid , lauric acid, myristic acid, palmtic acid, stearic acid, oleic acid and arachidic acid)

Juice Content, pH and Titratable Acidity (TA): High juice content (ml/100 g seeds) is a desirable attribute in pomegranate production [29,30]. There were varied significant differences in the juice content (ml per 100 g seeds) of the pomegranate cultivars, Nab El Gaml from Upper Egypt and Succari Red from North Sinai cultivars had the highest amount of juice (75.00-67.33) (ml per 100 g seeds) than the other cultivars (46.67 ml/100g) (Table1).

Table 1: Seed characterization of 32 Pomegranate (*Punica granatum* L.) germplasm grown in different locations of Egypt.

Characters									
*Germplasm		Juice content (ml/100g)		pH		TSS		Acidity	
				Assiut					
1	Assiuty Maragab	63.44	B-E	2.917	O	12.40	LM	2.753	A
2	Assiuty Abo shokaa	65.44	BCD	2.980	O	13.67	J-M	2.817	A
3	Nab-El-Gamal	75.00	A	3.893	F-J	14.07	H-M	0.667	H
4	Abo Lammam	67.33	B	3.653	I-L	14.67	D-L	0.933	EF
5	Balady	61.00	B-G	3.613	JKL	14.47	F-M	1.067	DE
6	Manfalouty	64.67	B-E	3.790	H-K	16.60	B-G	0.900	EF
7	Iranian	56.67	F-I	3.473	LM	13.20	KLM	1.167	D
8	Assiuty	50.67	IJK	3.107	NO	13.62	J-M	2.250	B
9	Manfalouty	66.33	B	3.777	H-K	13.60	J-M	0.733	GH
10	Nab-El-Gamal	59.33	C-H	4.320	A-E	13.40	J-M	0.733	GH
11	Assiuty	65.78	BC	3.250	MN	14.53	E-M	1.663	C
12	Manfalouty	63.00	B-F	3.723	I-L	14.40	F-M	0.600	HI
13	Hejazy	59.33	C-H	3.883	F-J	15.73	C-J	0.900	EF
14	Wardy	58.33	E-H	4.070	D-G	14.40	F-M	0.466	IJ
				Siwa Oasis					
15	Sour	53.33	HIJ	3.727	I-L	17.50	BC	0.833	FG
16	Sweet	60.67	B-G	4.237	B-E	14.27	G-M	0.366	J
17	Sweet	58.67	D-H	4.330	A-D	15.53	C-K	0.433	J
18	Sour	43.00	L	3.530	KL	20.33	A	1.067	DE
19	Sour	62.33	B-G	3.593	KL	14.20	H-M	0.400	J
20	Sour	64.00	B-E	4050	E-H	16.13	C-I	0.633	H
21	Sour	63.33	B-F	4.210	B-E	13.87	I-M	0.966	EF
22	Sweet	46.67	KL	4.350	ABC	18.67	AB	0.400	J
23	Sour	63.67	B-E	3.760	I-K	12.27	M	0.668	H
24	Sweet	56.00	GHI	4.357	ABC	14.40	F-M	0.366	J
25	Sour	49.33	JK	3.567	KL	17.00	BCD	1.033	DE
				Ismailia					
26	Manfalouty	63.33	B-F	3.730	I-L	15.67	C-J	1.00	E
				North Sinai					
27	Succari red (smooth)	66.67	B	4.453	AB	17.00	BCD	0.333	J
28	Succari red	67.33	B	4.530	A	17.40	BC	0.333	J
29	Succari white	64.00	B-E	3.907	F-I	16.67	B-F	0.400	J
30	Sour	65.00	B-E	3.777	H-K	16.87	B-E	1.167	D
31	Succari red	62.67	B-F	3.813	G-K	16.27	C-H	0.333	J
32	Succari red	67.33	B	4.110	C-F	15.67	C-J	0.300	J

* (1-14) pomegranate germplasm from Assiut (El-Badary, Sahel Selim and El Fatth), (15-25) from Siwa oasis, (26) from Ismailia and (27-32) from North Sinai.

Table 2: Seed characterization of 32 Pomegranate (*Punica granatum* L.) germplasm grown in different locations of Egypt.

