ORIGINAL ARTICLE



Antioxidant and physicochemical characteristics of unfermented and fermented pomegranate (*Punica granatum* L.) beverages

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Abstract The aim of this study was to evaluate the physicochemical, antioxidant (antioxidant activity, total phenolic compounds, flavonoids, anthocyanins), and sensory characteristics of fresh (FRJ) and fermented (FEB) pomegranate beverages. Three fermentation conditions were tested based on the total soluble solids (TSS: °Bx) content in pomegranate juice: (a) natural TSS (13.9 °Bx) in fresh juice (FEB1), (b) adjusted to 17.5 °Bx (FEB2) and (c) adjusted to 25 °Bx (FEB3). The antioxidant activity, total phenolic compounds, flavonoids and anthocyanins after fermentation in FEB3 were 262.61 \pm 0.12 mg Trolox, 188.60 ± 0.20 mg Gallic acid, 64.35 ± 0.09 mg $1.92\,\pm\,0.15$ and cyanidin-3-O-glucoside quercetin (C3OG)/100 mL, respectively. The final amounts of ethanol in FEB1, FEB2, and FEB3 were 6.82 ± 0.01 , 9.73 ± 0.01 , and $12.88 \pm 0.01\%$ (v/v), similar to that in wines. In general, the sensory characteristics of both FRJ and FEB beverages were well sensory accepted by consumers.

Keywords Pomegranate beverages · Fermentation · Antioxidant activity · Phenolic compounds · Anthocyanins · Flavonoids

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Introduction

Pomegranate (*Punica granatum* L.) is a fruit original at Iran, Asia. Today, the main growers and exporters are India, Iran, China, Turkey, and the United States. Mexico produced 4400 t of pomegranate in 2013 (SIAP 2014).

The pomegranate fruit is a large pome belonging to the Punicaceae family. Pomegranates have inside thin white membranes protecting seeds (called also arils or grains, the edible part of this fruit). The juice is in the arils (having a prismatic shape). The color of arils and juice varies from red to purple. Juice has a light astringent sensation (García and Pérez 2004). Pomegranate has become an important fruit because of its considerable nutritional and antioxidant characteristics. It contains sugars, organic acids, amino acids, polysaccharides, vitamins, minerals, and polyphenols. The pomegranate polyphenols include flavonoids (flavonols, flavanols and anthocyanins among others), condensed tannins (proanthocyanidins), and hydrolyzable tannins (Sepúlveda et al. 2010). High antioxidant activity has been reported in pomegranate arils (Tehranifar et al. 2010; Gil et al. 2000; Tzulker et al. 2007). Studies of pomegranate polyphenols have shown anticarcinogenic, anti-cardiovascular, antimicrobial, and anti-inflammatory properties (Sepúlveda et al. 2010; Tehranifar et al. 2010; Gil et al. 2000). Pomegranate juice has shown a threefold higher antioxidant activity than red wine or green tea (Sepúlveda et al. 2010) and 2-, 6- and 8-fold higher antioxidant activity than that detected in grape/cranberry, grapefruit, and orange juices, respectively (Gil et al. 2000). Pomegranate juice is a source of anthocyanins (red color), mostly anthocyanin-3-glucoside, 3,5-diglucoside, and derivatives of delphinidin, cyanidin, and pelargonidin (Gil et al. 2000).

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Products of pomegranate have great benefits to the human health due to the high content of polyphenols. Pomegranate is mostly consumed in a fresh fashion; however, it is also consumed as juice and processed products such as jams, syrups, jellies, among others (Gumienna et al. 2016). In the last few years, the consumption of pomegranate juice has increased because of its healthy properties; consequently, it is now a highly demanded product by consumers. This demand has increased its production and importations to countries where the fruit is not common (García and Pérez 2004).

