

DEVELOPMENT AND STANDARDIZATION OF TECHNOLOGY FOR PREPARATION AND STORAGE OF VALUE-ADDED PRODUCTS FROM *KENDU* (*DIOSPYROS MELANOXYLON ROXB.*) FRUIT

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ABSTRACT

Kendu is a highly nutritious seasonal fruit widely grown in India. It is a rich source of antioxidants, β -carotene, and phenols. Due to the lack of national recognition, this fruit remains underutilized. A significant portion of the fruit is lost due to its short shelf life and lack of processing technology. In the present work, the development and standardization of technology for various value-added products from *kendu* were explored to avoid spoilage and wastage of the fruit. Products like *kendu* nectar, jam, powder, and bar were developed, and the process was developed and optimized for extended shelf life. Chemical preservatives such as sodium benzoate (356.01 ppm) and potassium-meta bisulfate (602.81 ppm) were optimized for pulp preservation. Maximum consumer acceptability and microbial stability of the value-added products were obtained till 90 days of storage when stored at $4\pm 2^\circ$ C. Effect of microbial, sensorial, and physicochemical properties of products were also investigated for each of their storage conditions. A significant ($p < 0.05$) decrease in the total phenolic content was observed for all the products during the storage period. The results obtained from consumer acceptability and microbial load revealed that all products remained acceptable for consumption till the end of the storage period. Therefore, *kendu* fruit can be converted to value-added products during the available season to improve the variety of fruit-based products on the market shelf. This value-addition improves the commercial cultivation of the fruit, nutritional requirements of the people and enhances the livelihood of the locals in terms of economic sustainability.

Keywords: *Kendu* fruit, value-addition, process optimization, microbial stability, storage study

INTRODUCTION

Kendu (*Diospyros melaxoxylon Roxb.*) is a nutritious fruit abundantly available in India. It is a rich source of phenols and antioxidants. Due to lack of commercial cultivation, this fruit is grouped under minor forest produce (MFP). The fruit can also be seen in some south Asian countries (Hmar et al. 2017a). *Kendu* trees can be used for various applications like timber, food, and medicine (Orwa et al. 2009). The leaves of the trees are commonly used for manufacturing *bidi* (an indigenous traditional cigarette, which uses the *Kendu* leaf for rolling instead of paper cigarette roll). Its trade name is *ebony*, *Kendu* /*Tendu*, also called *coromandel ebony* (Saxena, 2003). It has an essential contribution to the socio-economic condition of the tribal people in India (Gupta et al. 2013). Apart from the commercial and economical utilization of *kendu* leaves, the *kendu* fruit has a good prospect for value addition and can be explored as an alternate nutritive food. The fruits are seasonally available in sufficient quantities in some South Asian countries during the summer months, May to June (Sadangi and Sahu, 2004; Hmar et al. 2017b).

Tribal individuals gather *Kendu* fruits during summer, and the fruit pulp is dried for utilization during off seasons for nourishment. The fruit has a pleasant and sweet taste when fully ripen. But the food value of the *Kendu* fruit has not been explored to its full potential. The fruit pulp is used as a traditional medicine due to its carminative and astringent properties by tribal communities in India. Very few works have been reported on the presence of several phytochemicals such as beta-carotene, terpenoids, flavonoids, saponin, tannin, and betulin in the fruit contributes to its high nutritive value (Natural Medicine Facts, 2015). The fresh pulp can be used as a recipe for the preparation of various organic foods such as *pakora*, *upama*, *pickle*, *paratha*, *jam*, etc., which are sold locally in the Indian and international market (Vivek et al. 2016; Vivek et al. 2018).

The nutritional value and energy-rich properties of *Kendu* pulp have not been utilized for value-addition purposes since the fruit is seasonal and has a lower shelf life. Therefore, preservation techniques are mandatory to increase the storability of the fruit pulp (Hmar et al. 2017b). Processing of fruits to juices

and other value-added products are the alternative ways in which excess fruits can be utilized to reduce wastage and bring economic returns. In this study, the process development and standardization of technology for the preparation of various value-added products and its storage requirements were explored for better utilization of the *Kendu* fruit in the off-season.

MATERIALS AND METHODS

Materials

Kendu fruits were procured from the local market of Rourkela ($22^\circ 14' 57''$ N $84^\circ 52' 58''$ E) of Sundergarh District, Odisha, India. The fruit samples obtained were cleaned in running tap water to remove any possible impurities and dirt. The clean whole fruit samples were packed in polyethylene pouches and stored at $4\pm 1^\circ$ C until extraction. All analytical grade chemicals were procured from sigma Alrich.