Characters *Germplasm		MI=TSS/Acidity		Seed oil (%)		Anthocyanin (mg/ml)		Vitamin C mg/100ml	
				Assiut					
1	Assiuty Maragab	4.51	M	20.29	D-H	0.2617	GHI	4.557	E-I
2	Assiuty Abo shokaa	4.86	LM	26.59	A-D	1.3720	A	7.320	BC
3	Nab-El-Gamal	21.20	FGH	18.01	E-H	0.1249	J	3.160	I
4	Abo Lammam	15.70	G-J	13.27	H	1.0820	B	7.900	AB
5	Balady	13.69	H-L	18.53	E-H	0.0818	J	3.710	GHI
6	Manfalouty	18.56	F-I	20.62	D-G	0.1041	J	2.770	I
7	Iranian	11.30	I-M	19.47	E-H	0.0582	J	4.350	E-I
8	Assiuty	6.05	KLM	29.45	ABC	0.6553	EF	5.217	E-H
9	Manfalouty	18.59	F-I	16.63	FGH	0.6753	EF	5.290	E-H
10	Nab-El-Gamal	18.37	F-I	20.46	D-H	0.2851	GH	3.000	I
11	Assiuty	8.79	J-M	28.05	ABC	0.5622	F	5.367	D-G
12	Manfalouty	24.47	EFG	14.15	GH	0.7712	DE	7.900	AB
13	Hejazy	17.48	F-J	6.47	I	0.5715	F	5.210	E-H
14	Wardy	31.23	DE	14.05	GH	1.3820	A	7.190	BCD
				Siwa Oasis					
15	Sour	21.09	FGH	17.49	FGH	0.0452	J	5.210	E-H
16	Sweet	40.17	C	5.00	I	0.1361	IJ	3.843	F-I
17	Sweet	36.23	CD	17.95	E-H	0.1183	J	3.790	GHI
18	Sour	19.11	F-I	18.41	E-H	0.3757	G	4.420	E-I
19	Sour	35.50	CD	20.36	D-H	0.3496	G	3.160	I
20	Sour	25.60	EF	5.75	I	0.1124	J	3.950	F-I
21	4	14.41	H-K	19.24	E-H	0.2580	GHI	4.350	E-I
22	Sweet	48.81	AB	18.33	E-H	0.1614	HIJ	3.320	HI
23	Sour	18.69	F-I	14.85	GH	0.8980	C	3.160	I
24	Sweet	40.17	C	4.75	I	0.0926	J	4.350	E-I
25	Sour	16.69	F-J	30.71	AB	0.3632	G	6.000	CDE
				Ismailia					
26	Manfalouty	16.10	F-J	23.23	C-F	0.8612	CD	5.530	C-G
				North Sinai					
27	Succari red (smooth)	51.94	A	25.06	A-E	0.1493	IJ	3.160	I
28	Succari red	53.16	A	23.69	B-F	0.0680	J	4.420	E-I
29	Succari white	41.67	BC	29.61	ABC	0.6613	EF	3.950	F-I
30	Sour	14.48	H-K	31.21	A	0.9214	C	5.820	C-F
31	Succari red	49.72	AB	27.67	ABC	1.4800	A	5.690	C-G
32	Succari red	52.22	A	27.76	ABC	0.7563	DE	9.480	A

Other studies, Martınez *et al.* [22] and Viswanath *et al.* [31] found percentages of juice content in Spanish and Indian varieties ranging from 44.96 to 68.55 which is closer to the values reported here. The acidity in Upper Egypt cultivars showed the highest value (2.75- 2.81 %).whereas, acidity in Siwa Oasis and North Sinai cultivars showed the lowest values (0.30), Table (1) Similar results were also reported by Fadavi *et al.* [32].While, acidity in Spanish variety ranged from (1.004 to 0.268 %) [22]. The pH in the studied cultivars ranged between 4.53% with Succari red from North Sinai to 3.10 in Assiuty from Upper Egypt (Table 1). These results are in agreement with those obtained by Al-Said *et al.* [14] and Tehranifara *et al.* [33]. However, the pH values obtained in the current study are greater than those reported by Cam *et al.* [34] on pomegranate cultivars grown in Turkey.

Total Soluble Solids Content (TSS): Total soluble solids displayed the highest variation of all parameter, both of sour and sweet cultivars from Siwa Oasis showed the highest TSS (20.33 and 18.67 °Brix) ,while, Assiuty Maragab cultivar from Upper Egypt had the lowest TSS(12.40 °Brix) (Table 1), while those varieties mentioned by Martınez *et al.* [22] varied from 12.36 to 16.32%.

