



Research Article

ANTIOXIDANT POTENTIAL OF POMEGRANATE LEMONADE

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ABSTRACT

Pomegranate (*Punica granatum*) belongs to the *Punicaceae* family and is used to cure diseases such as stomach ailments, cardiovascular disorders, skin allergies and inflammation. Pomegranate is used as fresh in very short time period due to scarcity of value added products however its juice can be prepared. So, current study is designed to prepare blend of pomegranate lemonade and to assess its sensorial and chemical composition. At first pomegranate aril powder was prepared and analyzed for composition including mineral contents and antioxidant potential. Afterwards, pomegranate lemonade was prepared and its acceptability was measured during storage period by consumers. The results showed that aril powder contained phenolic contents (TPC), ferric reducing antioxidant power (FRAP) and 2,2 diphenyl-1-picrylhydrazyl (DPPH) as 369.15±7.92 mg/100g GAE, 85.51±1.19 mM FRAP/g and 80.68±0.43 %, respectively. In the product development lemon juice was added at 5%, 10% and 15% to prepare pomegranate lemonade and sensory score were best at 5% addition of lemonade and then decreased linearly. Similar decreasing trend was observed during storage period with acceptable up to 5th day. Lemon juice results in decrease in pH, sugar content of lemonade while increase in acidity whereas I*, a* and b* values changed slightly with addition of lemon juice. So, it was be observed that pomegranate lemonade is acceptable at 5% level up to 5th storage day.

Keywords: Pomegranate lemonade, Phenolic contents, DPPH, FRAP, pH, Acidity

Introduction

Pomegranate (*Punica granatum*) is a minor fruit of Pakistan which belongs to the *Punicaceae* family and grows in tropical and sub-tropical regions. Pomegranate is round fruit with hard, shiny red-yellow skin. It is a delicious fruit consumed in both fresh and processed form. The word pomegranate comes from Latin word *pomum* (apple) and *granatum* (seeded). The shrub of pomegranate is up to 3-8 meters high. It can absorb the free radicals which are dangerous for human body (Waheed *et al.*, 2004). Phenolic, ascorbic acid, β-carotene and anthocyanins are bioactive components of pericarp of pomegranate. This fruit has various minerals like magnesium (Mg), sodium (Na), potassium (K), nitrogen (N) and (Ca) calcium (Holland *et al.*, 2009).

Pomegranate fruit is divided into three parts such as 15.8 % arils, 45 % juice and 39.2 % peel (Lansky and Newman, 2007). There are various nutrients present in per 100 grams of pomegranate arils. Pomegranate arils have 70 mg phosphorous, 10 mg calcium, 236 mg minerals, 3g protein, 18.70 g carbohydrates, 77.93 g moisture, 16 mg vitamin C, 0.12 g fats, 7 g fiber, 65 calorific value, 0.3 mg iron and 83 kcal energy (Adams *et al.*, 2006). Half cup of pomegranate arils consists of 1 gram fat, 16 grams carbohydrates, 1.5 grams protein and it provides 72 percent calories. Vitamin C which is helpful for immune

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system and clotting of blood present in high amount in arils. The most important mineral of the body i.e. potassium is present in the arils and is essential for muscle synthesis, normal regulation of blood pressure and nerve transmission system. Arils of pomegranate contain potassium, antioxidants and vitamins three times more than green tea. Pomegranate arils are dried in the form of anardana and used in curry development (Volkova *et al.*, 2000).

Fruit of pomegranate is used in the form of whole fruit, processed syrups and processed fruit juices. Pomegranate fruit is used to prepare many food products such as jellies, jam, anardana, juices, squashes & fruit salads. Pomegranate based value added products have gained a great status in the market for their high quality and nutritional value. Fruit of pomegranate has low quantity of fat while has high quantity of carbohydrates and protein contents as compared to fat (Fadavi *et al.*, 2005). Pomegranate reduces LDL cholesterol from the body (Basu and Penugonda, 2009). Various diseases can be prevented by the use of pomegranate (peel, juice and arils) such as stomach ailments, cardiovascular disorders, skin allergies and inflammation (Saxena and Vikram, 2004).

Due to high value, easy accessibility, health benefits and exceptional sensory characteristics processed pomegranate juice has become popular. Juices of pomegranate contain crude fiber, vitamins, crude fat, pectin, sugars, crude proteins and minerals (Viuda *et al.*, 2012). The blend of lime extract and pomegranate juice has been highlighted as positive method to preserve and improve the quality of the pomegranate juice (Gonzalez *et al.*, 2009). Pomegranate juices contain antioxidants which can maintain blood pressure and cholesterol level in the body (Guo *et al.*, 2008). So current study is designed to development of pomegranate lemonade and to assess its antioxidant

potential and acceptance by consumers through sensory evaluation.

Material And Methods

Procurement Of Raw Material

Pomegranate was obtained from local markets of Multan on the basis of quality characteristics such as equality in shape, size, color and scratch free. Reagents and chemicals such as ferrous sulphate, sodium carbonate (Na_2CO_3), 1,1-Diphenyl-2-picrylhydrazyl (DPPH), sodium hydroxide (NaOH), sulfuric acid, Folon-Ciocalteu reagent, digestion tablets, TPTZ, petroleum ether, gallic acid, methanol, ferric chloride, acetone, sodium acetate buffer and hydrochloric acid were gained from Merck, Germany.

