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Physicochemical and nutritional composition, volatile profile and antioxidant activity differences in Spanish jujube fruits

J. Reche[a], F. Hernández[b], M.S. Almansa[a], Á.A. Carbonell-Barrachina[c], P. Legua[b], A. Amorós[a,*]

[a] Department of Applied Biology, Universidad Miguel Hernández de Elche, Ctra. Beniel, km 3.2, 03312, Orihuela, Alicante, Spain
[b] Department of Plant Science and Microbiology, Universidad Miguel Hernández de Elche, Ctra. Beniel, Km 3.2, 03312, Orihuela, Alicante, Spain
[c] Department of Agro-Food Technology, Universidad Miguel Hernández de Elche, Ctra. Beniel, Km 3.2, 03312, Orihuela, Alicante, Spain

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A B S T R A C T

In Spain, jujube can be considered a minor crop; thus, its cultivars have not yet been investigated in depth. The objective of this study was to evaluate the composition, nutritional profile, and volatile composition of three cultivars of Spanish jujube fruits. The total antioxidant activity and phenolic compounds in peel and pulp of the fruits were studied. The obtained results proved that 'Isidro' cultivar presented the largest fruits, had the highest ascorbic acid content, with a good color intensity, total soluble solid content, and glucose and fructose. In addition, fruits of the 'Isidro' cultivar presented together with the 'Phoenix' ones, the highest protein content and juiciness. The cultivar 'Phoenix' behaved differently than the other two cultivars studied; its fruits had significantly lower contents of total phenols, flavonoids and flavonols and these compounds were mainly accumulated in the pulp. In the other two cultivars, bioactive compounds reached higher values and were mainly accumulated in their peels. 'Isidro' was the cultivar that presented maximum amounts of phenols and total flavonoids. Thus, both cultivars 'Isidro' and 'Phoenix' could be promising cultivars to be used in future breeding programs, to obtain fruit with high content of bioactive compounds as well as interesting organoleptic properties.

1. Introduction

Jujube (*Ziziphus jujuba* Mill.) tree has its origin in China, where it is popular for its high nutritional value and medicinal uses. Different parts of the jujube plant are used as pharmacological agents (Jiang, Huang, Chen, & Lin, 2007). It seems that there are significant differences among the contents of bioactive compounds in peel and pulp of the jujube fruit (Xue, Feng, Cao, Cao, & Jiang, 2009; Zhang, Jiang, Yue, Ye & Ren, 2010); however, the studies of Xue et al. (2009) and Zhang, Jiang, Ye, Ye, and Ren (2010) were conducted using Chinese jujube cultivars. This same trend has been previously reported in other fruits, such as nectarines, peaches, plums (Tomás-Barberán et al., 2001), orange, banana, and tangerine (Faller & Fialho, 2010).

In Western countries, there is great interest in antioxidant rich diets, ingredients and/or raw materials. There is a current trend towards replacement of chemical additives with "natural" antioxidants (Cheng, Liu, Zhang, Chen& Wang, 2018). Jujubes can be presented as an alternative additive in complex food, due to their high contents in antioxidants, phenolic compounds, potassium, iron, and vitamin C. Despite being a marginal crop in Spain and its low consumption, its demand is increasing (Hernández et al., 2016; Wojdyło, Carbonell-Barrachina, Legua, & Hernández, 2016).

However, there is scarce information on the phytochemical composition of Spanish jujubes and the effect of the cultivar factor on key nutritional (Hernández et al., 2016; Wojdyło et al., 2016). Therefore, the objective of this work was to study, for the first time, three cultivars of Spanish jujube and to establish their composition, the nutritional value and volatile profile, as well as the antioxidant capacity of the peel and the pulp of these fruits. This will be the first study describing the differences in the antioxidant capacity of the peel and pulp from Spanish jujube cultivars.

2. Materials and methods

2.1. Experimental conditions and plant material

The experiment was carried out in a commercial farm with 21-years-old jujube trees (latitude 38º10′22, 29″N x longitude 0º51′36, 138″W, 19 m above sea level) in Albatera (Alicante) from Spain. Trees were trained as a vase and spaced 4 m × 4 m. The jujube fruits under

* Corresponding author.
E-mail address: aamoros@umh.es (A. Amorós).

