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Investigation of some quality parameters of pomegranate, sumac and unripe grape sour products from Kilis markets

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RESEARCH ARTICLE

Abstract

The aim of the present work was to determine some quality parameters of pomegranate, sumac and unripe grape concentrate products sold in Kilis markets. The highest pH and titration acidity (TA) values were generally determined in homemade pomegranate concentrate (HPC) and homemade sumac concentrate (HSC) (values for pH were 3.29-3.51, values for TA were 13.19-29.59 g/100 ml). It was detected that the highest values for 5-hydroxymethylfurfural (HMF) and browning index were in the HPC sample (8,373.78 mg/l and 397.8 (abs.)). The sample having the highest and lowest HMF and browning index values was homemade unripe grape concentrate (HUC). The highest total phenolics, total flavonoids, ascorbic acid and antioxidant activity values were detected in HSC. However, a specific organic acid presence associated with spots was not detected. Retention factor values in samples belonging to spots recorded as 0.83 (HSC(1), commercial sumac concentrate; CSC(2), and HPC(3)) and 0.8 (commercial pomegranate concentrate; CPC(4), commercial unripe grape concentrate; CUC(5), and HUC(6)). None of them on *Candida albicans* were active. For the HPC and CPC sour samples, any inhibition were shown. But HSC, CSC, CUC and HUC samples are more active than vancomycin.

Keywords: pomegranate, sumac, unripe grape, total phenolics, antioxidants

1. Introduction

Sours derived from the lemon, grape and pomegranate are individually used in different food as flavour enhancers. Gaziantep cuisine (south-eastern region of Turkey) has a rich diversity of sours used for making food. Verjuice, obtained by lemon juice and pressed unripe grapes, is used as salad dressing and gravy during cooking process of local dishes. Another remarkable product, sumac, is commonly used in the Gaziantep cuisine providing a sour taste. Particularly, dried sumac powder is used in kebabs and salads. The sour sumac juice, derived by keeping it in water, is preferred in the cooking stage of some food. In addition, other sour widely used in the Gaziantep cuisine is sour pomegranate (Avcıkurt *et al.*, 2016).

Manthou *et al.* (2017) reported that the pomegranate is a cultivated fruit for centuries and indicated high nutritional value (such as vitamins and minerals) and therapeutic effect of its ingredients. In the extant literature, it has been mentioned that the biological and therapeutic properties of this important fruit are primarily attributed to the presence of polyphenols, (ellagitanins, flavonoids, phenolic acids, stilbenes, tannins and anthocyanins) and free radical scavenging compounds. Kahramanoglu and Usanmaz (2017) reported the antioxidant and antimicrobial capacity of pomegranate fruit, and its possible usage as potential agent towards some illness such as cancer and diabetic via decreasing blood pressure. In an other study, the antioxidant activity of pomegranate juices was compared with red wine and green tea (Gil *et al.*, 2000).

Rouhi-Boroujeni *et al.* (2016) declared that sumac (*Rhus coriaria* L.), which belong to the Anacardiaceae family, is used as one of the main medicinal plants since ancient times. Cakmak *et al.* (2017) noted that sumac grows widely

in Asian countries and uses as a traditional medicine. Researchers also said that the tannins and flavonoids are the main compounds of sumac extracts and it includes gallic acid and several B vitamins. Toghyani and Faghan *et al.* (2017) asserted that sumac is used as a herbal remedy because of assumed analgesic, antidiarrheal, antiseptic, anorectic, and antihyperglycemic properties. Capcarova *et al.* (2010) also stated the antifibrogenic, antifungal, anti-inflammatory, antimalarial, antimicrobial, antimutagenic, antioxidant, antithrombin, antitumorigenic, antiviral, cytotoxic, hypoglycaemic, leukopenic and atheroprotective effects of sumac.

Grapes (*Vitis vinifera*) belong to the Vitaceae family and are the most widely grown fruit in Kilis district, another settlement of south-east region of Turkey. Unripe grapes are locally named 'koruk'. It has long been processed in various traditional flavouring and condiment products, such as unripe grape juice (or koruk juice), unripe grape powder and unripe grape piece in Kilis. Unripe grape is also known as 'vertjus' or 'verjus' in French, 'verjons' or 'verjuice' in English, 'agraz' in Spanish and German, 'agresto' in Italian, 'koruk' in Turkish and 'abe-ghureh' in Persian (Ucan Türkmen *et al.*, 2017). Unripe grape juice is characterised by high acidity, low sugar content and sour/tart taste (Nikfardjam, 2008). De Matos *et al.* (2017) reported that unripe grape juice has received increasing attention in Western cuisine. It is a re-discovered food ingredient as an alternative to vinegar or lemon juice. Unripe grape juice has also been tested as a potential food preservative, due to its high organic acid content and phenolic compounds such as gallic acid, caffeic acid, catechin and quercetin glucoside (Nikfardjam, 2008).

