Changes in quality parameters and microbial stability of hog plum (Spondias mombin Linn.) juice during ambient and refrigerated storage

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Abstract

Purpose – Several factors may play critical roles in alterations to product quality during storage of hog plum juice. This study aims to evaluate variations to physicochemical, antioxidant, anti-nutritional properties and microbial stability of hog plum juice during storage.

Design/methodology/approach – Juice was produced from hog plum fruits and stored for eight weeks at refrigerated and ambient conditions. Physicochemical, antioxidant properties, antinutritional factors and microbial properties of juices were determined using standard procedures

Findings – Degradation of ascorbic acid was higher in juices stored at ambient conditions (64.4%) compared to those stored by refrigeration (44.4%). Trends were similar for total phenolic, total flavonoid and total carotenoid contents. Total phenolic, total carotenoid and lycopene contents of fresh juice were 3.9 mg

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GAE/mL, 4.0 mg/mL and 1.3 mg/mL, which were not significantly different (at \( p = 0.08, 0.07 \) and 0.08, respectively) from the values at two weeks of storage at refrigerated conditions (3.9 mg GAE/mL, 3.9 mg/mL and 1.3 mg/mL). A sharp decrease of more than 40% (\( p = 0.02 \)) in lycopene was recorded after four weeks, irrespective of storage temperature. Pasteurized hog plum juice showed no microbial growth until after four weeks of refrigerated storage when 1 CFU/mL each of bacterial and fungal growth were recorded. The juices, however, showed higher susceptibility to fungal growth as storage period increased.

**Research limitations/implications** – Other variables not considered in this study such as nature of packaging materials may have significantly contributed to the observed data set. Further studies may, therefore, widen the scope of discussion to evaluate the associated relationship of these variables. Hog plum juice retained a considerable amount of bioactive components during refrigerated storage, which makes it a viable nutraceutical drink with industrial potentials and possible positive health implications for consumers.

**Practical implications** – This study provides new information that support the possible classification and use of hog plum juice as a safe functional beverage for human consumption.

**Originality/value** – Although the effect of storage temperature was significant in most of the properties studied, storage duration seems to have a greater influence on the stability of quality parameters during the storage of hog plum juice.

**Keywords** Spondias mombin juice, Antioxidant properties, Microbial stability, Refrigerated storage, Antinutritional properties

**Paper type** Research paper

**Introduction**

Fruits are essential components of a nutritious and balanced diet. However, their high moisture contents make them prone to spoilage and deterioration and at risk of high post-harvest losses. In the tropical and sub-tropical regions of the world, which particularly has a rich biodiversity of underused indigenous fruit species, fruits post-harvest loss is usually associated with high ambient temperatures, poor handling practices and evapotranspiration (Sibanda and Workneh, 2020). This results in considerable qualitative and quantitative losses in these fruits, especially during peak season periods. Nonetheless, fruits are naturally low in calories and fats and are rich reserves of nutritive fiber, vitamins, minerals and other important micronutrients (Lim et al., 2007). Their consumption can be linked to reduced risk of chronic human degenerative diseases, and oxidative stress caused by free radicals and reactive oxygen species (Báez et al., 2007).

Hog plum (Spondias mombin L.) is widely grown in many parts of tropical Africa and the Americas (Mattietto and Matta, 2011). It possesses edible acidic fruits that are plum-like in appearance with a predominantly golden yellow color and an average of 37.9%, 24.5% and 37.6% composition of peel, pulp and seed (Dey et al., 2016). S. mombin is among such tropical fruits with considerable levels of essential micronutrients (Oladunjoye et al., 2021). It contains volatile aroma compounds and an exotic taste that has attracted industrial interest owing to its unique flavor and aroma characteristics (Adedeji et al., 1991). However, hog plum fruit is seasonal and underused, having a shelf life of 8–10 days (Sivaprasad et al., 2011).

