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Seed and juice characterization of pomegranate fruits grown in Tunisia: Comparison between sour and sweet cultivars revealed interesting properties for prospective industrial applications

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ABSTRACT

Tunisian pomegranate genetic resources consist of sweet and sour cultivars, showing large morphometric variability. In the present work we characterized seeds and juice contents of sugars and organic acids of 5 sour and 7 sweet pomegranate cultivars. Results showed that citric acid was predominant in sour pomegranates, while malic acid was the most prevalent in sweet ones. Paradoxically, sour cultivars have higher sugar content than the sweet ones. A strong correlation was found between sourness and citric acid content, which is assumed to be the major factor that determines sour taste in pomegranate fruits. Besides, some of the seed parameters showed a significant positive correlation with acidity. Sweet cultivars were appropriate for fresh consumption and juice production due to several attributes in addition to their sweetness. Equally, sour pomegranate showed several characteristics that could be of great interest for food and nutraceutical industries.

1. Introduction

In recent years, there has been an explosion of interest in pomegranate fruit, based on its high antioxidant activity as well as its several medical benefits (Caligiani et al., 2010; Gonzalez-Molina et al., 2009; Kasimsetty et al., 2010; Lansky and Newman, 2007; Panichayupakaranant et al., 2010; Ryan and Prescott, 2010; Tehranifar et al., 2010). It follows an increasing demand for industrial processing to make juice, jams, syrup, sauce, nutraceuticals, etc., in addition to the growing demand for fresh consumption. The edible part of pomegranate fruits (the arils) contains large amounts of organic acids, sugars, minerals, vitamins and polyphenols (Al-Maiman and Dilshad, 2002; Poyrazoglu et al., 2002; Tehranifar et al., 2010).

Mediterranean and Middle-East countries are the main regions of pomegranate cultivation and production (Jbir et al., 2008; Melgarejo et al., 2009). In Tunisia, pomegranate is one of the most important fruit trees. The total production exceeded 70,000 t in 2008 (Bchir et al., 2009). Almost all these productions are based on few cultivars, with interesting market characteristics, despite the quite large number of local ecotypes listed (Mars, 2001). In fact, in new plantations that are meant for exportation, many cultivars are becoming abandoned, despite their high potential of valorization. For instance, acidity (or sourness) could be an interesting trait for several purposes (blending juice of other fruits for example).

In the frame of this growing interest in this fruit species, it is highly important to knowledge fruit characteristics, particularly the edible part. This is a necessary step to get essential and useful information for fresh market and processing industry, as well as for cultivars classification. This will help for the best germplasm management and further utilization in breeding program and cultivar selection using cultivar with desirable traits.

Pomegranate preference characteristics are determined through the taste of juicy seed coat and the unpalatability due

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to the woody portion. These traits are of interest not only for consumer, but also for cultivators, breeders and industrials. Taste is determined mainly by organic acid–sugar content balance of the fruit, and these compounds serve as unequivocal markers for sensory attributes assessment and genotype characterization (Melgarejo et al., 2000; Poyrazoglu et al., 2002). As for the unpalatability of pomegranate arils, it is due to the seed hardness, which can be measured by the woody portion index (WPI) (Martínez et al., 2006). Obviously, these characteristics depend on many factors and the interaction between them: genotypes, climate, cultural practice, etc. (Borochov-Neori et al., 2009; Schwartz et al., 2009a,b).

Several reports have shown high variation in morphometric traits and chemical characteristics in different pomegranate accessions from several geographic regions (Martínez et al., 2006; Melgarejo et al., 2000; Poyrazoglu et al., 2002; Sarkhosh et al., 2009; Tehranifar et al., 2010; Zamani et al., 2010). However, in Tunisia there is scarcity in seed morphological and chemical characterization of local cultivars of pomegranate. In the current study we assess the variation of some seed and juice characteristics of Tunisian pomegranate cultivars, and we perform a comparison between sweet and sour pomegranate accessions originated from different locations.

2. Materials and methods

2.1. Fruit samples

Pomegranate fruits were sampled at ripe stage, *i.e.* ready for fresh consumption, from one, two or three trees depending on the tree's number per cultivar available. Twelve autochthonous cultivars, namely: 'Gabsi 1', 'Mezzi 1', 'Mezzi 2', 'Mezzi 3', 'Garoussi 2', 'Gabsi 5', 'Gabsi 9', 'Chelfi 1', 'Chelfi 3', 'Zehri 6', 'Garoussi 1' and 'Tounsi 4' are included in the present study. The five formers produce sour fruits and the seven latters have sweet fruits. Although pomegranate accessions are classified by several authors as sour, sour–sweet and sweet accessions (Poyrazoglu et al., 2002; Melgarejo et al., 2000), the cultivars included in the present study were carefully chosen to be either sour or sweet, to avoid unambiguous clear-cut grouping that may affect results and comparisons. All trees are maintained in the Tunisian national germplasm collection of pomegranate located at Zerkin (33°45′N, 10°16′E) (Mars, 2001), and cultivated under homogenous conditions, without any special management (no fertilizers, no irrigation except natural rainfall).