Changes to pomegranate juice might be done to modify its flavor by fermentation; therefore, to increase its shelf life. Fermented products, such as wines, probiotic beverages, yogurts, and plant products have shown benefits to the human health, being documented elsewhere by scientific researches (Gumienna et al. 2016). Due to its high content of sugars, pomegranate juice may be a good raw material to carry out an alcoholic fermentation (Zhuang et al. 2011). The fermentation process might provide new potential applications for pomegranate; the process may yield a liquid beverage that may keep its nutritional properties and be a functional beverage. Some researchers have suggested that this process increases the digestibility and bioavailability of nutrients and bioactive compounds found in the pomegranate juice (Gumienna et al. 2016). During fermentation, the changes taking place along the process, and until the end, modifies the sensory (flavor, aroma, color, and texture) and physicochemical characteristics of the pomegranate juice, improving its content in bioactive compounds. Some researchers have suggested that the chemical reactions taking place during fermentation generate different compounds for producing a completely different product with many metabolites (Gumienna et al. 2016), responsible of flavor, aroma, and textural characteristics. The differences in the content of bioactive compounds, before and after fermentation, are due to the reactions of polymerization, condensation, oxidation, hydrolysis, enzymatic activity and interactions of the yeasts with bioactive compounds. Other researchers have pointed out that the content of polyphenols decreased at the end of the fermentation process. They also ensure that the only bioactive compounds that maintains their concentrations, in fresh and fermented beverages were flavonoids (Zhuang et al. 2011; Mena et al. 2012; Ordoudi et al. 2014).

The aim of this study was to evaluate the physicochemical, antioxidant, and sensory characteristics of fresh pomegranate juice and fermented beverages.

Materials and methods

Materials

Fruit Pomegranate (*P. granatum* L.) fruits, "Apaseo" variety, were purchased at a local market in Puebla, Mexico. Pomegranates were chosen free from physical and microbiological injuries; then, washed, and disinfected for 1 min with a 150 μ L/L hypochlorite sodium solution. Arils were separated by hand from the fruit. The juice extraction from arils was performed using a Standard Turmix extractor (Switzerland). *Yeasts Saccharomyces cerevisiae* for wine fermentation was obtained from the Laboratory of Food Microbiology at Universidad de las Americas Puebla (UDLAP), Puebla, Mexico. *S. cerevisiae* was grown in yeasts broth at 25 °C until reaching the early stationary phase.

Fermentation

Three fermentation conditions were tested based on the total soluble solids (°Brix) in pomegranate fresh juice (FRJ). The three conditions were fresh juice with its initial total soluble solids (TSS) (13.9 °Bx) (FEB1) and adjusted to 17.5 (FEB2) or 25 (FEB3) °Brix using standard sugar (Zafra, S.A. de C.V., México). Each juice was filtered throughout cheesecloth and then centrifuged at 5500 rpm for 20 min. Finally, one litter of each juice was inoculated with five milliliters of S. cerevisiae inoculum $[(2.60 \pm 0.21) \times 10^6 \text{ CFU/mL})]$. The fermentation of juices was performed at 25 ± 1 °C in an incubator until reaching a constant TSS content. The physicochemical and antioxidant characteristics were analyzed immediately after the extraction of fresh juice and fermented products. All analyses were performed in triplicate.

Physicochemical characteristics

TSS An ATAGO refractometer (Atago Co. Ltd., Tokyo, Japan) was used for measuring the TSS content according to the 932.14C AOAC (2000) method. *pH* It was measured using a Conductronic pH-meter (Conductronic S. A., Puebla, Mexico) at 20 ± 5 °C. *Titratable (TA), volatile* (VA), and fixed (FA) acidities. TA, VA, and FA were determined by titration until reaching pH 8.1 using a 0.1 N NaOH solution according to the 942.15, 11.047, and 981.12 AOAC (2000) methods, respectively. TA and FA quantities were calculated as percentage (w/v) of citric acid (CA) and the VA content as percentage (w/v) of acetic acid (AA). *Ethanol* Ethanol content was measured by specific gravity according to the 10.023 AOAC (2000) method.

Color

A tri-stimulus Chroma Meter CR-400 (Konica Minolta Sensing Inc., Osaka, Japan) colorimeter was used for measuring the L^* , a^* , b^* color parameters (transmittance mode) in the *CIELab** scale. Hue (°), Chroma (*C*) and the total color change (ΔE^*) were calculated (Marcus 1998).