Except for small-scale production of hog plum pickle and preserves, to the best of the authors’ knowledge, hog plum has not been processed into any commercial industrial product. Hog plum is composed of about 25% of pulp (Dey et al., 2016), which may be exploited in industrial juice production. Hog plum juice powder, which can be rehydrated into juices, has been developed by spray drying on a small scale (Mishra et al., 2017). Processing of hog plum fruit into juices may be a critical agro-industrial venture, that makes its micronutrients and natural bioactive components available, improves its economic value, prevents its waste and minimizes post-harvest losses.
Several variables such as processing method (Olaoye et al., 2021), packaging materials, storage temperature and duration have been shown to play critical roles in alterations to product quality during storage (Mäkilä et al., 2017). Furthermore, the stability of the important microconstituents and other critical quality attributes of hog plum juice during storage at ambient and refrigerated temperatures have been sparsely studied to date. This study aimed to evaluate the variations in physicochemical, antioxidant, anti-nutritional properties and microbial stability of hog plum juice stored at ambient (26 ± 2°C) and refrigerated (4 ± 2°C) temperatures.

Materials and methods
Sources of materials
Matured and ripe hog plum fruits were collected into plastic crates from all five trees within the Faculty of Technology fruit garden of the University of Ibadan, Oyo State, Nigeria, in two replicates from August, 2020 harvest. The fruits were sorted to remove unwholesome ones, washed in chlorinated water (180 mg/L), rinsed in distilled water and air-dried using a Class II biosafety cabinet (BSC-I500II B2-X Labotech. Midrand 1685, South Africa) at ambient condition (26 ± 2°C) before further processing. The fruits were processed within 2 h of harvesting.

Preparation of hog plum juice
Hog plum juice was processed by feeding fruits into a juice extractor (TMG 410025; Mumbai, India) to separate the seeds and peels from the pulp. Distilled water (60% w/v) was added to aid efficient extraction. The juice was filtered and pasteurized at 95°C for 1 min. The pasteurized juice was hot-filled into sterilized bottles, corked and stored at refrigerated (4 ± 2°C) and ambient (26 ± 2°C) temperatures for eight weeks. All analyses were carried out in triplicates on the fresh pasteurized samples and the stored samples at two, four, six and eight weeks of storage.

Yield determination
The percentage yield of hog plum juice (mL/g) was obtained according to (1):

\[
Yield = \left( \frac{\text{Volume of juice} - \text{Volume of added water}}{\text{Original weight of fruit}} \right)
\]

Determination of pH
The pH of fresh and stored hog plum juice was determined using a pH meter (3505 Bench Jenway) standardized with buffers 4 and 7 before usage at room temperature.

Total titratable acidity
Juice (2 mL) was diluted with 20 mL of distilled water from which 5 mL was measured into a conical flask and two drops of phenolphthalein were added. The solution was titrated against 0.1 N of sodium hydroxide until the color changed to pink using the AOAC methods 967.21 and 942.15 (2000):
Total Titratable Acidity (TTA) = \frac{\text{Titre value} \times \text{Normality of NaOH} \times \text{Acid equivalent} \times 100}{\text{Volume of juice} \times 1000}

(2)

Total soluble solids (°Brix)
The total soluble solid (°Brix) of freshly extracted and stored hog plum juice was determined by placing two drops of the juice on the glass prism of a digital laboratory refractometer (HSR-500, Swastik Scientific, Mumbai, India) and noting the brix reading after allowing for temperature equilibration.

Ascorbic acid
The ascorbic acid contents of fresh and stored hog plum juices were determined as described by AOAC methods 967.21 and 942.15 (2000), using the 2,6-dichloroindophenol titrimetric method.

Total phenolic content
Total phenolic contents (TPCs) of freshly extracted and stored juices were determined using the Folin–Ciocalteu assay method as described by Oladunjoye et al. (2021). Absorbance was read on the spectrophotometer at 760 nm and the result was expressed as milligram of gallic acid equivalent (GAE) per milliliter of juice.

Total flavonoid content
The AlCl₃ colorimetry method was used in the determination of total flavonoids. Briefly, 1 mL of juice was added to 0.5 mL of 2% AlCl₃ in methanol (1:50 w/v). Exactly 0.5 mL of 1 M aqueous sodium acetate and equal volumes of double-distilled water, HCl, and ammonium acetate were added. The mixture was vigorously shaken and allowed 10 min of incubation before taking spectral readings at 425 nm. Results were expressed as milligram of quercetin equivalents per milliliter of the sample (mg QE/mL) (Liu et al., 2017).

