**Biochemical Studies of Pomegranate Coloured Pear Beverages on Storage**

**Abstract**

A study on the biochemical changes in pear beverages during storage was conducted in the Laboratory of the Department of Horticulture, Faculty of Agriculture, Guru Kashi University, Talwandi Sabo, Bathinda, Punjab, India, during the academic year 2024-25. Pear is widely utilized in the food processing industry, valued for its naturally appealing pulp colour, which significantly enhances the visual attractiveness of food products. Colour is a critical quality attribute in processed foods, playing a key role in consumer perception and marketability. In the present study, various quantities of pomegranate seeds were incorporated to naturally colour beverages prepared from white-fleshed pear, concentrating on Ready-to-Serve (RTS) beverages and squash. The results indicated that the use of 80 gm of pomegranate seeds per litre was optimal for achieving desirable colour in both RTS and squash formulations.

**Keywords:** RTS, squash, storage, pear, pomegranate

**Introduction**

Pear (*Pyrus communis* L.) is an important temperate fruit crop belonging to the family Rosaceae. It has been grown since ancient times and is said to have originated in parts of Western Asia and Eastern Europe. Pears are now grown extensively in temperate and subtropical climates in China, India, the United States, and Europe (**Gill *et al*. 2012**). The fruit is well-liked for its nutritious value, sweet taste, and juicy texture. They are high in antioxidants, vitamins (especially C and K), and dietary fiber. Pears and apples have many nutritional characteristics because of their botanical relationship. Pears are a good source of dietary fiber (4.1 g), calories (233 kg), protein (0.3 g), fat (0.03), carbohydrate (12.4 g), sugar (9.8 g), vitamin C (4 mg), and potassium (112 mg).

Additionally, pears contain a variety of phytochemicals or phytonutrients, including phenolic acid, flavonoids, stilbenes/lignans, and triterpenes. Phytochemicals are naturally occurring substances in plant-based foods that offer various health benefits, although they are not classified as essential nutrients like traditional vitamins and minerals. Pear skin, in particular, is rich in phenolic acids, which have been linked to several health-promoting effects, including the reduction of blood alcohol levels and the risk of conditions such as diabetes, cardiovascular diseases, and obesity. Pears also possess strong antioxidant properties, which contribute to wound healing, liver protection, and the management of allergic conditions such as rhinitis, asthma, and eczema (**Iqbal and Singh, 2018).** Pears are well-known for their high nutritional content and health benefits due to their abundance of minerals, fibers, amino acids, carbs, and bioactive substances.

Therefore, turning pears into pear wines or other fermented beverages, whether alcoholic or nonalcoholic, can reduce fruit waste and provide new value-added pear products. In our earlier work, we analyzed the chemical makeup of untreated juices from particular European pear cultivars and breeding selections that have not yet been commercialized, and the relationship between the primary phenolic traits and the usage grouping developed by breeders. In a previous study, providing breeders with chemical aims helped them create unique pear cultivars**.**

Pomegranate (Punica granatum L.) is an ancient fruit crop of significant horticultural and medicinal importance, belonging to the family Lythraceae. Native to the region from Iran to northern India, it has been cultivated for thousands of years and has now spread across tropical and subtropical regions worldwide. The plant is well-adapted to arid and semi-arid climates and is valued for its drought tolerance, hardy nature, and ability to thrive in a range of soil types. Morphologically, pomegranate is a deciduous or semi-deciduous shrub or small tree, typically reaching heights of 5-8 meters. It exhibits multiple stems with spiny branches and bears glossy, lanceolate leaves. The plant produces bright red, tubular flowers either singly or in clusters (**Singh, 2014; Mehta *et al*. 2018; Singh, 2017**). Pomegranate juice, extracted from the arils of the pomegranate fruit (*Punica granatum* L.), has gained significant commercial and nutritional importance in postharvest utilization. After harvesting, fruits that do not meet market standards due to size or minor blemishes are often processed into juice, reducing postharvest losses and increasing value addition. Pomegranate juice is known for its rich content of anthocyanins, tannins, vitamin C, and polyphenols, which contribute to its strong antioxidant properties.

