Research Article

Development of Amla (Emblica officinalis) ready to serve beverage fortified with dietary fiber

N. Surya, S. Jesupriya Poornakala, S. Kanchana, G. Hemalatha

Abstract

Fruit beverages are one of the popular categories of beverages that are consumed across the world. They are rich source of carbohydrates and have high availability of important vitamins and minerals. While preparation of juices the residues and the dietary fiber present in the fruits are removed during filtration as waste. Hence, the present investigation was carried out to standardize the fortification of dietary fiber into amla Ready to Serve (RTS) beverages. The amla RTS beverages were fortified with different concentration levels (0.5%, 1% and 3%) of dietary fiber extracted from the amla fruit with the aim of increasing the dietary fiber intake. The physicochemical characteristics such as pH, total soluble solids, acidity, ascorbic acid, total sugar, reducing sugar and total dietary fiber were analyzed in fruit juices and organoleptically evaluated to assess the maximum acceptability of the products. Juice characteristics such as clarity, turbidity, non-enzymatic browning (NEB) and color were analyzed. The standardized RTS beverages were packed in glass bottles and PET bottles and stored under refrigeration and storage behaviour was studied. RTS beverage fortified with 0.5 per cent dietary fiber was found to be best in their sensory characteristics and physical properties.

Keywords Ready to Serve (RTS) beverage, dietary fiber, amla, fortification

Introduction

Fruits are mostly water (75-90%), maximum located in vacuole producing turgor in the fruit tissue, and fruit juice is produced by infusing fresh fruits. Fruits cell wall contains of cellulose microfibrils entrenched in an amorphous matrix of hemicelluloses and pectin. Juice includes water; pectic substances; vitamins and minerals; pigments; aroma and flavor compounds and soluble solids (sugars and organic acids). The acidity and starch in fruits decrease while sugars increase during ripening [1]. It has been stated that regular consumption of fruit juices, rich in polyphenols, can enhance the protective effects against numerous degenerative diseases. Moreover, regular fruit juice consumption was reported to reduce the risk of several chronic diseases. Nowadays, there are many types of fruit juices available commercially in the market. The consumption of the fruit juices is popularizing rapidly as they are convenient, nutritious and ready-to-drink [2]. Although most of the fiber from whole fruit is removed during fruit juice processing, 100% fruit juices retain similar levels of other healthy vitamins, minerals and phytochemicals. The wastes of fruits and vegetables are inexpensive, abundantly available and are a good source of dietary fiber [3]. There is considerable epidemiological evidence that higher
dietary fiber intake reduces the risk of disease, including cardiovascular disease (CVD), type 2 diabetes, and cancer [4].

Amla [(Emblica oficinalis Gerth. Sync (Phyllanthus emblica)] also known as Indian gooseberry, is a minor fruit belongs to the family Euphorbiaceae and said to be native to India. In India, it is commercially cultivated in Uttar Pradesh followed by Gujarat, Tamil Nadu, Rajasthan, Karnataka and West Bengal [5]. The fruit is highly nutritious and the richest source of vitamin C. Amla fruit contains fiber (0.8-2.0 per cent), total soluble solids (10-64 per cent), vitamin ‘C’ (700-900 mg /100g), pectin (2.4 – 3.1 per cent) and phenols (2-3 %)[6]. Amla due to its strong antioxidant and biological properties prevent innumerable health disorders as it contains essential nutrients and highest amount of vitamin C. It can be used as a possible food additive or in nutraceuticals and biopharmaceutical industries. Fresh juice of amla contains nearly twenty times as much vitamin C as orange juice. A single tiny amla is equivalent to two oranges in vitamin C content. It is an ingredient of many Ayurvedic medicines and tonics, as it removes excessive salivation, nausea, vomiting, giddiness, spermatorrhoea, internal body heat and menstrual disorders [7]. Hence, the present study was undertaken to study the effect of dietary fiber fortification on the sensory and physico-chemical properties of amla RTS beverage.