Total carotenoid contents
Total carotenoid (TC) content of fresh and stored hog plum juices was determined according to the method of Castro-López et al. (2016) with slight modifications. About 0.5 mL of the juice was incubated in a 25 mL mixture of ethanol, hexane and acetone (1:2:1) in a beaker for 30 min at 37°C. The mixture was then centrifuged (5810R; Eppendorf Hamburg, Germany) at 5,000 × g for 10 min at 4°C. The absorbance of the decanted centrifuged supernatant was read at 453 nm in a spectrophotometer with n-hexane as blank.

Lycopene
The method of Isnaeni et al. (2019) was followed with slight modification. Precisely 2 mL of hog plum was weighed and mixed with 25 mL of hexane containing 2% dichloromethane, before an incubation period of 20 min at ambient temperature (26 ± 2°C) for lycopene extraction. The extract was separated, diluted with a solvent to 100 mL and read on a UV–VIS spectrophotometer at 505 nm. The lycopene content of the extract was determined using (3):
\[ Lycopene \text{ Content} \left( \frac{mg}{mL} \right) = \frac{A}{0.30} / (V \times d) \] (3)

where \( A \) is the absorbance at 505 nm, the slope of the standard curve is 0.30, while \( V \) represents the volume of hog plum juice, and \( d \) is the dilution ratio.

**Tannin**
To 25 mL of a mixture of acetone and acetic acid, 0.5 mL of juice was added and the mixture was allowed to stand for 4 h. From the solvent extract, absorbance was read at 500 nm on the spectrophotometer. The absorbance recorded was compared against that of a standard solution of tannic acid (Adepoju, 2009).

**Saponin**
In a boiling tube, 0.5 mL of the juice was measured, to which 10 mL of 80% ethanol and 20 mL of 40% magnesium carbonate were added. The mixture was filtered, and from the filtrate, 1 mL was measured into a 50-mL volumetric flask, and 47 mL of distilled water and 2 mL of 5% ferric chloride solution were added. The mixture was allowed to stand for 30 min until the color changed to red. The absorbance was read at 360 nm (Edema and Alaga, 2012).

**Phytate**
The phytate contents of fresh and stored juice were determined by titration with ferric chloride solution (Adepoju, 2009).

**Microbiological analysis**
Assessment of viable growths of total bacterial, yeast and mold and total coliform in the juice samples were carried out using plate count agar, potato dextrose agar and McConkey agar (Oxoid, Basingstoke, Hampshire, UK), respectively, by means of spread plate technique using samples dilution of 0.1 mL (37°C, 48 h). About 100 g/L of tartaric acid was used to regulate the media to a pH of 3.5 before sample inoculation and incubation (25°C, 120 h), after which samples were analyzed and colonies counted. Results were expressed as log CFU/mL.

**Statistical analysis**
All analyses on juice samples prepared were carried out in triplicates and the data obtained were analyzed by one-way analysis of variance, ANOVA using SPSS version 21 to determine the mean and standard deviation. Means were separated by Duncan’s post hoc test and at a statistical significance level set at < 0.05.

**Results and discussions**

**Yield and physicochemical properties of hog plum juice**
The yield and physicochemical properties of fresh and stored hog plum juice are summarized in Table 1. An average juice yield of 0.28 mL/g was obtained for hog plum fruit. This represents about 28.3% of the pulp portion of the fruit, which is higher than values for the pulp fraction of hog plum fruit reported earlier by Dey et al. (2016). Yield may be further improved using techniques such as enzyme liquefaction, ultrasound treatments, high-pressure processing or by soaking fruits prior to extraction. The pH of fresh and stored juices ranged from 2.5 to 2.7, showing no significant difference by storage temperature. These values were higher than the range (2.32– 2.36) reported for hog plum juice by Oladunjoye et al. (2021) but lower than the value (2.83) obtained by Tiburski et al. (2011) for
S. mombin. The difference may be associated with variation in species and maturity stage of fruit used. Low pH may confer enhanced sensory palatability on the juices as postulated by Deshpande et al. (2015).