2.2. Seed characters

Five characteristics were measured on a homogenized sample of 75 seeds (woody part of the arils) extracted from 3 fruits representing each cultivar (25 seeds per fruit): SL (seed length), ST1 and ST2 (seed thicknesses, which correspond to the two maximum thicks in the transverse section), SW (seed weight), and SS (seed shape). For seed shape, we identified 3 kinds of seeds: oblongs, ovate and globose, for which we gave the digit: 1, 2 or 3, respectively. In addition, the aril weight (AW) was also measured on 75 arils extracted from the same 3 pomegranate fruits, as well as the woody portion index (WPI). The latter was determined as: $[SW/AW] \times 100$ (Martínez et al., 2006). The measurements were carried out using digital calliper (Furiya, Japan) and the weights using an electronic balance (Mettler AJ50).

2.3. Juice extraction and HPLC analyses

In the laboratory, 3 ripe fruits representing each cultivar were hand-peeled and the arils were extracted. Subsequently they were juiced using a commercial blender. The crude juices were sieved to eliminate solid particles of pips. The pre-filtered juices were centrifuged at 15,000 rpm for 20 min. One millilitre of the centrifuged liquid was passed through a 0.45 μm Millipore filter and then injected into a Hewlett-Packard HPLC series 1100. The elution system consisted of 0.1% phosphoric acid with a flow rate of 0.5 mL/min. Organic acids were separated on a Supelcogel™ C-610H column (30 cm × 7.8 mm i.d., Supelco, Bellefonte, PA, USA) and detected by absorbance at 210 nm. Sugar analysis was performed on a μBondapak-NH2 column (30 cm × 3.9 mm i.d., Waters, Milford, MA, USA) using acetonitrile/water (85:15, v/v) as mobile phase. Sugars were detected with a refractive index detector. Standard curves for pure organic acids (oxalic, citric, tartaric, malic, acetic, fumaric, succinic and ascorbic acids) as well as for pure sugars (glucose, maltose, fructose, sucrose and sorbitol) purchased from Sigma (Poole, Dorset, UK), were used for quantification. Results were expressed as concentrations (g/100 g).

2.4. Statistical analysis

XLSTAT 2010 (http://www.xlstat.com) software is used to carry out descriptive statistics, as well as the correlation between similarity matrices (based on seed characters and juice parameters) using Mantel's test (Mantel, 1967) based on 10,000 random permutations. Histograms were built using Excel 2003 (Microsoft).

3. Results and discussions

3.1. Seed characteristics

The parameters measured on edible portion of pomegranate fruits are of economic interest and therefore they are of the most important targeted traits by growers, breeder and industrials. Fig. 1 shows obtained data concerning seed parameters measured on sour and sweet pomegranate cultivars. The woody portion index (WPI), the seed weight (SW) and the aril weight (AW) are the characteristics showing the highest variability. Their coefficients of variation were 42%, 29% and 25%, respectively (data not shown). Average WPI ranged between 2.16% ('Chelfi 3') and 7.33% ('Mezzi 1') with an overall mean of 4.43%. A significant difference was observed between sour and sweet accessions. The WPI mean in sour cultivars is twice higher than in sweet cultivars. Thus, the relatively high WPI, in addition to their sourness, increases the unpalatability of sour cultivars. Martínez et al. (2006) consider WPI as good parameter that reflects the wood quantity in edible part of pomegranate fruit. The WPI is significantly correlated with all other seed characters, except the seed length (SL). The remaining parameters presented less variation, and therefore were less discriminatory between cultivars, and between sour and sweet groups.

In comparison with similar previous studies, the Tunisian sweet pomegranate cultivars, for which the highest WPI was 3.35%, showed a very interesting trait dealing with fresh consuming preferences, since much higher WPI values were obtained from pomegranate cultivars from Spain (mean value ~8%) (Martínez et al., 2006) and from Iran (mean value ~7.5%) (Sarkhosh et al., 2009). It is also worth to report that this character had also the highest variation among all the other fruit traits in Iranian soft-seed cultivars (Sarkhosh et al., 2009). Thus, this character should be considered for any prospective selection (in addition to crude fibre and seed hardness). For instance, the cultivar 'Chelfi 3', which presents the lowest WPI and the highest AW, offers very promising genotypes for breeders to generate cultivars with greater agronomic potential.

3.2. Organic acids

HPLC was used to quantifying 8 organic acids: acetic, ascorbic, citric, fumaric, malic, oxalic, tartaric and succinic acids. Among