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Standardisation of Karonda incorporated Vadiyam and evaluation of nutritional quality during storage

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Abstract
The objective of research experiment was to develop a novel product by incorporating karonda fruit. Vadiyam (A traditional south Indian dish used as a side dish in the main meal) was tried by incorporating karonda fruit and the process was standardised successfully. Karonda Vadiyam prepared with equal proportion of karonda fruits and black gram recorded the highest overall acceptability (8.13) score and was in the "Like very much range". There was no bacterial and mould growth during storage for six months. The results revealed that sensory scores for colour, flavour, texture, taste and overall acceptability did not show any significant difference during storage for six months and showed good overall acceptability (8.00) at the end of six months storage period and was in the "Like very much" range. The nutritional value, microbial safety and overall acceptability revealed the possibility of incorporating karonda fruits for preparation of Vadiyam.

Keywords: Karonda, Processing, Value added products, Nutritional quality, storage studies

Introduction
Developing suitable processing and marketing strategies for the underutilized fruits creates demand in the domestic and international markets (Gajanana *et al.*, 2010) [4]. Most of the underutilized fruits are available in local markets and they are not known in other parts of the world. The nutritional value, nutraceutical and medicinal properties with resistance to biotic and abiotic stress conditions of these fruits, draws attention for providing solutions to fight hunger, poverty and malnutrition which finally results in rural development (FAO, 2018) [3]. Health-conscious consumers can be satisfied by these fruits due to the therapeutic and nutritive value. One such underutilized fruit is karonda (*Carissa carandas* L.). Karonda is a hardy, evergreen bush, grows well even on marginal and inferior land. It comes up well even in semiarid and arid regions. Karonda can be grown as hedge plant to serve as Bio fence. It can be grown successfully in tropical and subtropical climate. It can be grown on wide range of soils like laterite, sandy loams, alluvial and calcareous soil. It grows well even in stoney, rocky and less fertile soils. (Tripathi *et al*., 2014) [14].
Karonda. contains several phytochemical constituents and is popular in indigenous systems of medicine like Ayurveda, Unani and Homeopathy, useful in treatment of several illnesses such as intestinal worms, diarrohea, skin ailments etc. (Kumar *et al.*, 2013) [6]. It is not a popular table fruit because of highly acidic nature and astringency The unripe fruit harvested at maturity can be stored for 5 to 7 days at room temperature, but at ripe stage, it can be stored only up to two days (Srivastava *et al*., 2017) [12]. Value added products preparation from karonda may add income to rural community. It also offers food security and creates dietary diversity. The research was conducted to study the feasibility of Karonda (*Carissa carandas* L.) to process into Vadiyam.

Material and Methods
The experiment was conducted at Department of Fruit science laboratory, College of Horticulture, Rajendranagar, Hyderabad and Central Instrumentation Cell, PJTSAU. Mature karonda fruits (Half red half white) were obtained from Fruit Research Station, Sangareddy, SKLTSHU, for research work and all other ingredients from local market.

Karonda Fruits
Fresh karonda fruits based on colour and maturity without bruises were sorted for further processing. The selected fruits were washed with tap water and subjected to blanching for three minutes. Fruits were cut into halves and seeds were removed using stainless steel knives.

pH
pH was determined using pH meter. pH meter was calibrated with the help of standard buffer solutions (pH 4.0 and 7.0).10g sample was macerated with 100ml distilled water. The mixture was agitated and allowed to stand for 30 minutes. The supernatant was used to determine pH.

TSS (^{0}Brix)
Total soluble solids in samples were recorded using a digital Refractometer (Model: HI 96801 Refractometer 0-85 ^{0}Brix Hanna Instrument).

Total And Reducing Sugars (%)
The titrimetric method as described by Ranganna (1986) [10] was adopted for the estimation of total and reducing sugars using Fehling A and B.

$$\text{Reducing sugar (\%)} = \frac{\text{Mg of invert sugar} \times \text{dilution} \times 100}{\text{Titre} \times \text{weight of the sample} \times 100}$$

$$\text{Total sugars as invert sugars (\%)} = \frac{\text{Factor x Volume made up x Dilution x 1000}}{\text{Titre value x Weight of sample taken}}$$

Titrable Acidity (%)
Titrable acidity of the sample was measured by titrating a given sample against a standard alkali solution of known concentration using phenolphthalein as an indicator to a light pink colour. 10g of homogenized sample was taken and made up to 100ml in a volumetric flask. The contents were filtered through Whatman No.1 filter paper. 10ml of aliquot was taken into a conical flask and titrated against standard solution of sodium hydroxide (0.1 N NaOH), using phenolphthalein as an indicator. The acidity was expressed in terms of percent citric acid equivalent adopting the following formula (AOAC, 2000) [2].