Titratable acidity values for fresh juice (0.46%) increased insignificantly by the second week of storage and thereafter decreased sharply and significantly ($p = 0.02$) to slowly increase again till the end of the storage period at both storage conditions. The total titratable acidity (TTA) result obtained in this study compared favorably with the values reported by Oladunjoye et al. (2021) for freshly pasteurized hog plum juice (0.46%–0.49%), but higher than the values reported by Nonga et al. (2014) for mixed juice (0.01%) and tamarind juice (1.3%). The TTA of the juices increased as the storage period increased except at the fourth week of storage in both juices. The increase in TTA during storage may be attributed to hydrolysis of complex pectic polysaccharides in the fruit pulp, thereby increasing the release of inherent organic acids such as citric, malic, quinic and L-ascorbic acids occasioned by actions of inherent polymerase enzymes (Akter et al., 2013).

The total soluble solids progressively declined from 6.0 °Bx in the fresh juice to 4.5 and 3.9 °Bx in the juices stored at refrigerated and ambient conditions, respectively. Although Brugnoni et al. (2013) found that temperature and storage time had an insignificant effect on soluble solids concentration in their study of cloudy apple juice concentrate, contrarily, it was observed in this study that storage temperature may significantly affect solubilized solid contents in a sour acidic juice such as one from hog plum. This could possibly be related to the increased synthesis of non-volatile organic acids from the anaerobic degradation of sugars in the juice.

Antioxidant properties of hog plum juice
Ascorbic acid. The ascorbic acid (vitamin C) contents of fresh and stored hog plum juices are presented in Table 2. Values were expectedly highest when freshly processed and lowest after eight weeks of storage, decreasing progressively as storage time increased. Interestingly, ascorbic acid content was stable between weeks 2 and 4 of storage without any significant decrease during refrigerated storage. It was observed that vitamin C degradation was higher in juice stored at 26 ± 2°C (64.4%) compared to the juice stored at 4°C ± 2°C (44.4%) at the end of storage and at each time interval. This showed that refrigerated storage of hog plum juice preserved its vitamin C content better than storage at ambient temperature. This observation compared well with the result obtained for the

| Storage temperature | Duration of storage (weeks) | Yield (mL/g) | TTA (%) | pH | °Brix |
|---------------------|----------------------------|--------------|---------|----|-------|
| 4 ± 2°C             | 0                          | 0.28 ± 0.03  | 0.46 ± 0.02<sup>a</sup> | 2.6 ± 0.01<sup>b</sup> | 6.0 ± 1.00<sup>a</sup> |
|                     | 2                          |              | 0.47 ± 0.02<sup>a</sup> | 2.6 ± 0.01<sup>b</sup> | 5.5 ± 1.00<sup>a</sup> |
|                     | 4                          |              | 0.19 ± 0.02<sup>c</sup> | 2.7 ± 0.01<sup>a</sup> | 5.5 ± 1.00<sup>a</sup> |
|                     | 6                          |              | 0.24 ± 0.02<sup>c</sup> | 2.7 ± 0.01<sup>a</sup> | 4.9 ± 1.00<sup>b</sup> |
|                     | 8                          |              | 0.30 ± 0.03<sup>b</sup> | 2.5 ± 0.01<sup>c</sup> | 4.5 ± 1.00<sup>bc</sup> |
| 26 ± 2°C            | 0                          | 0.28 ± 0.03  | 0.46 ± 0.02<sup>a</sup> | 2.6 ± 0.01<sup>b</sup> | 6.0 ± 1.00<sup>a</sup> |
|                     | 2                          |              | 0.48 ± 1.01<sup>a</sup> | 2.7 ± 0.01<sup>a</sup> | 5.5 ± 1.00<sup>a</sup> |
|                     | 4                          |              | 0.22 ± 0.01<sup>c</sup> | 2.6 ± 0.01<sup>b</sup> | 5.0 ± 1.00<sup>b</sup> |
|                     | 6                          |              | 0.28 ± 0.02<sup>bc</sup> | 2.7 ± 0.01<sup>ab</sup> | 4.5 ± 1.00<sup>bc</sup> |
|                     | 8                          |              | 0.32 ± 0.02<sup>b</sup> | 2.5 ± 0.01<sup>c</sup> | 3.9 ± 0.10<sup>c</sup> |

