Physical characteristics, phytochemical content and antioxidant activity of cactus pear fruits growing in northeast Algeria

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ABSTRACT

The present work aims to evaluate the effect of locality on the quality of available orange-fleshed cactus pear cultivar in Souk-Ahras (Algeria). Samples collected from the study area were assessed for their physical characteristics, chemical composition and antioxidant properties. The cactus pear fruits originating from Remila site exhibited significantly higher fruit and seed weights. The juicy pulps were sweet (14.00~15.85°Bx) and showed high moisture content (85.04~86.80%), low acidity (0.038~0.049%) and a near neutral pH (5.91~6.25*)*. Phytochemical content analysis revealed significant higher levels of polyphenols, flavonoids and total carotenoids in pulp juice from Meridef and Chegaga sites, while pulp juice from Remila site had significant higher levels of vitamin C, flavonols and betalains contents. Results of principal component analysis suggest that vitamin C, betalains and flavonols content contributed efficiently to the radical scavenging activity, whereas polyphenols, flavonoids and carotenoids were rather associated with the reducing power activity.

Keywords: *Opuntia ficus indica*, betalains, carotenoids, flavonols, polyphenols.

INTRODUCTION

Opuntia ficus-indica is one of approximately three hundred species of the *Opuntia* genus, which belongs to the *Cactaceae* family. Only ten to twelve species are commonly cultivated for production of fruit (Yahia and Sáenz 2011). Cactus pear originated from arid and semi-arid regions of Mexico, and was introduced into North Africa in the 16th Century (Felker *et al*., 2005). Cactus contains a fruit known as cactus pear and the plant is referred to as nopal (pad). The edible portion of the fruit is made up of a number of funicles intermixed with juicy papillary cells. Funicles are produced as outgrowths from the internal fruit wall and involve many hard seeds. The variously colored fleshy pulp has a sweet flavor and is surrounded by a thick, fleshy skin (Yahia and Sáenz 2011). The presence of antioxidants (flavonoids, ascorbate), pigments (carotenoids, betalain such as indicaxanthin), or phenolic acids have been reported in all *Opuntia* products, including seeds, roots, pears, cladodes, or juice (Díaz *et al*. 2017).

The nutritional and health benefits of cactus fruit are currently thought to be related to their antioxidant properties, which in turn are due to their contents of ascorbic acid, phenolics including flavonoids, yellow betaxanthins and red betacyanins (Tesoriere *et al*., 2003; Kuti 2004).

Based on recent reports, *Opuntia ficus-indica* intake is another interesting nutritional strategy to stimulate insulin secretion and glycogen resynthesis after exercise. *Opuntia ficus-indica* has been found to lower blood glucose and increase basal plasma insulin levels in animals as well as in humans (Deldicque *et al*., 2013).

The antioxidant properties of the biocompounds in cactus pear plants makes the fruit an important product for preventing human health against degenerative diseases such as cancer, diabetes, hypercholesterolemia, arteriosclerosis or cardiovascular and gastric diseases, as is reported about other flavonol-rich foods (Jacob *et al*., 2008; Lampila *et al.,* 2009).

Opuntia species have adapted to grow and produce under low water regimes, high temperature and poor soils, adverse conditions for the production of many other crops. Previous studies have shown that the *Opuntia* species have a regional specificity (Bouzoubaâ *et al*., 2016; El Kharrassi *et al*., 2016). In fact, cactus pears exhibit both intra- and inter-site variability in the shape, color, weight, sugar, acids, antioxidants, etc. These parameters vary from one cultivar to another and are strongly influenced by the environment (Parish and Felker 1997)**.**

In Algeria, the cultivation of cacti in large areas began in 1994 through an implementation program initiated by the High Commission for the Development of the Steppe (HCDS) as a part of the program of development of pastoralism and countering desertification in steppe and agro-pastoral areas. Indeed, 52,000 hectares of cactus were planted in the four eastern Wilaya: Oum El Bouaghi, Tebessa, Khenchela and Souk-Ahras.

Besides, the cacti are present in the Algerian rural landscape, namely as hedges limiting plots of crops, orchards or trails. Many plantations continue to play a major role today not only in maintaining local populations and their livestock but also in the fight against soil erosion and the preservation of biodiversity. The cultivation of cacti is generally conducted in a traditional way and hardly receives attention or care from farmers, so it remains dependent on weather conditions and consequently provides fruit yields lower than real potential.

Recently the cactus cultivation receives a growing attention since the increasing demand on its products driven by the broadening of knowledge on the positive impact of *Opuntia* products consumption on health, in one hand, and the substantial commercial profit that can be earned from the industrial valorization of high added value products and by-products, on the other hand. Efforts are currently under way to develop cactus pear through implementation of an integrated agricultural pole for the *Opuntia* culture in Souk-Ahras region. This has stimulated the need to evaluate the quality and pharmaceutical properties of the cactus edible fruit growing in this region. Therefore, the present study was aimed to evaluate physical characteristics, antioxidant activity, phytochemical and nutritional properties of the fruits of four potential cactus pear cultivating sites in Souk-Ahras area.

MATERIALS AND METHODS

Ethics statement

The research conducted herein did not involve measurements with humans or animals. The study site is not considered a protected area. For locations/activities, no specific permissions were required and the field studies did not involve endangered or protected species.

Study area

The work was carried out on the region known as "Souk-Ahras", which is located in the eastern part of northern Algeria. The climate of this region is classified as of subhumid; known as an arid climate in summer and relatively mild winters. Depending on the site, annual average precipitation varies from 760 to 824 mm. Yearly average temperatures ranges from 32~35 °C as a maximum and 3.8~5.0 °C as a minimum (Table 1).

Table 1. Geographical coordinates and climate parameters of Souk-Ahras study sites.

Site	Latitude	Longitude	Altitude (m)	Precipitation* (mm)	Temperature* (°C)	
					Average Max	Average Min
Bouamoud	36.01649	8.144063	549	779	34.0	04.7
Chegaga	35.982088	8.219714	455	760	35.0	05.0
Meridef	36.04941	8.242078	589	787	34.0	04.5
Remila	36.2809	8.1046	771	824	32.7	03.8

*Source: Weather reports, statistics for 121 year. Weather data from the meteorological station of the city of Souk Ahras, located 655 m above sea level, were used. The values of precipitation and temperature at different altitudes were evaluated based on the altitudinal gradient of Seltzer *et al.* (1946).

Sample Material

Fruits of spineless cactus [*Opuntia ficus-indica (L.) Mill.*] were collected at full maturity, from non-irrigated orchards in Souk-Ahras area (Fig. 1). Four sites were considered, and forty healthy orange-skinned fruits from each site were taken. After manual separation of the peel from the pulp, fruits of each site were briefly homogenized in a laboratory-type blender (Waring, USA). Then the pulp was separated from the seeds by sieving (2 mm mesh size) and used to measure physical and chemical parameters. The seeds were washed, dried at room temperature and weighed. The juicy pulp homogenate was portioned and stored at −20 °C until analysis.