Extraction of *Kendu* pulp

The pulp of clean ripened *Kendu* fruits was extracted using a customized fruit pulper. Water was added at a ratio of 1:0.3 to achieve the maximum pulp recovery to extract the pulp that adhered to the seeds (Chakraborty et al. 2010). The extracted pulp was sieved to remove the fiber, packed in pouches, and was stored at -18° C till further use.

Optimization of preservatives

The extracted pulp was pasteurized at $80\pm 2^\circ$ C for 30 minutes in a water bath and packed in high-density polyethylene bags. The pasteurized pulp was chemically treated and kept at $4\pm 1^\circ$ C for further microbial, sensory, and physicochemical analysis. Different concentrations of chemical preservatives such as sodium benzoate (SB) and potassium meta bi-sulphite (KMS) was added within the

permissible limit from the chosen range of 300 to 500 ppm for SB and 500 to 700 ppm for KMS. The maximum allowable preservatives added for a fruit pulp is SB of 500 ppm and KMS of 1000 ppm for microbial stability as per FAO (2016a) standards and permissible limit of PFA (2004). One way factorial design was used to design the number of experiments with five (5) center points and triplicates for each reading. The minimum concentration of chemical preservatives was optimized for good quality and maximum microbial stability of pulp during storage conditions. Optimization was done on the basis of response such as pH, total soluble solids (TSS), titratable acidity (TA), and color (yellowness of the fruit, b*) using fresh pulp as control. ANOVA (Analysis of variance) was also performed to confirm the significance of each response to the experimental data for the chemically preserved *Kendu* pulp.

Value-added products from *kendu* fruit

Preparation of *Kendu* nectar

Kendu pulp was mixed with sugar syrup (Sugar + water + citric acid) according to the FAO (2016b) standards. The nectar prepared was poured into a sterilized bottle and was stored at a refrigerated temperature (4 ± 1 °C).

Preparation of *Kendu* jam

Fresh *Kendu* pulp was mixed with different ingredients (Sugar + water + citric acid + pectin) according to the FAO (2016b) standards for fruit jam preparation. The mix was boiled and stirred continuously to get the required jam consistency and acidity. Filling of the final product was done immediately under the sterile condition, and the jam bottles were stored at a refrigerated temperature of 4 ± 1 °C until further storage analysis (Hmar et al. 2017c).

Preparation of *Kendu* powder

Fresh *Kendu* pulp was dried in a tray dryer at a temperature of 60° C for the production of *Kendu* powder (Hmar et al. 2017c). The pulp was dried until a moisture content of 4 % (dry basis) was achieved. The dried product made from *Kendu* pulp was ground to powder by a ball mill. The powder was stored in airtight glass containers at ambient temperature (27–37° C) for different storage analysis.

Preparation of *Kendu* bar

Fresh *Kendu* pulp was mixed with ingredients (water + sugar + citric acid + Sodium benzoate (0.2 %) as explained with different according to the FAO (2016b) standards. The mix was heated at 70-80° C and was placed in a tray dryer and dehydrated until the product acquires a leather-like consistency (20 ± 2 % moisture). The product was sliced and wrapped in a cellophane bag and was stored at a refrigerated temperature of 4 ± 1 °C for further storage analysis.

Microbial and Sensory analysis

The shelf life of all the products was determined on the basis of both microbial stability and sensory evaluation for all the samples following the method suggested by Lawless and Heymann, (2010). Microbial load in the samples was found out using total viable count (TVC) with standard nutrient agar. The serial dilution and plate count method was used for the analysis (Vivek et al. 2017).

TVC gives a quantitative idea about the presence of all microorganisms such as heterotrophic bacteria, yeast, and mold in a sample (Hashmi et al. 2007). Sensory evaluation of all the value-added products was done by the most widely accepted scale for measuring food acceptability, a 9-point hedonic scale. It was accomplished by a panel of 10 judges for taste, flavor, appearance, and aroma. The sensory analysis of all the value-added products was at the end of their respective storage life.