Maturity Index: The maturity index (TSS/TA) is responsible for the taste and flavor of pomegranate, which some author used for classifying the pomegranate cultivars [21, 22, 34]. This classification has been optimized for Spanish cultivars: maturity index (MI) = 5-7 for sour, MI = 17-24 for sour-sweet and MI = 31-98 for sweet cultivars [22]. The maturity index varies considerably from 53.16 in Succari red (sweet variety) from North Sinai to 4.51 in Assiuty Maragab (sour variety) from Upper Egypt (Table 2). Similar values were obtained for some Spanish varieties evaluated by Martınez *et al.* [22]. Previous studies have also reported variable ranges of maturity index [22,31, 34, 35].According to the results, cultivar type plays an important role in terms of their total soluble solids, pH, titratable acidity and maturity index of

the pomegranate juice. All studied cultivars were suitable for direct consumption and production of pomegranate juice because they had the high levels of soluble solids.

Ascorbic Acid (Vitamin C) and Total Amount of Anthocyanins: The results for ascorbic acid of the pomegranate from the different cultivars are shown in Table 2 indicated that Succari red cultivar from North Sinai showed high value for ascorbic acid content (9.48 mg/100 ml),while both of Nab El Gamal and Manfalouty showed lowest value (2.77 mg/100 ml) of ascorbic acid content. Similar results obtained from variety Ganesh variety (>10 mg100/g) [7]. Anthocyanins are a member of phenolic compounds that contributes to the red, blue, or purple color of many fruits, including pomegranate juice and they are well-known for their antioxidant activity. There were significant differences in total anthocyanins content of pomegranate cultivars, which 'Both of Wardy and Assiuty Abo Shoka from Upper Egypt displayed high amounts of anthocyanin (1.37 mg/ml), while variability among the other cultivars were ranged from (0.8 -0.04 mg/ml), sour cultivar from Siwa Oasis showed the lowest value (0.045 mg/ml) (Table 2). Other study, Cam *et al.* [36] indicated that the levels of total anthocyanin varied between 8.1 and 36.9 mg100g/l of juice among different cultivars of pomegranate grown in Turkey. Also they indicated that the levels of ascorbic acid varied among different cultivars of pomegranate and there was a high genetic heterogeneity within the studied cultivars.

Seed Oil Content %: Data presented in Table 2 showed that Sour cultivar from North Sinai had the highest significant seed oil content (31.21 %), while the lowest (4.75 %) was obtained from fruits of Sweet cultivar from Siwa Oasis.

Ellagic Acid and Total Antioxidant Activity: Ellagic acid, which is an important phenolic acid with high antioxidant activity, has been reported in some fruit juices [37, 38] and was also, detected in pomegranate juice in the present study. The ellagic acid concentrations of the studied pomegranate cultivars are shown in Table 3. Significant differences were found among the ellagic acid levels of different cultivars. Assiuty from Upper Egypt showed the highest level of ellagic acid (10 mg/l) and Manfalouty cv. from Ismailia and Sweet cv. from Siwa Oasis contained the least amount of ellagic acid (0.99 and 0.84mg/l), respectively. In contrast, results of eight pomegranate juices from Iran showed lowest values ranged (8.0 to 0.7 mg/100g) than ellagic acid in Pomegranate juice [16].

Table 3: Eallgic acid content and antioxidant capacity (vitamin E) of 32 Pomegranate (*Punica granatum* L.) germplasm grown in different locations of Egypt.

