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RESEARCH ARTICLE

# Effect of Various Clarification Techniques on the Storage Studies of Carbonated Sugarcane Juice

Javed Abbas Bangash, Abdul Wajid Khalil* and Ghulam Mohi Uddin Paracha
Pakistan Council of Scientific & Industrial Research (PCSIR) Laboratories Complex, 25120. Peshawar, Pakistan

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***Corresponding Author:**
pcsir80@gmail.com

**ABSTRACT**

The carbonated sugarcane juice was prepared using various treatments like heating (85 ˚C for 10 minutes), adding citric acid (40 mg/100 ml) & potassium metabisulphite (350 ppm), microfiltration using cartridge filter (pore diameter: 0.4 µm & applied pressure: 10-40 psi) and was subjected to carbonation. The samples were stored at room and refrigeration temperatures in pre-sterilized bottles to evaluate the effect of the above mentioned treatments on physico-chemical (pH, total soluble solid (TSS), titratable acidity (TA), turbidity and clarity of juice), microbiological (total plate count, coliform bacteria, fecal coliform, yeast and mold) and sensory evaluation at every 30 days interval for 120 days. During storage studies at room temperature, the pH, TSS, turbidity and clarity juice were increased whereas mold/ yeast (cfu/ml) as well as total plate count (cfu/ml) was decreased moderately. Similarly, the pH and TSS were increased whereas, turbidity and clarity of juice were increased moderately, moreover, considerable decreased was observed in total plate counts and yeast/mold counts during storage at refrigeration temperature. The carbonated sugarcane juice showed minimum changes in sensory qualities at room and refrigeration temperature. It is concluded that the storage stability of carbonated sugarcane juice was 90 days at room temperature whereas it was 120 days at refrigeration temperature.

## INTRODUCTION

Sugarcane juice is highly nutritious having rich carbohydrates, iron, vitamins and minerals. It contains water (75-85%), reducing sugar (0.3-3.0%), non-reducing sugar (10-21%), iron (10 mg), carotene (6 µg) and 100 ml of sugarcane juice provides 40 Kcal of energy (Parvathy, 1983; Swaminathan, 1995). The high sugar content of the juice suggests that sugar-cane juice can potentially be developed into a natural energy drink (Easa, 2000; Tee et al., 1997; Yusof et al., 2000). Sugarcane juice is also possessing several medicinal and therapeutic properties like healing source for sore throat, cold, flu, good hydrating effect, cure of jaundice and also very useful in scanty urination (Banerji et al, 1997; Karthikeyan and Samipillai, 2010). Generally, sugarcane juice is fermented quickly by the presence of high sugars content and heavy load of microorganisms (Krishnakumar and Devadas, 2006). Development of effective treatments or procedures to delay the deterioration of sugarcane juice is the blanching before juice extraction, chemical treatment, heating, microfiltration followed by carbonation. But among these treatments, carbonation was consider effective as it does not destroy the nutrient contents, enhances the organoleptic properties and improves the shelf life of the product (Elahi, 1979; Alimulla, 1988; Shakeel et al., 2013). Nevertheless, carbonated sugarcane juice based beverage is a new concept with carbonation which can be promoted for commercial exploitation.

Therefore, the effect of various treatments of sugarcane juice on the physicochemical, microbiological, and sensory characteristics of carbonated sugarcane juice were investigated.

## MATERIALS AND METHODS

### Sample collection and preparation

The raw sugarcane was purchased from local market of Peshawar, Pakistan. Sugarcane was cleaned and washed with tap water to remove any foreign material. The skin and node of sugarcane stem was peeled, cut

into small pieces and blanched in boiling water for 5 minutes containing 0.1% potassium metabisulphite (Kapur et al., 1978).

**Extraction process**

The blanched small pieces of cane sugar stem were extracted using four-roller crusher to get the raw juice and filtered through double layer of muslin cloth. The filtered juice was used for further clarification treatments.