 Pear is rich in malic acid and citric acid and pomegranate is rich in folic acid, calcium and iron. So mixing pomegranate colour in pear beverage would not only increase the eye appeal but it may also increase the nutritional value of pear beverages. With nutritional properties, anti-microbial property is also found in pomegranate colour and in many research it is revealed that pomegranate colour is also helpful in increasing the storage life of food products. So, adding pomegranate in pear beverages may also increase the storage life of pear beverages. Therefore, keeping the above points in view, the following objectives have been formulated for the investigation:

1. To analyze and quantify the chemical constituents present in pear pulp.
2. To assess the quality and characteristics of pomegranate-coloured pear beverages formulated with different seed concentrations.
3. To study the colour stability of pomegranate-enriched pear beverages over the storage period.

**Materials and Methods**

The experiment comprising of five treatments, i.e., (i) 10% pear pulp + acidity 0.25% + pomegranate seed nil + sodium benzoate 100ppm, (ii) 10% pear pulp + acidity 0.25% + pomegranate seed 40g + sodium benzoate 100ppm, (iii) 10% pear pulp + acidity 0.25% + pomegranate seed 80g + sodium benzoate 100ppm (iv) 10% pear pulp + acidity 0.25% + pomegranate seed 120g + sodium benzoate 100ppm, (v) 10% pear pulp + acidity 0.25% + pomegranate seed 160g + sodium benzoate 100ppm) for ready to serve beverage and five treatments, i.e., (i) 25% pear pulp + acidity 1.1% + pomegranate seed nil + sodium benzoate 600ppm, (ii) 25% pear pulp + acidity 1.1% + pomegranate seed 40g + sodium benzoate 600ppm, (iii) 25% pear pulp + acidity 1.1% + pomegranate seed 80g + sodium benzoate 600ppm, (iv) 25% pear pulp + acidity 1.1% + pomegranate seed 120g + sodium benzoate 600ppm, (v) 25% pear pulp + acidity 1.1% + pomegranate seed 160g + sodium benzoate 600ppm for squash was conducted in Laboratory of the Department of Horticulture, Faculty of Agriculture, Guru Kashi University, Talwandi Sabo, Bathinda, Punjab, India during the year 2024-25. Pear fruits were thoroughly washed under a gentle stream of tap water, cut into small pieces, and ground in a blender using water in a 1:1 ratio. The resulting mixture was strained through muslin cloth to obtain the pear pulp. Sodium benzoate was then added as a preservative at a concentration of 600 mg per litre of pulp and mixed thoroughly. The prepared pulp was filled into glass bottles, sealed hermetically with crown corks, pasteurized, and labeled for further use. The natural colour from pomegranate seeds was extracted by boiling the seeds in 100 ml of water for 10 minutes and allowing the mixture to stand overnight. The next day, the solubilized colour was filtered through muslin cloth and incorporated into pear beverages at different concentrations. For Ready-to-Serve (RTS) beverages, pomegranate extract was added at 0, 40, 80, 120, and 160 g per litre. The same concentrations were also used for pear squash. The syrup was prepared by heating a mixture of sugar, water, pomegranate extract, and citric acid, followed by filtration through muslin cloth. The pear pulp was then blended with the prepared syrup and filled into 200 ml bottles, leaving a 2 cm headspace. These bottles were hermetically sealed with crown corks. The entire procedure was replicated three times for consistency. The sealed RTS bottles were pasteurized in boiling water for 20 minutes and cooled at room temperature. Both RTS and squash samples were stored under ambient room temperature conditions for further storage studies. A panel of 10 semi-trained members conducted a sensory evaluation using a nine-point Hedonic scale, where a score of nine indicated “Like Extremely” and one indicated “Dislike Extremely” (Amerine *et al*., 1965). The biochemical parameters, including acidity, ascorbic acid, reducing sugars, non-reducing sugars, and total sugars in pear pulp and pomegranate-coloured RTS and squash, were estimated using titrimetric methods as described by Ranganna (1986). Total Soluble Solids (TSS) were measured using a hand refractometer. Statistical analysis was carried out using the Complete Randomized Design (CRD).