Methodology

**Extraction of dietary fiber**

Dietary fiber from amla residue was extracted using the method mentioned by Yoshimoto et al., [8] with little modification. Amla residues after juice extraction were mixed with water (1: 20 w/v ratio). An alpha-amylase was added (0.1 ml/ g sample) to the residue. The sample was incubated at 95°C for 30 min. After cooling down to 60°C, an amyloglucosidase solution was added (0.1 ml/ g sample) and incubated at 60°C for 30 min. Finally, the mixture was filtered through Whatman No.4 filter paper and dried in the hot air oven at 50°C. The dried samples were then powdered in mill using a 1 mm sieve.

**Development of amla RTS fortified with dietary fiber**

Amla RTS beverage was prepared as per FSSAI specification. It was fortified with dietary fiber in different concentration levels with the aim of increasing the consumption of dietary fiber. The selected amla fruits were washed thoroughly in clean water. After washing, they were cut into pieces. Blanching process was carried out for amla fruits at the temperature of 100°C for 5 minutes. Pulping of the fruits was done with the help of mixer and the pulp was filtered through the muslin cloth to separate the residues. Sugar syrup was made with 13°Brix and citric acid was added. Then the sugar syrup was added to the filtered pulp after cooling. The dietary fiber extracted from amla residues was fortified each at 0.5 per cent (T1), 1.0 per cent (T2) and 3.0 per cent (T3) levels in amla RTS beverages. Amla RTS beverage prepared without fortification of dietary fiber was considered as control (T0). The prepared RTS beverage was poured immediately in sterilized glass and PET bottles leaving a head space of 3.0 cm and the bottles were corked with sterilized crown corks. The bottled RTS was pasteurized at a temperature of 90°C for 25 minutes and cooled to room temperature. RTS bottles were labeled and stored at refrigerated condition (4°C).

**Physico-chemical analysis**

Protein was analyzed by the amount of nitrogen available in the sample using Micro-Kjeldahl method [9]. The pH and acidity of the samples was estimated by the standard method [10]. The total soluble solids of the samples were observed by using a hand refractometer (0° to 32° Brix) [11]. The reducing and total sugar content of the samples was determined by Shaffer Somogyi micro-method [12].

**Sensory evaluation**

The quality attributes in terms of color and appearance, flavor, taste and overall acceptability were evaluated by a panel of 20 semi-trained judges using a score card with 9 point hedonic rating scale [13].
Analysis of juice characteristics

Clarity
Clarity is ascribed to particles in the juice, such as cellulose, hemicellulose, proteins, lipids and some other minor particles [14]. For fruit juices, stability of the clarity value is a visual quality factor associated with the flavor and color and is interconnected to consumer acceptance [15]. The clarity was determined by measuring the absorbance at 660 nm using a 2201UV-Visible Double Beam Recording Spectrophotometer (Systronics, India) [16].

Turbidity
Turbidity mainly occurs due to polysaccharides such as starch, pectin, cellulose, and hemicellulose [17]. The developed RTS beverages were subjected to turbidity measurements using a 2201UV-Visible Double Beam Recording Spectrophotometer (Systronics, India) with absorbance at 600 nm [18].

Non-enzymatic browning
Non-enzymatic browning (NEB) is one of the most important chemical reactions responsible for quality and color changes during the heating or prolonged storage of fruit juice [19]. The increase in absorbance of a sample extract at 440 nm was taken as a measure of non-enzymatic browning [20]. Color is a crucial characteristic in food as a consumer preference of acceptability. Measurement of color was done using Hunter L* a* b* color scale Chromometer, Lovibond.

Microbiological enumeration
The microbical load of the stored samples was enumerated by dilution plate method [21]. The media used for bacteria was nutrient agar media, for moulds and fungi Martin’s rose bengal agar and for yeast, yeast extract malt extract agar medium.

Storage studies
The amla RTS beverage with best dietary fiber fortification level on the basis of sensory evaluation were packed in glass bottles and PET bottles and kept at refrigerated temperature and changes in sensory, physico-chemical characteristics and microbial load were assessed during storage at 15 days intervals up to 45 days.

Statistical analysis
The data obtained were subjected to statistical analysis to find out the impact of treatments, storage periods and packaging materials on the quality of the samples during storage. Completely Randomized Design (FRBD) was applied for the analysis [22].