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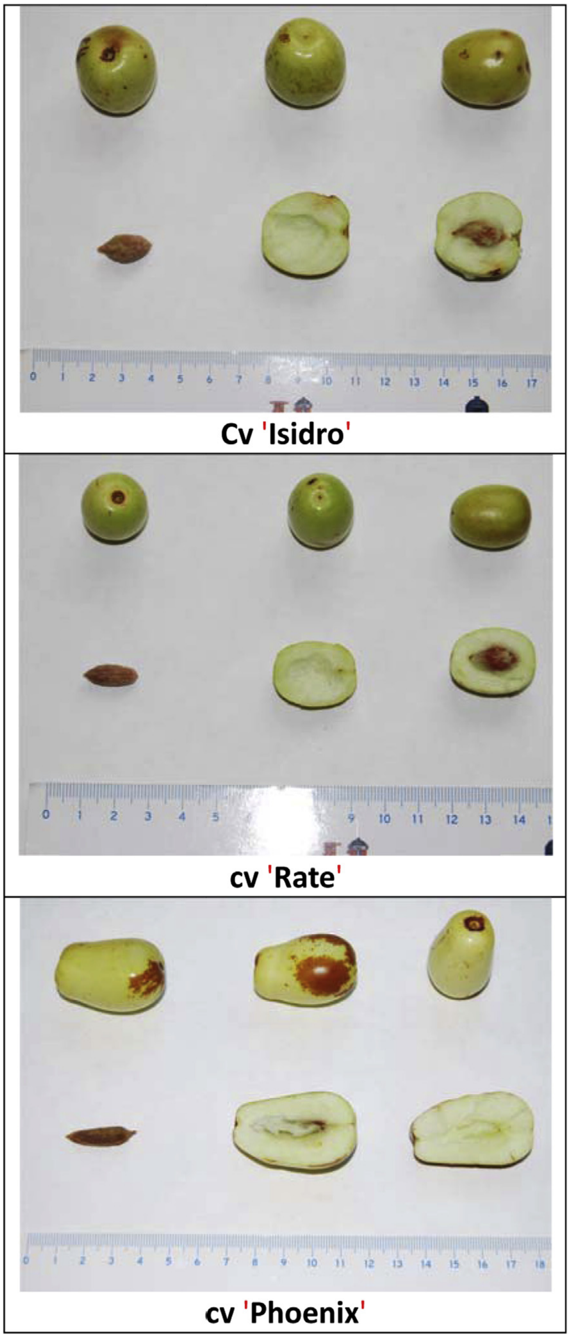

Fig. 1. Photographs of jujube fruits and stones of 'Isidro', 'Rate' and 'Phoenix' cultivars.

study belonged to three cultivars: (i) 'Phoenix', (ii) 'Isidro', and (iii) 'Rate' (Fig. 1). One hundred fruits from four trees (25 fruits per tree) of each cultivar were hand-harvested at commercial maturity (above 15 ºBrix). Fruits were immediately transported, under ventilated conditions, to the laboratory. Thirty fruits were taken for the physical analyses, while the other seventy fruits were used for the analyses of the biochemical parameters.

2.2. Physical parameters

The following physical parameters were measured in 30 fruits: equatorial diameter and fruit length (mm) using a digital caliper (model CD-15 DC; Mitutoyo (UK) Ltd, Telford, UK); fruit weight (g) was measured using a digital balance (model BL-600; Sartorius, Madrid, Spain). Instrumental color was on the surface of the jujubes at two opposite points of the equatorial zone. Color was assessed according to the *Commission Internationale de l'Eclairage* (CIE*Lab*) and expressed as L^*, a^*, b^*, chroma, and Hue angle, with a spectrophotometer Minolta C-300 Chroma Meter (Minolta Corp., Osaka, Japan) coupled to a Minolta DP-301 data processor. Then, the pulp was separated from the stone of each fruit, and in each stone the equatorial diameter, length and weight (g) were measured.

2.3. Chemical and biochemical parameters

All parameters included in this section were measured in triplicate.

Chlorophylls *a and b* were extracted from each sample using 85% acetone in a ratio 1:2 (w:v) (AOAC, 1990). The sample was crushed with sea sand in a mortar and, then, centrifuged at 12,000 rpm for 20 min at 4 °C. Absorbance of the supernatant was read at 664 and 647 nm, using a Helios Gamma spectrophotometer (model, UVG 1002E; Helios, Cambridge, UK). Results were expressed as mg 100 g^{-1} fresh weight (fw). Total carotenoids were extracted according to Valero et al. (2011), with acetone and diethyl ether; 10% NaCl was used to promote the phases separation. The lipophilic phase was used to estimate the total carotenoids content, by reading the absorbance at 450 nm, and results were expressed as mg of β-carotene equivalent 100 g^{-1} fw, taking into account the $\varepsilon^{1\%}_{cm} = 2560$.

The total soluble solids (TSS) were measured in triplicate using an ATAGO N20 refractometer (Minato-Ku, Tokyo, Japan).

The sugars and organic acids were extracted according to Almansa, Hernández, Legua, Nicolás-Almansa, and Amorós (2016). Five grams of jujube fruit were homogenized with 6 mL of 50 mM Tris-acetate buffer pH 6.0, 10 mM CaCl, and 6 mL of ethyl acetate, centrifuged, and, then, the aqueous and organic phases were separated. The aqueous phases was utilized for the identification and quantification of sugar and organic acid profiles, according to Almansa et al. (2016). One mL of the hydrophilic extract was filtered through a 0.45 mm Millipore filter and, then, used for high-performance liquid chromatography (HPLC) (Hewlett–Packard HPLC series 1100; Hewlett-Packard, Wilmington, DE, USA). The elution buffer consisted of 0.1% phosphoric acid with a flow rate of 0.5 mL min^{-1}. Organic acid was isolated using a Supelco column (Supelcogel TM C-610H column 30 cm × 7.8 mm) and a precolumn Supelguard (5 cm × 4.6 mm; Supelco, Inc., Bellefonte, PA, USA), and absorbance was measured at 210 nm using a diode-array detector (DAD). These same HPLC conditions (elution buffer, flow rate, and column) were used for the analysis of sugars. Standards of organic acids (L-ascorbic, oxalic, citric, tartaric, malic, quinic, shikimic, succinic, and fumaric acids) and sugars (glucose, fructose, sucrose, and sorbitol) were obtained from Sigma (Poole, Dorset, UK). Calibration curves were used for the quantification of organic acids, showing good linearity ($r^2 = 0.999$). Results for both organic acids and sugars were expressed as g 100 g^{-1} fw.