Physico-chemical analysis of value-added products

The physicochemical analyses of the chemically preserved pulp and all the value added products was investigated till the samples achieve unacceptable condition. The pH of the product was measured using a digital pH meter (Eutech, pH 700, USA). Titratable acidity (TA) was measured following standard titration protocol as was expressed with a percent of citric acid in accordance with AOAC (2016). Total soluble solids (TSS, ° Brix) were measured using a refractometer (ERMA Inc., Japan) (Hmar et al. 2017b). 2, 6-dichlorophenol indophenol method and 3, 5-dinitrosalicylic acid (DNS) method was used for measurement of ascorbic acid and total sugars, respectively. Standard procedures of the hot air oven method were followed by moisture content measurement, according to AOAC (2016). Total phenolic content (TPC) was determined using Folin Ciocalteu reagent (FCR), and the absorbance was taken at 765 nm using a spectrophotometer (Lutz et al. 2015). It was expressed as mg gallic acid equivalents/100g of fresh pulp (mg GAE/100g). *Kendu* pulp color was measured using Color Flex EZ (Hunter Lab, USA). Readings of each color index in the Hunter scale (L, a, b) were taken per sample with three replicates.

Statistical Analysis

Experimental design for optimization of chemical preservatives was done using One-way factorial design with five center points using Design Expert Software (Version 7.0). The selection of the optimized chemical preservative concentration was made by the desirability function. One-way analysis of variance (ANOVA) was performed to check the significant differences in the examined response for the chemically treated samples. For each treatment, three replicates were performed. The significance between the mean values of experimental data was analyzed using Turkey's t-test at $p < 0.05$ significance (95 % confidence level) by SPSS (Ver 22.0, USA).

RESULTS AND DISCUSSION

Optimization of preservatives

One-way ANOVA showed all responses have a significant effect ($p < 0.05$) on each different concentration of chemically preserved pulp samples as given in Table 1. The responses such as pH, TSS, TA, and color were targeted to be in range within the untreated fresh pulp values. The desirability function was used to select the optimized solution for the different chemical concentrations within the designed experiment. SB of 356.01 ppm and KMS of 602.81 ppm were chosen at the desirability of 0.87 and 0.89, respectively. Optimization was done to have the chemically preserved pulp properties in resemblance in terms of physicochemical properties of the fresh pulp.

Table 1 Total viable count (TVC) of optimised *Kendu* pulp

Sl. No	Name of the product	TVC (log CFU*/g),							
		Storage life (days)							
		0	15	30	45	60	75	90	105
1	Fresh pulp	3×10^1	17×10^3	42×10^5	-	-	-	-	-
2	Pulp ₃₅₀ SB	2×10^1	6×10^2	2×10^3	5×10^3	7×10^3	8×10^3	9×10^3	12×10^3
3	Pulp ₆₀₀ KMS	2×10^1	5×10^2	1×10^3	3×10^3	4×10^3	6×10^3	7×10^3	10×10^3

Legend: *CFU- colony forming unit; SB- sodium benzoate; KMS-potassium meta-bisulphite; NF- Not found. Values are the mean of three replicates.

Total viable count (TVC) of the fresh and both the optimized chemically preserved pulp was examined after every fifteen (15) days. It can be concluded that both the chemically preserved products could be stable in terms of total plate count up to nine (9) months while fresh *Kendu* pulp can be stored just for a maximum of three days at a refrigerated temperature of 4 ± 1 °C (Figure 1). An exponential increase can be seen for the fresh pulp after fifteen days of storage interval. This shows the instability of unpreserved pulp at ambient storage

temperature. TVC for both the chemical rested pulp followed a linear increase in the microbial count with their storage period and condition. Chemically preserved pulp at SB at 350 ppm and KMS at 600 ppm of the sample showed acceptable microbial stability after a storage period of ninety (90) days at 4 ± 1 °C, as shown in Table 1. The changes in the physicochemical properties of the preserved pulp can be to have a significant difference ($p < 0.05$) with different chemical preservatives used, as shown in Table 2.