Characters		Eallgic acid mg\l		DPPH Decolouration% At 100 µl(vitamin E)
*Germplasm				
			Assiut	
1	Assiuty Maragab	9.350	B	77.7
2	Assiuty Abo shokaa	10.53	A	72.8
3	Nab-El-Gamal	3.270	Ij	61.6
4	Abo-lammam	3.640	Hi	68.1
5	Balady	3.690	Hi	59.5
6	Manfalouty	4.880	Fg	68.1
7	Iranian	3.620	Hi	58.3
8	Assiuty	6.360	D	70.9
9	Manfalouty	3.600	Hi	66.0
10	Nab-El-Gamal	3.600	Hi	62.0
11	Assiuty	5.370	E	71.8
12	Manfalouty	3.840	H	68.1
13	Hejazy	2.810	K	44.7
14	Wardy	3.840	H	60.2
			Siwa Oasis	
15	Sour	4.430	G	68.6
16	Sweet	0.990	N	59.5
17	Sweet	1.470	Lm	51.7
18	Sour	6.650	Cd	62.0
19	Sour	0.840	N	54.0
20	Sour	5.210	Ef	63.7
21	Sour	7.050	C	60.7
22	Sweet	4.510	G	63.7
23	Sour	6.500	D	60.7
24	Sweet	1.550	L	69.0
25	Sour	6.545	Cd	50.0
			Ismailia	
26	Manfalouty	2.720	K	63.7
			North Sinai	
27	Succari red (smooth)	1.040	Mn	58.1
28	Succari red	3.050	Jk	66.2
29	Succari white	9.400	B	73.3
30	Sour	5.260	Ef	72.3
31	Succari red	1.640	L	61.6
32	Succari red	1.580	L	68.6

The DPPH radical scavenging assay is commonly employed to evaluate the ability of antioxidant to scavenge free radicals. The degree of discoloration indicates the scavenging potentials of the antioxidant extract. In this study, the differences in antioxidant activity among the pomegranate cultivars were statistically significant and the values ranged from 77.7 to 44.7%. Antioxidant capacity activity was higher in Red Succari cv. from North Sinai arils juice (77.7 % at 100 µl) than the other cultivars from other locations, while Hegazy cv. from Assiut showed the lowest antioxidant capacity activity (44.7 % at 100 µl). Decolouration% using standard concentration as vitamin E (0.1g/100ml methanol) was 89.9% at 100 µl while, decolouration% using standard concentration as vitamin C (0.1g/100 ml methanol) was 92.4% at 100 µl (Table 3).

Table 4: Fatty acid composition of 32 Pomegranate (*Punica granatum* L.) germplasm grown in different locations of Egypt.

Characters *Germplasm		Lauric		Myristic		Palmitic		Stearic	
				Assiut					
1	Assiuty Maragab	2.694	D-G	16.90	C	41.09	A	9.137	BC
2	Assiuty Abo shokaa	0.861	I-L	11.33	JK	21.07	GH	4.872	H-M
3	Nab-El-Gamal	0.989	H-L	7.22	MN	11.56	M	5.060	H-M
4	Abo Lammam	2.515	D-H	16.25	CD	24.29	EF	8.954	BC
5	Balady	1.086	G-L	12.50	HIJ	22.78	FG	7.864	C-F
6	Manfalouty	9.510	A	14.48	D-G	27.05	D	7.600	C-G
7	Iranian	3.068	CDE	12.29	HIJ	18.02	IJK	6.611	D-H
8	Assiuty	0.630	I-L	7.922	MN	19.16	IJ	4.473	J-N
9	Manfalouty	3.818	BCD	15.10	DEF	34.13	C	10.17	B
10	Nab-El-Gamal	1.868	E-K	13.45	F-I	35.16	BC	8.121	CD
11	Assiuty	0.373	JKL	8.75	LM	13.70	L	3.985	K-N
12	Manfalouty	1.561	E-L	11.77	IJK	19.00	IJ	6.138	E-J
13	Hejazy	4.952	B	15.38	CDE	33.72	C	7.589	C-G
14	Wardy	0.785	I-L	10.17	KL	19.46	HI	5.031	H-M
				Siwa Oasis					
15	Sour	0.543	JKL	7.019	MNO	5.68	PQ	3.651	LMN
16	Sweet	1.404	F-L	13.26	F-I	24.77	E	6.075	F-J
17	Sweet	1.986	E-J	18.68	B	8.091	NO	7.911	C-F
18	Sour	2.190	E-I	12.83	G-J	19.65	HI	3.953	K-N
19	Sour	1.203	F-L	6.501	NO	6.682	OP	3.537	MN
20	Sour	1.468	E-L	8.657	LM	22.08	G	5.454	H-M
21	Sour	1.468	E-L	8.657	LM	22.08	G	5.454	H-M
22	Sweet	4.345	BC	26.20	A	13.91	L	14.390	A
23	Sour	1.510	E-L	10.28	KL	12.01	M	4.853	H-M
24	Sweet	0.500	JKL	5.36	O	4.349	Q	2.670	N
25	Sour	4.519	BC	19.57	B	12.55	LM	9.085	BC
				Ismailia					
26	Manfalouty	2.773	DEF	8.73	LM	17.44	JK	5.477	H-L
				North Sinai					
27	Succari red (smooth)	2.731	DEF	13.77	E-H	22.81	FG	5.836	G-K
28	Succari red	0.326	KL	10.27	KL	9.258	N	4.709	H-M
29	Succari white	2.778	DEF	13.51	F-I	17.08	K	6.521	D-I
30	Sour	8.968	A	15.03	DEF	36.11	B	7.978	CDE
31	Succari red	8.795	A	18.82	B	16.36	K	4.627	I-M
32	Succari red	0.082	L	8.22	MN	9.475	N	4.050	K-N