Pomegranate arils powder preparation

Peeling of fruit was done manually and arils were separated from peel. The arils portion was dried and dried arils were subsequently ground for aril powder preparation. Ground pomegranate arils powder packed in polythene zip bags and stored at refrigerator.

Pomegranate characterization

Ground pomegranate arils powder was subjected to define nutritive profiling to following analytical methods including proximate analysis and mineral assay.

In pre-weighed china dish, 3-5 g of sample was placed at $105\pm 5^\circ\text{C}$ for 24 hour in hot air oven and dried at $105\pm 5^\circ\text{C}$ in hot air oven according to the procedure explained in AACC (2000). For measuring of ash contents, 3-5 g sample of an oven dried powder was obtained in pre-weighed crucible and placed in muffle furnace at 550 to 650 C after charring. At the end weight of residue was noted as ash content. To measure fat content aril powder was placed in filter paper and placed in soxhlet apparatus. Petroleum ether was used to was remove fat from sample after two to three washing sample was taken out and dried in oven at 60-70 C. Fat content was calculated by measuring the weight loss (AACC 2000) Crude fiber of

defatted and oven dried samples was measured by digesting sample (2-3g) in 250 mL 1.25% H₂SO₄ (simmering) and after washing the residue in distilled water, the residues again digested in 250 mL 1.25% NaOH (simmering). Afterwards, residues were filtered and washed with distilled water. The weight of residues was noted and placed in muffle furnace at 550-650 C and weight loss was measured as crude fiber content. In long neck flask, 3-5 g sample was digested by using digestion tablets with 30 mL H₂SO₄ for 3-4 hours till the samples turned to light yellow in color. A volume of 250 mL of digested sample was made by using distilled water. In the distillation tube, 10 ml sample was taken for the distillation and solution of 25 mL of 40% NaOH was added in it. Methyl red indicator in solution of 10 mL of 4% boric acid was added and ammonia was captured in it during distillation process. This ammonia borate was then titrated against 0.1N H₂SO₄. At the protein was calculated by multiplying N% with 5.71. Nitrogen free extracts (NFE) concentration was calculated by subtracting protein, fat, ash contents, fiber and moisture contents from 100 which was specified under resulting method.

For determination of mineral elements of dried pomegranate arils powder, Flame Photometer and Atomic Absorption Spectrophotometer were used by following the guidelines of AOAC (2006).

Sample of about 2g was taken and 3 mL HClO₄ and 7 mL HNO₃ were added in 100 mL small conical flask. On hot plate for heating conical flask was subjected and when 2-3 mL solutions was left stop the heating. With 100 mL distilled water samples were diluted and filtration was done for removing several suspended or un-dissolved solids. For measuring of Na, K and Ca samples were run on Flame Photometer and for Fe and Mn samples were run on Atomic Absorption Spectrophotometer.

Sample extraction

Sample (juice) was extracted from pomegranate arils with methanol as solvent by using mortars and pestles and shaking was continued at least for 3 hours. Filtered juice was stored in ultralow freezer (-4°C) for determining of total phenolics, free radical scavenging activity (DPPH) and FRAP of pomegranate arils juice (Elkar *et al.*, 2011).

Free radical scavenging activity (DPPH Method)

Pomegranate arils juice was extracted for determination of radical scavenging property and ascorbic acid was used as standard in DPPH method (Sharma and Bhat, 2009). Methanol (200µg/mL) was taken in the beaker and methanol was added in 2 mL of juice extracts. A volume of 3.0 mL of juice with methanol was taken in the test tubes and 1 mL DPPH solution (200µM) was added to start the reaction. Absorbance was measured at 517nm after the reaction mixture was kept in dark chamber at a temperature of 30°C for 30 min. Ascorbic acid standard curve was drawn or obtained at different concentrations (10 µM to 100 µM) against DPPH. Free radical scavenging properties of methanolic extracts was determined as per gram (mgAAE/g) of juice extracts.

Total phenolic contents

Extracted juice was analyzed through using gallic acid as a standard for total phenolic contents as used by Li *et al.*, (2006). Methanol-water solution was used and extract of juice was liquefied in this solution. Sample of about 0.5mL from this solution was pipetted in test tubes. For this purpose, 2.5 mL of ten times Folin-Ciocalteu reagents were added in sample and sample was dissolved in these reagents followed by the addition of Na₂CO₃ solution to a volume of 2 mL of 7.5% to the sample. Reaction mixture and sample was allowed to save at 25°C for 30 min and absorbance of sample was evaluated at 760nm by using UV/vis Spectrophotometer. For formation of

standard curve, a standard ranging from 10-100 mg/L of gallic acid was involved. Total polyphenols were calculated as gallic acid equal to (mg gallic acid/g)

Ferric reducing antioxidant power (FRAP)

Extracted juice of pomegranate was evaluated for ferric reducing antioxidant power through the process of Zahin *et al.*, (2010). In a ratio of 1:10:1, ferric chloride (20mM), sodium acetate buffer (pH 3.6) and 10mM of 2,4,6-Tripyridyl-s-Triazine was used for preparation of FRAP reagent. In the test tube, 300 µl reaction mixture was pipetted after heating of mixture for 10 min at a temperature of 37°C. At 593nm of UV/vis spectrophotometer samples absorbance was measured when in FRAP reagent tubes 25ml of extracted juice were taken. Ferrous sulphate was used as standard from 100- 1000µM. The obtaining results of the sample were compared as Fe in µM.