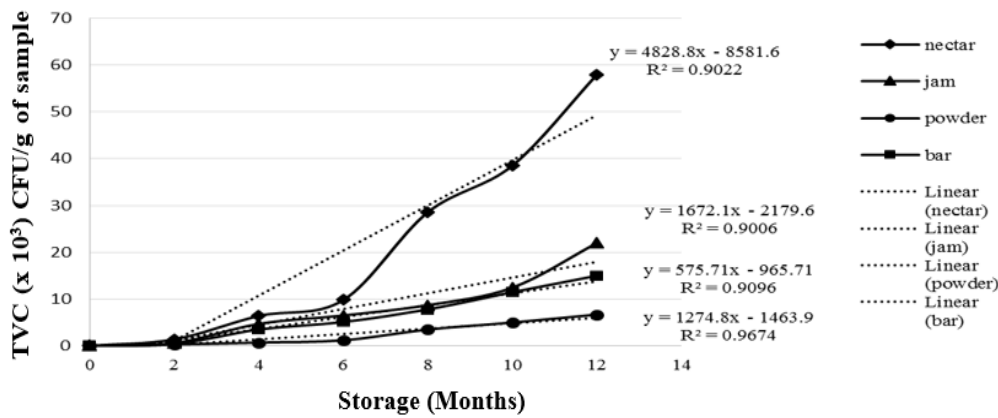


Figure 1 Average total viable count (TVC) on Kendu products during storage

Similar fruit pulp like mango concluded that chemical treatment containing KMS 1000 ppm and SB 500 ppm was effective against microorganisms without any significant changes in chemical composition and sensory characteristics up to 90 days of storage period. Hashmi et al. (2007) have also concluded that KMS has higher control over microbial growth for mango pulp as compared to SB. It was found that pasteurized mango pulp could be stored for an extended period of time as compared to the fresh unpasteurized pulp. The results recorded in this investigation are in agreement with other microbial qualities of pear and mango pulp (Saini et al. 2003; Sakhale et al. 2012).

Table 2 Results of Turkey’s significant test between responses of chemically preserved Kendu pulp after 90 days.

Chemical preservative	Concentration, (ppm)	Response			
		pH	TSS (°Brix)	Acidity (%)	Colour (b-value*)
SB	300	5.16 ^a	16.00 ^a	0.26 ^a	13.40 ^a
	350	5.03 ^b	16.66 ^b	0.30 ^b	13.44 ^b
	400	4.93 ^b	17.66 ^c	0.31 ^{bc}	13.49 ^{bc}
	450	4.89 ^c	18.33 ^d	0.31 ^{bc}	13.50 ^c
	500	4.86 ^c	18.39 ^d	0.32 ^c	14.45 ^d
KMS	500	5.13 ^a	16.66 ^b	0.24 ^a	12.58 ^a
	550	5.06 ^{ab}	17.33 ^{bc}	0.26 ^b	13.49 ^{bc}
	600	5.03 ^b	17.66 ^c	0.30 ^c	13.58 ^c
	650	4.96 ^b	17.68 ^c	0.31 ^{bc}	14.01 ^e
	700	4.86 ^c	18.66 ^d	0.32 ^c	14.35 ^{de}

Legend: *b-value -yellowness index for colorimeter reading (L, a, b); SB- Sodium Benzoate; KMS- potassium meta-bi sulphate; ppm-parts per million. Values are mean of three replicates. Means showing different letters in the same column are significantly different (p<0.05).

Value-added products

Microbial stability and sensory analysis of processed products during storage

The shelf life of all the processed Kendu pulp samples was determined both on the basis of total viable counts (TVC) and 9- point hedonic scale sensory analysis. TVC varies in different ranges for all products during their respective shelf life limit as was found to have a linear increase, as shown in Figure 1. The number of plate counts in Kendu nectar increased from 0.2 to 9 cfu×10³/g at the end of six (6) months than in refrigerated storage. The storage life of nectar was comparatively less than other products might be due to the incorporation of a large amount of water (Shahnawaz et al. 2012). The stability of Kendu jam bar product remains up to eight (8) months under given storage condition with plate counts increase linearly from 0.2 to 8.7 cfu×10³/g and 0.2 to 7.8 cfu×10³/g respectively within their storage condition. A negligible increase in the moisture

content of the Kendu bar was from 20.4 % to 22.0 % dry basis, which makes the product stable. Among all the samples, Kendu powder has the maximum storability up to twelve (12) months as the moisture that remains stable (4± 0.5 % dry basis) inside a sealed glass container. The average total viable count of powder increases linearly from 0.2 to 6.7 cfu/g of the sample at the end of ambient storage, as shown in Figure 1. Similar microbiological evaluation of processed products of minor fruits stored under both ambient and refrigerated conditions was earlier investigated by (Chakraborty et al. 2010; Chaurasiya et al. 2011). The average value of overall acceptability lies on and above 7 points on the scales (like moderately to like extremely), as shown in Figure 2. It was found that Kendu jam and bar has a maximum overall acceptability in terms of sensory analysis (Figure 2).