Notes: Values are means of duplicate samples, each analyzed in triplicates ($n = 6$) ± standard deviation. Means with different superscripts within the same column are significantly different at $p < 0.05$. TTA, total titratable acidity.
degradation rate of vitamin C at refrigerated temperatures in strawberry juice concentrate (Menevseoglu et al., 2020). Furthermore, ascorbic acid is readily oxidized, is heat-labile and highly sensitive to various processing and storage conditions of temperature and light (Tewari et al., 2017). Ascorbic acid retention has previously been used as indicator of quality in food and beverages; however, Sivaprasad et al. (2011) identified “spondiol” as a novel biomarker to benchmark the nutraceutical quality index of hog plum juice during storage.

**Total phenolic content.** The TPCs of fresh and stored hog plum juices are as shown in Table 2. Specifically, the phenolic contents of stored juices ranged from 3.9 to 1.7 mg GAE/mL and 3.9 to 2.1 mg GAE/mL at 26 ± 2°C and 4°C, respectively. There was a slight increase in TPC at the second week, and a retention level of 92.0% after four weeks in refrigerated storage, whereas only 61.5% of TPC was retained in juices stored at ambient conditions for the same duration. The rate of decrease in TPC was lower in the juice stored at refrigerated temperature compared to the one stored at room temperature. This is attributable to sensitivity of phenolic compounds to light and oxygen, as may be encountered during ambient storage. This is in agreement with the study by Touati et al. (2016) that reported a significant decrease in TPC of orange, pear and grape nectars after 7, 14 and 17 days of storage at 25°C and 4°C. Mishra et al. (2017) explained that refrigerated storage of spray-dried hog plum juice powder increased the retention of TPC and DPPH antioxidant activity. Generally, the level of TPC in fruit juices is dependent on the type and location of each phenolic compound in the fruit matrix (Minatel et al., 2017).

**Total flavonoid content.** Variations were noted in the flavonoid contents of fresh and stored juices. Values in stored juices ranged from 1.1 to 0.2 mg QE/mL and 1.1 to 0.3 mg QE/mL at ambient and refrigerated temperatures, respectively. It was observed that there was a gradual and progressive loss as storage duration increased, the highest loss occurring after six weeks until losses of 80.6% at 26 ± 2°C, and 76.6% at 4°C were recorded at the end of eight weeks. This indicated that storage of hog plum juice at refrigerated temperature preserved the flavonoid content better than storage at ambient temperature, considering that low-temperature storage may retard residual peroxidase activity, which otherwise could accelerate degradation of flavonoids in fruit juices (Igual et al., 2011). Phenolic compounds, flavonoids and ascorbic acids may react with free radicals created by aerial oxygen, to cause a reduction during the storage of fruits (García-Torres et al., 2009).