Results and Discussion

Chemical constituents of dietary fiber extracted from amla residues
The observed yield of dietary fiber was 16.05 per cent from amla residue. The dietary fiber contained 1.17 per cent ash, 2.62 per cent protein, pH 3.10 and 6.60 per cent moisture content.

Sensory characteristics of the amla RTS beverages
The sensory characteristics of the freshly prepared amla fortified RTS beverages with and without fortification of dietary fiber are presented in Table 1. It was observed that RTS beverage prepared with 0.5 % fortification of dietary fiber (T1) recorded high sensory score in all quality attributes as scored by control sample (T0) when compared to the other RTS beverages prepared with 1.0% (T2) and 3.0% (T3) fortification of dietary fiber. The high sensory scores for the sample implied higher preference of the particular sample.
Table 1. Sensory characteristics (mean scores) of the dietary fiber fortified amla RTS beverages

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Color and appearance</th>
<th>Flavor</th>
<th>Taste</th>
<th>Overall acceptability</th>
</tr>
</thead>
<tbody>
<tr>
<td>T₀</td>
<td>9.0</td>
<td>9.0</td>
<td>9.0</td>
<td>9.0</td>
</tr>
<tr>
<td>T₁</td>
<td>9.0</td>
<td>9.0</td>
<td>9.0</td>
<td>8.5</td>
</tr>
<tr>
<td>T₂</td>
<td>8.0</td>
<td>7.5</td>
<td>7.5</td>
<td>7.5</td>
</tr>
<tr>
<td>T₃</td>
<td>7.5</td>
<td>7.0</td>
<td>7.0</td>
<td>7.0</td>
</tr>
</tbody>
</table>

T₀ - Control; T₁- Amla RTS +0.5 % dietary fiber; T₂- Amla RTS +0.5 % dietary fiber; T₃- Amla RTS +0.5 % dietary fiber

Similar observations were made by Gandhi [23] who reported that the physiologically-effective clear beverage formulated with non-gel forming soluble fiber and a soluble salt of calcium and other mineral supplements along with pharmaceutically-active components had sensory properties similar to a regular beverage. Sweetened plain yogurt was fortified with insoluble dietary fibers of sugar beet, corn, oat, rice and soy. However, the addition of fiber caused a decrease in the flavor and texture scores. All the fiber fortified yogurts had a gritty texture and grainy flavor except the oat fiber fortified yogurt [24].

Physico-chemical characteristics of amla RTS beverages

The pH of amla RTS beverages ranged between 3.4 and 3.6, and acidity between 0.30 and 0.350 brix and TSS was 13° brix for all the samples. The vitamin C content was ranged from 53.0 to 54.80 mg per cent, reducing sugar ranged from 3.25 to 4.15 per cent, and total sugar 8.21 to 8.62 per cent. The total dietary fiber content ranged from 0.4 to 6.2 per cent [Table 2]. Thongsombat et al., [25] also stated that the guava juice developed with fortification of soluble dietary fiber and 0.50 per cent of pectin extracted from guava cake (peel, pulp, seeds) contained pH 4.20, TSS 15.270Bx, total acidity 0.56 per cent and viscosity 21.90 Cps.

Table 2. Physico-chemical characteristics of dietary fiber fortified amla RTS

<table>
<thead>
<tr>
<th>S. No.</th>
<th>Characteristics</th>
<th>T₀</th>
<th>T₁</th>
<th>T₂</th>
<th>T₃</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>pH</td>
<td>3.40</td>
<td>3.52</td>
<td>3.50</td>
<td>3.60</td>
</tr>
<tr>
<td>2.</td>
<td>Acidity</td>
<td>0.35</td>
<td>0.30</td>
<td>0.32</td>
<td>0.33</td>
</tr>
<tr>
<td>3.</td>
<td>TSS (0 Bx)</td>
<td>13.00</td>
<td>13.00</td>
<td>13.00</td>
<td>13.00</td>
</tr>
<tr>
<td>4.</td>
<td>Ascorbic acid (mg/100ml)</td>
<td>54.80</td>
<td>53.0</td>
<td>53.0</td>
<td>53.24</td>
</tr>
<tr>
<td>5.</td>
<td>Reducing Sugar (%)</td>
<td>4.15</td>
<td>3.42</td>
<td>3.25</td>
<td>3.32</td>
</tr>
<tr>
<td>6.</td>
<td>Total sugar (%)</td>
<td>8.62</td>
<td>8.54</td>
<td>8.36</td>
<td>8.21</td>
</tr>
<tr>
<td>7.</td>
<td>Total dietary fiber (%)</td>
<td>0.40</td>
<td>2.30</td>
<td>3.40</td>
<td>6.20</td>
</tr>
</tbody>
</table>