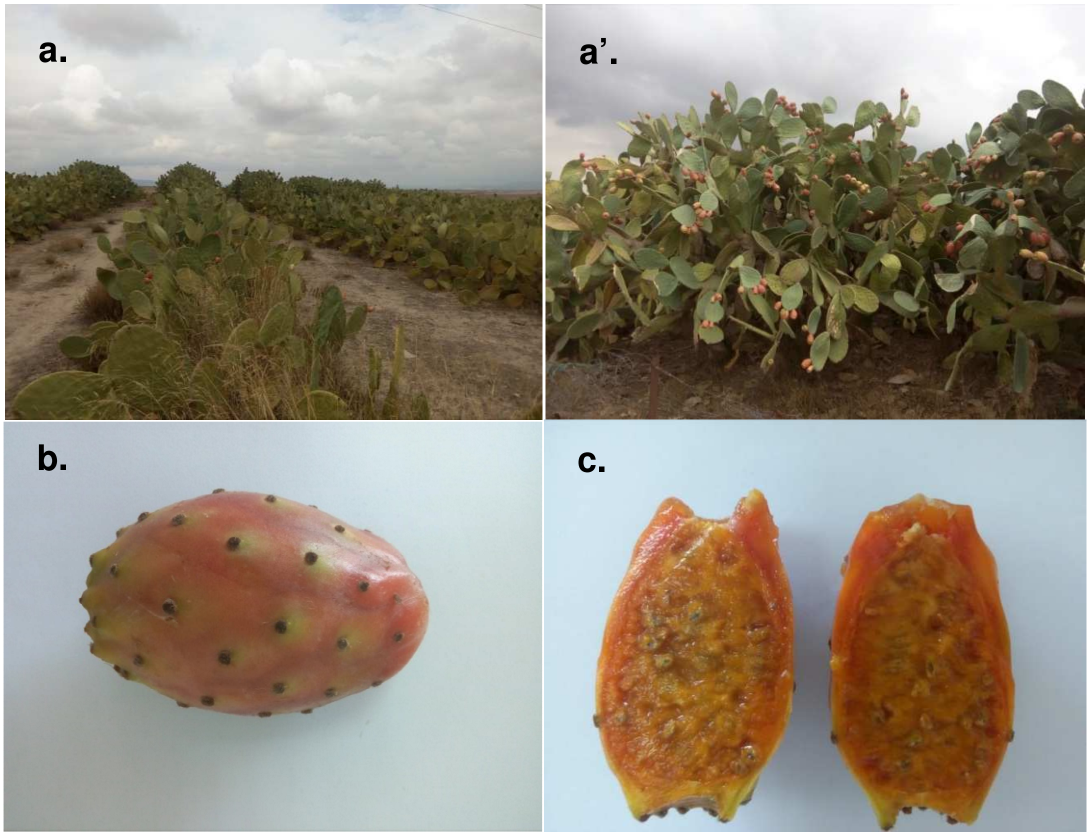

Figure 1. Cactus pear fields in Souk-Ahras region **(a.** and **a'.)**; Cactus pear fruit **(b.)**; longitudinal cross section of cactus pear fruit from Souk-Ahras area **(c.)**

Reagents

All reagents were of analytical grade. Folin Ciocalteu's reagent, gallic acid, sodium carbonate, hexane, ethanol, 2,2-diphenyl-1-picrylhydrazyl, trichloro-acetic acid and ammonium molybdate were purchased from Sigma-Aldrich (St. Louis, USA). Sulfuric acid (H_2SO_4 96%) and sodium hydroxide (NaOH 98%) were from Cheminova internacional, SA (Madrid, Spain). Aluminium trichloride, acetone, sodium thiosulfate, diiode were purchased from Fluka (Buchs, Germany). Potassium ferricyanide, ferric chloride, sodium dihydrogen phosphate and sodium hydrogen phosphate were from Biochem Chemopharma (Quebec, Canada). Methanol was purchased from Honeywell (Seelze, Germany).

Physical and Chemical Parameters

Fruit length and diameter were determined over forty fruit unit per site using a digital caliper. Whole fruit peel and dried seed weights were determined using laboratory precision scale (OHAUS, Explorer). Moisture was determined on three replicates by desiccation at 105°C for 24 h. Soluble solids and the pH were measured using a temperature-adjusted refractometer (Krüss, Germany) at 20°C, and a pH meter (Crison, Basic 20, Spain), respectively. Titratable acidity was determined by titration with NaOH (0.1 N) and expressed as percent of citric acid (Cámara *et al*., 1994).

Phytochemicals content assessment

Total phenolics

The total phenolic compounds present in the cactus pear juice were quantified using a method adapted from Abdel-Hameed *et al*. (2014). An aliquot of 100 µl of pulp juice diluted in distilled water (100 µl/ml, v/v) was added into a test tube containing 0.5 ml Folin-Ciocalteu reagent. The mixture was shaken thoroughly then allowed to react for 5 min, after which 1.5 ml of 20% Na_2CO_3 was added. The mixture was incubated in dark for 2 hours at room temperature, and the absorbance was recorded at a wavelength of 765 nm using UV-2005 spectrophotometer (Selecta, Spain). For the calibration curve, different concentrations of the standard gallic acid were prepared with the same conditions. Total phenolics content of the sample was expressed as gallic acid equivalent (GAE, mg gallic acid/100 ml juice). The data were reported as mean ± standard deviation (SD) for at least three independent replications.

Flavonoids content

Total flavonoid content was measured using a spectrophotometric assay as reported by Abdel-Hameed *et al*. (2014). An aliquot 100 µl of diluted cactus pear juice in distilled water (100 µl/ml, v/v) or standard solution of quercetin was added 100 µl of 2% aluminum trichloride solution in ethanol and a drop of acetic acid. The final volume was adjusted to 5 ml with methanol and thoroughly mixed. After 40 minutes, the absorbance of the mixture was determined at 415 nm. Total flavonoid content was deduced through the calibration curve of quercetin and was expressed as mg quercetin equivalent/100 ml pulp juice (mg QE/100 ml juice). All samples were analyzed in triplicates.

Flavonols content

Total flavonols in the pulp juice were estimated using the method reported by Abdel-Hameed *et al*. (2014) with slight modification. To 1 ml of diluted pulp juice sample in distilled water (500 µl/ml, v/v), 1 ml of $AlCl_3$ (20 g/L) in ethanol and 3 ml (50 g/L) sodium acetate solutions were added. The mixture was shaken and incubated for 2.5 h at room temperature. The absorbance was read at 440 nm. For the calibration curve, different concentrations of quercetin were prepared with the same conditions. Total flavonols content was expressed as mg of quercetin equivalents per 100 ml of pulp juice (mg QE/100 ml juice). Three replicates were applied for each sample analysis.

Vitamin C

The determination of ascorbic acid was realized according to the iodometric method. A 5 ml aliquot of pulp juice was mixed with an excess of iodine solution (0.005 M) and some drops of starch paste. The vitamin C reacted with the iodine and excess iodine was titrated with sodium thiosulfate until disappearance of the dark color (Farrington *et al*., 1994). All determinations were carried out in triplicate.