The mineral contents were analyzed using an atomic absorption-emission spectrometer (Solaar 969, Unicam Ltd, Cambridge, UK), according to Hernández et al. (2016). Macro-elements (Ca, Mg, K, and Na) were expressed as g kg^{-1} dry weight (dw) and micro-elements (Fe, Cu, Mn, and Zn) were expressed as mg kg^{-1} dw.

The protein content was analyzed by the Bradford (1976) method using the Bio-Rad reactive. A standard curve of pure bovine serum albumin (BSA) was used for quantification according to Almansa et al. (2016). Results were expressed as mg 100 g^{-1} fw.

The dry matter was determined by drying at 55 °C until constant weight and was expressed as g 100 g^{-1} fw.

The volatile composition of the jujubes was studied by using headspace solid phase micro-extraction (HS-SPME), according to the methodology previously described by Hernández et al. (2016). A gas chromatograph (GC-MS) Shimadzu GC-17A (Shimadzu Corporation, Kyoto, Japan) coupled with a Shimadzu mass spectrometer detector GC-MS QP-5050A was used in the identification and semi-

Table 1
Morphology of jujube fruits as affected by cultivar.

Cultivar	Fruit			Stone		
	Weight (g)	Equatorial diameter (mm)	Length (mm)	Weight (g)	Equatorial diameter (mm)	Length (mm)
'Isidro'	12.7 ± 0.5 b[†]	29.08 ± 0.38 b	29.55 ± 0.36 b	0.62 ± 0.02 c	9.21 ± 0.13 c	17.80 ± 0.27 a
'Rate'	10.4 ± 2.5 ab	22.98 ± 0.22 a	28.43 ± 0.20 a	0.53 ± 0.01 b	8.12 ± 0.08 b	19.55 ± 0.27 b
'Phoenix'	9.1 ± 0.3 a	23.90 ± 0.49 a	39.59 ± 0.51 c	0.36 ± 0.02 a	6.81 ± 0.10 a	26.97 ± 0.36 c

[†] Different letters next to a value in each column within cultivar indicate significant differences according to Fisher's LSD test ($p < 0.05$) ($n = 30$).

quantification of the volatile compounds of jujube. A TRACSIL Meta X5 column (Telnolroma S. Co. Ltd., Barcelona, Spain) was used. The identification of each volatile compound was reached using three methods: 1) retention index of the problem compound, 2) GC-MS retention time of authentic standard, and 3) mass spectra of authentic chemical and database Wiley 229. Results were expressed as percentage of the total area represented by each compound.

Total phenols, flavonoids and flavonols and total antioxidant activity (TAA) were quantified in both pulp and peel of jujube, separated with a vegetable peeler.

Total phenolic compounds were quantified in the hydrophilic phase according to Singleton, Orthofer, and Lamuela-Raventos (1999), and using the Folin–Ciocalteu reagent. Briefly, 25 μL of hydrophilic extracts were mixed with 2.5 mL of Folin–Ciocalteau. The mixture was incubated for 2 min at room temperature and 2 mL of sodium carbonate (75 g L^{-1}) were added and vortexed. Finally, the mixture was incubated at 50 °C for 5 min and the absorbance was measured at 760 nm. A calibration curve was performed with gallic acid and results were expressed as mg GAE 100 g^{-1} fw.

Flavonoids and flavonols were extracted using 80% methanol from the peel and pulp of jujubes following the method of Zhuang (1992). The analysis of total flavonoids (mg rutin equivalents 100 g^{-1} fw) was performed by spectrophotometry after using 5% $NaNO_2$, 10% $AlCl_3$ and 1 M NaOH; absorbance was measured at 512 nm on a Helios Gamma spectrophotometer (model, UVG 1002E; Helios, Cambridge, UK). Quantification of total flavonols (mg rutin equivalents 100 g^{-1} fw) was done according to Kumaran, Kutty, Chatterji, Subrayan, and Mishra (2007), using $AlCl_3$ (2 mg mL^{-1}) and sodium acetate (50 mg mL^{-1}), and absorbance was measured at 440 nm.

For the antioxidant activity determination, a methanol extract was prepared as described by Wojdyło, Oszmiański, and Bielicki (2013). The free radical scavenging capacities were determined by three methods, $ABTS^{\bullet+}$ method (Re et al., 1999), $DPPH^{\bullet}$ radical (2,2-diphenyl-1-picrylhydrazyl) method (Brand-Williams, Cuvelier, & Berset, 1995), and FRAP (ferric reducing antioxidant power) method (Benzie & Strain, 1996). Calibration curves, in the range 0.5–5.0 mmol Trolox L^{-1}, were prepared for all three methods and showed good linearity ($R^2 = 0.998$). All antioxidant capacity analyses were run in triplicate, and results were expressed as mmol Trolox kg^{-1} fw.

The activity of H-TAA (hydrophilic-TAA) and L-TAA (lipophilic-TAA) of jujube fruit (peel or pulp) were determined in the aqueous and organic extracts used for the sugar and organic acid analyses. The reaction mixture contained 10 mM ABTS, 1 mM hydrogen peroxide, and 10 mM peroxidase in a total volume of 1 mL of 50 mM glycine-HCl buffer (pH 4.5) for H-TAA, or ethyl acetate for L-TAA. The reaction was monitored at 730 nm until a stable absorbance was obtained using a UNICAM Helios spectrophotometer (Cambridge, UK). After that, a suitable amount of jujube fruit extract was added and the observed decrease in absorbance was determined. A calibration curve was performed with Trolox as standard antioxidant for both H-TAA and L-TAA. (Arnao, Cano, & Acosta, 2001). The results were expressed as mg Trolox equivalent 100 g^{-1} fw.