T₀ - Control; T₁- Amla RTS +0.5 % dietary fiber; T₂- Amla RTS +0.5 % dietary fiber; T₃- Amla RTS +0.5 % dietary fiber

Juice characteristics of dietary fiber fortified amla RTS beverages

The physical properties such as clarity, turbidity and NEB in amla RTS beverages are given in Table 3. The clarity values of amla RTS beverages were 0.057, 0.154, 0.165 and 0.181 for T₀, T₁, T₂ and T₃ samples respectively. Cesar et al., [26] also reported that the clarity value of acai fruit juice was 0.016 (OD). The turbidity values of amla RTS beverages were 0.120, 0.131, 0.142 and 0.188 for T₀, T₁, T₂ and T₃ samples respectively. Turbidity of dietary fiber fortified RTS samples were reported to be greater than the control RTS sample. Similar findings were observed by Bornare and Khan [27] who determined that the turbidity of banana blended with pseudo stem RTS sample was reported to be greater than banana RTS sample. The non-enzymatic browning (NEB) of amla RTS beverages was 0.020, 0.027, 0.032 and 0.041 for T₀, T₁, T₂ and T₃ samples respectively. It was observed that control RTS beverage sample (T₀) developed
low level of non-enzymatic browning (0.020 OD) followed by RTS prepared with 0.5% fortification of dietary fiber

Table 3. Juice characteristics of dietary fiber fortified amla RTS beverages

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Clarity (OD Value)</th>
<th>Turbidity (OD Value)</th>
<th>NEB (OD Value)</th>
</tr>
</thead>
<tbody>
<tr>
<td>T₀</td>
<td>0.057</td>
<td>0.120</td>
<td>0.020</td>
</tr>
<tr>
<td>T₁</td>
<td>0.154</td>
<td>0.131</td>
<td>0.027</td>
</tr>
<tr>
<td>T₂</td>
<td>0.165</td>
<td>0.142</td>
<td>0.032</td>
</tr>
<tr>
<td>T₃</td>
<td>0.181</td>
<td>0.188</td>
<td>0.041</td>
</tr>
</tbody>
</table>

T₀ - Control; T₁ - Amla RTS +0.5 % dietary fiber; T₂ - Amla RTS +0.5 % dietary fiber; T₃ - Amla RTS +0.5 % dietary fiber, OD – Optical Density

(T1) (0.027 OD). Ramachandran and Nagarajan [28] observed that the NEB in spiced papaya RTS beverage was 0.13 ± 0.01 (OD) and in spiced aloe gel RTS beverage 0.14 ± 0.01(OD).

Color value of dietary fiber fortified amla RTS beverages

In Table 4, the color values of RTS beverages have been summarized. Amla RTS obtained 30.90, 28.80, 27.85 and 26.05 of (L*) values, -1.54, -1.58, -1.53 and -1.56 of a* values and -3.32, -3.34, -3.37 and -3.41 of b* values for T₀, T₁, T₂ and T₃ samples respectively. Low (L*) values indicated that fortification of dietary fiber into the amla RTS samples (T₁, T₂ and T₃) affected the color properties of L* values. T₃ samples had darker colour than other samples. These results are in agreement with the findings of Cesar et al., [26] who reported that (L*) values of pulpy açai juice and clear juice was 19.76 and 34.34 respectively. Valero et al., [29] stated that the changes in color parameter of L*, a* and b* may due to the particle size changes in the juice.