Antioxidant characteristics

Antioxidant activity The antioxidant activity was analyzed according to the DPPH (2,2-diphenyl-1-picrylhydrazyl) method reported by Brand-Williams et al. (1995) with some modifications (Baqueiro-Peña and Guerrero-Beltrán 2017). The antioxidant activity was calculated as Trolox equivalents (TE) per 100 mL of beverage (Eq. 1).

$$TE\left(\frac{\mathrm{mg}}{100\,\mathrm{mL}}\right) = \left(\frac{A-b}{m}\right) * DF * 100\tag{1}$$

where A is the absorbance of the sample, b is the intercept (1.3634), m is the slope (3215.2 1/mg) of the standard curve, and DF is the dilution factor of the sample. For pomegranate juice, 5 μ L of pomegranate juice was diluted with 1995 μ L of ethanol (99%); then, 2.0 mL of DPPH solution (0.038 mg/mL) added, thoroughly mixed and analyzed by spectrophotometry at 517 nm.

Total phenolic compounds Total phenolic compounds were evaluated using the Folin–Ciocalteu method (Singleton et al. 1999) with some modifications (Baqueiro-Peña and Guerrero-Beltrán 2017). The total phenolic compounds content was calculated as Gallic acid (GA) per 100 mL of juice (Eq. 2).

$$GA\left(\frac{\mathrm{mg}}{100\,\mathrm{mL}}\right) = \left(\frac{A-b}{m}\right) * DF * 100\tag{2}$$

where A is the absorbance of the sample, b is the intercept (0.077), m is the slope (20.425 1/mg) of the standard curve and DF is the dilution factor of the sample. For pomegranate, 10 μ L of beverages, 3990 μ L of water, 250 μ L of Folin–Ciocalteu regent, 750 μ L of 20% NA₂CO₃ were mixed, left for 2 h, and analyzed by spectrophotometry at 765 nm.

Flavonoids Flavonoids were analyzed according to the Dewanto et al. (2002) method with some modifications. Concentrations in juice were calculated as mg of quercetin (QU) equivalents per 100 mL of juice (Eq. 3).

$$QU\left(\frac{\mathrm{mg}}{100\,\mathrm{mL}}\right) = \left(\frac{ABS - b}{m}\right) * DF * 100 \tag{3}$$

where *ABS* is the absorbance of the sample, *b* is the intercept (0.004), *m* is the slope (2.6727 1/mg) of the standard curve and *FD* is the dilution factor. For pome-granate, 10 μ L of beverage, 3265 μ L of distilled water,

75 μ L of 10%NaOH were mixed, left for 5 min. Then, 150 μ L of 10% AlCl₃·6H₂O added, left for 6 min, 500 μ L of 40 g/L NaOH added and finally analyzed by spectrophotometry at 510 nm.

Total anthocyanins

The total anthocyanins content was analyzed using the pH differential method (Giusti and Wrolstad 2001). Results were calculated as mg cyanidin-3-*O*-glucoside (C3OG) per 100 mL of juice.

Sensory evaluation

For the sensory evaluation of FRJ and FEBs, appearance, color, aroma, sweetness, taste and general acceptability were evaluated. A nine-point hedonic structured scale was used. The sensory evaluation was performed with 20 untrained judges (Wichchukit and O'Mahony 2015).

Statistical analysis

Experimental data were analyzed using a Minitab v.17 Statistical Software (Minitab Inc., State College, PA, USA). Differences within means of treatments were considered significant for $p \le 0.05$ using ANOVA and Turkey's tests.

Results and discussion

Pomegranate juice

Physicochemical characteristics

The physicochemical characteristics of pomegranate juice are shown in Table 1. These characteristics are important in pomegranate juice for the fermentation process, highlighting the content of TSS.

TSS The TSS content found in this researched for pomegranate juice was $13.9 \pm 0.02\%$ (w/w). This TSS content in pomegranate juice was an adequate medium to carry out the alcoholic fermentation. Zarei et al. (2011) reported a TSS in pomegranate juice in the range 9.56-10.30% (w/w) and Sepúlveda et al. (2010) and Tehranifar et al. (2010) reported values of TSS in the range 11.37-15.07 °Bx. These changes in the TSS are mainly due to the variety and maturity of pomegranate.

pH It is an important factor in the fermentation process due to its effect with the yeasts and quality attributes of the product such as flavor, color, and aroma. It is recommended that the pH in the fermentation wort be in the range 2.8–4.0 (Sepúlveda et al. 2010; Zarei et al. 2011).