Total carotenoids

Carotenoid extraction and determination was performed according to procedures previously described (Fernández-López *et al.*, 2010). Approximately 5 g of homogenized fruit pulp were extracted twice with 50 ml of hexane/acetone/ethanol (50:25:25, v/v) before being centrifuged for 5 min at 3,900 rpm in a 2-6E centrifuge (Sigma, Germany). The top layer of hexane, containing the color, was recovered and transferred to a 50 ml volumetric flask and adjusted to 50 ml with hexane. Total carotenoid determination was performed on an aliquot by measuring absorbance at 450 nm and using an extinction coefficient of β-carotene, $E^{1\%}$ = 2.505 (Kuti, 2004). The results were expressed as µg of β-carotene equivalent per 100 g of fresh pulp.

Betalain content

Betalain pigments were extracted by stirring 10 g of the pulp homogenate with 50 ml of distilled water for 5 min. Extracts were then clarified by centrifugation at 1,660 × g for 10 min in a 2-6E centrifuge (Sigma, Germany) and the supernatant was passed through a 0.45 µm nylon filter (Fernández-López *et al*., 2010). The betalain content (BLC) was calculated as described earlier (Stintzing *et al*., 2003): [BLC (mg/L) = (A × DF × MW × 1000) / (ε × l)], where A is the absorption value at the absorption maximum, the dilution factor (DF) and l the path-length (1 cm) of the cuvette. For quantification of betacyanins (Bc) and betaxanthins (Bx), the molecular weights (MW) and molar extinction coefficients (ε) of betanin (MW = 550 g/mol; ε = 60,000 L/(mol cm) in H_2O; λ = 538 nm) and indicaxanthin (MW=308 g/mol; ε = 48,000 L/(mol cm) in H_2O; λ = 480 nm) were applied, respectively.

Antioxidant Activity

Three methods were used for the evaluation of the antioxidant properties of cactus pear fruit juice collected from the study sites: 1,1-diphenylpicrylhydrazyl radical scavenging activity, reducing power activity and total antioxidant capacity. All experiments were performed in triplicate.

Radical scavenging activity of 1,1-diphenylpicrylhydrazyl (DPPH)

The ability of cactus pear juice to scavenge 1,1-diphenyl-2-picryl-hydrazyl (DPPH) free radicals was estimated as previously described by (Abdel-Hameed *et al*., 2014). Various dilutions of pulp juice or ascorbic acid (2 ml) were mixed with 2 ml of a 0.2 mM methanolic solution of DPPH. Reaction mixtures were incubated in a dark place at room temperature for 30 minutes. Then the absorbance was measured at 517 nm. The disappearance of DPPH was recorded and the percent inhibition of the DPPH radicals by sample is calculated as follows:

$$\text{Radical scavenging (\%)} = [(A_0 - A_1 / A_0) \times 100]$$

Where A_0 is the absorbance of the control and A_1 is the absorbance of the sample. All determinations were carried out in triplicates.

Reducing power activity

The reducing power was determined according to procedure based on the method of Oyaizu (1986). Aliquots (2 ml) of different cactus pear juice dilutions in distilled water or standard ascorbic acid (1~20 µg/ml) were added phosphate buffer (2 ml, 0.2 M, pH 6.6) and were mixed with potassium ferricyanide (2ml, 1%). The mixture was then incubated at 50°C for 20 min. The reaction was terminated by trichloroacetic acid solution (2 ml, 10%) and centrifuged at 3,900 rpm for 10 min. Two milliliters of the supernatant was taken out immediately and added 2 ml of ultrapure water and mixed with ferric chloride (0.5 ml, 0.1%). The absorbance was measured at 700 nm against a blank. Three replicates were performed on control (ascorbic acid) and each tested sample.

Total antioxidant capacity (Phosphomolybdenum method)

The total antioxidant capacity (TAC) of cactus pear juice was spectrophotometrically determined by the phosphomolybdenum assay using the method described by (Abdel-Hameed *et al.*, 2014). Briefly, 0.3 ml of diluted pulp juice (0.1 ml/ml; v/v) solution in distilled water was mixed with 3 ml phosphomolybdenum reagent (28 mM sodium phosphate and 4 mM ammonium molybdate in 0.6 M sulfuric acid) in capped test tubes. Incubation was then carried out for 90 min in a water bath at 95°C. After cooling to room temperature, the absorbance of the solutions was measured using a UV-visible spectrophotometer at 695 nm against a blank. For the calibration curve, the same procedure was applied to various concentrations of ascorbic acid (5~80 µg/ml). TAC results were expressed as ascorbic acid equivalents (mg AAE/100 ml juice).

Statistical analysis

All the statistics were performed by means of the XLSTAT software for Windows. Mean values obtained for the variables studied in the different groups were compared by one-way ANOVA, assuming that there were significant differences among them when the statistical comparison gives $p<0.05$. Factor analysis, using principal components as the method of factor extraction was employed to summarize information in a reduced number of factors.

RESULTS AND DISCUSSION:

Physical and chemical analysis

Fruit physical characteristics of *Opuntia ficus-indica* growing in different sites of Souk-Ahras are reported in Table 2. The fruit length ranged from 6.04 to 6.91 cm while the fruit diameter oscillated from 4.40 to 5.02 cm. These data show that among the study sites, Remila is characterized by the largest mean fruit size, whereas, Meridef site had significantly the lowest mean fruit size. The distinction of the Remila site would probably be related to the geographical location (mountainous area) which receives higher precipitations compared to the other sites (Table 1). According to the shape coefficient C_f (Temagoult *et al.,* 2017), which ranged from 1.37 to 1.44, the fruits of the study sites were classified in the category of elongated shape.

Among the studied sites, Remila presented a significantly higher fruit mass per unit (93.02 g), followed by Bouamoud with 82.90 g. Chegaga and Meridef sites showed the lower fruit mass per unit with 70.18 and 67.96 g, respectively. In a study of the *Opuntia* fruit gathered from different Mexico and Argentina regions, the commercial fruit weight was ranged from 112 g to 212 g (Felker *et al*. 2005). In North African studies, Chougui *et al.* (2013) reported an average fruit weight of 76.28 g for the orange variety grown in Bejaia (Algeria), whereas, Temagoult *et al.* (2017) reported the value of 63.38 for cactus pears growing in Batna (Algeria). Furthermore, Bouzoubaâ *et al.* (2016) reported an average weight range of 43.25~64.55 g for spineless orange variety growing in Morocco.

Table 2. Physical characteristics of *Opuntia ficus indica* fruits growing in different sites of Souk-Ahras.

Site	Dimension (cm)		Weight	Peel	Pulp	Seed	
	Length	Diameter	(g)	(%)	(%)	Seed per fruit	Weight (g)
Bouamoud	6.71±0.84[a]	4.81±0.43[b]	82.90±22.03[b]	40.40±5.77[a]	59.60±5.77[b]	187.10±25.28[b]	2.51±0.36[b]
Chegaga	6.68±0.55[a]	4.61±0.28[c]	70.18±11.66[c]	37.21±5.02[b]	62.79±5.02[a]	173,80±23.21[b]	2.28±0.30[b]
Meridef	6.04±0.59[b]	4.40±0.43[d]	67.96±14.52[c]	40.18±5.94[a]	59.82±5.94[b]	189.33±17.65[b]	2.20±0.26[b]
Remila	6.91±0.60[a]	5.02±0.32[a]	93.02±17.04[a]	36.29±4.74[b]	63.71±4.74[a]	254.63±13.66[a]	3.46±0.27[a]

The physical characteristics measures were obtained from forty fresh fruit selected in a random way (n=40). For seeds n=10.
Values having same letter within the column did not differ significantly from each other according to LSD test at $p<0.05$.