2.4. Statistical analysis

Statistical analyses were performed using the software package SPSS 18.0 for Windows (SPSS Science, Chicago, IL, USA). A basic descriptive statistical analysis was followed by an analysis of variance test (ANOVA) for mean comparisons. The method used to discriminate among the means (multiple range test) was Fisher's LSD (Least Significant Difference) procedure at a 95.0% confidence level.

3. Results and discussion

3.1. Dimensions of jujube fruit

Data showed that there was a significant difference in size among cultivars (Table 1). The 'Isidro' cultivar had a higher equatorial diameter than the other two cultivars; however, the one with the longest length was 'Phoenix', followed by 'Isidro' and 'Rate'. These differences in the size led to differences in the weight, with the cultivar 'Isidro' (thickest fruit) having the highest value and 'Phoenix' (longest fruit) the lowest one.

The morphology of the stones followed the same trend than that described for the whole fruits (Table 1), with the highest equatorial diameter and weight being found in the 'Isidro' samples, and the longest length in the 'Phoenix' one; the 'Rate' fruits and stones had intermediate values of all morphologic parameters. The fruit weight and dimensions, including the edible portion and the non-edible stones, might be influenced by several factors, such a cultivar genotype, also depends on crop load (Gao et al., 2012b).

The pulp yield of the three cultivars was very similar and ranged between 94.90% ('Rate') and 96.04% ('Phoenix'), without significant statistical differences. These final weights, dimensions and pulp yields were within the normality ranges reported previously in other Spanish cultivars 'Jínjoles Grandes' and 'Jínjoles Medianos' (Almansa et al., 2016) and 'Grande de Albatera', 'MSI', 'PSI' and 'Dátil' (Hernández et al., 2016), but also in Korean (Choi, Ahn, Kozukue, Levin, & Friedman, 2011), Chinese (Gao et al., 2012b, 2011; Wang et al., 2012) and Ukrainian (Grygorieva, Abrahamová, Karnatowská, Bleha, & Brindza, 2014) jujube cultivars.

3.2. Color of jujube fruit

Color is one of the main quality parameters of agricultural products as it determines consumers' acceptance (Pathare, Opara, & Al-Said, 2013). The fruits modify their coloration due to the synthesis and degradation of pigments throughout their development and maturation (Almansa et al., 2016; Pék, Helyes, & Lugasi, 2010). All jujubes were harvested at their commercial maturity; however, slight changes in the reflection color were observed. The 'Rate' fruits presented a more reddish and darker color, due to their highest values of the green-red coordinate and the lowest of lightness. The other two cultivars, 'Isidro' and 'Phoenix' followed the same trend regarding external color, and had the highest values of lightness, blue-yellow coordinate, chroma, and Hue angle (Table 2).

The color parameter values reported here were very similar to those previously reported by other researchers for Spanish cultivars 'Jínjoles

Table 2
External color of jujube fruits as affected by cultivar.

Cultivar	L^*	a^*	b^*	C	H^o
'Isidro'	71.82 ± 0.37 b[†]	−0.36 ± 1.01 a	34.48 ± 1.02 b	35.30 ± 0.33 b	89.40 ± 2.76 b
'Rate'	63.54 ± 0.63 a	3.49 ± 0.41 b	30.94 ± 0.51 a	31.23 ± 0.47 a	83.27 ± 0.84 a
'Phoenix'	72.10 ± 0.32 b	−0.20 ± 0.21 a	34.86 ± 0.19 b	34.94 ± 0.19 b	90.29 ± 0.42 b

[†]Different letters next to a value in each column within cultivar indicate significant differences according to Fisher's LSD test ($p < 0.05$) ($n = 60$). L^*, lightness; a^*, green/red coordinate; b^*, blue/yellow coordinate; C, chroma; $H^°$, hue angle.

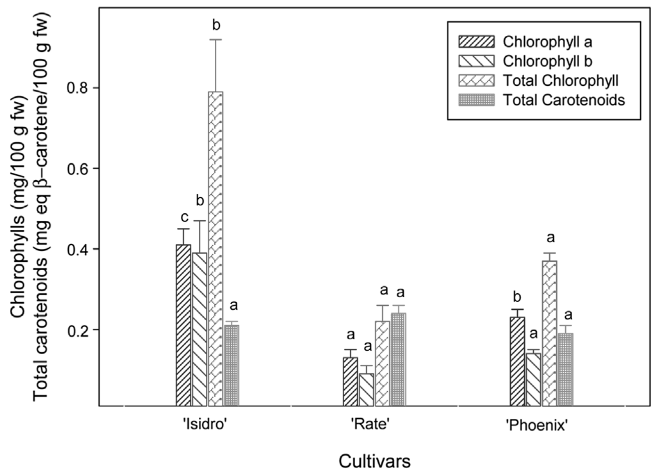

Fig. 2. Chlorophylls and carotenoids of jujube fruits 'Isidro', 'Rate' and 'Phoenix' cultivars. Different letters on top of bars indicate significant differences according to Fisher's LSD procedure at 95% confidence level ($n = 3$).

Grandes' and 'Jínjoles Medianos' (Almansa et al., 2016), 'Grande de Albatera' (Collado-González et al., 2014; Galindo et al., 2015) and even Chinese jujube cultivars (Wang et al., 2012).