**Clarification treatments of juice**

The extracted juice was clarified by adding citric acid (40mg/100ml) to adjust pH at 4.0, heated (85°C) and kept for overnight to settle the suspended materials. After keeping overnight, the supernatant layer was separated from the remaining sample. The supernatant juice was filtered with cartridge filter made with polypropylene (pore diameter: 0.4 µm & applied pressure: 10-40 psi).

**Carbonation process**

After filtration of sugarcane juice, Carbon dioxide ($CO_2$) was added to clarified juice using a Soda stream machine.

**Analytical work**

The pH was determined using a digital meter analyzer, the total soluble solids content (expressed as °Brix) was determined using digital refractometer and the titratable acidity (expressed as % citric acid) determined by titration with 0.1 N NaOH (AOAC, 2006). The sample (5 ml) was centrifuged at 3000 rpm for 10 min at room temperature. Turbidity (Cloud value) was measured as supernatant absorbance at 660 nm (UV/VIS Double Spectrophotometer). The absorbance of distilled water was considered as a blank (Versteeg et al., 1980). The clarity of the juice was measured by measuring the transmittance at a wavelength of 570 nm using UV-VIS spectrophotometer (Arsad et al., 2015).

**Microbiological Analysis**

Total Plate Count was determined by pour plate method. The Total Coliform /Fecal Coliform bacteria were determined by multiple tube fermentation technique. The yeast and mold were also analyzed according to the procedure as described by the American public health association (APHA, 2013).

**Sensory evaluation**

Juice samples were also evaluated for sensory characteristics namely appearance, color, flavor/taste and sharpness ($CO_2$ gas) through 10 member's panel using 9-point Hedonic scale (Larmond, 1977).

**Statistical analysis**

All analyses were conducted in replicate, and the results were presented as means ± standard error (SE).

## RESULTS AND DISCUSSION

**Physicochemical analysis**

The results of the pH, TSS, acidity, turbidity and juice clarity of carbonated sugarcane juice during storage at room as well as refrigeration temperature are shown in Table 1. The pH was increased whereas acidity decreased of sample during storage at room and refrigeration temperature. The TSS was increased while turbidity and juice clarity were decreased slightly during storage. The extent of increase in pH, TSS and decrease in acidity, turbidity and juice clarity were found higher in room as compared to refrigeration temperature. Similar trend was observed by Dhinesh et al., (2016). Moreover, in the present study, the clarification of sugarcane juice through microfiltration was higher in declining the turbidity were found similar results as reported by Nogueira et al (2007), Hervé et al (1995) and Kishihara et al. (1981). The increased pH was occurred due to the decreased acidity of carbonated sugarcane juices. It is also possible to have biochemical reaction taking place during storage periods together with microbial action in the juices (Makanjuola et al., 2013). The rise in TSS could be partially due to the increment of soluble sugars, which may result from the conversion of insoluble pectin and cellulose by pectinolytic and cellulase enzymes to produce soluble sugars (Schobinger et al., 1981).

**Microbiological analysis**

The values of the mold/yeast, total plate count (TPC), total coliforms and fecal coliforms of carbonated sugarcane juice were stored at room as well as refrigeration temperature as shown in Table 2. The count of yeast and mold was noted (55 cfu/ml) in fresh juice and decreased was occurred up to 17 cfu/ml after 3 month storage studies at room temperature while at refrigeration temperature the value of mold/yeast was not detected after four months. Similarly, the TPC value was recorded (820 and 822 cfu/ml) in fresh juice and decreased was observed significantly from 28 to 07 cfu/ml during storage at room and refrigeration temperature. The total coliforms (MPN/100 ml) and fecal Coliforms were found within the maximum permitted limit of carbonated sugarcane juice at room as well as refrigeration temperature (Gulf Standards, 2000). Similar observations were observed by Moreno et al., (2012). Masse et al., (2013) who's reported the size of yeast (3 to 4 µm) and bacterial cells (0.1 µm) whereas, the cartridge filter used for filtration in this study had a pore size of 0.4 µm which was capable to retain all the microorganisms resulting in extension of shelf life of carbonated sugarcane juice. Moreover, it has been reported by Moreno et al., (2012) that microfiltration technique is more effective in controlling microorganism's growth than conventional method in clarification of sugar cane juice.