The average percentage fruit pulp in the four sites was above 59%. Chegaga and Remila's average percentages fruit pulp were noticeably higher than those of Meridef and Bouamoud. Pulp percentage should not be below 55–60% for export markets (Inglese *et al*., 1995). The relatively smaller size observed in this study could probably due to many factors including: the overcrowding orchards, the lack of cultural practices like pruning, thinning and fertilization in one hand, and in the other hand despite these sites are situated mostly in a subhumid area, the culture is suffering from a lack of precipitation particularly during the last two months prior to maturation. About 200 mm in the 6 weeks prior to harvest are required for commercial fruit sizes greater than 130 g. In a study comparing fruits of cactus pear growing in Argentina to those growing in USA, Felker *et al.* (2002), have observed that the rainfall in the last two months of fruit maturation in Argentina was greater than that of the U.S.A. They concluded that the greater rainfall in the later stages of fruit development in Argentina is responsible for the greater fruit size, greater pulp to total fruit percent and less seeds to pulp content in some clones. The authors suggested that at the correct stage of fruit maturation irrigation or ample water, can merely increase fruit size and pulp percentages without having a detrimental effect on decreasing fruit sugar concentrations.

Remila site exhibited the higher content and weight of seed per fruit unit (Table 2). Meridef, Chegaga and Bouamoud sites did not differ significantly in terms of seed content and seed weight per fruit unit.

Chemically, the cactus pears juicy pulp of the four sites enclose a high moisture content (85.04~86.80%). Their pH values were determined in the range of 5.91 to 6.25 confirming that this fruit is low acidic food. During ripening, the pH of cactus pear fruit usually rises from 5 to a range around 5.5–6.5 (Albano *et al.*, 2015).

The titratable acidity of the fruit pulp of cactus pears from different sites analyzed in this study ranged from 0.038 to 0.049%. The highest titratable acidity was measured in fruit pulp of cultivar from Meridef and the lowest was in that from Remila (Table 3). These values are lower than those reported by (Temagoult *et al.*, 2017; Bouzoubaâ *et al.*, 2016; Díaz Medina *et al.*, 2007; Jiménez-Aguilar *et al.*, 2015). Overall, fruits from Remila and Bouamoud sites exhibited a significantly lower titratable acidity and higher pH values, whereas those from Chegaga and Meridef sites displayed a significantly higher titratable acidity and lower pH values (Table3).

The four sites showed fruit pulp Brix (%) value equal or greater than 14; however they did not differ significantly from each other (Table 3). These values are higher than those reported by Stintzing *et al.* (2005), Albano *et al.* (2015), El Kharrassi *et al.* (2016) and Temagoult *et al.* (2017); but are in good agreement with those reported by Chougui *et al.* (2013), Bouzoubaâ *et al.* (2016) and Díaz-Medina *et al.* (2007).

Total soluble solids (TSS) is a variable parameter depending on maturity stage and fruit metabolism. Sugar content is an important criterion of fruit quality for consumers who prefer sweet fruit (Inglese *et al.*, 1995). TSS is highly influenced by crop management and environment since cactus pear fruit grown in dry areas are sweeter than those grown in humid areas or being irrigated (Inglese *et al.*, 1995).

Table 3. Relevant chemical characteristics of *Opuntia ficus indica* fruit pulp grown in different sites of Souk-Ahras.

Site	Moisture content (%)	pH	Acidity (%)	Brix (%)
Bouamoud	85.08±0.15^{b}	6.06±0.07^{b}	0,039±0,002^{a}	14.75±1.06
Chegaga	86.63±0.10^{a}	5.93±0.06^{c}	0,046±0,002^{b}	14.00±0.00
Meridef	85.04±0.16^{b}	5.91±0.07^{c}	0,049±0,004^{b}	15.85±0.92
Remila	86.80±0.11^{a}	6.25±0.09^{a}	0,038±0,003^{a}	14.75±0.35

Values were expressed as mean of triplicate determinations ± standard deviation.
Values having same letter within the column did not differ significantly from each other according to LSD test at $p<0.05$.

Phytochemical Content

The amount of total phenolics, flavonoids, flavonols, vitamin C, total carotenoids and betalains content, differed significantly between sites of the study (Table 4). Meridef and Chegaga sites showed a significantly higher content of total phenolics, flavonoids and carotenoids; while Remila site had significant higher values of flavonols, vitamin C and betalains. The total phenolics content ranged from 493.56 to 618.56 mg GAE/100 ml pulp juice. These values are higher than those previously published (Stintzing *et al*., 2005; Saénz *et al*.,. 2009) and are close to data published for yellow pulp cactus pears (Abdel-Hameed *et al*., 2014) and pulp of Mexican cactus pear varieties (Jiménez-Aguilar *et al*., 2015).

Table 4. Pulp juice phytochemical content (mg/100 ml) of *Opuntia ficus indica* grown in different sites of Souk-Ahras.

Site	Phenolic content[1]	Flavonoids[2]	Flavonols[2]	Vitamin C	Total carotenoids[3]	Betacyanin	Betaxanthin	Total Betalains
Bouamoud	503.31±57.45[b]	064.14±12.02[b]	07.65±0.07[c]	71.62±0.92[c]	093.15±12.20[b]	1.66±0.43[b]	09.10±0.43[c]	10.76±0.86[c]
Chegaga	611.44±45.92[a]	089.86±06.55[a]	06.92±0.06[c]	69.42±1.55[d]	122.42±06.10[a]	1.64±0.25[b]	08.42±0.29[d]	10.06±0.54[c]
Meridef	618.56±53.24[a]	102.00±04.34[a]	11.23±0.03[b]	76.91±1.02[b]	123.75±06.91[a]	2.43±0.10[a]	12.05±0.18[b]	14.47±0.28[b]
Remila	493.56±16.97[b]	072.48±05.02[b]	12.84±1.22[a]	78.96±0.25[a]	095.81±06.91[b]	2.49±0.03[a]	14.16±0.12[a]	16.65±0.12[a]

Values were expressed as mean of triplicate determinations ± standard deviation (n = 3).
Values having same letter within the column did not differ significantly from each other according to LSD test at p<0.05.
[1] Expressed by mg gallic acid equivalent/100 ml juice.
[2] Expressed by mg quercetin equivalent/100 ml juice.
[3] Expressed by µg/100 g pulp.

Flavonoids content measured for cactus pears from Meridef and Chegaga sites exhibited the values 102 and 89.86 mg QE/100 ml of pulp juice respectively; whereas Remila and Bouamoud sites had the lower values 72.48 and 64.14 mg QE/100 ml pulp juice, respectively.

Fernández-López *et al.* (2010) reported for different *Opuntia spp.* tested, significant amounts of flavonoids, quercetin being the predominant type followed by isorhamnetin, luteolin and kaempferol. Flavonoids are associated with a broad spectrum of health-promoting effects and are an indispensable component in a variety of nutraceutical, pharmaceutical, medicinal and cosmetic applications. This is because of their antioxidative, anti-inflammatory, anti-mutagenic and anti-carcinogenic properties coupled with their capacity to modulate key cellular enzyme functions (Panche *et al*., 2016).