Preparation of pomegranate lemonade drinks

For the preparation of pomegranate lemonade drinks, lemon and fully ripe pomegranate fruits were used. Running water was used carefully for washing of both fruits. Arils of pomegranate were gained by cutting pomegranate fruit into four quarters and juice was extracted by using the juicer. Extraction of lemon juice was done manually. Muslin cloth was used for filtration of juices. Lemon juice was added in pomegranate juice by setting different percentage as shown in Table 1. Food grade preservatives such as citric acid and carboxymethyl cellulose (CMC), 0.5 g food grade color, 2 drops of pomegranate flavor and 150 g sugar were added in the juice. Lemonade to examine including total sugars, pH, color, titerable acidity and sensory score of 0, 5 and 10 days was observed (Rosenblat *et al.*, 2006).

Table: 1. Treatment plan or pomegranate drinks development

| Treatments | Description |
|----------------|--|
| T ₀ | Whole pomegranate juice |
| T ₁ | Whole pomegranate juice +5% lemon juice |
| T ₂ | Whole pomegranate juice +10% lemon juice |
| T ₃ | Whole pomegranate juice +15% lemon juice |

Analysis of pomegranate drinks

Samples of pomegranate drinks from every four treatments were separately taken in a 50 mL small beaker. For washing of pH meter electrodes distilled water was used and on the digital pH meter reading of pH was noted by dipping electrode in the sample as described in AOAC (2006). This process was continued 3 times for each treatment. Every time, before running of the new sample, electrode was washed with distilled water.

Pomegranate drinks sample were evaluated for color values i.e. L*, a* and b* by using CIE-Lab Color Meter according to the guidelines explained by Duangmal *et al.* (2008). Therefore, sample of about 10 mL was taken and one-to-one values of color were determined as L* (lightness), a* (-a greenness; +a redness) and b* (-b blueness; +b yellowness). Color readings were noted at least 3 times of each drink.

Samples of juice were studied for estimation of total acidity following the procedure of AOAC (2006). In a small 50 mL beaker, 10 mL sample was taken for determination of acidity and phenolphthalein indicator was added in it. Against 0.1N NaOH solution

samples were titrated for persisting of light pink color.

Approximately, by using the following formula total acidity was calculated:

$$\text{Total acidity \%} = \frac{0.0064 \times 0.1N\text{NaOH} \times \text{mL of NaOH}}{\text{mL of sample}} \times 100$$

Total sugars of pomegranate lemonade were determined through refractometer by using the methods of AOAC (2006). For determination of total sugars, at first refractometer was cleaned with wet tissue and then dry tissue was used for drying it. Two drops of pomegranate lemonade were taken on it. Reading button was pressed and reading was noted at least three times on refractometer.

Sensory evaluation

For sensory characteristics of pomegranate lemonade drinks i.e. aroma, taste, appearance, color, overall acceptance and after taste by using 9-point Hedonic Scale system was evaluated (Meilgaard *et al.*, 2007). At least twenty five sensual panelists were selected for sensory evaluation. Evaluation performas were given to the judges and the aim of the study was informed to the panelists to record their scores.

Statistical analysis

Statistical difference between data was computed through Statistix 8.1 by using analysis of variance techniques (ANOVA) under CRD as per the instructions given by Monotgomery (2008). Mean values & graphs were prepared by using MS Excel 2013 and data was represented as mean±S.D.

RESULTS AND DISCUSSION

Chemical composition of Pomegranate aril powder

Nutritional profiling of pomegranate fruit arils clarified that ash contents, crude protein, crude fiber, moisture contents, nitrogen free extract (NFE) and crude fat were 77.67±2.52%, 2.15±0.86%, 1.44±0.30%, 3.76±1.89%, 1.43±0.72% and 13.55±7.33%, respectively shown in table 2. The arils of pomegranate contain high contents of moisture, fiber and ash as compared to fat and protein of arils of pomegranate.

The results are related to the results of Khan, (2009), Wang *et al.*, (2010) and Viuda *et al.*, (2010) who estimated moisture contents from 72.6 to 84.6 %, fat contents from 0.9 to 1.5 %, fiber contents from 3.4 to 5.0%, ash contents from 0.9 to 2.5 % respectively. The findings of protein are similar to the results of Dumas *et al.*, (2003), Toor *et al.*, (2006), Raffo *et al.*, (2006), Zarei *et al.*, (2011) and Borochoy *et al.*, (2009) who reported the protein contents from 0.05 to 1.6 % respectively. Similarities in the nutritional composition of pomegranate fruit were due to same variety, growing area, climatic condition, culture practices and maturity stage of fruit. Similarities in the protein contents are also dependent on the harvesting time.