Unripe grape products, in case of direct intake or as an additive in foods, may contribute to the functional and natural food products due to their physicochemical and antioxidant properties (Öncül and Karabıyıklı, 2015).

In the present study it was aimed to determine some quality parameters of pomegranate, sumac and unripe grape concentrates sold in Kilis markets. The following abbreviations were used to describe the different commercial and homemade sour samples in our study: homemade pomegranate concentrate (HPC); commercial pomegranate concentrate (CPC); homemade sumac concentrate (HSC); commercial sumac concentrate (CSC); homemade unripe grape concentrate (HUC); commercial unripe grape concentrate (CUC). The physicochemical analysis of sour samples has also been performed by measuring pH and total soluble solids and by titrimetric assays of titration acidity. Following, moisture content, browning index, colour and 5-hydroxymethylfurfural (HMF) analyses were performed. Afterwards, total phenolics, total flavonoids and ascorbic acid contents, antioxidant and antimicrobial activity were determined.

2. Materials and methods

Homemade (pomegranate, sumac and unripe grape) sour samples were purchased from Kilis located in eastern Mediterranean region of Turkey. In order to perform a detailed comparison, the commercial sour samples (pomegranate, sumac and unripe grape) were also provided from a local market (Kilis, Turkey).

Sour samples were diluted to their original brix values. The brix values were set at 5.83, 4 and 3.5° for pomegranate, unripe grape and sumac sour, respectively. All treatments and analyses were carried out in triplicates. Sour samples were subjected to the following analyses:

Physicochemical analysis (total soluble solids, pH, titratable acidity)

The total soluble solids (or the °Brix) and pH of samples were analysed by an Abbe refractometer (WYA-25 model; J.P. Selecta, Barcelona, Spain) and WTW pH-meter (Weilheim, Germany), (Cemeroğlu, 2007). Total acidity analyses of juice samples were carried out according to Sánchez-Moreno *et al.* (2003). Neutralisation of titratable acidity by means of an end point titration at pH 8.1 with 0.1 N NaOH. The total acidity was calculated as citric acid and the result was expressed as g/100 ml.

Determination of browning index

5 ml of sample was mixed with 5 ml ethyl alcohol (95%) in Teflon tubes and then the mixture was centrifuged (4,000 rpm, 10 min, at 4 °C). The supernatant was passed through a 0.45 μm Teflon membrane filter and the absorbance of the supernatant was obtained at 420 nm in a spectrophotometer (Libra S60; Biochrom, Cambridge, UK) (Meydav, 1977).

Colour measurement

Colour (CIE L*, a*, b*) analysis were conducted by the HunterLab Spectrofotometer (HunterLab miniscan, Hunter Associates Laboratory Inc, Virginia, VI, USA). 50 ml of sample was transferred into 20 mm glass optical cell light path and then analysed. The results were given according to the CIELAB colour system. In this system, L* defines lightness (0 = black; 100 = white), a* denotes the red/green value (+ = red; - = green) and b* the yellow/blue value (+ = yellow; - = blue). In addition, the following formulas were used for the calculations of hue* and chroma values (C*) (Ucan Türkmen and Mercimek Takci, 2018):

$$\text{Hue}^* = \arctan\left(\frac{b^*}{a^*}\right)$$

$$C^* = \sqrt{(a^*)^2 + (b^*)^2}$$

HMF content

The HMF content was quantitatively determined following the procedure described by Cemeroglu (2007), which is based on the colorimetric reaction between barbituric acid, *p*-toluidine and HMF, forming a red colour complex.

Total phenolic contents

Total phenolic content in samples was measured by using a spectrophotometric method (Stankovic, 2011). 0.5 ml of the samples was mixed with 2.5 ml of 10% Folin-Ciocalteu's reagent and 2.5 ml 7.5% $NaHCO_3$. The reaction mixtures were incubated in a water bath at 45 °C for 45 min. Thereafter the absorbance of samples was spectrophotometrically measured at 765 nm (Libra S60; Biochrom). A standard curve was prepared by using standard gallic acid solution in different concentrations. The content of phenolic in samples was expressed as gallic acid equivalent (mg/l), according to the measured absorbance.

Total flavonoid contents

The total flavonoid content of samples was determined by the aluminium chloride colorimetric method (catechol as a standard). In brief, 1 ml of test sample was diluted (1:6) and mixed with 0.3 ml 5% $NaNO_2$. Then the mixture was mixed with a vortex and thereafter was incubated for 5 min. At the end of time, 0.6 ml of 10% $AlCl_3.6H_2O$ solution was added and after incubated (5 min), the obtained reaction mixture by adding 2 ml of 1 M NaOH solution, brought to 10 ml with double-distilled water. The mixture was allowed to stand for 15 min, and absorbance was measured at 510 nm with UV-VIS spectrophotometer (Libra S60; Biochrom). The total flavonoid content was calculated from a calibration curve and the result was expressed as mg/l catechol (Sharm and Vig, 2013).