Table 4. Colour value of dietary fiber fortified amla RTS beverages

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Lightness (L*)</th>
<th>Chromatically coordinates</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>a*(red (+a) to greenness (-a))</td>
</tr>
<tr>
<td>T₀</td>
<td>30.90</td>
<td>-1.54</td>
</tr>
<tr>
<td>T₁</td>
<td>28.80</td>
<td>-1.58</td>
</tr>
<tr>
<td>T₂</td>
<td>27.85</td>
<td>-1.53</td>
</tr>
<tr>
<td>T₃</td>
<td>26.05</td>
<td>-1.56</td>
</tr>
</tbody>
</table>

T₀ - Control; T₁ - Amla RTS +0.5 % dietary fiber; T₂ - Amla RTS +0.5 % dietary fiber; T₃ - Amla RTS +0.5 % dietary fiber

Storage studies

The RTS beverage with best fortification level (0.5%) (on the basis of sensory evaluation) was packed in glass bottles and PET bottles and kept at refrigerated temperature and changes in sensory characteristics, physical and chemical characteristics were determined during storage at 15 days intervals up to 45 days.

Changes in the sensory characteristics of the dietary fiber fortified RTS beverages during storage

The sensory characteristics of dietary fiber fortified amla RTS beverages during storage are given in Table 5. It depicts the sensory characteristics based on the mean scores for all the quality attributes (color and appearance, flavor, taste and overall acceptability) of the samples stored at refrigeration temperature. Initially, both the samples T₀ and T₁ obtained a score of 8.9 to 9.00. During storage, it was observed that the sensory characteristics of both the samples T₀ and T₁ slightly decreased and scored 8.4 to 8.9. However, at the end of storage period both the samples (T₀ and T₁) packed in both the packaging materials (P₁ and P₂)
### Table 5. Changes in the sensory characteristics of amla RTS beverages during storage

<table>
<thead>
<tr>
<th>Quality attributes</th>
<th>P₁</th>
<th>T₀</th>
<th>T₁</th>
<th>P₂</th>
<th>T₀</th>
<th>T₁</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Storage period (days)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Color and appearance</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Initial day</td>
<td>9.0</td>
<td>9.0</td>
<td>9.0</td>
<td>9.0</td>
<td>9.0</td>
<td>9.0</td>
</tr>
<tr>
<td>45</td>
<td>8.9</td>
<td>8.5</td>
<td>8.8</td>
<td>8.4</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Flavor</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Initial day</td>
<td>8.9</td>
<td>8.9</td>
<td>8.9</td>
<td>8.9</td>
<td>8.9</td>
<td>8.9</td>
</tr>
<tr>
<td>45</td>
<td>8.8</td>
<td>8.5</td>
<td>8.8</td>
<td>8.4</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Taste</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Initial day</td>
<td>9.0</td>
<td>9.0</td>
<td>9.0</td>
<td>9.0</td>
<td>9.0</td>
<td>9.0</td>
</tr>
<tr>
<td>45</td>
<td>8.7</td>
<td>8.4</td>
<td>8.6</td>
<td>8.3</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Overall acceptability</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>Initial day</td>
<td>9.0</td>
<td>8.9</td>
<td>9.0</td>
<td>8.9</td>
<td>8.9</td>
<td>8.9</td>
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<tr>
<td>45</td>
<td>8.8</td>
<td>8.5</td>
<td>8.7</td>
<td>8.4</td>
<td>8.4</td>
<td>8.4</td>
</tr>
</tbody>
</table>

P₁ – Glass bottle, P₂ – PET bottle, T₀ – Control, T₁ – RTS +0.5 % dietary fiber

remained under the consideration of “Like extremely” by panel members. RTS beverage packed in PET bottle obtained lowest scores than glass bottle because of slower rate of chemical reactions in product packed in glass bottle as a result of difference in their thermal conductance properties [30].

**Physico-chemical characteristics of amla RTS beverages fortified with dietary fiber during storage**

The pH, TSS and acidity of the amla RTS beverages fortified with dietary fiber during storage is shown in Table 6.