Flavonols are one of the important subgroups of flavonoids. Unlike the other phenolic content the higher flavonols amount was observed for Remila site with 12.84 mg QE/100 ml followed by the Meridef site with 11.23 mg QE/100 ml pulp juice. Lower values were recorded for Chegaga and Bouamoud sites (6.92 and 7.65 mg QE/100 ml), respectively.

Cactus pear fruits show a relatively high flavonol content, phytochemicals that contribute to antioxidant capacity. Intake of flavonols is found to be associated with a wide range of health benefits, which includes antioxidant potential, and reduced risk of vascular disease (Panche *et al*., 2016). These compounds are more efficient antioxidants than vitamins, since flavonoids, and phenolic compounds in general, are able to delay the prooxidative effects of proteins, DNA and lipids through the generation of stable radicals (Feugang *et al*., 2006).

Ascorbic acid content was checked for cactus pears of the study sites. Concentrations ranged from 69.42 to 78.96 mg/100 ml pulp juice. The highest concentration was registered for pulp of cactus pears from Remila site followed by that of Meridef site. Chegaga and Bouamoud sites had lower values of ascorbic acid content (Table 4). These values are higher than those published by Stintzing *et al*. (2005), Díaz-Medina *et al*. (2007), and Albano *et al*. (2015). However, they are lower than those reported by Abdel-Hameed *et al*. (2014) and Jiménez-Aguilar *et al*. (2015).

Ascorbic acid is an electron donor and acts as a cofactor for fifteen mammalian enzymes (Padayatty and Levine 2016). It is the most potent concentration dependent water-soluble antioxidant in the body and the principal antioxidant that quenches aqueous peroxyl radicals and lipid peroxidation products in plasma *ex vivo* (Frei *et al*., 1990). The recommended dietary allowance (RDA) for vitamin C is 75 mg for women and 90 mg for men.

Compared to other phytochemical compounds total carotenoids amounts were lower for all the studied sites. Their concentration ranged from 93.15 to 123.75 µg/100 g fresh pulp. The data are lower than those reported by Chougui *et al*. (2013) for orange cactus pear variety grown in Bejaia (2.65 mg/100 g), and those reported by Kuti (2004) for Texan (USA) yellow-skinned cactus pear varieties with orange juice (0.6~1.2 mg/100 g). However they are much higher than those reported by El Kharrassi *et al*. (2016) for Moroccan verities (5.4~20.8 µg/L) and Fernández-López *et al*. (2010) for the Murcia (Spain) *Opuntia ficus-indica* fruit (2.58 µg/100 g fresh fruit).

Carotenoid content in cactus pear fruits is not very high (Fernández-López *et al*., 2010). The carotenoid composition of foods are affected by factors such as cultivar or variety, part of the plant, stage of maturity, climate or geographic site of production, harvesting and postharvest handling, processing and storage (Rodriguez-Amaya, 1993). Carotenoids are not evenly distributed in the food itself. In this study the total carotenoid content was checked in the fruit pulp. Various investigators found that carotenoids are usually more concentrated in the peel than in the pulp of fruits and fruit vegetables. Greater exposure to sunlight and elevated temperature heighten carotenoid biosynthesis in fruits (Rodríguez-Amaya 2001). However, according to other reports, total carotenoid were significantly higher in the winter than in the summer in the harvested fruit, which appears compatible with greater destruction of leaf carotenoids on exposure to higher temperature and greater sunlight (Young and Britton 1990).

Betalains quantification shows that the pulp juice of fruits collected from Remila site contains the highest content of total betalains (16.65 mg) followed by the Meridef site with 14.47 mg/100 ml. The lowest betalains contents were recorded for Chegaga and Bouamoud sites with 10.06 mg and 10.76 mg/100 ml pulp juice, respectively. Almost the same order in terms of betacyanins and betaxanthins concentrations in the fruit juice of the study area was observed. Remila and Meridef sites exhibited a significant higher content in betacyanins (2.49 and 2.43 mg) and betaxanthins (14.16 and 12.05 mg), respectively. In all the cases, the betacyanins/betaxanthins ratio was close to 1:5 (w/w).

Accordingly, Castellano-Santiago & Yahia (2008) reported that the yellow/orange Mexican cultivars/lines had higher levels of betaxanthins than betacyanin content. They concluded that the variations in the content of the yellow-orange betaxanthins and the red-violet betacyanins as well as the variations in the levels of certain structurally different betacyanins are responsible for the cactus pear fruit color.

Cactus pears from cv. Orange exhibit a higher ratio of red to yellow pigments than the yellow fruits (Stintzing *et al*., 2005). Findings by Tesoriere *et al*. (2005) provide evidence that human red blood cells incorporate dietary betalains, which may protect the cells and avoid oxidative hemolysis. Indicaxanthin and betanin have been shown also to bind to human low-density lipoproteins *in vitro* (Tesoriere *et al*., 2003) and *in vivo* (Tesoriere *et al*., 2004), increasing their resistance to oxidation. Stintzing *et al*. (2005) indicated that the contribution of betalains on *in vitro* antioxidant activity of cactus pear is even much greater than that provided by ascorbic acid. In Algeria, the orange fruits cultivar is the most abundant and the most consumed (Chougui *et al*. 2013). Orange fruits are especially beneficial to health because they are provided with yellow pigment betaxanthin. The latter modulates the macrophage activation process leading to anti-inflammatory mediators thus contributing to the control of metabolic responses during inflammation (Allegra *et al.,* 2014).

Antioxidant activity

The radical scavenging activity as considered by the SC_{50} parameter, which is the concentration needed for inhibition 50% of DPPH radicals, varied from 33.45 to 49.62 µl pulp juice/ml. The Best SC_{50} was observed for the pulp juice of cactus pears from Remila and Meridef sites (33.45 and 33.49 µl/ml) corresponding to 28.32 and 28.26 mg AAE/100 ml, respectively; followed by Chegaga site with 41.68 µl/ml, corresponding to 22.75 mg AAE/100 ml, while pulp juice of cactus pears from Bouamoud site had the lowest radical scavenging activity (49.62 µl/ml) equivalent to 19.09 mg AAE/100 ml (Table 5).

Table 5. Free radical scavenging activity, total antioxidant capacity and reducing power of fruit pulp juice of *Opuntia ficus indica* grown in different sites of Souk-Ahras.