DPPH radical scavenging activity

The antioxidant capacity of the samples was detected by the stable 2,2-diphenyl 1-picrylhydrazyl (DPPH) radical scavenging assay. This assay is based upon the destruction of DPPH radical, a pink stable compound, and measuring the colour decrease spectrophotometrically. 100 µl of the samples and 3.9 ml of the DPPH (0.025 g/l in methanol) solution were mixed. The mixtures were incubated in dark at room temperature for 120 min. The remaining DPPH amount was determined by measuring at 515 nm absorbance. In test samples, the inhibition of DPPH was calculated as percent according to the formula (Huang *et al.*, 2005; Yılmaz, 2011):

$$I\% = \frac{Ablank - Asample}{Ablank} \times 100$$

Ascorbic acid content

The amounts of ascorbic acid in the samples were tested by spectrophotometric absorbance measurements at 518 nm, using 2,6-diclorophenol-indophenol as colour reagent. Ascorbic acid content in the samples was calculated by comparison a standard curve prepared with L-ascorbic acid (Hışıl, 2004).

Sensory analysis

In the sensory evaluation of the samples, the taste profile analyses (colour, clarity, taste, smell and general impression) was performed by a panellist group composed of 7 people using the graph scale method (Altuğ, 1993; Watts *et al.*, 1989).

Analysis of organic acids (TLC procedure)

Thin layer chromatography (TLC) was used to determine active fractions of organic acids in the sour samples. TLC silica gel 60 F_{254} aluminium sheets were used for analyses. Standard citric acid and the sour samples at the concentration of 10% (v/v; w/v) were subjected to an acetone : distilled water : chloroform : ethanol : ammonium hydroxide (60:2:6:10:22) phase and 5 µl was transferred to plates (Lee *et al.*, 2001). The samples were migrated to 16 cm plates for approximately 60 min using the executive phase (n-butanol : acetone : 25% ammonium hydroxide : distilled water (35:25:20:10). The air-dried plates were dyed with the colouring solution (4% dimethyl amino benzaldehyde in acetic acid) and heated at 150 °C for 3 min to observe the fractions red-pink stains were evaluated as citric acid in the pale back plate (Sočič and Gaberc-Porekar, 1981). The presence of citric acid in the sample was determined by comparing the retention (Rf) values of the citric acid standard only as the organic acid.

Determination of antimicrobial activity

Antimicrobial activities of the sour samples were tested on *Escherichia coli*, *Staphylococcus aureus* and *Candida albicans* samples (obtained from Microbiology Department of Kilis State Hospital) by Kirby Bauer disc diffusion method (Bauer *et al.*, 1966). Mueller hinton agar (MHA) medium for bacteria and potato dextrose agar (PDA) medium for fungi were used. Following inoculation by spreading, the sterile antibiotic discs absorbed 50 µl sour samples were transferred to the plates. MHA and PDA plates were incubated at 37 °C and 25-28 °C for 24-48 hours. Antibiotic Vancomycin 5 (mcg/disc) (HIMEDIA Laboratories Pvt., Mumbai, India) was used as positive control. Thereafter, the inhibition zones around the discs were measured and calculated in mm.

Statistical analysis

The software SPSS 21.0 for Windows (SPSS Inc., Chicago, IL, USA) was used for analysis of variance (ANOVA) and Duncan's multiple comparison tests in order to determine significant differences between the samples. Each experiment was repeated at least three times.

3. Results and discussion

Physicochemical analysis

Results obtained for the physicochemical analysis of samples are given in Table 1. The pH values of sour samples, which can be a real indication of quality, ranged from 2.84-3.51 and the difference between homemade and commercial samples was statistically found to be significant (P<0.05). The highest and the lowest pH values were determined to 3.51 and 2.84 in the HPC and CSC, respectively. According to the sours pH values; it can be classified as high acidity (pH<3.7), acid (pH=3.7-4.6), medium acid (pH=4.6-5.3) and low acid (pH>5.3) group (Ucan *et al.*, 2014). In our study, sour samples were at high acidity group because of pH values between 2.84 and 3.51.

Ucan *et al.* (2014) found that pH values of lemon sour samples in vacuum and atmospheric conditions were 1.12 and 1.18, respectively. Akcalıoglu *et al.* (2014) detected that pH values of bitter orange sour samples in vacuum and atmospheric conditions were 1.24 and 1.28, respectively. Metin (2014) studied that determination of hydroxymethylfurfural levels of pomegranate sour, pomegranate sour sauce and grape molasses sold in Ankara markets. In the current study, pH values of pomegranate sour and pomegranate sour sauce samples ranged from 2.7 to 3 and 1.74 to 2.62. In another study, Özkanli and Tekin (2008) reported that pH value for 70 °Brix sumac concentrate was 3.2.