**pH**

Amla RTS beverages had an initial pH of 3.45 in T₀ and 3.52 in T₁. During storage the pH of the RTS beverages increased to 3.61 and 3.66 in T₀ and T₁ samples stored in glass bottles, 3.60 and 3.56 in T₀ and T₂ samples in PET bottles respectively. Sasikumar and Vivek [31] also observed that the pH in aloe vera and pine apple blend RTS beverages increased from 4.5 to 5.4 during the storage period of 60 days. The pH of RTS could be correlated inversely with the acidity of RTS and found to decrease with increase in storage periods as reported by AOAC [32].

**Total Soluble Solids**

It was observed that the TSS content of the amla RTS beverages were significantly increased during the storage period. The initial TSS was 13.00 Bx in all beverages. The values of amla RTS beverages were increased to 13.36 in T₀ and 13.29 in T₁ samples, which were stored in glass bottles and the values increased to 13.29 in T₀ and 13.34 in T₁ which were stored in PET bottles. Similar observations were reported by Bhardwaj and Mukherjee [33] that there was a gradual increase in TSS of kinnow juice blended with aonla juice and ginger juice from 11.5 to 14.790 Brix at the end of six months of storage. This might be due to hydrolysis of polysaccharides into monosaccharide and oligosaccharides [34].

**Acidity**

The decreasing trend in acidity of amla RTS beverages during storage was observed. The acidity of amla RTS beverages was 0.35 and 0.33 per cent in T₀ and T₁ respectively. After storage of forty five days the
acidity was significantly decreased to 0.21 (T<sub>0</sub>) and 0.20 (T<sub>1</sub>) per cent in glass bottled (P<sub>1</sub>) samples, 0.20 (T<sub>0</sub>) and 0.21 (T<sub>1</sub>) per cent in PET bottled (P<sub>2</sub>) samples. Similar results were also reported by Gaikwad et al., [35] that there was a decrease in the acidity level of low calorie herbal aonla ginger RTS beverages and it was ranged from 0.3 to 0.42 per cent at the end of storage period. These changes in the decrease of acidity might be due to the acidic hydrolysis of the polysaccharides, where the acid is utilized for converting non reducing sugar into reducing sugar [36].

**Changes in the chemical characteristics amla RTS beverages during storage**

The ascorbic acid, reducing sugar, total sugar and dietary fiber of the amla RTS beverages fortified with dietary fiber during storage is given in Table 7.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Method</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>pH</td>
<td>Metrohm 774 pH-meter</td>
<td>pH meter</td>
</tr>
<tr>
<td>TS</td>
<td>Method 2540 C</td>
<td></td>
</tr>
<tr>
<td>VS</td>
<td>Method 2540 E</td>
<td>[13]</td>
</tr>
<tr>
<td>COD</td>
<td>Method 5220</td>
<td></td>
</tr>
<tr>
<td>Biomethane estimation</td>
<td>via COD</td>
<td>[14], [15]</td>
</tr>
<tr>
<td>Percentage of CH₄, CO₂ and H₂S</td>
<td>BMP analysis</td>
<td></td>
</tr>
</tbody>
</table>

**Ascorbic acid**

Ascorbic acid is considered to be a heat-labile vitamin and most sensitive towards light and higher temperature. The initial ascorbic acid content was found to be 54.80 and 54.0 mg/100ml in T<sub>0</sub> samples stored in glass and PET bottles respectively. The ascorbic acid content was 53.00mg/100ml in T<sub>1</sub> of both packaged samples of amla RTS beverages. At the end of forty five days of storage, the ascorbic acid content of amla RTS beverages was significantly reduced to 53.29 for T<sub>0</sub> and 52.30 mg/100 ml for T<sub>1</sub> in glass bottled samples and 52.00 for T<sub>0</sub> and 51.46 mg/100 ml for T<sub>1</sub> in PET bottled samples respectively. Official Methods of Analysis [32] observed this same trend that there was a reduction in vitamin C content of pineapple juice blended with aloevera. Deka et al., [37] also studied that there was linear decrease in vitamin C content in mango- pineapple spiced beverage during storage of six months. A gradual decrease in vitamin C content of low calorie herbal aonla ginger RTS beverage was observed during the storage period of sixty days [36].
Reducing sugar
The initial reducing sugar content was 4.15 for T₀ and 3.42 for T₁ of amla RTS beverages respectively. An increasing trend in the reducing sugar content of RTS beverages was observed in all the samples. At the end of the forty five days of storage the reducing sugar content was significantly increased to 4.41 for T₀ and 4.98 for T₁ glass bottled samples, and 4.42 for T₀ and 4.95 for T₁ PET bottled samples of amla respectively. The increase in reducing sugars in juice could be due to the gradual inversion of non-reducing sugars during storage [38]. The results are comparable to the findings reported by Yadav et al., [39] reported that reducing sugar content of whey- banana herbal beverage was gradually increased during 20 days of storage period.