Site	DPPH free radical scavenging activity SC_{50}		Reducing power EC_{50}		Total antioxidant capacity
	(µl/ml)	(mg AAE/100 ml juice)	(µl/ml)	(mg AAE/100 ml juice)	(mg AAE/100 ml juice)
Bouamoud	49.62±2.03^{d}	19.09±0.76^{c}	59.76±0.63^{d}	28.53±0.30^{c}	1444.00±52.00^{a,b}
Chegaga	41.68±2.41^{c}	22.75±1.36^{b}	40.59±4.36^{b}	42.35±4.83^{a}	1340.00±28.00^{b}
Meridef	33.49±0.94^{b}	28.26±0.78^{a}	49.23±0.33^{c}	34.64±0.23^{b}	1472.00±94.06^{a}
Remila	33.45±1.53^{b}	28.32±1.31^{a}	48.70±2.01^{c}	35.05±1.46^{b}	1346.67±32.33^{b}
	09.46±0.85^{a*}	-	17.05±1.22^{a*}	-	-

Values were expressed as mean of triplicate determinations ± standard deviation.
Values having same letter within the column did not differ significantly from each other according to LSD test at $p<0.05$.
SC_{50} (µl/ml): effective concentration at which 50% of DPPH radicals (0.2 mM) are scavenged.
EC_{50} (µl/ml), effective concentration at which the absorbance is 0.5.
*Ascorbic acid concentration (µg/ml).

Chougui *et al*. (2013) reported values of SC_{50} varying from 6.33 to 4.31 µl juicy pulp extract/µg DPPH with best values being recorded for orange cultivar. Abdel-Hameed *et al*. (2014) reported the SC_{50} value of 30.96 µl for yellow pulps that scavenged 50% of DPPH radicals (0.1 mM). The activity exhibited by the juicy pulp extracts, seems to be mainly exerted by the water-soluble components betalains and vitamin C; the linear and significant positive correlations r = 0.83 and r = 0.77, respectively, were noticed between the amount of these compounds and SC_{50} values. Moreover, flavonols fraction of phenols content had contributed (r = 0.81) to the overall radical scavenging activity (Table 6).

These results agree with Stintzing *et al*. (2005) who found that betacyanins, betaxanthins and vitamin C, as well as the phenolic content, all contributed to cactus pear pulp antioxidant activity. The good antioxidant performance of the orange variety could be related to its high betaxanthin content (Albano *et al*., 2015).

Table 6. Matrix correlation of evaluated pulp juice parameters of *Opuntia ficus indica* growing in different sites of Souk-Ahras.

Variables	Polyphenols	Flavonoids	Flavonols	Betacyanins	Betaxanthins	Total Betalains	Vitamin C	Total Carotenoids	DPPH SC_{50}	Reducing Power EC_{50}	TAA-Capacity
Polyphenols	**1**										
Flavonoids	**0,812***	**1**									
Flavonols	-0,281	0,075	**1**								
Betacyanins	-0,239	0,214	0,853	**1**							
betaxanthins	-0,378	0,029	0,966	**0,892***	**1**						
Total Betalains	-0,361	0,060	0,962	**0,923***	**0,997***	**1**					
Vitamin C	-0,243	0,104	0,955	**0,873***	**0,963***	**0,963***	**1**				
Total Carotenoids	**0,663***	**0,753***	-0,136	0,055	-0,200	-0,160	-0,209	**1**			
DPPH SC_{50}	0,045	0,473	0,814	**0,805***	**0,829***	**0,837***	**0,777***	0,241	**1**		
Reducing Power EC_{50}	0,466	0,438	-0,137	-0,148	-0,157	-0,158	-0,279	0,517	0,287	**1**	
TAA-Capacity	0,028	0,307	-0,004	0,081	-0,023	-0,006	0,074	0,049	-0,025	-0,454	**1**

*Significant correlation values ($p<0.05$).
TAA-Capacity: Total Antioxidant Capacity.

The reducing power activity was assessed for the different study sites. In the investigated amount range, the reducing power increased proportionally with concentration. The reducing power of each pulp juice sample was estimated by the EC_{50} parameter that is the effective concentration at which the absorbance is 0.5. Cactus pear juice from Chegaga site showed the highest reducing power (40.59 µl/ml) corresponding to 42.35 mg AAE/100 ml, followed by the juice from Remila and Meridef sites with the values 48.70 and 49.23 µl/ml, corresponding to 35.05 and 34.64 mg AAE/100 ml, respectively.

The weakest reducing power was registered for pulp juice from Bouamoud site with the value of 59.76 µl/ml, equivalent to 28.53 mg AAE/100 ml (Table 5). These values are higher than those reported by Chougui *et al*. (2013) for red and orange cactus cultivars from Bejaia region (Algeria) that showed highest reducing power at more than 19 mg AAE/100 g; However, still lower than those reported by Abdel-Hameed *et al*. (2014) for pulps of red and yellow cultivars from Saudi Arabia. The correlation matrix suggest that the reducing power is partially influenced to some extent by the content in polyphenols, flavonoids and carotenoids with a positive correlation r = 0.46, 0.43 and 0.51, respectively (Table 6).

Total antioxidant capacity was investigated through the phosphomolybdenum test. This assay depends on the reduction of Mo (VI) to Mo (V) by the antioxidants and subsequent formation of a green phosphate/Mo (V) complex at acid pH. Total antioxidant capacity was significantly higher in pulp juices of cactus pears from Meridef and Bouamoud sites with the values 1,472 and 1,444 mg AAE/100 ml, respectively. Slightly weaker values were recorded for pulp juices from Remila and Chegaga sites, with 1,346.37 and 1,340 mg AAE/100 ml, respectively (Table 5). The total antioxidant capacity was negatively correlated to reducing power r = -0.45 (Table 6).

Eventhough no clear correlation with the individual phytochemical content among cactus pears, the high antioxidant capacity observed in this study, may be due to the combination of the appropriate proportions of antioxidants producing synergistic effect. A multivariate PCA was applied to data obtained for all the study sites in order to establish a more simplified view of the relationship among the analyzed components (Figure 2).

The first factor that explains the higher percentage of variance (51.01%) is strongly related to DPPH radical scavenging activity (SC_{50}), and is concomitant with total betalains, betacyanins, betaxanthins, ascorbic acid, and flavonols content. The second factor is related to reducing power (RP EC_{50}) and is associated with total phenolics, flavonoids and total carotenoids content. From the representation of the scores plot on the first and second factor it can be observed that the samples of cactus pears of each study site are well separated from each other.

From the result, it appears that cactus pears from Remila site are richer in betalains, flavonols and vitamin C phytochemicals, which contributed markedly to the efficiency of radical scavenging activity as estimated by DPPH test. While cactus pears from Meridef and Chegaga sites are richer in polyphenols, flavonoids and total carotenoids and bear a higher reducing power activity; whereas, Bouamoud site exhibited the least phytochemical content and the lowest radical scavenging and reducing power activities.