The total soluble solids (TSS/°Brix value) is routinely used to measure the rough sugar levels in fruit juices. TSS values of samples ranged from 3.67 to 78.20 °Brix and the difference between the treatments was found to be statistically significant (P<0.05). The samples having the highest and lowest sugar content were CPC and CUC, respectively. According to Ucan *et al.* (2014), the TSS values of lemon sour samples in vacuum and atmospheric condition were determined 65 and 64%, respectively. Akcalıoglu *et al.* (2014) declared that the TSS values of bitter orange sour samples in vacuum and atmospheric conditions were detected (as 65.50 and 68.50%).

Titratable acidity (TA), which measures the total acid concentration in a food, is another vital quality parameter that can influence the storage behaviour in fruit juices (Bhat, 2016). In most fruits, TA is responsible for the distinct taste and flavours and hence, is also a reliable indicator to evaluate the overall quality of fruits (George *et al.*, 2015). As seen in Table 1, the titratable acidity values of samples, ranged from 1.71 to 29.59 g/100 ml and the difference between the samples was found to be statistically significant (P<0.05). The highest and the lowest titration acidity values were determined to 29.59 and 1.71 g/100 ml in the HSC and CUC, respectively. Ucan *et al.* (2014) reported that the TA values of lemon sour samples in vacuum and atmospheric conditions were determined as 43.86 and 49.26 g/100 ml. Akcalıoglu *et al.* (2014) reported that the TA values of bitter orange sour samples in vacuum and atmospheric conditions were detected as 41.85 and 42.24 g/100 ml

According to Eyigün (2012) the TA values of pomegranate sour samples in vacuum and atmospheric conditions ranged from 2.09 to 21.36 g/100 ml and range from 18.73 to 21.25 g/100 ml, respectively. Nikfardjam (2008) studied seven different verjuice and the titratable acidity ranged between 1.96 and 3.96% among samples.

Table 1. Results of physicochemical analysis belonging to sour samples.[1]

Samples[2]	pH	Total soluble solid (°Brix)	Titratable acidity (g/100 ml)	Moisture (%)
HPC	3.51±0.00^{a}	75.53±0.35^{b}	13.19±0.12^{b}	24.90±0.14^{e}
CPC	3.06±0.05^{c}	78.20±1.04^{a}	3.58±0.02^{c}	22.34±0.55^{f}
HSC	3.29±0.01^{b}	64.50±0.30^{d}	29.59±1.91^{a}	28.19±0.61^{c}
CSC	2.84±0.04^{d}	67.17±0.23^{c}	12.06±0.15^{b}	28.92±0.08^{c}
HUC	3.06±0.05^{c}	4.20±0.10^{e}	3.10±0.01^{cd}	95.50±0.44^{b}
CUC	3.07±0.05^{c}	3.67±0.12^{e}	1.71±0.02^{d}	97.15±0.01^{a}

[1] Values followed by different superscripted letter within the same column are significantly different from each other (P<0.05).
[2] HPC = homemade pomegranate concentrate; CPC = commercial pomegranate concentrate; HSC = homemade sumac concentrate; CSC = commercial sumac concentrate; HUC = homemade unripe grape concentrate; CUC = commercial unripe grape concentrate.

Öncül and Karabıyıklı (2016) analysed five verjuice and sour grape sauce samples and the revealed showed that the mean of the values were 2.41 for pH, 5.63 °Brix for soluble solid content and 3.83% for titratable acidity. These researchers mention that the variation in the results caused by some factors such as variety differences, maturation stage, harvesting time, genotypic differences and environmental stress.

Browning index, colour and HMF contents of sour samples

Browning index is the browning of juice due to maillard reactions, subsequently causing colour changes and loss of nutrients (Caminiti *et al.*, 2012; Santhirasegaram *et al.*, 2015). It is an index of the dark-coloured components that are formed by the heat-based processes applied to the product and the severity of these processes. As shown in Table 2, the difference between all homemade and commercial samples in browning index values were found to be statistically significant ($P<0.05$). Browning index values ranged between 1.15 and 397.8 (abs.) and the highest and the lowest values were detected in the HPC and HUC, respectively. As the accumulation of these components increases in the environment, the absorptive light and the browning index value increases in the same proportion.

Ucan *et al.* (2014) stated that the browning index values of lemon sour samples in vacuum condition and atmospheric condition were determined 0.98 and 2.48 (abs.), respectively. Akcalıoglu *et al.* (2014) detected that the browning index values of bitter orange sour samples in vacuum condition and atmospheric condition were detected 0.77 and 2.99 (abs.), respectively.