Table 1 Physicochemicalcharacteristics of FRJ¹ andFEB² products

	Beverage				
	FRJ	FEB1	FEB2	FEB3	
TSS (% w/w)	13.90 ± 0.02	$4.30\pm0.01a^3$	$5.90\pm0.01\mathrm{b}^3$	$11.70 \pm 0.02c^3$	
рН	$3.30\pm0.02a$	$3.41\pm0.01\mathrm{b}$	$3.60\pm0.02c$	$3.70\pm0.01\text{d}$	
TA (% citric acid)	$0.35\pm0.02a$	$0.40\pm0.03b$	$0.62\pm0.01c$	$0.67\pm0.01\rm{d}$	
FA (% citric acid)	-	$0.30\pm0.02a$	$0.52\pm0.03b$	$0.57\pm0.05\mathrm{c}$	
VA (% acetic acid)	-	$0.09\pm0.01\mathrm{a}$	$0.10\pm0.01\mathrm{b}$	$0.10\pm0.01\mathrm{b}$	
Ethanol ⁴	_	$6.82\pm0.01\mathrm{a}$	$9.73\pm0.01\mathrm{b}$	$12.88\pm0.01\mathrm{c}$	
L^*	$31.74\pm0.02a$	$32.54\pm0.05\mathrm{b}$	$35.47\pm0.02c$	$30.83\pm0.04d$	
<i>a</i> *	$34.02\pm0.05a$	$32.04\pm0.01\mathrm{b}$	$31.62\pm0.01c$	$31.98\pm0.06\mathrm{c}$	
b^*	$14.11\pm0.04a$	$11.23\pm0.03\mathrm{b}$	$13.50\pm0.03c$	$10.01\pm0.08\mathrm{d}$	
Hue (°)	22.50a	19.30b	23.10c	17.40d	
Chroma	36.83a	33.95b	34.38c	33.51b	
ΔE^*	-	3.59a	4.48b	4.67c	

Different letters within rows indicate significant differences ($p \le 0.05$)

¹FRJ: pomegranate fresh juice

²FEB: pomegranate fermented beverage

³At the end of fermentation

⁴Percentage (v/v)

TA Total acidity, calculated as percentage of citric acid (CA) was $0.35 \pm 0.02\%$ in FRJ. Tehranifar et al. (2010) reported a TA content in the range 0.33-2.44% CA in 20 samples of pomegranate from different Iranian cultivars. Results of this study showed a lower percentage of acidity than that reported by other researchers (Sepúlveda et al. 2010; Tehranifar et al. 2010; Zarei et al. 2011) which could be due to the variety and maturity of the fruit.

Color

The color parameters of pomegranate juice were $L^*= 31.74 \pm 0.02, a^*= 34.02 \pm 0.05, b^*= 14.11 \pm 0.04,$ hue = 22.5° and C = 36.83 (Table 1). The L* value indicate a dark pomegranate juice (being 0 = white and 100 = black). The a^* (red-green) value (34.02 \pm 0.05) is characteristic of the red color which gives important information about the content of red natural pigments (anthocyanins) responsible for the typical color of pomegranate juice (Tzulker et al. 2007). Regarding the b^* color parameter, the value (14.11 ± 0.04) was found in the redyellow color segment of the color space; the value indicates a yellowish hue. The hue value, found in the red-yellow segment of the color space, had a tendency to an intense red color. The chroma parameter indicates a high saturation of red, which is mainly due to the content of anthocyanins. All values of the color parameters are located in the redyellow segment of the color space (Baqueiro-Peña and Guerrero-Beltrán 2017).

Antioxidant characteristics

The antioxidant characteristics of pomegranate juice are shown in Table 2.