| Storage temperature | Duration of storage (weeks) | Ascorbic acid (mg/mL) | TPC (mg GAE/mL) | TFC (mg QE/mL) | TCC (mg/mL) | LYC (mg/mL) |
|---------------------|----------------------------|-----------------------|----------------|---------------|-------------|-------------|
| 4 ± 2°C             | 0                          | 0.45 ± 0.01<sup>a</sup> | 3.9 ± 0.09<sup>b</sup> | 1.1 ± 0.03<sup>a</sup> | 4.0 ± 0.04<sup>a</sup> | 1.3 ± 0.05<sup>a</sup> |
|                     | 2                          | 0.41 ± 0.01<sup>ab</sup> | 3.9 ± 0.02<sup>a</sup> | 1.1 ± 0.04<sup>a</sup> | 3.9 ± 0.02<sup>a</sup> | 1.3 ± 0.02<sup>a</sup> |
|                     | 4                          | 0.37 ± 0.01<sup>ab</sup> | 3.6 ± 0.04<sup>ab</sup> | 0.9 ± 0.01<sup>b</sup> | 3.5 ± 0.03<sup>ab</sup> | 0.43 ± 0.03<sup>c</sup> |
|                     | 6                          | 0.31 ± 0.02<sup>b</sup> | 2.4 ± 0.05<sup>c</sup> | 0.76 ± 0.01<sup>cd</sup> | 1.8 ± 0.03<sup>cd</sup> | 0.31 ± 0.04<sup>cd</sup> |
|                     | 8                          | 0.25 ± 0.00<sup>c</sup> | 2.1 ± 0.02<sup>d</sup> | 0.38 ± 0.00<sup>e</sup> | 0.93 ± 0.02<sup>de</sup> | 0.3 ± 0.02<sup>cd</sup> |
| 26 ± 2°C            | 0                          | 0.45 ± 0.01<sup>a</sup> | 3.9 ± 0.09<sup>a</sup> | 1.1 ± 0.03<sup>a</sup> | 4.0 ± 0.04<sup>a</sup> | 1.3 ± 0.05<sup>a</sup> |
|                     | 2                          | 0.33 ± 0.02<sup>b</sup> | 3.3 ± 0.05<sup>b</sup> | 0.92 ± 0.01<sup>b</sup> | 2.8 ± 0.03<sup>b</sup> | 0.69 ± 0.02<sup>b</sup> |
|                     | 4                          | 0.35 ± 0.00<sup>b</sup> | 2.4 ± 0.08<sup>c</sup> | 0.81 ± 0.02<sup>d</sup> | 2.0 ± 0.05<sup>d</sup> | 0.38 ± 0.01<sup>cd</sup> |
|                     | 6                          | 0.22 ± 0.01<sup>c</sup> | 2.3 ± 0.04<sup>c</sup> | 0.68 ± 0.01<sup>d</sup> | 1.3 ± 0.04<sup>d</sup> | 0.26 ± 0.01<sup>d</sup> |
|                     | 8                          | 0.16 ± 0.01<sup>d</sup> | 1.7 ± 0.04<sup>e</sup> | 0.22 ± 0.01<sup>e</sup> | 0.78 ± 0.01<sup>e</sup> | 0.21 ± 0.01<sup>e</sup> |

**Notes:** Values are means of duplicate samples, each analyzed in triplicates (n = 6) ± standard deviation. Means with different superscripts within the same column are significantly different at p < 0.05. TPC, total phenolic content; TFC, total flavonoid content; TCC, total carotenoid content; LYC, lycopene content.
Total carotenoid content. Carotenoids are likely accountable for the intense yellow color of ripe and mature hog plum fruits and may be typically related to the visual quality perception of food products because color affects consumer sensory preferences. In an earlier study, a mix of lycopene precursor and β-carotene was also considered to be responsible for the yellow color of some tomato cultivars (Sánchez-Moreno et al., 2008). Values for TC contents of fresh and stored hog plum juices ranged from 4.0 to 0.78 mg/mL and 4.0 to 0.93 mg/mL with a total of 80.3% and 76.5% percentage degradation at ambient and refrigerated temperatures, respectively, at the end of eight weeks of storage (Table 2). Notably, only 11% of the TC was lost after four weeks of refrigerated storage, in contrast to 49% loss recorded in juices stored at ambient temperature. Tiburski et al. (2011) reported a considerably lower content of TCs of 4869 µg per 100 g of S. mombin pulp, while Touati et al. (2016) reported a reduction in TC contents of fruit nectars after 28 days of storage at 4°C, 25°C and 37°C. Oxidation is a possible major cause of carotenoid loss, which is a spontaneous free-radical chain reaction catalyzed by oxygen, light, metals, enzymes and/or peroxides (Saini et al., 2020).