Table 5: Fatty acids characterization of 32 Pomegranate (*Punica granatum* L.) germplasm grown in different locations of Egypt.

Characters *Germplasm		Oleic		Linoleic		Linolenic		Arachidic		Arachidonic	
					Assiut						
1	Assiuty Maragab	16.24	G-J	5.410	JK	3.108	B-G	0.763	FGH	None	
2	Assiuty Abo shokaa	17.08	FGH	10.20	CD	2.398	C-I	2.201	E	20.08	I
3	Nab-El-Gamal	9.397	QRS	6.928	HIJ	1.572	E-J	0.337	GH	50.71	A
4	Abo Lammam	15.35	HIJ	6.274	IJ	2.527	C-I	0.772	FGH	3.546	LM
5	Balady	19.33	E	9.208	D-G	3.721	A-D	0.880	E-H	16.55	J
6	Manfalouty	17.61	EFG	13.36	B	2.973	B-H	1.108	E-H	None	
7	Iranian	14.90	IJK	14.76	AB	2.341	C-I	0.661	FGH	19.27	I
8	Assiuty	12.85	LMN	7.974	F-I	1.698	E-J	1.918	EF	35.50	E
9	Manfalouty	16.62	GHI	4.463	K	3.851	ABC	1.211	E-H	1.823	N
10	Nab-El-Gamal	18.55	EF	6.465	HIJ	1.798	E-J	0.950	E-H	6.602	K
11	Assiuty	10.13	PQR	7.743	GHI	1.838	D-J	0.776	FGH	48.63	B
12	Manfalouty	18.42	EF	5.259	JK	3.418	B-E	1.313	E-H	23.34	H
13	Hejazy	12.01	MNO	10.64	CD	2.163	C-I	1.838	EF	4.625	L
14	Wardy	8.390	RS	6.728	HIJ	1.050	HIJ	1.215	E-H	35.55	E
					Siwa Oasis						
15	Sour	11.25	NOP	9.689	C-F	1.272	F-J	51.97	B	None	
16	Sweet	8.30	S	9.000	D-G	2.221	C-I	0.937	E-H	26.21	G
17	Sweet	17.83	EFG	15.97	A	2.418	C-I	17.77	D	None	
18	Sour	10.78	OPQ	9.188	D-G	2.833	B-I	1.212	E-H	26.71	G
19	Sour	10.15	PQR	9.123	D-G	0.888	IJ	47.99	C	None	
20	Sour	9.03	QRS	7.143	HIJ	1.111	HIJ	0.745	FGH	37.78	D
21	Sour	9.03	QRS	7.143	HIJ	1.111	HIJ	0.745	FGH	37.78	D
22	Sweet	24.54	C	7.123	HIJ	5.349	A	0.911	E-H	None	
23	Sour	14.50	JKL	11.38	C	1.523	E-J	0.372	GH	38.56	D
24	Sweet	6.68	T	6.113	IJK	1.006	IJ	59.32	A	None	
25	Sour	30.81	A	10.36	CD	2.138	C-I	1.514	EFG	None	