Antioxidant activity The antioxidant activity of pomegranate FRJ was 482.23 ± 0.20 mg TE/100 mL. Gil et al. (2000) reported that the antioxidant activity of commercial pomegranate juice was higher than that in red wine (354.89 mg TE/100 mL) and green tea (276.02 mg TE/ 100 mL). Çam et al. (2009), on the other hand, analyzed the antioxidant activity of eight cultivars of pomegranate from Turkey. They reported an antioxidant activity in the range 221.2–418.3 mg TE/100 mL of juice.

Total phenolic compounds Pomegranate is rich in phenolic compounds such as ellagic acid, *p*-coumaric acid, tannins, anthocyanins and catechin. The total phenolic compounds content in this study was 393.78 ± 0.13 mg GA/100 mL. Poyrazoglu (2002) reported Gallic acid as the major phenolic compound in pomegranate fruit. Sepúlveda et al. (2010) reported 123.6 mg GA/100 mL. Other researchers have reported lower amount of total phenolic compounds (275.0 mg GA/100 mL) from arils of pomegranate "Mollar" variety.

Flavonoids The flavonoid content in FRJ was 172.10 ± 0.15 mg QU/100 mL. El Kar et al. (2011) analyzed different varieties of pomegranate juice and reported a total flavonoids content in the range 13.5–63.6 mg QU/100 mL. It has been reported kaempferol, luteolin, quercetin and catechin in pomegranate juice (Van Elswijk et al. 2004).

Table 2 Antioxidant profile of FRJ^1 and FEB^2 products

	Beverage				
	FRJ	FEB1	FEB2	FEB3	
Antioxidant activity ³	$482.23 \pm 0.20a$	$281.65 \pm 0.11b$	$273.35 \pm 0.09c$	$262.61 \pm 0.12d$	
Total phenolics ⁴	$393.78 \pm 0.13a$	$229.28\pm0.13b$	$191.14 \pm 0.14c$	$188.60 \pm 0.20d$	
Flavonoids ⁵	$172.10\pm0.15a$	$119.70 \pm 0.08b$	$86.80\pm0.06\mathrm{c}$	$64.35 \pm 0.09d$	
Anthocyanins ⁶	$4.61\pm0.38a$	$2.22\pm0.06\mathrm{b}$	$1.96\pm0.09\mathrm{c}$	$1.92 \pm 0.15d$	

Different letters within rows indicate significant differences ($p \le 0.05$)

¹FRJ: pomegranate fresh juice

²FEB: pomegranate fermented beverage

³mg ET/100 mL

⁴mg GA/100 mL

5mg QU/100 mL

6mg C3OG/100 mL

Total anthocyanins

The content of anthocyanins in pomegranate juice was 4.61 ± 0.38 mg of C3OG/100 mL (Table 2). Gil et al. (2000) reported that C3OG was the main anthocyanin in pomegranate juice (12.8 mg/100 mL). Sepúlveda et al. (2010) reported a total content of anthocyanins in pomegranate juice of ten different genotypes in the range 17.0–134.2 mg of C3OG/100 mL. Tzulker et al. (2007) reported 10–30 mg of C3OG/100 mL of juice from arils of 29 Israeli pomegranate cultivars.

Fermented beverages

Physicochemical properties

Table 1 shows the physicochemical characteristics of fermented beverages.

TSS Soluble solids were metabolized by yeasts to ethanol. Ethanol also has refractive properties; therefore, ethanol is detected as a "soluble solid" along with residual sugars in FEB beverages.

pH

It increased in the three FEB beverages, being higher in the FEB3 beverage. In fermented beverages, pH should be in a range 2.8–4.0; pH for the three FEBs was in this range. Berenguer et al. (2016) reported a pH of 3.4 at the beginning and 3.5 at the end of fermentation of pomegranate juice. Zhuang et al. (2011) reported 3.2 at the start and 3.3 at the end of the fermentation. All pH values were higher in FEBs than in FRJ. This increase was very probably due to the metabolites delivered during the fermentation. A significant difference ($p \le 0.05$) in pH was observed in FEB beverages.