**Results**

**Standardization of Pomegranate Seed for Colour and Quality of pear RTS**

Organoleptic evaluation was conducted to measure the quality of pomegranate-enriched pear RTS beverages prepared with various concentrations of pomegranate seed extract. Sensory attributes such as appearance, flavour, and overall acceptability were evaluated. The sensory data revealed that pear RTS supplemented with 80 g of pomegranate seed extract per litre received the highest overall acceptability score of 8.9 on the nine-point Hedonic scale, indicating a rating of "Liked Very Much." This formulation was preferred over other concentrations in terms of sensory quality. In addition to enhancing the sensory appeal, the incorporation of pomegranate seed extract also contributed to extending the shelf life of pear RTS.

**Standardization of pomegranate Seed for Colour and Quality of pear Squash**

Pear squash enriched with varying concentrations of pomegranate seed extract was evaluated for its quality attributes. The data indicated that the addition of 80 g of pomegranate seed extract per litre resulted in the best colour and taste, receiving the highest sensory score of 8.8 on the nine-point Hedonic scale and rated as "Liked Very Much," compared to other treatments. Key sensory parameters-including colour, flavour, appearance, and overall acceptability-were recorded for both pear RTS and squash throughout the experiment. The organoleptic evaluation revealed that overall acceptability was highest when pomegranate seed extract was added at a concentration of 80 g/litre in both RTS and squash formulations, with both products being rated as "Liked Very Much." Currently, limited research has been conducted on the application of pomegranate seed extract in non-dairy beverages. This study highlights its potential as a natural colourant and flavour enhancer in fruit-based beverages.

**Table: 1** Pear RTS's organoleptic qualities, enhanced with pomegranate colour from various seed amounts

|  |  |  |  |
| --- | --- | --- | --- |
| **Treatments** | **Quantity of pomegranate****seed (g per litre)** | **Organoleptic quality** |  |
| **Score** | **Rating** |
| **T1** | Nil | 7.7 | LM |
| **T2** | 40 | 7.8 | LM |
| **T3** | 80 | 8.9 | LVM |
| **T4** | 120 | 7.5 | LM |
| **T5** | 160 | 6.8 | LM |

**Table: 2** Pear squash's organoleptic qualities enhanced by pomegranate colour derived from various seed amounts

|  |  |  |
| --- | --- | --- |
| **S. No.** | **Amount of pomegranate seed(g/litre)** | **Organoleptic quality** |
| **Score** | **Rating** |
| **T1** | Nil | 7.6 | LM |
| **T2** | 40 | 7.9 | LM |
| **T3** | 80 | 8.8 | LVM |
| **T4** | 120 | 7.7 | LM |
| **T5** | 160 | 7.3 | LM |

 Note: LM- Liked Much; LVM- Liked Very Much

**Table 3.** Changes in pomegranate seed enriched pear RTS during storage

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| **Storage (Month)** | **Total Soluble Solids (%)** | **Acidity (%)** | **Ascorbic acid** | **Reducing sugars (%)** | **Non-reducing sugar (%)** | **Total sugars (%)** | **Non-enzymatic browning** |
| 0 | 12.00 | 0.23 | 2.40 | 6.39 | 3.90 | 11.22 | 0.13 |
| 1 | 12.00 | 0.23 | 2.40 | 6.39 | 3.90 | 11.22 | 0.13 |
| 2 | 12.00 | 0.23 | 2.20 | 6.39 | 3.90 | 11.22 | 0.13 |
| 3 | 12.00 | 0.23 | 2.20 | 6.39 | 3.90 | 11.22 | 0.13 |
| 4 | 12.20 | 0.24 | 2.15 | 6.67 | 3.98 | 11.38 | 0.13 |
| 5 | 12.40 | 0.24 | 2.14 | 6.67 | 3.98 | 11.38 | 0.13 |
| 6 | 12.40 | 0.25 | 2.10 | 6.67 | 3.98 | 11.38 | 0.13 |
| 7 | 12.50 | 0.26 | 2.10 | 6.94 | 3.60 | 11.46 | 0.13 |
| 8 | 12.50 | 0.26 | 2.00 | 6.94 | 3.60 | 11.46 | 0.13 |
| 9 | 12.70 | 0.27 | 1.90 | 7.18 | 3.40 | 11.62 | 0.14 |
| 10 | 12.70 | 0.29 | 1.90 | 7.18 | 3.40 | 11.62 | 0.15 |
| SEm± | 0.012 | 0.18 | 0.194 | 0.001 | 0.004 | 0.001 | 0.005 |
| CD (5%) | 0.004 | 0.006 | NA | 0.002 | 0.001 | 0.003 | NA |