Ascorbic acid (mg/100g)
Ascorbic acid was estimated by 2, 6 dichlorophenol - indophenol visual titration method (AOAC, 2000) [2]. The method is based on reduction of 2, 6 dichlorophenol – indophenol dye. The dye, which is blue in alkaline solution and red in acidic solution, is reduced by ascorbic acid to a colourless form. The reduction is quantitative and specific for ascorbic acid in solutions in the pH range of 1.0 - 3.5. 5 ml of 3% metaphosphoric acid extract of the sample was taken in a conical flask and titrated with standard dye. The end point was pink, which existed for at least 15 seconds.

$$\text{Ascorbic acid (mg/100g)} = \frac{\text{Titre value x Dye factor x Volume made x100}}{\text{Aliquot taken x Volume of sample taken}}$$

$$\text{Total Phenols (GAE mg/100g)} = \frac{\text{Standard concentration x Sample OD x Volume made up x 100}}{\text{Standard OD x Aliquot taken x sample weight (g) x 1000}}$$

Microbial load
The media used were Nutrient agar (NA) for Total Bacterial Count and Rose Bengal Agar (RBA) for Total Mould count. All media used were prepared according to the manufacturer's instructions.

Total Bacterial Count
One gram of each sample was homogenized with 9ml of sterilized distilled water. Thereafter, 1ml homogenized sample was serially diluted for 10 times (10^{-1} - 10^{-10}). From each dilution test tube, one ml liquid was spread on to the Nutrient Agar plate. The inoculated plates were inverted and incubated at 37 ^{o}C for 48 hr. The bacterial colonies were counted with the help of colony counter. Individual colonies were counted and multiplied with the dilution factor to get the microbial population in gram of sample. The plates giving a range between 30 and 300 colonies were considered to be taken into account. The total colony count, referred as colony forming units (cfu) was calculated as below:

$$\text{cfu} = \frac{y}{dx}$$

Where
y = Number of colonies formed
d = Dilution
x = Volume of sample taken

Total Mould Count
One gram of food sample was homogenized with 9ml of sterilized distilled water. Thereafter, 1ml homogenized sample was serially diluted for 10 times (10^{-1} - 10^{-10}). From each dilution test tube, one ml sample was spread on to the Rose Bengal agar plate. The inoculated plates were inverted and incubated at 24^{o}C for 3 - 5 days and the numbers of colonies were counted.

Organoleptic evaluation
Organoleptic scoring was done by a panel of 15 members on a 9 point hedonic scale with corresponding descriptive terms ranging from 9 'like extremely' to 1 'dislike extremely', for colour, flavour, taste, texture, and overall acceptability (Jones, 1955; Marek *et al.*, 2007) [5, 7] developed for the purpose.

Statistical analysis
The analysis of variance of the data obtained was done by using Completely Randomized Design (CRD) and interpreted.

Results and Discussion
Trials were conducted to develop a novel product by incorporating karonda fruit. Vadiyam (A traditional South Indian dish used as a side dish in the main meal) was tried by incorporating karonda fruit and the process was standardised successfully. The best acceptable three variations were subjected to organoleptic evaluation by a panel consisting of 15 members on a 9 point Hedonic scale and the results are presented in Table 1 and Table 2. The product was appealing and acceptable which can be used as a side dish in a meal. The best variation was selected based on sensory evaluation data (Table 2).
Karonda vadiyam prepared as per Trial-II recorded the highest sensory score for colour (8.27) and differed significantly from other two variations. Highest sensory score

for flavour was reported in Trial-III (8.13) and was on par with Trial-II (8.0) whereas it differed significantly from Trial-I. Organoleptic evaluation for taste and overall acceptability showed the highest score in Trial-II (8.27and 8.13respectively) and differed significantly from the other two variations and is in "Like very much range" on 9 point Hedonic scale. However, no significant variation was observed for texture in Trial-II and Trial-III. The highest overall acceptability of Karonda Vadiyam as per Trial-II might be attributed to best acceptable colour, flavour, texture and taste. The results reveal the possibility of promising future for addition of Karonda Vadiyam to the consumer food list.