The colour is a vital parameter for preferability of a foodstuff and therefore admitted upper attention in food industries and by consumers. Colour change in a fruit juice indicates the microbial activity and so the quality decreases due to insufficient treatments (Bhat and Stamminger, 2015). As presented in Table 2, differences were found to be statistically significant in all colour values of commercial and homemade sour samples ($P<0.05$).

The L* values varied between 1.14-21.75. The highest L* value was determined in CUC and followed by HUC. L* is a parameter that indicates the brightness of the measured object, having a range between 0 and 100. It is understood that when the L* value is 0, the object is excessively matt or black, while when it is 100, it indicates an extremely bright or white colour. A low L* value indicates a loss of brightness (Ucan, 2013).

The a* values of sour samples were found between 0.86 and 7.41. The highest and lowest values were determined in CUC and CSC. The b * values of the samples varied between 1.87-21.73. The highest and lowest b* values were determined in CUC and CPC, respectively. The a* value represents greenness (-a*) and redness (+a*) of a colour. The b* value represents blueness (-b*) and yellowness (+b*) of a colour. As expected, a* values and b* values were found in positive values. The hue angle (°) represents a specific red, blue, yellow, or green colour, or any combination of colours (Mohd-Hanif *et al.*, 2016). Hue values of sour samples ranged from 44.32 to 79.43 and the highest and the lowest values were detected in HUC and HSC, respectively. Chroma indicates the intensity of a colour (Mohd-Hanif *et al.*, 2016). Chroma values of samples ranged from 2.55 to 22.95 and the highest and the lowest values were determined in CUC and CPC.

According to Öncül and Karabıyıklı (2016), regarding the heating processes applied on a foodstuff, some ingredients, such as protein and polysaccharide may, aggregate. In another study, Maskan (2006) reported that several heating processes (microwave, rotary vacuum and atmospheric heating concentration processes) significantly decreased the colour parameters (L*, a*, and b* values) of pomegranate

Table 2. Results of browning index, colour (L*, a*, b*, hue and chroma) and 5-hydroxymethylfurfural (HMF) belonging to sour samples.[1]

Samples[2]	Browning index (abs.)	L*	a*	b*	Hue	Chroma	HMF (mg/l)
HPC	397.8±3.00^{a}	3.42±1.34^{c}	2.38±0.32^{b}	3.67±0.79^{c}	56.72±3.07^{b}	4.38±0.82^{c}	8,373.78±14.58^{a}
CPC	26.16±0.24^{c}	1.14±0.15^{e}	1.72±0.42^{c}	1.87±0.19^{e}	47.76±7.92^{bc}	2.55±0.30^{d}	210.92±2.91^{c}
HSC	98.09±8.14^{b}	2.59±0.40^{cd}	2.26±0.40^{b}	2.33±1.07^{de}	44.32±8.12^{c}	3.27±1.04^{d}	446.51±4.25^{b}
CSC	22.23±0.63^{c}	1.96±0.31^{de}	0.86±0.27^{d}	2.94±0.25^{cd}	73.77±5.30^{a}	3.07±0.24^{d}	209.95±2.91^{c}
HUC	1.15±0.06^{d}	11.08±0.33^{b}	2.27±0.06^{b}	12.17±0.24^{b}	79.43±0.07^{a}	12.37±0.24^{b}	19.20±0.40^{e}
CUC	2.71±0.03^{d}	21.75±0.04^{a}	7.41±0.02^{a}	21.73±0.22^{a}	71.18±0.14^{a}	22.95±0.21^{a}	46.90±1.53^{d}

[1] Values followed by different superscripted letter within the same column are significantly different from each other ($P<0.05$).

[2] HPC = homemade pomegranate concentrate; CPC = commercial pomegranate concentrate; HSC = homemade sumac concentrate; CSC = commercial sumac concentrate; HUC = homemade unripe grape concentrate; CUC = commercial unripe grape concentrate.

juice. The extent of colour degradation increased with soluble solids content and lightness (L* value) decreased time-dependently.

HMF content of homemade and commercial sour samples, ranged from 19.20-8373.78 mg/l. The highest and lowest values were detected in HPC and HUC, respectively. Changes in the HMF content of the samples were found as statistically significant (P<0.05). It was mentioned in the extant literature that the HMF contents of lemon sour samples in vacuum and atmospheric conditions were determined as 1.61 and 960.16 mg/l, respectively (Ucan *et al.*, 2014). It was reported that the HMF contents values of bitter orange sour samples in vacuum and atmospheric conditions were detected as 1.21 and 1,548.21 mg/l (Akcalıoglu *et al.*, 2014). In another study, it was stated that the average HMF value of pomegranate sour samples was 2,875.72 mg/kg and this value was above of pomegranate sour standard (50 mg/kg) by (Metin, 2014).