**Table 4.** Changes in pomegranate seed enriched pear squash during storage

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| **Storage (Month)** | **Total Soluble Solids (%)** | **Acidity (%)** | **Ascorbic acid** | **Reducing sugars (%)** | **Non-reducing sugar (%)** | **Total sugars (%)** | **Non-enzymatic browning** |
| 0 | 50.00 | 1.09 | 2.45 | 12.14 | 7.82 | 32.05 | 0.37 |
| 1 | 50. 00 | 1.09 | 2.45 | 12.14 | 7.82 | 32.05 | 0.37 |
| 2 | 50. 00 | 1.09 | 2.40 | 12.14 | 7.82 | 32.05 | 0.37 |
| 3 | 50. 00 | 1.09 | 2.40 | 12.14 | 7.82 | 32.05 | 0.37 |
| 4 | 50. 00 | 1.09 | 2.37 | 12.14 | 7.82 | 32.05 | 0.37 |
| 5 | 50.30 | 1.10 | 2.37 | 12.96 | 7.51 | 32.53 | 0.37 |
| 6 | 50.50 | 1.11 | 2.33 | 12.96 | 7.51 | 32.53 | 0.37 |
| 7 | 50.50 | 1.11 | 2.33 | 13.75 | 7.25 | 34.12 | 0.37 |
| 8 | 50.80 | 1.12 | 2.12 | 13.75 | 7.25 | 34.12 | 0.37 |
| 9 | 51.10 | 1.12 | 2.12 | 14.26 | 7.00 | 34.34 | 0.38 |
| 10 | 51.20 | 1.13 | 1.09 | 14.26 | 7.00 | 34.34 | 0.39 |
| SEm± | 14.907 | 0.024 | 0.193 | 0.020 | 7.006 | 0.012 | 0.006 |
| CD (5%) | NA | 0.008 | NA | 0.007 | 7.002 | 0.035 | NA |

**Changes in** **Pomegranate Enriched Pear RTS During Storage**

No significant changes were observed in total soluble solids (TSS), reducing sugars, total sugars, and acidity of pomegranate-enriched pear RTS during the first three months of storage. However, all four parameters showed a significant increase from the fourth month onward until the end of the experiment. Ascorbic acid content in the RTS declined non-significantly during the first two months, followed by a significant reduction in subsequent months. The concentration of non-reducing sugars remained relatively stable up to six months of storage, after which a significant decline was recorded. No signs of non-enzymatic browning were observed in pomegranate-enriched pear RTS during the first eight months of storage; however, browning increased significantly thereafter. The organoleptic quality of pomegranate-enriched pear RTS remained unchanged for the first three months. A gradual, non-significant decline in sensory scores was observed from the fourth to the eighth month of storage.