Table. 2. Proximate composition (%) of pomegranate fruit arils

| Component Analyzed | Arils powder |
|--------------------|--------------|
| Moisture | 77.67±2.52% |
| Ash | 2.15±0.86% |

| | |
|------------------------------|----------------------|
| Fat | 1.44±0.30% |
| Fiber | 3.76±1.89% |
| Protein | 1.43±0.72% |
| Nitrogen free extracts (NFE) | 13.55±7.33% |
| Calcium (Ca) | 3.10±3.38 (mg/100g) |
| Sodium (Na) | 1.33±0.57 (mg/100g) |
| Potassium (K) | 54.57±7.00 (mg/100g) |
| Manganese (Mn) | 10.13±0.83 (mg/100g) |
| Iron (Fe) | 1.06±0.15 (mg/100g) |
| Total phenolic (mg/100g GAE) | 145.15±2.18 |
| DPPH (%) | 80.68±0.43 |
| FRAP (mM FRAP/g) | 85.51±1.19 |

Mineral composition

Pomegranate arils mineral contents were given away in table 2. In the current study, minerals such as Iron (Fe), Sodium (Na), Calcium (Ca), Manganese (Mn) and Potassium (K) were recorded as 3.10±3.38, 1.33±0.57, 54.57±7.00, 10.13±0.83 and 1.06±0.15 respectively. Potassium (K) and manganese (Mn) concentration of pomegranate arils was found the highest as compared to the calcium (Ca), sodium (Na) and iron (Fe). Important physiological component in human beings is sodium (N) that regulates the normal fluid balance in and out of the cells. Deficiency of potassium (K) may cause muscle cramp cardiac diseases, hypertension and mental diseases. Potassium helps in the transportation of glucose into the muscle cells. Calcium (Ca) is essential for development of bones, dental tissues and hormonal release. Manganese helps in bone growth and iron (Fe) is essential for hemoglobin formation.

Values for calcium, potassium, manganese, iron and sodium were estimated 3.0-12 mg/100g, 16.0-56.0 mg/100g, 9.0-14 mg/100g, 0.3-1.2 mg/100g and 1.0-5.0

mg/100g respectively related to the outcomes of Mirdehagan and Rahemi. (2007). Results of calcium, sodium and iron are similar to the findings of Yilmaz, (2007). The findings of potassium are similar to the findings of Davidson *et al.*, (2009) and the findings of manganese are similar to the findings of Tezcan *et al.*, (2009).

Antioxidant potential of pomegranate juice

Total phenolic contents observed values for, 2,2-diphenyl 1-1- picrylhydrazyl (DPPH) and ferric reducing antioxidant power (FRAP) were identified 369.15±7.92 (mg/100g GAE), 80.68±0.43 (%) and 85.51±1.19 (mM FRAP/g) respectively (Table 2). Pomegranate juice contains high amount of total phenolics as compared to DPPH and FRAP. Components and levels of phenolics improved from natural products depending on the type of solvent, particle size of plant material, extraction temperature and solvent-solid ratio. FRAP are considered constant and generally accepted analyzed techniques for the determination of total antioxidants

activity. Free radical scavenging give electrons to free radicals for converting in reasonably stable form based on their logical speed and simplicity (Akhter *et al.*, 2014).

Total phenolic contents observed values are similar to the results of Tzulker *et al.* (2007) who stated total phenolic levels 144 ± 2.13 mg/100g GAE of juice prepared from arils. The findings for (DPPH) are similar to the results of Gil *et al.*, (2000) that described activity of antioxidant (DPPH) 79.46 ± 0.54 %. The outcomes are related with the outcomes of Ozgen *et al.*, (2008) who identified the first time ferric reducing antioxidant power (FRAP) was 86.89 ± 1.76 mM FRAP/g. Pomegranate arils juice has higher total phenolics.

Sensory evaluation of pomegranate lemonade drinks during storage

Sensory evaluation determines the marketability of juice. Color, flavor and aroma generally reduce with increase in storage period. The color, flavor, aroma and appearance of the blended juice were found to be superior as compared to the juice prepared from individual fruit.

Flavor

Flavor of pomegranate lemonade drinks in T_0 showed significantly lowest score (4.87 ± 0.31) made by 0% lemon juice. While flavor showed highest score in T_1 , T_2 and T_3 were 6.80 ± 0.38 , 6.33 ± 0.35 and 5.07 ± 0.46 prepared by 5%, 10% and 15% lemon juice respectively (Table 3). Mean value of pomegranate drinks flavor was decreased by increase the level of lemon juice. Similarly, storage period also gives opposite effect on flavor. Mean value of storage at 0, 5 and 10 days were 6.00 ± 0.13 , 5.70 ± 0.43 and 5.60 ± 0.56 respectively as can be seen in Table 3. Day's effects of pomegranate lemonade drinks indicated that score of flavor were slowly reduced with rise period of storage on flavor score.

The outcomes of pomegranate lemonade drinks are similar to the results of Bhardwaj and Mukherjee, (2012) who justified a declining trend of drinks on flavor quality during storage days due to increasing in unpleasant smell of flavor drinks with the passage of time and while working on factors such as sensory, microbiological and physico-chemical quality of kinnow juice blends. Blending of orange juice and pomegranate juice found to be rejected and undesirable.