**Sensory evaluation**

Hedonic scale tests were used to evaluate the appearance, color, flavor/taste and sharpness ($CO_2$ gas) of carbonated sugarcane juice during storage conditions as shown in Table 3. The appearance and color scores were found 8.4 & 7.5 in fresh juice while decreased was

**Table 1: Physicochemical analysis of carbonated sugarcane juice during storage study**

| | | | | | | |
|---|---|---|---|---|---|---|
| | | **Storage at room temperature** | | | | |
| **S. No.** | Parameters | **Fresh juice** | **After 1 Month** | **After 2 Months** | **After 3 Months** | |
| 1. | pH | 3.8 ± 0.01 | 4.0 ± 0.02 | 4.1± 0.033 | 4.2 ± 0.000 | * |
| 2. | T.S.S (°Brix) | 11 ± 0.07 | 11.8 ± 0.01 | 12 ± 0.000 | 12.3 ± 0.003 | * |
| 3. | Acidity (%) | 0.32 ± 0.09 | 0.31 ± 0.04 | 0.29 ± 0.006 | 0.28 ± 0.04 | * |
| 4. | Juice turbidity (%) | 9.7 ± 0.04 | 8.1 ± 0.05 | 8.0 ± 0.003 | 7.2 ± 0.115 | * |
| 5. | Juice clarity (%) | 84 ± 0.01 | 81± 0.02 | 75 ± 0.057 | 70 ± 0.067 | * |
| | | **Storage at refrigeration temperature** | | | | |
| | Parameters | **Fresh juice** | **After 1 Month** | **After 2 Months** | **After 3 Months** | **After 4 months** |
| 1. | pH | 3.9 ± 0.28 | 4.2 ± 0.12 | 4.2 ± 0.09 | 4.2 ± 0.003 | 4.2 ± 0.64 |
| 2. | T.S.S (°Brix) | 11.3 ± 0.35 | 11.7 ± 0.14 | 12 ± 0.04 | 12 ± 0.115 | 12 ± 0.135 |
| 3. | Acidity (%) | 0.30 ± 0.64 | 0.29 ± 0.16 | 0.28 ± 0.01 | 0.27 ± 0.06 | 0.26 ± 0.133 |
| 4. | Juice turbidity (%) | 9.8 ± 0.82 | 9.7 ± 0.17 | 9.7 ± 0.07 | 9.7 ± 0.12 | 9.2 ± 0.136 |
| 5. | Juice clarity (%) | 84 ± 0.01 | 83 ± 0.18 | 83 ± 0.02 | 83 ± 0.04 | 82 ± 0.149 |

Values are means of 30 replicates ± standard error.