Table 1: Standardization trials of Karonda Vadiyam

Ingredients	Trial 1	Trial 2	Trial 3
Karonda fruit	50g	100g	150 g
Black gram dal	100 g	100g	100 g
Green chilli	30g	30g	30g
Cumin seeds	15g	15g	15g
Salt	10g	10g	10g
Curry leaf	5g	5g	5g
Remark		Accepted for further study based on organoleptic scores	

Table 2: Organoleptic evaluation of different variations of Karonda Vadiyam during standardization

Karonda Vadiyam Variations	Mean Scores of Sensory evaluation				
	Colour	Flavour	Texture	Taste	Overall acceptability
Trial I - Karonda Vadiyam- Karonda fruit 50g keeping all other ingredients constant	7.60	7.07	7.53	7.60	7.53
Trial II - Karonda Vadiyam- Karonda fruit 100g keeping all other ingredients constant	8.27	8.00	8.13	8.27	8.13
Trial III - Karonda Vadiyam -Karonda fruit 150g keeping all other ingredients constant	8.13	8.13	8.00	6.87	7.07
S.Em+	0.17	0.17	0.15	0.15	0.17
C.D. at (5%)	0.49	0.49	0.42	0.43	0.49
C.V.	8.24	8.62	7.18	7.69	8.73

Storage studies

The best acceptable Karonda Vadiyam was studied for changes in chemical composition, moisture content, microbial count and organoleptic evaluation during storage for six months.

pH

The changes occurred in pH during the storage of Karonda Vadiyam during six months storage are presented in Table 3. The pH of Karonda Vadiyam was 3.86 immediately after preparation. Product pH was stable upto two months of storage and thereafter pH decreased to 3.66.

Total Soluble Solids (ºBrix)

The changes recorded in TSS during the storage for six months of Karonda Vadiyam are presented in Table 3. Highest TSS (6.55 ºBrix) was recorded at the beginning of storage but significant decrease in TSS was seen only after three months of storage. TSS (6.42 ºBrix) was recorded at the end of storage for six months. Similar results were reported by Ankush *et al.* (2019) [1] in dried aonla, ber and bael fruits.

Titrable acidity (%)

The titrable acidity during the storage of Karonda Vadiyam for six months was recorded and presented in Table 3. Titrable acidity of Karonda Vadiyam at beginning of storage (2.35%). There was no significant difference observed in titrable acidity during storage.

Total Sugars (%)

The changes in total sugar content in Karonda Vadiyam during six months of storage was recorded and presented in Table 3. Total sugars of 3.54% was recorded in freshly prepared Karonda Vadiyam. There is no change in total sugars upto three months of storage however, there is a decrease in total sugars (3.51%) in later part of storage under study. This might be due to the inter conversion of sugars, nonspecific hydrolysis of macromolecules and aggregation of monomers during storage (Patter, 1985) [9].

Reducing Sugars (%)

The reducing sugars during the storage of Karonda Vadiyam was recorded for a six months period and presented in Table 3. Highest reducing sugar content (2.33%) was recorded at the beginning of storage and decreased significantly after four months of storage and showed decrease during later period of storage upto six months (2.28%).

Table 3: Changes in Chemical properties of Karonda Vadiyam during storage

Storage Period	Chemical properties of Karonda Vadiyam					
	pH	TSS (ºBrix)	Titrable acidity (%)	Total Sugars (%)	Reducing Sugars (%)	Ascorbic acid (mg/100g)
0 Months	3.86	6.55	2.35	3.54	2.33	26.49
1 Month	3.85	6.50	2.35	3.53	2.32	25.63
2 Months	3.84	6.50	2.37	3.53	2.32	24.77
3 Months	3.83	6.48	2.37	3.53	2.32	23.23
4 Months	3.80	6.46	2.38	3.52	2.31	22.34
5 Months	3.79	6.44	2.39	3.52	2.30	21.69
6 Months	3.66	6.42	2.43	3.51	2.28	19.60
S.Em+	0.01	0.03	0.02	0.01	0.01	0.35
C.D. at (5%)	0.02	0.08	NS	0.01	0.02	1.07
C.V.	0.24	0.67	1.51	0.21	0.30	2.58

Ascorbic acid (mg/100g)

The changes in ascorbic acid content during the storage of Karonda Vadiyam are presented in Table 3. Ascorbic acid content was highest immediately after preparation (26.49mg/100g), whereas lowest was recorded after a six months storage period (19.60 mg/100g). This might be due to degradation of ascorbic acid into dehydro ascorbic acid as well as to furfural during storage. Similar results of decrease in ascorbic acid content were reported by Pareek and Kaushik (2012) [8] in dried aonla powder and Suhasini *et al.* (2015) [13] in osmotically dehydrated karonda. Saxena *et al.* (2016) [11] reported decrease in ascorbic acid content throughout storage of karonda powder for four months which ranged from 56 to 69 percent.

Moisture content (%)

Moisture content of Karonda Vadiyam was studied at monthly intervals and presented in Table 4. Moisture content of freshly prepared Karonda Vadiyam was found to be 5.44%. Significant increase in moisture content was observed only after 3 months of storage and increased upto 5.66% by the end of six months storage period. This might be due to proper hygienic preparation, drying and storage conditions. Similar results were reported by Suhasini *et al.* (2015) [13] in osmotically dehydrated karonda and Saxena *et al.* (2016) [11] in karonda powder.