Total phenolics, total flavonoids, DPPH (inhibition%) and ascorbic acid contents of sour samples

Total phenolic content of samples ranged from 775.26 to 9,3543.81 mg/l and the difference between treatments were found statistically significant (P<0.05) (Table 3). The highest and the lowest values were determined in HSC and HUC, respectively.

Karakaplan and Özcan (2017) reported that the phenolic content of freshly squeezed pomegranate juice was higher than that of pomegranate juice concentrate. Researchers also mentioned that the boiling and pasteurisation processes decrease the phenolic compounds of the juices. It was reported that the total phenolics of lemon sour samples in vacuum and atmospheric conditions were 2,651.88 and 2,924.03 mg/l (Ucan *et al.*, 2014). In another study, total phenolic content of bitter orange sour samples in vacuum and atmospheric conditions were detected to be 1,628.28 and 1,956.80 mg/l (Akcalıoglu *et al.*, 2014).

Total flavonoid contents of samples ranged from 8.18 to 211.77 mg/l and the difference between homemade and commercial sour samples. The difference between values was found to be statistically significant (P<0.05) (Table 3). The highest and the lowest values were determined in HSC and CSC, respectively. Grape berries and derived products known to be a good source of phenolic compounds, particularly flavonoids, at high concentrations of 1000-1.800 mg/ml (Öncül and Karabıyıklı, 2016).

Antioxidants (such as vitamin E, C and β-carotene) and polyphenol content are essential components of the human diet. Antioxidants are known to play a key role in the prevention of oxidative damage in the cells. Antioxidants are also used as food additive in order to prevent food degradation (Aloqbi *et al.*, 2016). According to revealed results (Table 3) inhibition values ranged from 1.44 to 94.69% and the difference between treatments was found to be statistically significant (P<0.05). The highest and the lowest values were determined in HSC and CPC, respectively. Aloqbi *et al.* (2016) determined the inhibition (%) of pomegranate juice as 14.4, 27.5 and 37.9% for three concentrations (0.05, 0.1 and 0.15 mg/ml), respectively.

It was reported that the antioxidant activities of lemon sour samples in vacuum and atmospheric conditions were ranged between 90.17 and 77.83% (Ucan *et al.*, 2014). The highest values were detected in vacuum condition. The loss of ascorbic acid content in the lemon sour sample produced under atmospheric conditions was (48.36%) higher than the sample produced under vacuum. In another study, antioxidant activities of bitter orange sour samples in vacuum and atmospheric conditions were examined and antioxidant activity values of 84.67 and 70.85%, respectively, were found (Akcalıoglu *et al.*, 2014).

Ascorbic acid contents of sour samples ranged from 49.87 to 839.23 mg/l and the highest and the lowest values were detected in CSC and HUC, respectively. Furthermore, the difference of between treatments was found to be

Table 3. Results of total phenolics, total flavonoids, antioxidant activity and ascorbic acid belonging to sour samples.[1]

Samples[2]	Total phenolics (mg/l)	Total flavonoids (mg/l)	DPPH (% inhibition)	Ascorbic acid (mg/l)
HPC	6,785.57±290.72^{b}	93.45±2.27^{b}	37.30±1.38^{b}	656.92±28.85^{b}
CPC	3,946.39±49.48^{c}	12.73±1.67^{cd}	1.44±0.37^{e}	524.62±52.34^{c}
HSC	93,543.81±1,533.51^{a}	211.77±2.62^{a}	94.69±0.17^{a}	824.52±6.73^{a}
CSC	3,098.97±74.23^{c}	8.18±5.90^{d}	13.14±1.33^{d}	839.23±12.12^{a}
HUC	775.26±48.45^{d}	11.65±0.65^{cd}	28.94±0.99^{c}	49.87±0.87^{d}
CUC	896.39±10.82^{d}	14.48±0.43^{c}	35.00±6.05^{b}	63.65±1.20^{d}

[1] Values followed by different superscripted letter within the same column are significantly different from each other (P<0.05).
[2] HPC = homemade pomegranate concentrate; CPC = commercial pomegranate concentrate; HSC = homemade sumac concentrate; CSC = commercial sumac concentrate; HUC = homemade unripe grape concentrate; CUC = commercial unripe grape concentrate.

statistically significant (P<0.05) (Table 3). It is known that the ascorbic acid is used as an additive and also a chelating agent in juice industry in order to prevent unwanted colour change and Maillard's reaction in a food product (Torkamani and Niakousari, 2011). This acid is a heat-sensitive bioactive compound in the presence of oxygen and it easily degrades by oxidative processes (Alothman *et al.*, 2009).

The ascorbic acid content of sour samples in vacuum and atmospheric conditions were detected to be 1.53 and 0.79 g/l, respectively by (Ucan *et al.*, 2014). For the bitter orange sour samples, ascorbic acid contents in vacuum and atmospheric conditions were determined as 2.44 and 1.43 g/l by (Akcalıoglu *et al.*, 2014).