Fruits from the cultivar 'Isidro' had the highest contents of chlorophylls, with the other two cultivars being grouped and with significantly smaller contents; this trend was valid for chlorophyll *a*, chlorophyll *b*, and total chlorophylls (Fig. 2). All three cultivars presented equivalent contents of total carotenoids (~0.20 mg eq β-carotene 100 g^{-1} fw).

3.3. Biochemical properties of jujube fruit

The 'Rate' cultivar had the lowest value of TSS, with the other two cultivars ('Isidro' and 'Phoenix') having significantly higher and statistically equivalent contents (Table 3). The reported values of TSS agreed with those found in Spanish 'Grande de Albatera' (Galindo et al., 2015) and Chinese cultivars (Gao et al., 2012b, 2011; Wu, Gao, Guo, Yu, & Wang, 2012).

The trend showed in TSS was corroborated by the sugars profiles, with fruits from the cultivars 'Isidro' and 'Phoenix' having significantly higher contents of glucose, fructose and consequently total sugars, than fruits of the cultivar 'Rate' (Table 3). These sugar contents were higher than those found in Spanish cultivars 'Jínjoles Grandes' and 'Jínjoles Medianos' (Almansa et al., 2016) and 'Grande de Albatera', 'MSI', 'PSI' and 'Dátil' (Hernández et al., 2016), and also Chinese cultivars (Gao et al., 2012a; b; Wu et al., 2012). The predominant sugars were sucrose and fructose, being glucose the less abundant sugar. This trend agreed with the one reported by Hernández et al. (2016). However, Wu et al. (2012) obtained a maximum content of fructose followed by glucose and almost no sucrose was found; while, Gao et al. (2012b) reported a predominance of glucose, with low contents of sucrose and fructose. These differences may be due to the fact that during the development of the jujubes, sucrose degradation occurs with a parallel increase in the synthesis of glucose and fructose, leading to slightly higher contents of fructose than glucose (Almansa et al., 2016). Therefore, small differences in the jujubes harvest time can lead to relatively high differences in the ratios of these three sugars.

The predominant organic acid was succinic (Table 3). Regarding the effect of the factor cultivar, the 'Isidro' fruits had the highest concentration of all identified acids and consequently of the total content; while, 'Phoenix' fruits had the lowest contents. This study showed higher ascorbic acid content, ranging from 0.33 ('Phoenix') to 0.65 g 100 g^{-1} ('Isidro'), as compared to other common fruits which are well-known for their high ascorbic acid content, such as strawberries (0.046 g 100 g^{-1}), oranges (0.031 g 100 g^{-1}) (Roberts & Gordon, 2003), and kiwi fruits (0.029–0.080 g 100 g^{-1}) (Nishiyama et al., 2004). These three jujube cultivars showed higher ascorbic acid values than Spanish, 'Grande de Albatera', 'MSI', 'PSI' and 'Dátil' (Hernández et al., 2016) and Chinese (Gao et al., 2012b) cultivars. Ascorbic acid is a powerful water soluble antioxidant, which plays an important role in the suppression of free radicals (Zhang et al., 2010). Therefore, it appears that these Spanish jujube cultivars are a good source of vitamin C in the diet.

The total sugars/total acids ratio is very interesting because it gives an idea of the potential fruit taste. The cultivars 'Isidro' and 'Rate' had practically the same value, around 5 (Table 3), which indicates that although they were sweet, they presented a moderate sour taste; on the other hand, the sweet taste clearly predominated in the cultivar 'Phoenix', with a ratio of 13.7. These values were similar those reported for Chinese and Spanish jujube cultivars with a similar maturity stage (Hernández et al., 2016; Wu et al., 2012), with the exception of the cultivar 'Phoenix', which had the highest value reported in the literature up to now and shows the high interest of this cultivar for fresh consumption.

The content of proteins in jujubes ranged from 0.37 ('Rate') to 0.61 mg g^{-1} fw ('Phoenix'), with statistically significant differences between these two cultivars (Table 4). These protein contents were within the normal range for Spanish cultivars 'Jínjoles Grandes' and 'Jínjoles Medianos' (Almansa et al., 2016), but were lower than those reported in Chinese (Li, Fan, Ding, & Ding, 2007) and Korean (Choi et al., 2012) cultivars.

The 'Rate' fruits had the highest dry matter content (Table 4), and can be considered as the less juicy ones. The range of this parameter found in the literature fluctuated between 15.3 and 25.7 g 100 g^{-1} fw in Spanish cultivars 'Grande de Albatera', 'MSI', 'PSI' and 'Dátil' (Hernández et al., 2016), and Chinese (Li et al., 2007) and Korean jujube fruits (Choi et al., 2012).

Jujubes were a good source of K and Ca (Hernández et al., 2016). Potassium was the predominant mineral with contents of ~5 g kg^{-1} dw (Table 4). This content was low compared to other Spanish cultivars, but higher than that reported in Chinese ones. The mean contents of Mg and Na were 1.3 and 1.0 g kg^{-1} dw, while those of Zn, Cu, Mn, and Fe were 4.7, 1.6, 3.1, and 11.9 mg kg^{-1} dw, respectively. These contents proved that the three studied jujube cultivars had interesting contents of both macro- and micro-elements.

No significant differences were found in any mineral element among the three cultivars studied, except for the Ca content, which was higher in the 'Isidro' cultivar. Several studies showed that Ca is an effective pressure lowering agent (Osborne et al., 1996); thus, a high Ca content

Table 3
Total soluble solids, TSS (ºBrix), sugars (g 100 g^{-1} fw) and organic acids (g 100 g^{-1} fw) profiles of jujube fruits as affected by cultivar.