Aroma

Aroma of pomegranate lemonade drinks in T_0 showed significantly lowest score (5.07 ± 0.40) made by 0% lemon juice. While aroma showed highest score in T_1 , T_2 and T_3 were 7.07 ± 0.36 , 6.87 ± 0.42 and 5.20 ± 0.28 prepared by 5%, 10% and 15% lemon juice respectively. Mean value of pomegranate drinks aroma was slowly reduced with rise period of storage on aroma score by increasing the level of lemon juice. Similarly, storage period also gives opposite effect on aroma. Mean value of storage at 0, 5 and 10 days were 6.00 ± 0.396 , 6.05 ± 0.30 and 6.10 ± 0.41 respectively (Table 3). Day's effects of pomegranate lemonade drinks indicated that score of aroma were slowly reduced with rise period of storage on aroma score.

Results of pomegranate lemonade drinks are similar to the findings of Tiwari, (2000) who justified a declining trend in aroma quality of pomegranate lemonade drinks during storage days due to addition of acidity of lemon juice in pomegranate juice while working on the effect of sugarcane juice on pomegranate juice shelf life during storage days with the passage of time.

Color

Color of pomegranate lemonade drinks in T_1 showed significantly lowest score (5.87 ± 0.66) made by 5% lemon juice. While color showed highest score in T_0 , T_2 and T_3 were 6.13 ± 0.67 , 6.07 ± 0.61 and 5.93 ± 0.57 prepared by 0%, 10% and 15% lemon juice

respectively can be verified from Table 3. Treatments and day's interaction effects were slowly increased in T₁, T₂ and T₃ during period of storage days by increasing the level of lemon juice. Mean value of storage at 0, 5 and 10 days were 6.05±0.59, 6.00±0.71 and 5.95±0.58 respectively (Table 3). Day's effects of pomegranate lemonade drinks indicated that the color of drinks was slowly reduced with rise period of storage on color score by increasing the level of lemon juice. Results of pomegranate lemonade drinks are related to the results of Deka *et al.*, (2000) who described a rising trend in color quality of pomegranate lemonade drinks during storage days due to addition of different amount of color in all drinks while working on the effect of pear juice, ethanolic extract of ginger and garlic on pomegranate juice shelf life.

Appearance

Appearance of pomegranate lemonade drinks in T₀ showed significantly lowest score (4.87±0.47) made by 0% lemon juice. While

appearance showed highest score in T₁, T₂ and T₃ were 7.53±0.38, 7.00±0.41 and 5.20±0.31 prepared by 5%, 10% and 15% lemon juice respectively (Table 3). Day's effects of pomegranate lemonade drinks indicated that the appearance of drinks was slowly reduced with rise period of storage on color score by increasing the level of lemon juice. Treatments and days interaction effects of pomegranate lemonade drinks indicated that the appearance of drinks was slowly reduced in all treatments during period of storage days. Mean value of storage at 0, 5 and 10 days were 6.25±0.27, 6.15±0.43 and 6.05±0.47, respectively.

The outcomes of pomegranate lemonade drinks are similar to the results of Khare *et al.*, (2012) who justified a declining trend of pomegranate lemonade drinks on appearance quality of drinks with the passage of time while working on effects of sugarcane juice on the shelf life of pomegranate juice during storage.

Table 3. Sensorial Characteristics of pomegranate lemonade drinks during storage

| | Treatments | Storage | | | |
|--------|----------------|-----------|-----------|-----------|-----------|
| | | 0 | 5 | 10 | Mean |
| Flavor | T ₀ | 5.00±0.00 | 4.60±0.52 | 5.00±0.41 | 4.87±0.31 |
| | T ₁ | 7.00±0.00 | 6.80±0.48 | 6.60±0.66 | 6.80±0.38 |
| | T ₂ | 6.80±0.26 | 6.40±0.32 | 5.80±0.48 | 6.33±0.35 |
| | T ₃ | 5.20±0.26 | 5.00±0.41 | 5.00±0.71 | 5.07±0.46 |
| | Mean | 6.00±0.13 | 5.70±0.43 | 5.60±0.56 | |
| Aroma | T ₀ | 4.80±0.48 | 5.00±0.41 | 5.40±0.32 | 5.07±0.40 |
| | T ₁ | 7.40±0.32 | 7.20±0.26 | 6.60±0.52 | 7.07±0.36 |
| | T ₂ | 6.60±0.52 | 7.20±0.26 | 6.80±0.48 | 6.87±0.42 |
| | T ₃ | 5.20±0.26 | 4.80±0.26 | 5.60±0.32 | 5.20±0.28 |
| | Mean | 6.00±0.39 | 6.05±0.30 | 6.10±0.41 | |
| Color | T ₀ | 7.40±0.78 | 5.80±0.75 | 5.20±0.48 | 6.13±0.67 |
| | T ₁ | 5.20±0.63 | 6.20±0.48 | 6.20±0.86 | 5.87±0.66 |
| | T ₂ | 5.80±0.48 | 5.80±0.75 | 6.20±0.48 | 5.93±0.57 |
| | T ₃ | 5.80±0.48 | 6.20±0.86 | 6.20±0.48 | 6.07±0.61 |
| | Mean | 6.05±0.59 | 6.00±0.71 | 5.95±0.58 | |
| Taste | T ₀ | 5.00±0.41 | 6.40±0.32 | 5.20±0.26 | 5.53±0.33 |
| | T ₁ | 7.80±0.26 | 7.00±0.71 | 7.40±0.52 | 7.40±0.49 |