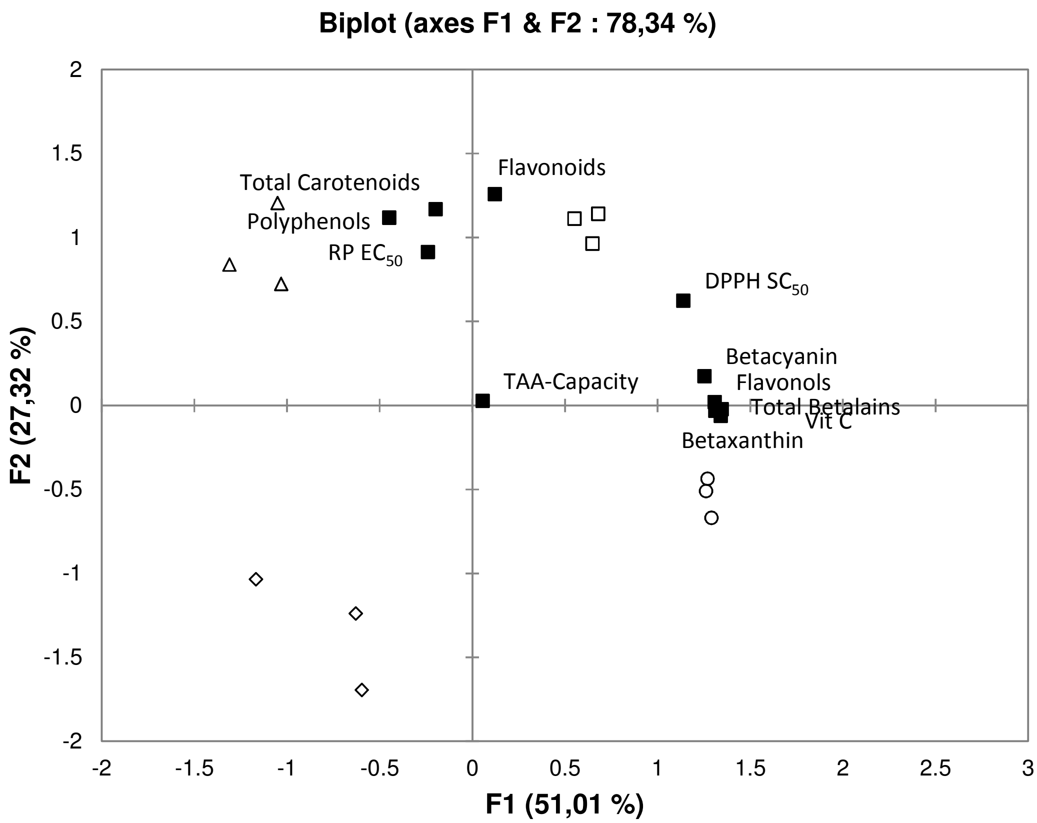

Figure 2. Score and loading plot of principal component analysis applied to the data set of phytochemical content and antioxidant activities for pulp juice of cactus pears from Souk-Ahras. Data were presented as filled squares, while the origin sites were presented with empty symbols: Meridef (squares), Remila (circles), Chegaga (triangles) and Bouamoud (diamond).

CONCLUSIONS

In the present study, we observed that cactus pears of Souk-Ahras differentiated significantly following the growing site. Fruits from Remila site presented appreciable biometric parameters, including size, weight and seed content. The fruits of the study sites were characterized by their relative high pulp percentage and TSS and low acidity. The multivariate analysis implied that the content of phytochemical classes and the level of antioxidant activity were markedly influenced by the growing site. Cactus pear fruits growing in Remila site had a significant higher content of flavonols, vitamin C and betalains, while fruits growing in Meridef and Chegaga sites were significantly higher in phenolics, flavonoids and carotenoids content. The consumption of cactus pears represents an important contribution to the intake of antioxidants. The results obtained in this study may contribute to the sensitization of local authorities and socioeconomic operators on the potential of the *Opuntia* sector in the region of Souk-Ahras. Despite the relative abundance of phytochemicals content of the cactus pear fruits, cactus pear growers should pay more attention to cultural practices in order to improve fruit size, especially in the perspective of accessing the international market.

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REFERENCES

Abdel-Hameed, El-S.S., Nagaty, M.A., Salman, M.S,, Bazaid, S.A. 2014. Phytochemicals, nutritionals and antioxidant properties of two prickly pear cactus cultivars (Opuntia ficus indica Mill.) growing in Taif, KSA. Food Chemistry 160: 31–38. doi: 10.1016/j.foodchem.2014.03.060.

Albano, C., Negro, C., Tommasi, N., Gerardi, C., Mita, G., Miceli, A., De Bellis, L., Blando, F, 2015. Betalains, Phenols and Antioxidant Capacity in Cactus Pear [Opuntia ficus-indica (L.) Mill.] Fruits from Apulia (South Italy) Genotypes. Antioxidants 4: 269-280. doi: 10.3390/antiox4020269.

Allegra M., D'Acquisto F., Tesoriere L., Attanzio A., Livrea M.A. 2014. Pro-oxidant activity of indicaxanthin from *Opuntia ficus indica* modulates arachidonate metabolism and prostaglandin synthesis through lipid peroxide production in LPS-stimulated RAW 264.7 macrophages. Redox Biology 2: 892–900.

Bouzoubaâ, Z., Essoukrati, Y., Tahrouch, S., Hatimi, A., Gharby, S., Harhar, H. 2016. Phytochemical study of prickly pear from southern Morocco. Journal of the Saudi Society of Agricultural Sciences 15: 155–161. Doi: doi.org/10.1016/j.jssas.2014.09.002.

Cámara, M.M., Diez, C., Torija, M.E., Cano, M.P. 1994. HPLC determination of organic acids in pineapple juices and nectars. Zeitschrift für Lebensmittel-Untersuchung und-Forschung 198: 52-56. doi: 10.1007/BF01195284.

Castellanos-Santiago, E. and Yahia, E.M. 2008. Identification and Quantification of Betalains from the Fruits of 10 Mexican Prickly Pear Cultivars by High-Performance Liquid Chromatography and Electrospray Ionization Mass Spectrometry. Journal Agricultural and Food Chemistry 56: 5758–5764. doi: 10.1021/jf800362t.

Chougui, N., Louaileche, H., Mohedeb, S., Mouloudj, Y., Hammoui, Y., Tamendjari, A. 2013. Physico-chemical characterisation and antioxidant activity of some Opuntia ficus-indica varieties grown in North Algeria. African Journal of Biotechnology 12(3): 299-307. doi:10.5897/AJB12.1946.

Deldicque, L., Van Proeyen, K., Ramaekers, M., Pischel, I., Sievers, H., Hespel, P. 2013. Additive insulinogenic action of Opuntia ficus-indica cladode and fruit skin extract and leucine after exercise in healthy males. Journal of the International Society of Sports Nutrition 10(45): 1-6. doi: 10.1186/1550-2783-10-45.

Díaz, M.E.M., Rodríguez, R.E.M., Díaz, R.C., 2007. Chemical characterization of Opuntia dillenii and Opuntia ficus indica fruits. Food Chemistry 103 : 38–45. doi: 10.1016/j.foodchem.2006.06.064.

Díaz, M. del S.S., Barba de la Rosa, A.-P., Héliès-Toussaint, C., Guéraud, F., Nègre-Salvayre, A. 2017. Opuntia spp.: Characterization and Benefits in Chronic Diseases. Oxidative Medicine and Cellular Longevity 2017: 1-17. doi: 10.1155/2017/8634249.

El Kharrassi, Y., Mazri, M.A., Benyahia, H., Benaouda, H., Nasser, B., El Mzouri, E.H., 2016. Fruit and juice characteristics of 30 accessions of two cactus pear species (Opuntia ficus indica and Opuntia megacantha) from different regions of Morocco. LWT-Food Science and Technology 65: 610-617. doi: 10.1016/j.lwt.2015.08.044.

