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Phytonutritional and Sensorial Assessment of a Novel Functional Beverage Formulated from an Underutilized Fruit of *Carissa spinarum* L

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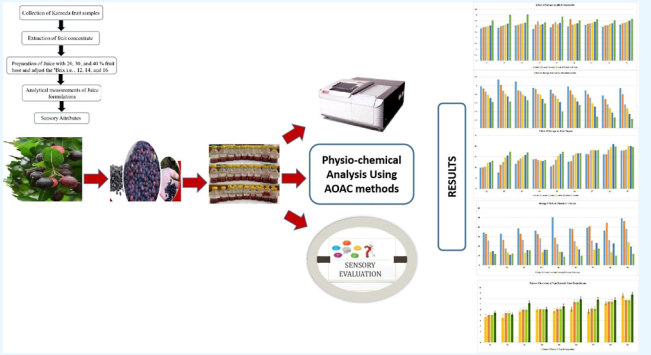
ABSTRACT: *Carissa spinarum* L. belongs to the family Apocynaceae. It is a native shrub of Asia, locally known as Karonda or Karanda, and is an underutilized crop throughout the Asian region. The Karonda fruit is a rich source of vitamin C, minerals, phenolics, antioxidants, flavonoids, and other biofunctional compounds. The lack of awareness and knowledge among the community results in the wastage of fruits. Therefore, the present research was designed to formulate an easy-to-prepare beverage drink using *C. spinarum* fruit to evaluate the nutritional potential of the undervalued Karonda fruit. A beverage drink was formulated with three pulp concentrations: 20, 30, and 40%, each having 12, 14, and 16 °Brix, respectively. A total of nine treatments were prepared and stored for up to 10 weeks in refrigerated storage. The physicochemical parameters, such as pH, titratable acidity, vitamin C, total sugars, anthocyanin, total phenolics, flavonoids, and antioxidants, were measured at two-week intervals from 0 to 10 weeks. Additionally, a sensory assessment of the beverage was conducted. A decreasing trend in titratable acidity was exhibited among all the treatments (from treatment 1 to treatment 9), with the values decreasing from 0.815 to 0.556 as the fruit concentration increased. On the other hand, an increasing trend was observed for pH (from 3.04 to 3.37), vitamin C (from 22.2 to 31.48), reducing and non-reducing sugars, anthocyanin (from 31.95 to 110), total phenolics (from 19.86 to 32.16), flavonoids (from 0.64 to 0.77), and antioxidants (from 48.8 to 67.6) from treatment 1 to treatment 9, respectively. The sensory studies of the beverage formulations revealed that treatment 9, which consisted of a 40% fruit base and 16 °Brix, was the most acceptable for further development of the beverage at a commercial scale. This study represents a novel scientific contribution toward the utilization of the undervalued fruit of *C. spinarum* L. for the development of a beverage product. Ultimately, it has the potential to address food insecurity issues worldwide while offering its associated health benefits.



1. INTRODUCTION

Currently, beverages are significantly the most dynamic functional foods due to the ease of dissolving their functional compounds, coupled with the convenience they offer in terms of consumption.[1] In the food industry, functional beverages have received growing interest among consumers as they hold a significant position in research and the biofunctional development.[2] Functional beverages, which are essentially synthesized from fruits, are a crucial source of antioxidants, polyphenols, vitamin C, and minerals. The antioxidative composition of such functional beverages is beneficial in providing a shield against free radicals and enhancing cells' resistance against any reactive oxygen species present in the system. Moreover, antioxidants are well-known for their essential role in human health.[3−6]

For synthesizing novel functional beverages, numerous advanced techniques are being used, such as the valorization of fruit byproducts, the utilization of indigestible food constituents that render healthful benefits, the use of natural constituents and their optimization, the utilization of microbes to increase functional gains, and the use of nonthermal treatment options for the preservation of the functional qualities of the beverages.[7−9] To enhance the health benefits in the modern era, a synergistic formulation and a distinctive approach to combining the fundamental ingredients used in

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the synthesis of functional beverages, especially for eliminating free radicals, are critically required.