Total Bacterial Count (TBC)

Karonda Vadiyam was tested for bacterial growth at monthly intervals and presented in Table 4. The results reveal that there was no bacterial growth throughout the study period. This may be due to low moisture content, hygienic processing and storage conditions. Similar results were reported by Suhasini *et al.* (2015) [13] in osmotically dehydrated karonda and Saxena *et al.* (2016) [11] in karonda powder.

Total Mould Count (TMC)

Karonda Vadiyam was tested for mould growth at monthly intervals and presented in Table 4. The results reveal that there was no mould growth throughout the study period. This may be due to low moisture content of the product, hygienic processing and storage conditions. Similar results were also reported by Saxena *et al.* (2016) [11] in karonda powder and reported no yeast and mould growth during storage of sun-dried and cabinet-dried samples for 4 months.

Table 4: Moisture content, total bacterial and Mould count of Karonda Vadiyam during storage

Storage Period	Moisture content, Total Bacterial and Mould count of Karonda Vadiyam		
	Moisture Content (%)	Total Bacterial Count (Log CFU/g)	Total Mould Count (Log CFU/g)
0 Months	5.44	0.00	0.00
1 Month	5.46	0.00	0.00
2 Months	5.47	0.00	0.00
3 Months	5.50	0.00	0.00
4 Months	5.57	0.00	0.00
5 Months	5.62	0.00	0.00
6 Months	5.66	0.00	0.00
S.Em+	0.03	0.00	0.00
C.D. at (5%)	0.08	0.00	0.00
C.V.	0.85	0.00	0.00

Sensory evaluation of Karonda Vadiyam

After confirming microbiological safety of Karonda Vadiyam through TBC and TMC at monthly intervals, the Karonda Vadiyam was subjected to organoleptic evaluation by a panel of 15 members, on a 9 point hedonic scale at monthly intervals and presented in Fig.1. The results revealed that sensory scores for colour, flavour, texture, taste and overall acceptability did not show any significant difference during storage for six months.

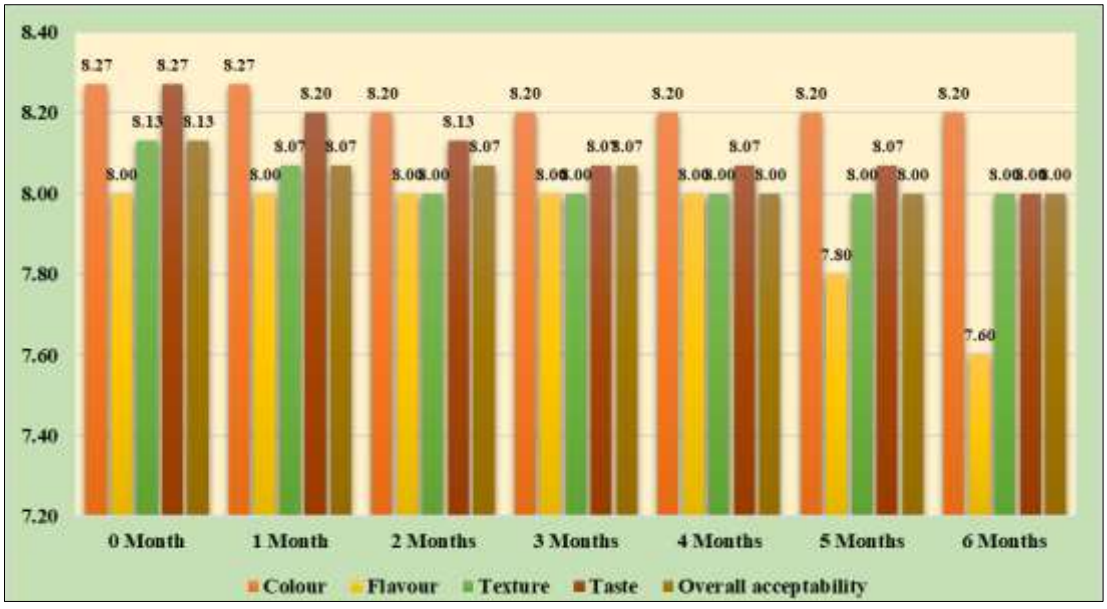

Fig 1: Organoleptic evaluation of Karonda Vadiyam during storage

Conclusion

The results revealed that karonda fruit can be processed into Vadiyam which can be stored safely for about six months with acceptable sensory score. Nutritional quality, Organoleptic evaluation and microbiological safety results show a promising future of incorporating karonda in preparation of value added products like Vadiyam in the processing industry.

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