Farrington, A.M., Jagota, N., Slater, J.M., 1994. Simple Solid Wire Microdisc Electrodes for the Determination of Vitamin C in Fruit Juices. Analyst 119: 233-238. doi: 10.1039/AN9941900233.

Felker, P., Soulier C., Leguizamon G. & Ochoaw J., 2002. A comparison of the fruit parameters of 12 Opuntia clones grown in Argentina and the United States. Journal of Arid Environments 52: 361–370. doi:10.1006/jare.2002.1001.

Felker, P., Rodriguez, S. del C., Casoliba, R.M., Filippini, R., Medina, D., Zapata, R. 2005. Comparison of Opuntia ficus indica varieties of Mexican and Argentine origin for fruit yield and quality in Argentina. Journal of Arid Environments 60: 405–422. doi: 10.1016/j.jaridenv.2004.06.003.

Fernández-López, J.A., Almela, L., Obón, J.M., Castellar, R., 2010. Determination of Antioxidant Constituents in Cactus Pear Fruits. Plant Foods for Human Nutrition 65: 253–259. doi: 10.1007/s11130-010-0189-x.

Feugang, J.M., Konarski, P., Zou, D., Stintzing, F.C., Zou, C., 2006. Nutritional and medicinal use of cactus pear (Opuntia spp.) cladodes and fruits. Frontiers in Bioscice 11: 2574–2589. doi: 10.2741/1992.

Frei, B., Stocker, R., England, L., Ames, B.N., 1990. Ascorbate: the most effective antioxidant in human blood plasma. *Advances in* Experimental Medicine and Biology 264: 155–63. doi: 10.1007/978-1-4684-5730-8_24.

Inglese, P., Barbera, G., La Mantia, T., 1995. Research strategies for the improvement of cactus pear (Opuntia ficus-indica) fruit quality and production. Journal of Arid Environments 29: 455–468. doi: 10.1016/S0140-1963(95)80018-2.

Jacob, J.K., Hakimuddin, F., Paliyath, G., Fisher, H., 2008. Antioxidant and antiproliferative activity of polyphenols in novel high-polyphenol grape lines. Food Research International 41: 419–428. doi: 10.1016/j.foodres.2008.02.009.

Jiménez-Aguilar, D.M., López-Martínez, J.M., Hernández-Brenes, C., Gutiérrez-Uribe, J.A., Welti-Chanes, J., 2015. Dietary fiber, phytochemical composition and antioxidant activity of Mexican commercial varieties of cactus pear. Journal of Food Composition and Analysis 41: 66–73. doi: 10.1016/j.jfca.2015.01.017.

Kuti, J.O., 2004. Antioxidant compounds from four Opuntia cactus pear fruit varieties. Food Chemistry 85: 527–533. doi: 10.1016/S0308-8146(03)00184-5.

Lampila, P., Lieshout, M., Germmen, B., Lähteenmäki, L., 2009. Consumer attitudes towards enhanced flavonoid content in fruit. Food Research International 42: 122–129. doi: 10.1016/j.foodres.2008.09.002.

Oyaizu, M., 1986. Studies on products of browning reactions: Antioxidative activities of browning reaction prepared from glucosamine. Japanese Journal of Nutrition 44: 307–315. doi: 10.5264/eiyogakuzashi.44.307.

Padayatty, S.J., Levine, M., 2016. Vitamin C physiology: the known and the unknown and Goldilocks. Oral Diseases 22: 463–493. doi: 10.1111/odi.12446.

Panche, A.N., Diwan, A.D., Chandra, S.R., 2016. Flavonoids: an overview. Journal of Nutritional Science 5(e47): 1-15. doi: 10.1017/jns.2016.41.

Parish, J. & Felker, P., 1997. Fruit quality and production of cactus pear (Opuntia spp.) fruit clones selected for increased frost hardiness. Journal of Arid Environments 37: 123 – 143. doi: 10.1006/jare.1997.0261.

Rodriguez-Amaya, D.B., 1993. Nature and distribution of carotenoids in foods. In Charalambous G. (ed.), Shelf life studies of foods and beverages. Chemical, biological, physical and nutritional aspects, (pp. 547-589). Elsevier Science Publishers, Amsterdam.

Rodríguez-Amaya, D.B., 2001. A guide to carotenoid analysis in foods. ILSI Human Nutrition Institute Press, Washington D.

Saénz, C., Tapia, S., Chávez, J., Robert, P. 2009. Microencapsulation by spray drying of bioactive compounds from cactus pear (Opuntia ficus-indica). Food Chemistry 114: 616–622. doi : 10.1016/j.foodchem.2008.09.095.

Seltzer, P., Lasserre, A., Grandjean, A., Auberty, R. 1946. Le climat de l'Algérie. Travaux de l'Institut de météorologie et de physique du globe de l'Algérie, (pp. 219). Editeur La Typo-litho" & J. Carbonel.

Stintzing, F.C., Herbach, K.M., Mosshammer, M.R., Carle, R., YI, W., Sellappan, S., Akoh, C.C., Bunch, R., Felker, P. 2005. Color, Betalain Pattern, and Antioxidant Properties of Cactus Pear (Opuntia spp.) Clones. Journal of Agricultural and Food Chemistry 53: 442-451. doi: 10.1021/jf048751y.

Stintzing, F.C., Schieber, A., Carle, R. 2003. Evaluation of colour properties and chemical quality parameters of cactus juices. European Food Research and Technology 216: 303–311. doi: 10.1007/s00217-002-0657-0.

Temagoult, A., Zitouni, B., Noui, Y. 2017. Algerian Prickly Pear (Opuntia ficus-indica L.) Physicochemical Characteristics. International Journal of Scientific Research & Engineering Technology 5: 14-17. In: International Conference on Green Energy and Environmental Engineering (GEEE-2017), ISSN 2356-5608.

Tesoriere, L., Allegra, M., Butera, D., Livrea, M.A. 2004. Absorption, excretion, and distribution of dietary antioxidant betalains in LDLs: Potential health effects of betalains in humans. American Journal of Clinical Nutrition 80: 941–945. doi: 10.1093/ajcn/80.4.941.

Tesoriere, L., Butera, D., D'Arpa, D., Di Gaudio, F., Allegra, M., Gentile, C., Livrea, M.A. 2003. Increased resistance to oxidation of betalain-enriched human low density lipoproteins. Free Radical Research 37: 689–696. doi: 10.1080/1071576031000097490.

Tesoriere, L., Butera, L., Allegra, M., Fazzari, M., Livrea, M.A. 2005. Distribution of betalain pigments in red blood cells after consumption of cactus pear fruits and increased resistance of the, cells to ex vivo induced oxidative hemolysis in humans. Journal of Agricultural and Food Chemistry 53: 1266–1270. doi: 10.1021/jf048134+.

Yahia, E.M. & Sáenz, C. 2011. Cactus pear (Opuntia species). in book: Postharvest biology and technology of tropical and subtropical fruits. Volume 2. Chapter 14, (pp. 290-329). Acai to citrus Publisher: Woodhead Publishing, England.

Young, A. & Britton, G. 1990. Carotenoids and stress. In Alscher R.G., Cumming J.R. (eds), Stress responses in plants: adaptation and acclimation mechanisms. Plant Biology, vol 12. Wiley-Liss Inc, New York.