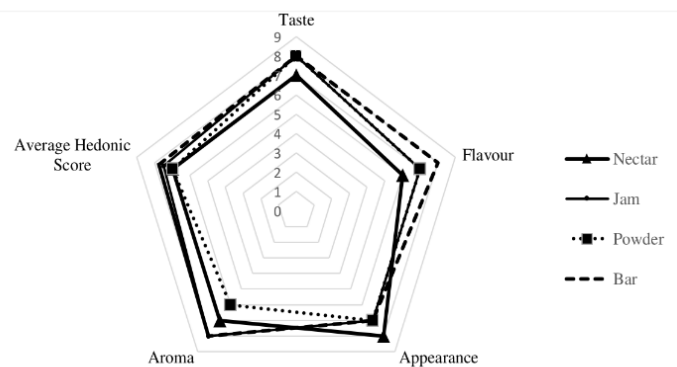


Figure 2 Radar graph showing the sensory profile of different Kendu products at the end of their respective storage life 9-Like Extremely; 8-Like Very Much; 7-Like Moderately; 6-Like Slightly; 5- Neither Like nor Dislike; 4-Dislike Slightly; 3-Dislike Moderately; 2-Dislike Very Much; 1-Dislike Extremely

Physico-chemical analysis of value-added products

The value-added Kendu pulp products were tested for physicochemical, microbiological, and sensory characteristics. The changes in the physicochemical properties of value-added products can be seen in the bar graph represented in Figure 3. The turkey’s t-test performed for comparison of the means between the initial products and products after the end of storage life shows non-significant differences (p > 0.05). Minor changes can be reported, such as a slight increase in total soluble solids (TSS), titrable acidity (TA), and total sugar, while a small decrease in ascorbic acid, pH processed products have been observed for the storage condition (Figure 3 and 4).

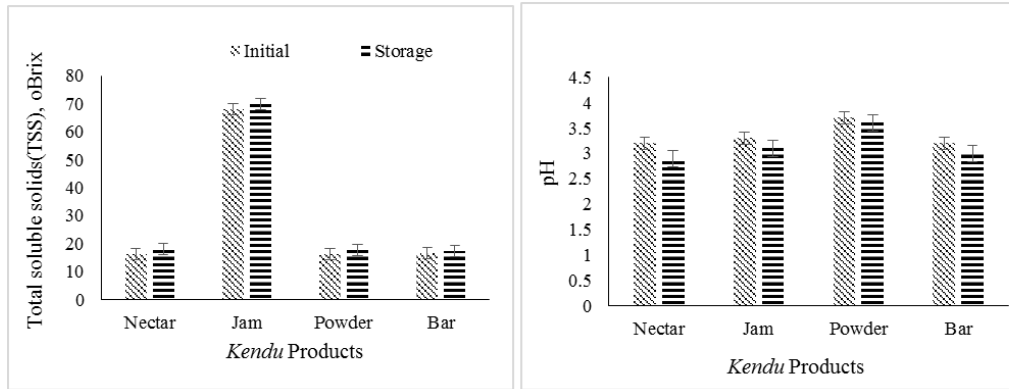


Figure 3 Effect of storage on Tss and pH graph of various value-added products from *kendu*

The increase in TSS might be due to loss in moisture and hydrolysis of polysaccharides into simple sugar (Athmaselvi et al. 2014). The reason behind the increase in total sugar content during storage might be due to the partial hydrolysis of complex carbohydrates (Sharma et al. 2011). The increase in acidity can be due to the addition of preservatives that converts into sulfurous

acid in products. The decrease in ascorbic acid can also be observed due to exposure to heat and oxygen that leads to enzymatic degradation during processing and storage (El-Sohaimy and Hafez, 2010; Mooz et al. 2012).