Sensory analysis of sour samples

Sensory quality plays a crucial role in consumer preferences. A panel was composed of seven experienced assessors having ages from 25 to 45 years from our department. For sensory evaluation, five criteria were identified in sour samples; colour, clarity, taste, smell and general impression.

The sensory analysis results are depicted in Table 4. The colour values of samples ranged from 2.21 to 5.50; clarity values ranged from 2.57 to 6.43; taste values 3.36 to 5.43; smell values 3.57 to 5.43 and general impression values ranged from to 4.93 to 5.57 and the changes were statistically not significant (P>0.05). The best scores for colour property were obtained from CSC sample (5.50). Clarity and taste criteria were unpopular in HSC sample. Regarding the smell of sour samples, the most preferred one was HPC. The general impression is a sensory property which describes the appreciation of a product; therefore, this property is very important for a foodstuff. In general impression, the highest score was the CPC sample.

Eyigün (2012) reported that the pomegranate juice produced in the atmospheric condition had better sensory properties rather than the vacuum packed The pomegranate juice is originally quite dark and thick. Due to this reason, the pomegranate produced under vacuum is not liked because of its unusual colour and consistency. However, the colour and natural properties of the product produced under vacuum are better protected than other production methods. It is thought that the consumers will embrace the pomegranate sour in new features when production is transferred at the industrial level.

Thin layer chromatography (determination of active fractions)

Thin layer chromatography profiles of sour samples are depicted in Figure 1. The red-pink spots on the pale back plate evaluated as citric acid (0.84 Rf). As seen Figure 1, Rf values of yellow spots in CSC(2), HPC(3), and CPC(4) examples were 0.14, 0.3 and 0.08, respectively. However, a specific organic acid presence associated with spots was not detected. Rf values in samples belonging to spots recorded as 0.83 (HSC(1), CSC(2) and HPC(3)) and 0.8 (CPC(4), CUC(5) and HUC(6)).

Microbial analysis of sour samples

Antimicrobial activity on test microorganisms are given in Table 5.According to the results, sour samples were not effective against all micro-organisms tested. None of them were active on *C. albicans* (Figure 2). However, HSC, CSC, CUC and HUC samples were more active than vancomycin (Figure 3).

The highest antimicrobial activity on *E. coli* was recorded with 15 mm inhibition zone in HSC sample. The antimicrobial activity on *S. aureus*, the highest antimicrobial activity (23 mm) detected in HSC sour sample (Figure 4). However, the CUC and HUC sour samples affecting *E. coli*, did not show inhibition against *S. aureus*. On the other hand, HPC and CPC samples were inactive against *E. coli*, effective against *S. aureus.*

Table 4. Results of sensory analysis belonging to sour samples.[1]

Samples[2]	Colour	Clarity	Taste	Smell	General impression
HPC	5.14±2.61^{ab}	3.21±2.48^{ab}	4.43±2.92^{a}	5.43±2.23^{a}	4.93±3.21^{a}
CPC	4.93±2.81^{ab}	4.86±3.47^{ab}	5.29±2.91^{a}	3.57±2.98^{a}	5.57±2.71^{a}
HSC	4.57±3.02^{ab}	2.57±2.09^{b}	3.36±2.95^{a}	4.79±2.16^{a}	4.21±2.45^{a}
CSC	5.50±2.66^{a}	4.79±3.16^{ab}	5.43±3.25^{a}	3.79±2.96^{a}	5.50±2.81^{a}
HUC	2.21±1.55^{b}	6.43±3.37^{a}	5.00±3.01^{a}	3.57±2.89^{a}	5.14±3.05^{a}
CUC	3.43±2.64^{ab}	5.79±2.78^{ab}	5.21±2.31^{a}	4.29±2.23^{a}	5.21±1.95^{a}

[1] Values followed by different superscripted letter within the same column are significantly different from each other (P<0.05).
[2] HPC = homemade pomegranate concentrate; CPC = commercial pomegranate concentrate; HSC = homemade sumac concentrate; CSC = commercial sumac concentrate; HUC = homemade unripe grape concentrate; CUC = commercial unripe grape concentrate.

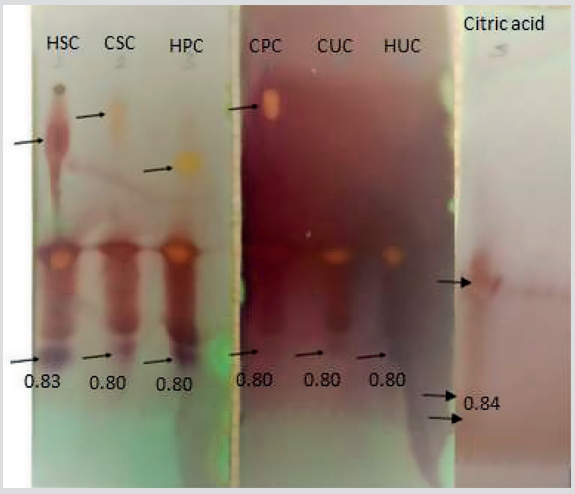

Figure 1. Thin layer chromatographic profiles of sour samples.