**Table 2: Microbiological analysis of carbonated sugarcane juice during storage study**

| | | | | | | |
|---|---|---|---|---|---|---|
| | | **Storage at room temperature** | | | | |
| **S. No.** | Parameters | **Fresh juice** | **After 1 Month** | **After 2 Months** | **After 3 Months** | |
| 1. | Mold/ Yeast (cfu/ml) | 55 ± 0.12 | 48 ± 0.18 | 38 ± 0.10 | 17 ± 0.11 | * |
| 2. | Total Plate count (cfu/ml) | 820 ± 0.13 | 182 ± 0.16 | 160 ± 0.12 | 28 ± 0.18 | * |
| 3. | Total Coliforms (MPN/100 ml) | 16 ± 0.13 | <1.1 | <1.1 | <1.1 | * |
| 4. | Fecal Coliforms (Present/Absent) | Present | Absent | Absent | Absent | * |
| | | **Storage at refrigeration temperature** | | | | |
| | Parameters | **Fresh juice** | **After 1 Month** | **After 2 Months** | **After 3 Months** | **After 4 months** |
| 1. | Mold/ Yeast (cfu/ml) | 56 ± 0.11 | 26 ± 0.16 | 18 ± 0.15 | 06 ± 0.13 | Absent |
| 2. | Total Plate count (cfu/ml) | 822 ± 0.14 | 176 ± 0.18 | 73 ± 0.18 | 61± 0.19 | 07 ± 0.15 |
| 3. | Total Coliforms (MPN/100 ml) | 16 ± 0.12 | <1.1 | <1.1 | <1.1 | <1.1 |
| 4. | Fecal Coliforms (Present/Absent) | Present | Absent | Absent | Absent | Absent |

Values are means of 30 replicates ± standard error.

**Table 3: Sensory evaluation of carbonated sugarcane juice during storage study**

| | | | | | | |
|---|---|---|---|---|---|---|
| | | **Storage at room temperature** | | | | |
| **S. No.** | Parameters | **Fresh juice** | **After 1 Month** | **After 2 Months** | **After 3 Months** | |
| 1. | Appearance | 8.4 ± 0.09 | 8.2 ± 0.03 | 7.5 ± 0.02 | 7.0 ± 0.05 | * |
| 2. | Color | 7.5 ± 0.07 | 7.2 ± 0.04 | 7.0 ± 0.03 | 6.0 ± 0.04 | * |
| 3. | Flavor/Taste | 6.4 ± 0.08 | 5.8 ± 0.05 | 5.0 ± 0.02 | 4.0 ± 0.06 | * |
| 4. | Sharpness ($CO_2$ gas) | 5.6 ± 0.07 | 5.4 ± 0.06 | 5.0 ± 0.04 | 4.0 ± 0.07 | * |
| | | **Storage at refrigeration temperature** | | | | |
| | Parameters | **Fresh juice** | **After 1 Month** | **After 2 Months** | **After 3 Months** | **After 4 months** |
| 1. | Appearance | 8.4 ± 0.09 | 8.2 ± 0.08 | 8.0 ± 0.03 | 7.8 ± 0.02 | 7.6 ± 0.07 |
| 2. | Color | 7.5 ± 0.07 | 7.3 ± 0.06 | 7.0 ± 0.02 | 6.8 ± 0.04 | 6.6 ± 0.08 |
| 3. | Flavor/Taste | 6.4 ± 0.08 | 6.4 ± 0.05 | 6.2 ± 0.04 | 6.1 ± 0.05 | 6.0 ± 0.06 |
| 4. | Sharpness ($CO_2$ gas) | 5.6 ± 0.07 | 5.5 ± 0.07 | 5.3 ± 0.05 | 5.2 ± 0.06 | 5.0 ± 0.04 |

Values are means of 30 replicates ± standard error.

occurred moderately from 7 to 7.6 & 6.0 to 6.6 during storage studies at room and refrigeration temperature of carbonated sugarcane juice. The sharpness ($CO_2$ gas) scores were decreased gradually at room as well as refrigeration temperature with the advancement of storage. However, the rate of increase in sensory scores of samples stored at room temperature was greater than those stored at refrigeration temperature. Similar findings were reported by Dhinesh et al., (2016). In this study, the higher scores of appearance and color of clarified juice was due to the removal of suspended and gummy materials as reported by Dziezak (1990).

## Conclusion

It is concluded from the current study that the shelf life of carbonated sugarcane juice was extended up to 90 days at room temperature whereas 120 days at refrigeration temperature. Furthermore, the carbonation of sugarcane juice is considered effective in enhancing the organoleptic properties as well as improves the shelf life of the product.

## Authors' Contribution

All authors contributed equally to this manuscript.

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