TA

TA of FEBs is shown in Table 1. Significant differences $(p \le 0.05)$ were observed in TA of pomegranate beverages. Due to the fermentation process, there are many changes in organic acids in the juice, which may increase, decrease, or be generated as new acids (citric, malic, lactic or acetic acids). Berenguer et al. (2016) reported TA contents of 0.28 \pm 0.03 and 0.59 \pm 0.04% of CA in fresh and fermented pomegranate beverages, respectively. Zhuang et al. (2011) reported TA values of 0.52 ± 0.05 and 0.73 ± 0.02 g/100 mL CA in fresh and fermented juices, respectively. Ordoudi et al. (2014) reported TA values of 0.36 ± 0.02 and $0.52\pm0.04\%$ CA in unfermented and fermented juices, respectively. Acidity has a decisive impact in aroma and flavor of fermented drinks. The increase in TA may be due to the production of α -ketoglutaric and succinic acids in the glyceropyruvic fermentation pathway and pyruvic acid in the glycolytic pathway.

VA and FA

There was a greater production of VA and FA in FEB3 than in FEB1 and FEB2 beverages. According to the International Code of Oenological Practices (2015), the maximum permitted limit of VA in wine is 0.12 g/100 mL (expressed as acetic acid) and the minimum permitted limit of FA is 0.4 g/100 mL (expressed as tartaric acid). The acidities, found in this research were within the limits permitted by the International Code of Oenological Practices (2015).

Ethanol

The concentrations of ethanol in FEB1, FEB2, and FEB3 were 6.82 ± 0.01 , 9.73 ± 0.01 and $12.88 \pm 0.01\%$ (v/v),

respectively. The Mexican Norm number NOM-142-SSA1/ SCFI-2014 (DGN 2014), for alcoholic beverages, classifies an alcoholic beverage when the alcohol content is in the range 6.1–20.0% by volume; therefore, the three FEBs had an average alcoholic content within this range.

Color

Differences in the color parameters were observed within all fermented pomegranate beverages (Table 1). In general, the a^* and b^* color values were higher in FRJ than in FEBs. It is important to notice that the a^* value (red color) of the FEB1 was higher than the values for FEB2 and FEB3. The hue (°) was lower in the FEB1. The FEB2 beverage presented a greater angle (23.1°), indicating a more intense red color, according to the red-yellow color space segment. The Chroma color parameter decreased during fermentation, showing a less saturated color. For the total change in color (ΔE^*), the FEBs beverages had lighter changes, being barely clearer than FRJ. All ΔE^* values were similar. The color of a fermented beverage is an important parameter for the consumer's acceptance. Anthocyanins are labile to physical factors such as pH, temperature, metals among others. In this work, the pH in the three FEB beverages barely changed; therefore, the color barely changed in fermented products.

Antioxidant activity

The antioxidant activity, total phenolics, flavonoids and anthocyanins content for FEB beverages are shown in Table 2.

The antioxidant activity decreased from 482.23 ± 0.20 (in FRJ) to 262.61 ± 0.12 (in FEB3) mg TE/100 mL. After fermentation, the antioxidant activity was reduced about 45%. The decrease in the antioxidant activity could be due to the oxidation reactions that occurs along the fermentation process. The pomegranate compounds may interact with each other and oxidize, decreasing the antioxidant activity. Each phenolic compound and their interaction in the pomegranate beverages might contribute differently to the change in the antioxidant activity. Zhuang et al. (2011) and Ordoudi et al. (2014) reported changes in antioxidant activity in the rage 16-39% in pomegranate beverages.

Total phenolic compounds

The amount of these compounds was 393.78 ± 0.13 and 188.6 ± 0.20 mg GA/100 mL, before (FRJ) and after fermentation (FEB3), respectively (Table 2). Significant differences ($p \le 0.05$) were observed in the total phenolic compounds content. The content of Gallic acid in FEBs decreased as the initial TSS increased in the initial

fermentation system. Ordoudi et al. (2014) studied the effect of fermentation on the total phenolic compounds; they reported 138.7 and 139.5 mg GA/100 mL in unfermented and fermented products, respectively. Lantzouraki et al. (2015) compared the total phenolic compounds content of pomegranate ("Wonderful" variety) fermented beverages with red wine; they pointed out that the total phenolic compounds content was significantly higher in the pomegranate beverage (383 mg GA/100 mL) than in red wine (296 mg GA/100 mL).