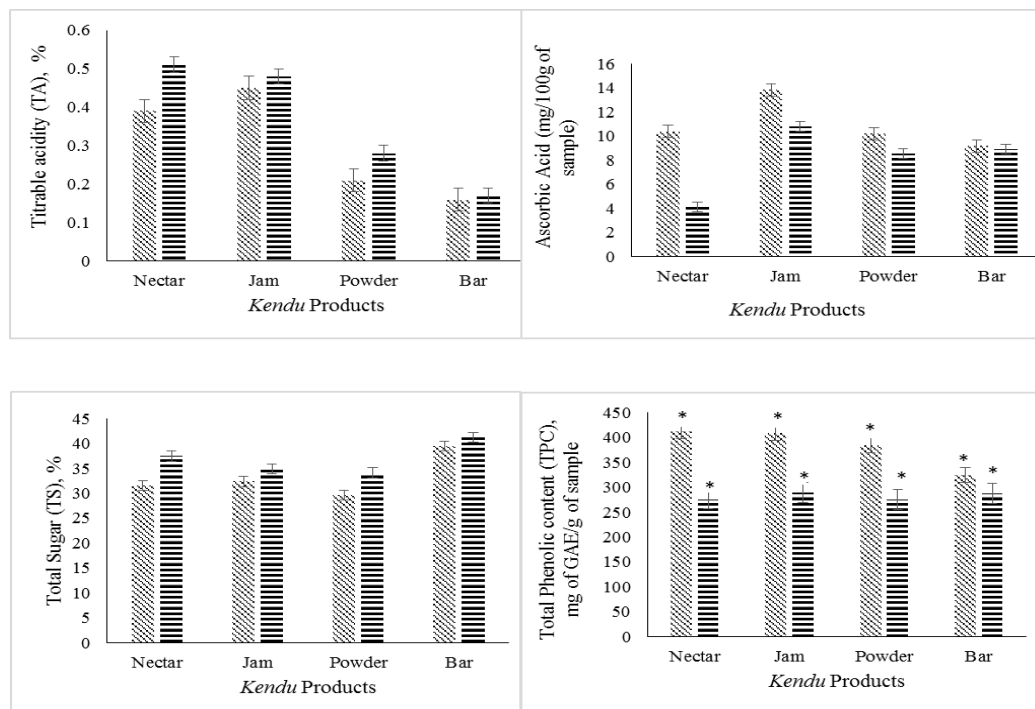


Figure 4 Effect of storage on TA, AA, TS and TPC graph of various value-added products from *kendu*

Total phenolic content (TPC) was found to decrease significantly ($p = 0.024$) with storage (Figure 4). This may be due to the sensitivity of phenolic compounds to heat treatment. Many factors affect the stability of phenolic content, including temperature, pH, oxygen, enzymes, ascorbic acid, etc. Loss of phenolic pigments is probably due to oxidation as well as due to condensation of phenolic pigments with ascorbic acid even in a short period. (Kalt et al. 1999). Similar results were found where a decrease in the content of phenols and ascorbic acid in orange

juice during storage were most affected by both duration and temperature of storage (Klimczak et al. 2007). The changes in all the physicochemical properties of these value-added products were within the acceptable limits for each product as given by FSSAI, (2010). The ANOVA table for various physicochemical properties for both the preservatives was presented in Table 3.

Table 3 ANOVA between sodium benzoate (SB) and potassium meta bi-sulphate (KMS) treated samples and each response

Concentration			SS	df	MS	F	Sig.
SB	pH	Between Groups	.183	4	.046	21.215	.000
		Within Groups	.022	10	.002		
		Total	.205	14			
	TSS	Between Groups	15.067	4	3.767	14.125	.022
		Within Groups	2.667	10	.267		
		Total	17.733	14			
	Acidity	Between Groups	.015	4	.004	19.914	.000
		Within Groups	.002	10	.000		
		Total	.017	14			
	Colour	Between Groups	6.074	4	1.519	315.046	.000
		Within Groups	.048	10	.005		
		Total	6.122	14			

KMS	pH	Between Groups	.124	4	.031	9.300	.002
		Within Groups	.033	10	.003		
		Total	.157	14			
TSS		Between Groups	6.267	4	1.567	4.700	.022
		Within Groups	3.333	10	.333		
		Total	9.600	14			
Acidity		Between Groups	.030	4	.008	22.820	.000
		Within Groups	.003	10	.000		
		Total	.034	14			
Colour		Between Groups	5.330	4	1.333	1537.508	.000
		Within Groups	.009	10	.001		
		Total	5.339	14			

Legend: SB-Sodium Benzoate; KMS-Potassium meta bi-sulphate; SS-sum of squares; df-degree of freedom; MS-mean square, Sig-significance, TSS- Total soluble solids.

CONCLUSION

It can be concluded that the various value-added products can be successfully developed from underutilized *Kendu* fruit like jam, nectar, bar, and powder. Slight changes for physicochemical properties were observed but were in the acceptable range till the end of the storage period. Physico-chemical composition, sensory evaluation, and microbial status revealed that all these products remained acceptable. The storage studies indicated that *Kendu* products could easily be stored from six months to twelve months for each of their storage conditions. Thus, value-addition improves the commercial cultivation of the fruit, nutritional requirement of the people and enhances the livelihood of the locals in terms of economic sustainability

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