| | | | | | |
|------------------------------|----------------------|-----------|-----------|-----------|-----------|
| Appearance | T₂ | 7.80±0.48 | 6.80±0.48 | 6.40±0.52 | 7.00±0.49 |
| | T₃ | 5.40±0.52 | 5.40±0.32 | 6.00±0.41 | 5.60±0.41 |
| | Mean | 6.50±0.42 | 6.40±0.46 | 6.25±0.43 | |
| | T₀ | 4.80±0.26 | 4.80±0.75 | 5.00±0.41 | 4.87±0.47 |
| | T₁ | 7.60±0.32 | 7.60±0.32 | 7.40±0.52 | 7.53±0.38 |
| | T₂ | 6.80±0.26 | 7.20±0.26 | 7.00±0.71 | 7.00±0.41 |
| | T₃ | 5.80±0.26 | 5.00±0.41 | 4.80±0.26 | 5.20±0.31 |
| Overall Acceptability | Mean | 6.25±0.27 | 6.15±0.43 | 6.05±0.47 | |
| | T₀ | 6.60±0.66 | 5.00±0.71 | 5.00±0.41 | 5.53±0.59 |
| | T₁ | 5.60±0.66 | 6.20±0.63 | 5.00±0.71 | 5.60±0.67 |
| | T₂ | 7.20±0.63 | 6.00±0.71 | 4.80±0.75 | 6.00±0.70 |
| | T₃ | 5.60±0.32 | 5.60±0.66 | 4.20±0.95 | 5.13±0.64 |
| | Mean | 6.25±0.57 | 5.70±0.68 | 4.75±0.71 | |

T₀: Whole pomegranate juice, T₁: Whole pomegranate juice +5% lemon juice, T₂: Whole pomegranate juice +10% lemon juice, T₃: Whole pomegranate juice +15% lemon juice

After taste

After taste of pomegranate lemonade drinks in T₀ showed significantly lowest score (5.53±0.33) made by 0% lemon juice. While after taste showed highest score in T₁, T₂ and T₃ were 7.40±0.49, 7.00±0.49 and 5.60±0.41 prepared by 5%, 10% and 15% lemon juice respectively. Treatments and days interaction effects of pomegranate lemonade drinks indicated that the after taste of drinks was slowly reduced in all treatments during period of storage days. Mean value of storage at 0, 5 and 10 days were 6.50±0.42, 6.40±0.46 and 6.25±0.43, respectively (Table 3).

The outcomes of pomegranate lemonade drinks are same to the outcomes of Lawless *et al.*, (2013) who justified a reducing trend on after taste quality during days of storage due to disagreeable smell of lemon juice and flavor while working on the effect of grape juice on pomegranate juice shelf life.

Overall acceptance

Overall acceptance of pomegranate lemonade drinks in T₃ showed significantly lowest score (5.13±0.64) made by 15% lemon juice. While overall acceptance showed highest score in T₀, T₁ and T₂ were

7.40±0.49, 7.00±0.49 and 5.60±0.41 prepared by 0%, 5% and 10% lemon juice respectively. Day's effects of pomegranate lemonade drinks indicated that score of overall acceptance were slowly reduced with rise period of storage on overall acceptance score. Treatments and day's interaction effects of pomegranate lemonade drinks indicated that overall acceptance of drinks was slowly increased in T₀, T₁ and T₂ treatments during period of storage days. Mean value of storage at 0, 5 and 10 days were 6.25±0.57, 5.70±0.68 and 4.75±0.71 respectively (Table 3).

The outcomes of drinks are similar to the results of Jeong *et al.*, (2009) who justified a rising trend of pomegranate lemonade drinks on overall acceptance quality due to addition of different quantity of lemon juice in pomegranate juice while working on effects of orange juice on the shelf life of pomegranate juice during storage.

Analysis of pomegranate lemonade drinks during storage pH

Treatments effects of pomegranate lemonade drinks on pH value of drinks indicated that significantly the lowest value was given to the T₃ drink for pH made by 15% lemon juice

with mean values 2.89 ± 0.00 . Whereas, the highest value was given to the T₀ T₁ and T₂ drinks prepared by 0%, 5% and 10% lemon juice for pH with mean values 3.80 ± 0.00 , 3.21 ± 0.00 and 3.01 ± 0.00 , respectively (Table 4). Day's effects of pomegranate lemonade drinks indicated that pH was slowly reduced with rise period of storage on pH value. Treatments and day's interaction effects of pomegranate lemonade drinks indicated that pH value of drinks was slowly reduced in all treatments during period of storage days. Mean value of storage at 0, 5 and 10 days were 3.44 ± 0.00 , 3.29 ± 0.00 and 2.95 ± 0.00 , respectively.

Results of drinks are same to the results of Akponah *et al.*, (2013) who stated a declining trend of value of pH due to addition of lime acidity while working on garlic juice effects on pomegranate juice shelf life during storage days. Reason is that pH values reduced in the mixtures of pomegranate and lemon juice when 25% of lemon juice was added in 75% of pomegranate juice and when 75% of lemon juice was added in 25% of pomegranate juice ranging from 2.80 to 2.54, respectively.