Flavonoids

The flavonoids content was reduced in FEBs (Table 2) by about 60%. In addition to the significant differences ($p \le 0.05$) of flavonoids within fresh juice (FRJ) and fermented beverages, the beverage with the lower flavonoids content was the FEB3 (64.35 \pm 0.09 mg QU/100 mL). The amount of flavonoids in FEB3 was similar to that reported by El Kar et al. (2011) in pomegranate fresh juice (63.6 mg QU/100 mL).

Total anthocyanins The total anthocyanins content in FEBs was in the range 2.22–1.92 mg of C3OG/100 mL (Table 2). Significant differences ($p \le 0.05$) were observed within the anthocyanins content in FRJ and FEB beverages, being lower in FEB3. The amount of anthocyanins was barely reduced in beverages that had the higher amount of initial TSS but produced the higher amount of ethanol. Therefore, the low change observed of anthocyanins was actually low; the reduction was about 13.5%. The reduction of anthocyanins in beverages, as the initial content of TSS increased, might be due to the reactions of polymerization with acetaldehyde, generated by the metabolic activity of yeasts, with the formation of complex compounds causing loses of the red color (Gil et al. 2000).

Sensory evaluation

In Table 3 are reported the results of the sensory evaluation of FRJ and FEBs. Significant differences ($p \le 0.05$) were found within different samples. In general, the best-liked sample was the FRJ beverage. None of the samples obtained a score higher than 8 or less than 5. The FEB1 beverage was the best scored in appearance and color, this could be due to its red-bright color (a^* value of 32.04 ± 0.01), suitable for red color. For aroma, sweetness, taste, and general acceptability, the FEB3 beverage was very well accepted. FEB3 was the beverage with the highest content of TSS (11.7 ± 0.02 °Bx) and ethanol content ($12.88 \pm 0.01\%$ v/v). According to the International Code of Oenological Practices (2015), a beverage with more than 5 g sugars/100 mL is classified as a

Table 3 Sensory evaluation ofFRJ1 and FEB2 products

	Beverage				
	FRJ	FEB1	FEB2	FEB3	
Appearance	$7.75\pm0.786ab$	$8.05\pm0.686a$	$7.45 \pm 1.432a$	$6.95 \pm 1.877 \mathrm{b}$	
Color	$7.05\pm0.999\mathrm{b}$	$8.15\pm0.933a$	$6.80\pm1.795b$	$7.20\pm1.240ab$	
Aroma	$7.80\pm0.768a$	$6.45\pm1.986b$	$6.75 \pm 1.916b$	$6.85 \pm 1.461 \mathrm{ab}$	
Sweetness	$7.50\pm0.827a$	$5.05\pm1.877\mathrm{b}$	$5.70\pm2.296\mathrm{b}$	$7.25 \pm 1.372 a$	
Taste	$7.80\pm0.834a$	$5.30\pm2.515b$	$6.05\pm2.395\mathrm{b}$	$6.60\pm1.957 ab$	
General acceptability	$7.85 \pm 0.671 a$	$5.25\pm2.403b$	$5.30\pm2.812\mathrm{b}$	$6.90 \pm 1.971 { m ab}$	

Different letters within rows indicate significant differences ($p \le 0.05$)

¹FRJ: pomegranate fresh juice

²FEB: pomegranate fermented juice

fermented sweet-beverage. The FEB3 of the three FEB beverages was the one with the highest content of ethanol and total acidity $(0.67 \pm 0.01\%$ citric acid). Ethanol is the most abundant volatile compound in fermented beverages and it may modify both the sensory perception of aromatic attributes and detection of volatile compounds (Goldner et al. 2009). Ethanol also influence the "body" of the fermented beverage and the perception of astringency, acidity, sweetness, and flavor.

Conclusion

Saccharomyces cerevisiae changed the physicochemical composition of fresh pomegranate juice, contributing to the modification of their physicochemical and antioxidant properties. The analyses indicated that the pomegranate juice and alcoholic beverages are products rich in antioxidants. Antioxidant activity, total phenolic compounds, flavonoids, and anthocyanins were lightly reduced at the end of the fermentation process. Regardless of this reduction, the antioxidants were still maintained in fermented products. All pomegranate beverages were well sensory accepted.

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