Total sugar
As the storage period increased the total sugar content was increased in all the samples. The initial total sugar contents of the RTS beverages was 8.62 per cent in T₀ samples and 8.54 per cent in T₁ samples of amla RTS respectively. At the end of the forty five days of storage the total sugar content was significantly increased to 8.95 in T₀ and 8.87 in T₁ glass bottled samples, and 8.96 in T₀ and 8.80 in T₁ in PET bottled samples of amla RTS respectively. The rise in total sugars could be due to hydrolysis of starch to sugars in juice during storage. The results of the present study are in accordance with the results of [37] that there was a gradual increase in total sugar content of kinnon juice blended with aonla juice and ginger juice.

Total dietary fiber
A slight change in total dietary fiber content was observed during storage. The initial total dietary fiber content was 0.4 in T₀ and 2.3 in T₁ samples of amla RTS beverages respectively. At the end of the storage period the total dietary fiber content was 0.6 per cent in T₀ and 2.41 per cent in T₁ glass and 0.5 and 2.41 in PET bottled samples of amla RTS respectively.

Changes in the microbial population of dietary fiber fortified RTS beverages during storage
At the start, no microbial count was observed in the prepared RTS beverage samples. At the end of storage period amla RTS T0 sample indicated as below detectable level but T1 sample was increased to 0.22 × 10⁵ cfu/ml in both glass and PET bottled samples respectively. Similar pattern of increase in bacterial count 0 to 1.7 × 10⁵ cfu/ml was observed by Jan and Masih [35] in RTS made with pineapple juice, carrot juice and orange juice blend (60:10:30) at the end of storage of 21 days.

The fungal count of was increased to 0.33 × 10⁴ cfu/ml in T₀ and T₁ for samples stored in glass bottles and 0.33 × 10⁴ cfu/ml and 0.44 × 10⁴ cfu/ml for T₀ and T₁ for samples stored in PET bottles respectively. Similar observations were made by Tamilselvi et al., [40] who reported that there was a gradual increase in total number of fungal count during storage of lime based RTS beverage.

Yeast population was increased to 1.00 and 1.30 × 10³ cfu/ml for T₀ and T₁ samples stored in glass bottles and 1.22 and 1.33 × 10³ cfu/ml for T₀ and T₁ samples stored in PET bottles respectively. The increase in microbial load for 45 days of storage was negligible and safe for consumption. These results were in conformity with the finding of Pant et al., [41] who reported that yeast population of RTS beverage from soy whey blended with kiwi fruit pulp have 4.1 × 10⁵ cfu/ml at 45th day of storage.

Conclusion
In the present study, dietary fiber extracted from amla residues was fortified into amla RTS beverages at three different concentration levels with the aim of improving the dietary fiber content. The formulated amla RTS beverages contained more dietary fiber and ascorbic acid. Among the three treatments experimented, amla RTS beverage formulated with 0.5 % dietary fiber fortification was found to be highly acceptable based on the sensory characteristics and physical properties. The RTS beverages stored under refrigeration conditions for forty five days were found to be possessed good sensory characteristics, negligible microbial load and fair amounts of dietary fiber and ascorbic acid. The physico chemical characteristics were significantly different among treatments, storage period and packaging materials. Among the two different packaging materials, glass bottles retained more chemical constituents when
compared to PET bottles. On the basis of the above results, it is concluded that amla residues thrown as waste after juice extraction may be utilized for the production of dietary fiber and it can be successfully utilized for the production of good quality and healthy RTS beverages.

References