Lycopene content. The lycopene content of fresh hog plum juice was 1.3 mg/mL, which significantly ($p = 0.01$) decreased until the end of storage. Remarkably, there was no significant difference in the values recorded for fresh juice and at two weeks of refrigerated storage, but a sharp decline was observed between the second and fourth week. At the end of the eight weeks of storage, the lycopene contents of the stored juices degraded to 0.21 and 0.3 mg/mL at ambient and refrigerated temperatures, respectively. Higher degradation of lycopene in the juices stored at ambient conditions (possible increase in light and oxygen exposure) may be attributed to autooxidation of the cis-isomers of lycopene which may have caused fragmentation of the lycopene molecules in solution (Xianquan et al., 2005).

Anti-nutritional properties of hog plum juice

Tannin content. Anti-nutrients are compounds that act to limit nutrient digestion, absorption and utilization in the human body and may produce detrimental health effects (Aberoumand, 2012). The anti-nutritional properties of hog plum juice are presented in Table 3. There was no significant difference between the tannin content of juices stored at ambient and refrigerated temperatures at the end of two weeks of storage. The tannin content of fresh juice was 0.36 mg/mL, while those of stored hog plum juices ranged from

| Storage temperature | Duration of storage (weeks) | Tannin (mg/g) | Saponin (µg/g) | Phytate (mg/g) |
|---------------------|-----------------------------|---------------|----------------|----------------|
| 4 ± 2°C             | 0                           | 0.36 ± 0.01b  | 30.1 ± 0.01a   | 0.15 ± 0.01c   |
|                     | 2                           | 0.35 ± 0.02b  | 30.0 ± 0.02a   | 0.13 ± 0.01c   |
|                     | 4                           | 0.36 ± 0.03b  | 30.0 ± 0.05a   | 0.58 ± 0.08c   |
|                     | 6                           | 0.40 ± 0.02a  | 30.1 ± 0.04b   | 0.64 ± 0.01e   |
|                     | 8                           | 0.38 ± 0.04ab | 29.9 ± 0.01c   | 0.68 ± 0.10f   |
| 26 ± 2°C            | 0                           | 0.36 ± 0.01b  | 30.0 ± 0.03a   | 0.15 ± 0.01c   |
|                     | 2                           | 0.34 ± 0.01b  | 29.9 ± 0.04a   | 0.10 ± 0.01f   |
|                     | 4                           | 0.41 ± 0.00a  | 29.9 ± 0.01a   | 0.85 ± 0.08b   |
|                     | 6                           | 0.44 ± 0.20a  | 30.0 ± 0.02a   | 0.91 ± 0.02a   |
|                     | 8                           | 0.43 ± 0.00a  | 30.0 ± 0.03a   | 0.91 ± 0.08a   |

Table 3.
Variations in anti-nutritional properties of hog plum juice during ambient and refrigerated storage

Notes: Values are means of duplicate samples, each analyzed in triplicate ($n = 6$) ± standard deviations. Means with different superscripts within the column are significantly different at $p < 0.05$. 

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0.35 to 0.4 mg/mL in the refrigerated juice, and 0.34 to 0.44 mg/mL in the ambient-stored juices. These values were higher compared to 3.4 mg/100 g for banana fruit juice and similar to 48.2 mg/100 g for sweet orange juice as reported by Onibon et al. (2007). The tannin contents of hog plum juices were less than the permissible limit of 20 mg/g as reported by Ndidi et al. (2014). High tannin contents may affect organoleptic properties in terms of astringency, color and turbidity of juices (Prommajak et al., 2020).

Saponin content. The saponin content of hog plum juice was stable at 30 μg/mL at both ambient and refrigerated temperatures throughout the eight weeks of storage. High concentrations of saponin may impart a bitter taste, a major limiting factor to its use. However, saponins have been shown to also possess anticarcinogenic properties, immune modulation activities, active in the regulation of cell proliferation and cholesterol-lowering activity (Seigler, 1998).