Table 5: Continued

Characters *Germplasm		Oleic		Linoleic		Linolenic		Arachidic		Arachidonic	
					Ismailia						
26	Manfalouty	8.57	RS	9.984	CDE	1.238	F-J	0.924	E-H	32.42	F
					North Sinai						
27	Succari red (smooth)	27.00	B	14.02	B	4.461	AB	0.600	FGH	3.204	M
28	Succari red	13.31	KLM	9.390	D-G	1.839	D-J	0.760	FGH	42.97	C
29	Succari white	21.28	D	10.23	CD	1.394	F-J	0.788	FGH	18.97	I
30	Sour	16.25	G-J	6.909	HIJ	3.154	B-F	1.052	E-H	0.358	O
31	Succari red	27.21	B	13.41	B	2.310	C-I	1.33	E-H 50.70	A	
32	Succari red	11.81	M-P	8.232	E-H	1.188	G-J	1.337	E-H	51.71	A

* (1-14) pomegranate germplasm from Assiut (El-Badary, Sahel Selim and El Fatth), (15-25) from Siwa oasis, (26) from Ismailia and (27-32) from North Sinai.

Antioxidant activity has been reported for seven commercial pomegranate juices from Turkey 10.37-67.46% (17) and eight pomegranate juices from Iran 18.6-42.8% [16].According to the results presented in Table 3, cultivars from Assiut had the highest levels of ellagic acid and antioxidant activity, while the lowest levels were in Siwa Oasis cultivars. Thus it can be concluded that there was a close relationship between the phenolic component and antioxidant activity.

Total Fatty Acid: The total nine fatty acid compositions in 32 pomegranate cultivars growing in different four locations (Upper Egypt, North Sinai, Ismailia and Siwa Oasis) was determined; it was found some differences among the studied cultivars. Palmitic acid was determined to be the pre-dominant fatty acid in 32 pomegranates varieties, its amounts ranged between 41.09-9.47%and followed by oleic acid. Regarding the content in fatty acids, Sweet cultivars from Siwa Oasis showed the greatest percentage of Arachidic, Myristic, Linoeic, Stearic and Linolenic acids (59.32, 26.2, 15.9 14.3 and 5.3% ,respectively). Manfalouty cv. from Upper Egypt displayed the highest content of Lauric acid (9.5 %) and Assiuty Maragab showed the highest content of Palmitic acid (41.09 %). Sour cv. from Siwa Oasis displayed the highest value of Oleic acid (30.81%). Enormous differences were found among cultivars regarding content of Arachidonic acid where many cultivars did not contain Arachidonic acid, whereas Nab El-Gamal cv. showed the highest value (50.7 %) and Sour cv. from Siwa Oasis had the lowest value (0.35%). These results are in agreement with the findings of Hernandez *et al.* [39], who found that some differences among the studied varieties, where one variety had the lowest linoleic acid content in comparison with the rest of the varieties and the three studied varieties present very few differences in palmitic, oleic and stearic acid content. Our results confirm the presence of Oleic (C18:1), linoleic (C18:2), linolenic (C18:3) and arachdonic (C20:4) acids as unsaturated fatty acids where lauric (C12:0), myristic(C14:0), palmitic (C16:0), stearic(C18:0) and arachidic(C20:0) as saturated fatty acids in all seed oils of pomegranates, while unsaturated fatty acids were pre-dominant in all varieties as previously reported by Melgarejo and Artes [40], Fadavi *et al.* [41] and Parashar *et al.* [42].

CONCLUSION

This study showed considerable variation in some of the chemical and antioxidant properties of pomegranate cultivars grown in different regions of Egypt. Among the thirty two cultivars studied, Siwa Oasis cultivars showed the highest content of fatty acids content, while Assiut cultivars displayed the highest content of antioxidant activity, total anthocyanins and ellagic acid which are suitable for health benefits. Also, the results provide important information of the physic-chemical properties of pomegranate cultivars which can be useful for developing fruit processing industry and selection of superior desirable pomegranate genotypes for bringing to commercial cultivation. It is important to evaluate and conserve local genetic materials, not only for general consumption, but also for their health advantages particularly; the physiological effects of pomegranate juice constituents are remarkable for their preventive potential against heart disease and certain cancers. The variation could originate from the pomegranate cultivar and agro-climatic as well as environmental conditions. This study provides important data for composition information of the fruits (e.g. vitamin C, titratable acidity, antioxidant activity etc.), highlighting that the pomegranate fruit can be a good source of nutrients. In conclusion, a comparison of the results obtained by us with those found in other studies reveals that pomegranate fruit contains important amounts of antioxidant and high amount of nutrients both in arils and peel that play a valuable role in people's daily diet.

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