Total acidity

Treatments effects of pomegranate lemonade drinks on total acidity value of drinks indicated that significantly the lowest value was given to the T₀ drink for total acidity value made by 0% lemon juice with mean values 0.47 ± 0.01 . Whereas, the highest value was given to the T₁, T₂ and T₃ drink prepared by 5%, 10% and 15% lemon juice for total acidity with mean values 0.62 ± 0.02 , 0.54 ± 0.0 and 0.50 ± 0.03 , respectively as can be seen in Table 4. Treatments and day's interaction effects of pomegranate lemonade drinks indicated that total acidity value of drinks was slowly increased in all treatments during period of storage days. Mean value of storage at 0, 5 and 10 days were 0.50 ± 0.01 , 0.54 ± 0.01 and 0.56 ± 0.02 , respectively.

Results are related to the outcomes of Aradhya and Kulkarni, (2005) who described that acidity value increased in the mixture of pomegranate and lemon juice through storage days when working on the effect of juice of lemon on pomegranate juice shelf life. Pomegranate lemonade drinks acidity increased during storage by the addition of citric acid which is present in lemon juice.

Total sugars

Treatments effects of pomegranate lemonade drinks on total sugar value of drinks indicated that significantly the lowest value was given to the T₃ drink for total sugar value made by 15% lemon juice with mean values 12.59 ± 0.03 . Whereas, the highest value was given to the T₀, T₁ and T₂ drink prepared by 0%, 5% and 10% lemon juice for total sugar with mean values 14.00 ± 0.07 , 13.70 ± 0.04 and 12.81 ± 0.06 , respectively (Table 4). Treatments and day's interaction effects of pomegranate lemonade drinks indicated that total sugar value of drinks was slowly reduced in all treatments during period of storage days. Day's effects of pomegranate lemonade drinks indicated that total sugar value was slowly reduced with rise period of storage days on total sugar value. Mean value of storage at 0, 5 and 10 days were 14.78 ± 0.07 , 13.65 ± 0.08 and 11.39 ± 0.26 , respectively.

Outcomes of pomegranate lemonade drinks are related to the results of Melgarejo *et al.*, (2000) who stated that sugar contents of all drinks reduced with the passage of time through increasing of acidity in pomegranate juice when effects the pomegranate juice shelf life.

L* value

Treatments effects of pomegranate lemonade drinks on the color L* value of drinks indicated that significantly the lowest value was given to the T₀ drink for color L* value made by 0% lemon juice with mean values 22.59 ± 0.38 . Whereas, the highest value was given to the T₁, T₂ and T₃ drink prepared by

5%, 10% and 15% lemon juice for color L* value with mean values 25.03±0.25, 25.00±0.25 and 23.94±0.41, respectively. Treatments and days interaction effects of pomegranate lemonade drinks indicated that the color L* value of drinks was slowly reduced in all treatments during period of storage days. The lowest value was given to the drinks at 10 days of period of storage with mean value 23.21±0.54 and the highest value was given to the drink at the start of storage for color L* value with mean value

25.12±0.56. Mean value of storage at 0, 5 and 10 days were 25.12±0.56, 24.09±0.57 and 23.21±0.54, respectively which can be viewed in Table 4.

Outcomes of pomegranate lemonade drinks are related to the results of Boulton, (2001) who stated that color L* value of all drinks increased with the passage of time due to addition of different compounds such as flavanones of lemon juice in pomegranate juice when effects the pomegranate juice shelf life.