Cultivar	TSS	Sucrose	Glucose	Fructose	Total sugars	Citric acid	Malic acid	Ascorbic acid	Succinic acid	Total acids	Sugars/acids
	(ºBrix)	(g 100 g^{-1} fw)									
'Isidro'	22.40 ± 0.81 b[†]	8.84 ± 0.31 a	6.06 ± 0.25 b	8.40 ± 0.33 b	23.30 ± 0.98 b	0.95 ± 0.02 c	0.13 ± 0.02 a	0.65 ± 0.01 c	2.87 ± 0.08 c	4.60 ± 0.13 c	5.06
'Rate'	17.73 ± 0.18 a	7.38 ± 0.33 a	4.81 ± 0.14 a	6.76 ± 0.28 a	18.95 ± 0.74 a	0.83 ± 0.05 b	0.17 ± 0.01 a	0.52 ± 0.02 b	1.94 ± 0.08 b	3.46 ± 0.16 b	5.48
'Phoenix'	24.07 ± 0.34 b	9.51 ± 0.14 a	6.02 ± 0.10 b	8.84 ± 0.09 b	24.37 ± 0.35 b	0.36 ± 0.04 a	0.26 ± 0.01 b	0.33 ± 0.06 a	0.82 ± 0.02 a	1.77 ± 1.00 a	13.77

[†]Different letters next to a value in each column within cultivar indicate significant differences according to Fisher's LSD test ($p < 0.05$) ($n = 3$).

Table 4
Protein (mg g^{-1} fresh weight, fw), dry matter (g 100 g^{-1} fw), and minerals content (g or mg kg^{-1} dry weight, dw) in jujube fruits as affected by cultivar.

Cultivar	Protein (mg g^{-1} fw)	Dry matter (g 100 g^{-1} fw)	Macro-elements (g kg^{-1} dw)				Micro-elements (mg kg^{-1} dw)			
			Potassium	Sodium	Magnesium	Calcium	Zinc	Copper	Manganese	Iron
'Isidro'	0.56 ± 0.032 b	22.17 ± 0.48 a	5.0 ± 0.5 a	0.02 ± 1.04 a	1.4 ± 0.08 a	4.0 ± 0.34 b	5.68 ± 1.13 a	1.95 ± 0.20 a	4.26 ± 0.17 a	7.53 ± 1.15 a
'Rate'	0.37 ± 0.014 a	25.50 ± 0.27 b	5.2 ± 0.1 a	2.00 ± 3.42 a	1.5 ± 0.29 a	2.6 ± 0.23 a	3.69 ± 0.69 a	0.79 ± 0.39 a	2.40 ± 0.29 a	10.95 ± 5.16 a
'Phoenix'	0.61 ± 0.017 b	22.15 ± 0.53 a	5.0 ± 0.4 a	1.10 ± 3.38 a	0.9 ± 0.15 a	2.0 ± 0.48 a	4.81 ± 1.95 a	1.95 ± 0.38 a	2.55 ± 0.38 a	17.32 ± 5.32 a

[†]Different letters next to a value in each column within cultivar indicate significant differences according to Fisher's LSD test ($p < 0.05$) ($n = 3$).

Table 5
Retention time (min), retention indexes (Exp. = experimental, and Lit. = literature) used for identification of the volatile compounds (μg kg^{-1}) found in jujube fruits as affected by cultivar.

Compound	Retention time (min)	Retention Indexes		ANOVA	Concentration (μg kg^{-1})		
		Exp.	Lit.		'Isidro'	'Rate'	'Phoenix'
Hexanal	7.24	1071	1075	*†	13.4 a‡	15.2 a	57.4 b
α-Phellandrene	9.42	1148	1158	**	126 b	134 b	8.0 a
β-Myrcene	9.55	1152	1156	NS	14.4	16.1	0.7
Heptanal	10.29	1177	1176	NS	7.1	6.7	14.4
Limonene	10.48	1183	1189	NS	40.4	39.4	16.6
β-Phellandrene	10.76	1192	1196	*	59.6 b	72.7 b	7.1 a
trans-2-Hexenal	11.30	1210	1211	**	53.8 a	64.8 a	484 b
p-Cymene	12.80	1260	1268	***	97.6 b	179 c	5.6 a
Octanal	13.50	1284	1288	NS	14.5	10.5	15.3
1-Octen-3-one	13.84	1295	1300	NS	5.7	7.2	4.2
2-Heptenal	14.49	1317	1320	**	13.5 a	18.5 a	81.6 b
6-Methyl-5-hepten-2-one	14.92	1331	1337	NS	19.5	19.2	10.6
Nonanal	16.58	1387	1389	*	119 b	45.1 a	129 b
2-Octenal	17.63	1422	1416	**	53.5 a	78.1 a	112 b
Benzaldehyde	20.39	1515	1508	***	257 a	1024 b	106 a
6-Methyl-1-heptanol	20.84	1530	1520	**	21.3 a	10.2 a	105 b
Hexanoic acid	30.05	1846	1843	*	210 b	215 b	64.4 a
Methyl hexadecanoate	39.54	2210	2217	NS	36.5	19.9	27.6
Total				**	1162 a	1976 b	1250 a

†NS = not significant at $p < 0.05$; *, **, and ***, significant at $p < 0.05$, 0.01, and 0.001, respectively.
‡Values (mean of 3 replications) in each row followed by the same letter, within the same factor, were not significantly different ($p < 0.05$).

can be beneficial for health. It is important to mention that the Fe content (7.5–17.3 g kg^{-1} dw) was typical of other Spanish cultivars but lower than those of other countries, such as China (Li et al., 2007). This fact can be due to the immobilization of Fe in Mediterranean soils with high pH and low content of organic matter (Hernández et al., 2016).