**Changes in the Pomegranate Enriched Pear Squash During Storage**

The total soluble solids (TSS), acidity, reducing sugars, non-reducing sugars, and total sugars in pomegranate-enriched pear squash remained stable during the first four months of storage. However, from the fifth month onward, all parameters-except non-reducing sugars-showed a gradual increase until the end of the experiment. A significant increase in TSS was recorded after four months of storage. Non-reducing sugar content decreased non-significantly up to the eighth month. Ascorbic acid content declined non-significantly during the initial two months, followed by a significant reduction in the subsequent storage period. Non-enzymatic browning in the squash remained unchanged up to eight months but showed a non-significant increase thereafter. The organoleptic quality of pomegranate-enriched pear squash remained stable for the first four months. After this period, a slight, non-significant decline in sensory scores was observed. Despite the minor decrease, the squash remained organoleptically acceptable throughout the entire ten-month storage period, with an overall rating of "Liked Very Much."

**Discussion**

During storage at room temperature, the pomegranate-enriched pear beverages' total soluble solids, acidity, reducing sugars, and total sugars all marginally increased. Similar outcomes were noted by **Rashid *et al*. (2018)** in the RTS of bio-coloured guava. The observed increase in total soluble solids (TSS) in pomegranate-enriched pear RTS and squash during storage may be attributed to the hydrolysis of polysaccharides and oligosaccharides into monosaccharides, particularly reducing sugars. The increase in acidity over time is likely due to the breakdown of pectic substances present in the fruit pulp. The increase in reducing and total sugars can be explained by the inversion of non-reducing sugars into reducing sugars, as well as the hydrolysis of complex carbohydrates into simpler sugar forms. The increase in non-enzymatic browning during storage is likely a result of complex Maillard-type reactions involving nitrogenous compounds and sugars, nitrogenous compounds and organic acids, sugars and organic acids, and interactions among organic acids themselves. Additionally, browning may occur due to reactions involving the carbinol group of cyclic sugars with basic proteins or amino acid complexes. Over the storage period, a decline was noted in ascorbic acid content, non-reducing sugars, and the organoleptic quality of pomegranate-enriched pear beverages, reflecting the natural degradation of nutritional and sensory attributes with time.

The loss of ascorbic acid, non-reducing sugar and organoleptic quality was also observed by **Bal *et al*. (2014)** in nectar of guava cv. Lalit, Jakhar *et al*. (2012) in guava-barbados cherry blend RTS, **Selvi *et al*. (2013)** in guava, lime and ginger blended RTS, **Sarkar and Bulo (2017)** in guava and pineapple blended RTS and **Ravi *et al*. (2018)** in RTS and squash of different varieties of guava. The loss of ascorbic acid, non-reducing sugars, and organoleptic quality during storage has also been reported in earlier studies. **Choudhary *et al*. (2008)** observed similar trends in guava nectar of cv. Lucknow-49, while **Bal *et al*. (2014)** reported such losses in guava nectar of cv. Lalit. Comparable findings were noted by **Jakhar *et al*. (2012)** in guava-Barbados cherry blended RTS, and by **Kumar *et al*. (2023)** in guava, lime, and ginger blended RTS. **Sarkar and Bulo (2017)** also recorded quality degradation in guava-pineapple blended RTS.

The degradation of ascorbic acid in pomegranate-enriched pear RTS and squash may be attributed to the use of transparent bottles, as ascorbic acid is highly sensitive to both light and temperature. Additionally, the reduction in ascorbic acid content could be due to oxidative reactions involving trapped oxygen within the bottles, leading to the formation of dehydroascorbic acid. The gradual decline in non-reducing sugars observed during storage is likely a result of their conversion into reducing sugars. The organoleptic scores of pomegranate-enriched pear RTS and squash began to decline after three and four months of storage, respectively. However, pomegranate-enriched RTS remained acceptable for up to 10 months, and the squash maintained its acceptability throughout the entire experimental period (up to 10 months). The findings of the present study suggest that pomegranate not only enhanced the visual appeal of the pear beverages but also contributed to their extended shelf life.

**Conclusion**

The current study used various amounts of pomegranate seeds to add colour to white-fleshed pear-based beverages, specifically squash and Ready-to-Serve (RTS) beverages. The findings showed that in both RTS and squash formulations, using 80 grams of pomegranate seeds per litre was the best way to achieve the required colour.

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