Table 4. Chemical parameters of pomegranate lemonade drink during storage

| | Treatments | Storage | | | |
|--------------------|----------------------|------------|------------|------------|------------|
| | | 0 | 5 | 10 | Mean |
| pH | T₀ | 4.07±0.00 | 4.06±0.00 | 3.28±0.00 | 3.80±0.00 |
| | T₁ | 3.45±0.00 | 3.29±0.00 | 2.89±0.00 | 3.21±0.00 |
| | T₂ | 3.21±0.00 | 2.99±0.00 | 2.84±0.00 | 3.01±0.00 |
| | T₃ | 3.04±0.00 | 2.85±0.00 | 2.77±0.00 | 2.89±0.00 |
| | Mean | 3.44±0.00 | 3.29±0.00 | 2.95±0.00 | |
| Acidity | T₀ | 0.47±0.01 | 0.48±0.00 | 0.47±0.01 | 0.47±0.01 |
| | T₁ | 0.48±0.00 | 0.49±0.01 | 0.55±0.02 | 0.50±0.03 |
| | T₂ | 0.52±0.01 | 0.55±0.01 | 0.54±0.01 | 0.54±0.01 |
| | T₃ | 0.55±0.01 | 0.62±0.02 | 0.68±0.05 | 0.62±0.05 |
| | Mean | 0.50±0.01 | 0.54±0.01 | 0.56±0.02 | |
| Total Sugar | T₀ | 15.27±0.03 | 14.43±0.12 | 12.30±0.06 | 14.00±0.15 |
| | T₁ | 15.13±0.03 | 14.37±0.15 | 11.60±0.26 | 13.70±0.08 |
| | T₂ | 14.43±0.15 | 13.13±0.03 | 10.87±0.37 | 12.81±0.06 |
| | T₃ | 14.30±0.07 | 12.67±0.03 | 10.80±0.35 | 12.59±0.03 |
| | Mean | 14.78±0.07 | 13.65±0.08 | 11.39±0.26 | |
| L* | T₀ | 23.62±0.41 | 22.57±0.38 | 21.58±0.30 | 22.59±0.38 |
| | T₁ | 24.95±0.58 | 23.85±0.53 | 23.01±0.56 | 23.94±0.41 |
| | T₂ | 25.87±0.17 | 25.04±0.25 | 24.09±0.43 | 25.00±0.17 |
| | T₃ | 26.03±1.10 | 24.89±1.13 | 24.16±0.86 | 25.03±0.25 |
| | Mean | 25.12±0.56 | 24.09±0.57 | 23.21±0.54 | |
| a* | T₀ | 0.82±0.04 | 0.91±0.06 | 0.69±0.04 | 0.81±0.04 |
| | T₁ | 1.10±0.04 | 1.19±0.03 | 1.27±0.08 | 1.19±0.03 |
| | T₂ | 2.04±0.10 | 2.09±0.06 | 2.01±0.14 | 2.05±0.10 |
| | T₃ | 2.26±0.06 | 2.27±0.07 | 2.31±0.05 | 2.28±0.07 |
| | Mean | 1.56±0.06 | 1.61±0.06 | 1.57±0.08 | |
| b* | T₀ | 1.13±0.07 | 1.15±0.07 | 1.17±0.10 | 1.15±0.07 |
| | T₁ | 0.24±0.05 | 0.31±0.06 | 0.30±0.07 | 0.28±0.05 |
| | T₂ | 0.43±0.10 | 0.46±0.12 | 0.45±0.12 | 0.45±0.12 |

| | | | | | |
|--|----------------------|-----------|-----------|-----------|-----------|
| | T₃ | 0.35±0.08 | 0.45±0.18 | 0.39±0.11 | 0.40±0.11 |
| | Mean | 0.54±0.07 | 0.59±0.11 | 0.58±0.10 | |

T₀: Whole pomegranate juice, T₁: Whole pomegranate juice +5% lemon juice, T₂: Whole pomegranate juice +10% lemon juice, T₃: Whole pomegranate juice +15% lemon juice

a* value

Treatments effects of pomegranate lemonade drinks on the color a* value of drinks indicated that significantly the lowest value was given to the T₀ drink for color a* value made by 0% lemon juice with mean values 0.81±0.04. Whereas, the highest value was given to the T₁, T₂ and T₃ drink prepared by 5%, 10% and 15% lemon juice with mean values 2.28±0.07, 2.05±0.10 and 1.19±0.03, respectively. Treatments and days interaction effects of pomegranate lemonade drinks indicated that the color a* value of drinks was slowly increased in all treatments during storage days. Suggestively the highest value was given to the T₃ drink for color a* value made by 15% lemon juice at 10 and 5 days of storage with mean values 2.31±0.05, 2.27±0.07, correspondingly. Mean value of storage at 0, 5 and 10 days were 1.56±0.06, 1.61±0.06 and 1.57±0.08, respectively (Table 4).

Outcomes of pomegranate lemonade are same to the results of Boulton, (2001) who described that color a* value of all drinks generally rised up within storage days when effects the pomegranate juice shelf life.

b* value

Treatments effects of pomegranate lemonade drinks on the color b* value of drinks indicated that significantly the lowest value was given to the T₁ drink for color b* value made by 5% lemon juice with mean values 0.28±0.05. Whereas, the highest value was given to the T₀, T₂ and T₃ drink prepared by 0%, 10% and 15% lemon juice for color b* value followed with mean values 1.15±0.07, 0.45±0.18 and 0.40±0.11, respectively. Treatments and days interaction effects of pomegranate lemonade drinks indicated that the color b* value of drinks was slowly

quadrated in all treatments during storage days. Considerably, the lowest value was given to the drinks at 0 days of storage with mean value 0.54±0.07 and the highest value was given to the drink at 5 days of storage for color b* value with mean value 0.59±0.11. Mean value of storage at 0, 5 and 10 days were 0.54±0.07, 0.59±0.11 and 0.58±0.10, respectively (Table 4).

Outcomes of pomegranate lemonade drinks of color b* value (yellowness) are similar to the results of Boulton, (2001) who justified that color b* value of all drinks reduced during storage days due to the variation in temperature and presence of citric acid of lemon juice.

Conclusion

Fruit juices industry is the fastest growing area in the world of the beverage industry. Fruit juices are important for human health. Pomegranate juice has been used due to its beneficial effects. It has high antioxidant activity as compared to other fruit juices. Pomegranate arils being a byproduct of fruit processing industry, stands among the most explored commodities for its biological, pharmacological and nutritional features. The outcomes of the current study highlighted role of pomegranate arils powder and pomegranate arils juices as bio-preservatives against product associated physicochemical and microbiological spoilage and as functional food ingredient to serve the purpose of food security. The study further discussed the improvement of food quality. Pomegranate fruit plays an important role in disease chemoprevention.

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