Table 5. Results of antimicrobial activity belonging to sour samples (mm).[1]

Samples	*Escherichia coli*	*Staphylococcus aureus*	*Candida albicans*
(1) HSC	15	23	–
(2) CSC	10	20	–
(3) HPC	–	12	–
(4) CPC	–	17	–
(5) CUC	10	–	–
(6) HUC	10	–	–
Vancomycin (5 mcg/disc)	6	16	–

[1] HPC = homemade pomegranate concentrate; CPC = commercial pomegranate concentrate; HSC = homemade sumac concentrate; CSC = commercial sumac concentrate; HUC = homemade unripe grape concentrate; CUC = commercial unripe grape concentrate.

Figure 2. Antimicrobial effect of sour samples against *Candida albicans.*

Figure 3. Antimicrobial effect of sour samples against *Escherichia coli.*

Figure 4. Antimicrobial effect of sour samples against *Staphylococcus aureus.*

Antimicrobial mechanism of sour samples and their effects on bacteria is not clear. Different behaviours related to the characteristic mechanisms such as penetration into the cell and/or membrane stability and permeability degradation, may be explained against the tested Gram positive and Gram negative bacteria. The HSC and CSC sour samples showed antimicrobial activity on both of test bacteria. For, these samples the highest inhibition zone against *S. aureus* was determined (23 mm). These results supported the use of sour samples in traditional treatments against the food infection due to the synthesised enterotoxins from bacteria.

In light of the results obtained, the most effective sour sample on the test bacteria was a homemade sumac sour (23 mm) and it showed a higher inhibition zone than the commercial product (20 mm). There was no difference in the antimicrobial behaviour of the homemade and commercial unripe grape juices. Compared to the homemade pomegranate juice, the highest inhibitory effect on *S. aureus* observed only in the commercial sample.

Mainly, the antimicrobial effects of unripe grape products depend on their organic acid contents. The acidity of the samples was affected by the type (acetic, benzoic, etc.) and concentration of the organic acid. Xiong *et al.* (1998) reported that the grape seed extract reduced to the *Salmonella typhimurium* attached on chicken skin between 1.6 and 1.8 log at 0.1% and 0.5% concentrations. In another study, the grape bagasse extracts had no inhibitory effects

on the tested 15 bacteria, while the grape seed extracts inhibited all the bacteria except *Bacillus amyloliquefaciens* at 20% concentration (Baydar *et al.*, 2004). Jayaprakasha *et al.* (2003) reported that Gram-positive bacteria were inhibited at the lower concentrations of grape seed extracts than Gram-negative bacteria (Karapinar and Sengun, 2007). In another research, the inhibitory effect of neutralised unripe grape products on foodborne pathogen due to their rich phenolic properties was indicated (Karabiyikli and Öncül, 2016).

Öncül and Karabıyıklı (2016) reported that the tested pathogens in unripe grape products could not survived after 5 min treatment at low inoculation doses. The inhibitory effect of the samples was associated with initial dose and application time. Gram positive (*S. aureus* and *Listeria monocytogenes*) and Gram negative (*E. coli* and *S. typhimurium*) bacteria strains were tested. The cell wall structure of micro-organisms was not affected. Rouhi-Boroujeni *et al.* (2016) reported along with antimicrobial and antioxidant effects of sumac, the level of serum lipids can be effectively reduced by following use of this agent, especially in combination of anti-lipidemic drugs.

4. Conclusions

In this study, some quality parameters of pomegranate, sumac and unripe grape concentrate sour samples sold in Kilis markets were investigated. The samples having the highest pH and TA values were determined in HPC and HSC samples. The highest and the lowest HMF contents and browning index values were detected in HPC and HUC, respectively. The highest total phenolics, total flavonoids, ascorbic acid and antioxidant activity values were found in HSC. TLC results stated that a specific organic acid presence associated with spots was not detected. The antimicrobial activity results supported the use of sour samples in traditional treatments against the food infection factors due to the synthesised enterotoxins from bacteria. Besides their flavour and condiment uses of the sour products, it could be said that they are good source for human health. In the recent years, there is an increasing interest in functional and natural foods. As traditional food additives and flavourings, sour products should be evaluated as natural food flavouring agents in the food industry.

Conflict of interest statement

The authors declare that there is no conflict of interests regarding the publication of this paper.

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