Phytate content. The phytate content of fresh hog plum juice was 0.15 mg/mL, while those of stored juices ranged from 0.10 to 0.91 mg/mL and 0.13 to 0.68 mg/mL at ambient and refrigerated temperatures, respectively. The values obtained in this study were lower compared to 2.88 mg/100 g in banana fruit juice and 10.71 mg/100 g in sweet orange as was reported by Onibon et al. (2007). These values are also lower than the maximum tolerable limit (2.5–5 mg/g) for human consumption (Ndidi et al., 2014). Phytate may bind essential minerals such as zinc, calcium, copper, magnesium and iron to form insoluble complexes, resulting in a deficiency of such minerals even if consumed from diets.

Microbiological properties of hog plum juice
There was no growth of bacteria nor fungi in the fresh, pasteurized juice and after two weeks of refrigerated storage. This shows the effectiveness of the pasteurization treatment to eliminate any primary and secondary microbial contaminations in the juice. The values for bacterial count however increased from 1 CFU/mL at four weeks to 2 CFU/mL at the end of eight weeks, whereas, a single fungal growth was also noted at Week 4 but increased to 2.5 CFU/mL after eight weeks (Table 4). For the juice stored at ambient temperature, the total bacterial and total fungal counts ranged from 1.1 to 3.3 CFU/mL and 1.1 to 5.0 CFU/mL from the second week to the end of eight weeks of storage. Hog plum juice was observed

| Storage temperature | Duration of storage (weeks) | TBC (CFU/mL) | TFC (CFU/mL) | TCC (CFU/mL) |
|---------------------|-----------------------------|--------------|--------------|--------------|
| 4 ± 2°C             | 0                           | ND           | ND           | ND           |
|                     | 2                           | ND           | ND           | ND           |
|                     | 4                           | 1.0 ± 0.02^c | 1.0 ± 0.82^d | ND           |
|                     | 6                           | 1.2 ± 0.03^c | 2.5 ± 0.05^c | ND           |
|                     | 8                           | 2.0 ± 0.04^b | 2.5 ± 0.05^c | ND           |
| 26 ± 2°C            | 0                           | ND           | ND           | ND           |
|                     | 2                           | ND           | ND           | ND           |
|                     | 4                           | 1.1 ± 0.01^c | 1.1 ± 0.04^d | ND           |
|                     | 6                           | 1.5 ± 0.02^c | 2.3 ± 0.05^c | ND           |
|                     | 8                           | 1.6 ± 0.01^c | 3.2 ± 0.05^b | ND           |
|                     | 8                           | 3.3 ± 0.04^a | 5.0 ± 0.05^a | ND           |

Notes: Values are means of duplicate samples, each analyzed in triplicate (n = 6) ± standard deviation. Means with different superscripts within the same column are significantly different at p < 0.05. TBC, total bacterial count; TFC, total fungal count; TCC, total coliform count; ND, not detectable.
to be more susceptible to fungal contamination than bacterial, supporting the report of Moss (2008) that fruits having low pH and high simple carbohydrates components like hog plum may be ideal substrates for fungal growth. Some mold species such as *Byssochlamys fulva* and *Neosartorya fischeri* can survive high pasteurization temperatures, low pH of about 3–4.5 and low oxygen tension to produce pectinolytic enzymes which affect the stability of juice during storage (Tournas et al., 2006). Coliform was not detected in all samples at all storage duration intervals and at both storage temperatures considered. Fruit juices are high-risk foods whose microbiological status may negatively influence safety, organoleptic and physicochemical attributes of the product.

**Conclusion**

Hog plum juice has considerable levels of bioactive compounds such as ascorbic acid, total phenolic, flavonoid and carotenoid contents, and low contents of antinutritional factors, which makes it potentially promising as a natural functional beverage in the fruit juice industry. Ambient and refrigerated storage had varying effects on the stability of the bioactive compounds, showing significant decline as storage duration increased. However, total phenolic, flavonoid and lycopene contents were stable in the first 14 days of storage at refrigerated conditions. The pasteurization treatment sufficiently eliminated all secondary microbial contaminants as revealed by the absence of bacterial, fungal or coliform growth in the fresh juice. The low total bacteria and fungi counts recorded at the end of storage with an absence of coliform showed that hog plum juice would possess considerable shelf life even without the use of chemical preservatives. Storage duration, however, had greater influence on stability of hog plum juice quality.

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