Eighteen volatile compounds were found in the volatile profile of jujube fruits of the three studied cultivars (Table 5). The low number of volatile compounds (18) seemed to indicate that the odor and aroma (perception of volatile compounds outside or inside the mouth, respectively) of these fruits is not one of their most relevant sensory attributes (Galindo et al., 2015). The predominant compound was benzaldehyde, followed by hexanoic acid, α-phellandrene, nonanal, and *p*-cymene. The quantitative analysis showed that 'Rate' jujubes had higher total concentration of volatile compounds that the other two cultivars. This trend was basically linked to the benzaldehyde content, which was about four time higher in the 'Rate' fruits than in the cultivar 'Isidro' and ten times than in 'Phoenix'. But, in general, the volatile profiles of fruits of the cultivars 'Isidro' and 'Rate' were similar between them and different from that of 'Phoenix' fruits. In this way, the most abundant compounds in 'Phoenix' were *trans*-2-hexenal, nonanal, 2-octenal, and benzaldehyde only occupied the fourth position in the abundance order.

3.4. Phenol compounds of jujube fruit

The cultivar factor significantly affected the total phenols content (TPC) in peel and pulp of jujubes (Table 6). The content of total phenols was 1.18 and 1.45 times higher in peel than in pulp in cultivars 'Isidro' and 'Rate', respectively; while, in the cultivar 'Phoenix' the content was 1.35 times higher in pulp than in peel. Xue et al. (2009) reported higher values in peel than pulp, with the only difference that the pulp contents were lower than the current ones. Zhang et al. (2010) reported a similar trend ($TPC_{peel} > TPC_{pulp}$) in two cultivars, but in another cultivar contents were similar ($TPC_{peel} \cong TPC_{pulp}$). This trend is not specific of jujubes and, for instance, Faller and Fialho (2010) found more soluble phenols in peel than in pulp in oranges, bananas, and tangerines, although in apples, papayas and mangos, they found equivalent contents. Therefore, it seems that phenolic compounds accumulate in epidermal plant tissues, because they have a potential role in the protection against ultraviolet rays, also because they act as attractants in the dispersion of fruits, and as defensive chemicals against pathogens and predators (Xue et al., 2009). Polymeric proanthocyanidins were the predominant compounds among the flavan-3-ols phenolics in Spanish jujubes cultivars (Wojdyło et al., 2016). The flavan-3-ols (monomer, dimer, and polymeric proanthocyanidins) represented 89–94% of the total phenolic contents (Collado-González et al., 2013; Wojdyło et al., 2016).

Table 6
Total phenols, flavonoids, flavonols contents of jujube fruits as affected by cultivar.

Cultivar		Total phenols (mg GAE 100 g^{-1} fw)	Total flavonoids (mg eq. rutin 100 g^{-1} fw)	Total flavonols (mg eq. rutin 100 g^{-1} fw)
'Isidro'	Peel	457 ± 20 e†	101 ± 3 d	6.18 ± 0.56 a
	Pulp	386 ± 17 d	19.82 ± 0.10 a	9.55 ± 0.32 b
'Rate'	Peel	453 ± 15 e	31.82 ± 1.77 b	ND
	Pulp	313 ± 21 c	70.60 ± 0.20 c	ND
'Phoenix'	Peel	178 ± 24 a	16.23 ± 0.35 a	6.18 ± 0.26 a
	Pulp	241 ± 21 b	29.23 ± 0.60 b	ND

†Different letters next to a value in each column within cultivar indicate significant differences according to Fisher's LSD test ($p < 0.05$) (n = 3).

This study showed higher TPC values in jujubes as compared to other fruits, such as persimmon (112 mg GAE 100 g^{-1} fw), pomegranates (147 mg GAE 100 g^{-1} fw), and apples (73.9 mg GAE 100 g^{-1} fw) (Fu et al., 2011). It is known that phenolic compounds have inhibitory effects on mutagenesis and carcinogenesis in humans when ingested in the form of a diet rich in fruits and vegetables (Tanaka, Kuei, & Nagashima, 1998). Therefore, our results have demonstrated the jujubes, and especially their peel, can be considered a good source of phenolic compounds and, therefore, be good for health. It is recommended to consume jujubes with peel, but after proper washing to ensure maximum intake of phenolic compounds.

The total flavonoids content (TFC) was 5 times higher in peel than in pulp in 'Isidro' jujubes (Table 6). However, TFC was almost double in the pulp than in peel in 'Rate' and 'Phoenix' fruits. The trend found in these last two cultivars ($TFC_{peel} < TFC_{pulp}$) is new; Zhang et al. (2010) found more flavonoids in peel than in jujube pulp, between 1.27 and

Table 7
Antioxidant activity of jujube fruits as affected by cultivar.

Cultivar		FRAP	$ABTS^{\bullet +}$	$DPPH^{\bullet}$	H-TAA	L-TAA
		(mmoles Trolox kg^{-1} fw)			(mg Trolox 100 g^{-1} fw)	
'Isidro'	Peel	165 ± 27 b[†]	43.63 ± 2.75 c	67.84 ± 1.22 c	562 ± 13 cd	363 ± 4 c
	Pulp	13.42 ± 1.61 a	37.37 ± 2.13 b	61.70 ± 0.56 b	606 ± 23 d	60.15 ± 1.68 b
'Rate'	Peel	372 ± 55 c	46.54 ± 0.22 c	70.10 ± 1.80 c	513 ± 10 c	427 ± 4 d
	Pulp	9.24 ± 0.07 a	32.99 ± 0.62 b	61.45 ± 2.58 b	595 ± 17 d	52.87 ± 0.81 b
'Phoenix'	Peel	18.93 ± 0.87 a	11.45 ± 0.59 a	11.05 ± 0.35 a	246 ± 28 a	367 ± 27 c
	Pulp	14.82 ± 0.95 a	10.08 ± 1.22 a	9.10 ± 0.31 a	374 ± 11 b	18.34 ± 1.33 a

[†]Different letters next to a value in each column within cultivar indicate significant differences according to Fisher's LSD test ($p < 0.05$) ($n = 3$).

4.7 times higher. The flavonoids found in *Ziziphus jujuba,* such as spinosin and its derivatives, are its main pharmacologically active compounds, and have been proven to be responsible for its sedative properties, protection against NMDA-induced neuronal cell damage, among other effects (Bai, Wang, Liu, & Li, 2010).

Total flavonols content (TFoC) were low and not detectable in several cases, especially in fruits of cultivars 'Rate' and 'Phoenix' (Table 6). Wojdyło et al. (2016) found that flavonols (6–11%) were the second major group of the total phenolic compounds in Spanish jujubes cultivars 'Grande de Albatera', 'MSI', 'PSI' and 'Dátil' (only 0–2.5% in the current study); these same authors reported that quercetin and its derivatives, especially the quercetin-3-*O*-rutinoside, were the predominant flavonols.

3.5. Antioxidant activity of jujube fruit

The total antioxidant activity was quantified in jujubes peel and pulp by three methods, FRAP, $ABTS^{\bullet +}$, and $DPPH^{\bullet}$. In general, the TAA was higher in peel than in pulp, although the differences were not statistically significant in the 'Phoenix' fruits (Table 7). These differences between cultivars were also found by other authors in jujubes (Choi et al., 2012; Gao et al., 2012b). Factors such as geographical source, genotype, but mainly maturity stage at harvest may account for the observed divergence. Besides, the differences between peel and pulp values were higher in the FRAP method, implying that this method is better suited to this specific fruit. Xue et al. (2009) and Zhang et al. (2010) also observed significantly higher FRAP values in peel than in pulp, but their differences were smaller (1.4–3.9 higher in peel than in pulp) as compared to results of the current experiment (12.2–40.2 higher in peel than in pulp).

The $ABTS^{\bullet +}$ and $DPPH^{\bullet}$ methods showed lower values in peel activity but higher in that of pulp, as compared to results of the FRAP method in 'Isidro' and 'Rate' cultivars, and again no differences were found in the 'Phoenix' cultivar (Table 7). The variation of antioxidant capacity between peel and pulp extracts may be due to differences in antioxidant compounds and their effectiveness (Robards, Prenzler, Tucker, Swatsitang, & Glover, 1999). The jujube peel of fruits of the cultivar 'Rate' showed the highest values of FRAP and L-TAA.

In the three studied cultivars, the peel had higher L-TAA than the pulp (Table 7), and showed significant differences among cultivars. The H-TAA activity was very high in both peel and pulp in the three cultivars, being in most of the cases significantly higher in the pulp than in the peel.

It is interesting to note that the cultivar 'Phoenix' presented significantly lower TPC contents, both in peel and pulp, than in the other two cultivars, leading to lower H-TAA and L-TAA, in peel and pulp, and, lower FRAP, $ABTS^{\bullet +}$, and $DPPH^{\bullet}$ activities.

A significant positive correlation was obtained among TPC and H-TAA in both peel and pulp ($R^2 = 0.8595$), while between TPC and L-TAA this correlation was only significant for the pulp ($R^2 = 0.873$). These positive correlations seemed to indicate that phenols are soluble in water, and that different compounds should be responsible for the antioxidant activity of the lipophilic fraction.

4. Conclusion

Physico-chemical and bioactive compounds in three cultivars of Spanish jujubes were compared. Due to the high antioxidant properties and the presence of important bioactive compounds in these fruits, it is considered that their cultivation has a great interest. The high resistance of this crop to arid conditions existing in the Mediterranean region of Spain, suggested it can be a useful crop in the Mediterranean countries and the cultivars studied can be promising for cultivar selection. The only advantage of the cultivar 'Rate' was that had the highest amount on volatile compounds. The cultivar 'Isidro' had the biggest and heaviest fruits, and together with 'Phoenix' fruits had the better color characteristics, as well as the highest contents of sugars, organic acids, proteins, and moisture. In this study, the content of H-TAA, L-TAA, FRAP, $ABTS^{\bullet +}$, $DPPH^{\bullet}$, total phenols, total flavonoids, and total flavonols was studied for the first time in the peel and pulp of these three jujube cultivars. It can be concluded that the cultivar 'Isidro' can be considered as the most interesting one, and have a huge potential for its fresh consumption, besides the antioxidant activity, total phenols and total flavonoids were higher generally in peel than in pulp; thus, it is recommended to eat this fruit without peeling. Thus, both cultivars 'Isidro' and 'Phoenix' could be promising cultivars to be used in future breeding programs, to obtain fruit with high content of bioactive compounds as well as attractive organoleptic properties.

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