Underutilized fruits are mostly found in rural areas worldwide and have distinct aromas and tastes. They provide a multitude of impressive health benefits due to their high nutrient and bioactive content. They also possess numerous genes that confer increased resistance to abiotic stresses. Many nutrient-dense wild edible plants, especially those rich in vitamins and minerals, can be utilized to meet the nutritional needs of both humans and livestock. However, their full potential is not being exploited. Underutilized plant species have enormous potential to contribute to food security, health (in the nutritional and/or medicinal industries), revenue generation, and environmental benefits.[10−12] In order to minimize the undesirable effects of recent ecological shifts and technological developments in the world, it is now inevitable that people consume health-promoting foods. Therefore, the demand for fruits with valuable metabolites is continually increasing.[13]

Carissa spinarum L. is a wild-flowering shrub that belongs to the Apocynaceae family. The crop plant is known as "wild Karanda" and is an underutilized wild fruit closely related to *Carissa carandas* L. Its various parts, such as fruit, leaf, and root, have medicinal potential that is beneficial for humans, and berries are a rich source of iron and vitamin C.[14−16] The plant is native to Pakistan and is also distributed in Sri Lanka, Indonesia, Malaysia, Myanmar, India, Nepal, Afghanistan, Australia, and South Africa.[12] Karanda fruit is technically a berry that grows in clusters of three to ten. The plant produces small, round fruits that are green at first but acquire a reddish-purple color when fully ripened.[17] Most of the information on the phytochemical composition of Karanda fruit has focused on flavonoids and phenolic content; however, there is no information on the fruit's anthocyanin content.[18] Fruit juice from ripened Karanda fruits comprises high levels of vitamin C, DPPH scavenging activity, flavonoids, tannins, and anthocyanins. Karanda has a high phenolic content, indicating that it is a high-quality and nutrient-rich food with additional health advantages. Almost every part of the plant is utilized in some way. Iron and calcium are particularly abundant in its fruit, with higher pectin content. Notably, it has more vitamin C than an apple or a banana.[19]

Since its fruit possesses health benefits and the ability to prevent non-communicable diseases, it has the potential to be used as a functional food as well as a therapeutic food, offering benefits to both customers and farmers. Unripe Karanda fruits have a sour taste and can be used to treat liver disease, break a fever, and prevent blood putrefaction. Ripe Karanda fruits, on the other hand, have a sweeter taste and are used as an antiscorbutic and a cure for biliousness.[20] Karanda has long been used to cure a variety of ailments, such as flatulence, indigestion, acidity, and sores, which are all symptoms of a weak immune system.

The fruit, leaf, and root of *C. spinarum* have numerous medicinal attributes that are beneficial for both human beings and animals as well. Various plant parts are utilized for their potential antimicrobial actions.[21−23] The entire plant is used as an anthelmintic and antidiarrheal antidote, whereas the stem is used to strengthen tendons. Fruits and leaves are used to treat skin infections as well as fevers and syphilitic discomfort. The alcoholic extract of the root material lowers blood pressure, whereas the aqueous extract of roots has histamine-releasing, anthelmintic, and cardiotoxic properties.[24] The plant's popularity has skyrocketed in the market because it possesses antiscorbutic properties and has been discovered to have a beneficial effect in the treatment of anemia. It has been used in a variety of Ayurvedic medicines. Moreover, chest discomfort is also treated with root extract.[25] Nowadays, millennial consumer behavior is influenced by a strong focus on healthy, natural, and nutritious food products, leading to new developments. Karonda is an appetizer. Before the fruit ripens, it is usually pickled. Due to the high pectin content of ripe Karonda fruit, it is also used to make jellies, jams, squashes, sauces, pies, syrup, tarts, and chutney, all of which are in high demand in the worldwide market.[26] When the ripe fruit is cooked, it produces gummy latex, but when it cools, it yields a rich red juice that becomes clear, making it a delightful summer drink. The juice of ripe Karanda is easily digested, extremely pleasant, thirst-quenching, delicious, and nutritionally superior to many synthetic and aerated beverages.[27] These fruits are less expensive and fresher and have a shorter shelf life. As a result, fruit processing is required to extend its shelf life.[27]

Kashmir is blessed with immense plant diversity. Most of the land is mountainous, with diverse microclimatic zones, making it unsuitable for commercial-level cultivation of cereal crops. However, fruits and vegetables, along with the cultivation of ornamental and herbal plants, offer huge economic potential. Therefore, Karonda fruits are cultivated and consumed locally. Karonda fruits have many advantages, such as being easy to grow and producing a crop even under adverse soil and climatic conditions. Unfortunately, they have received little attention, and there are few reports on their nutritional composition. Since there is a disturbingly prevailing situation of food and health insecurity among underdeveloped mountain communities, it is imperative that academia, industry, and the government pay attention to neglected and underutilized crops like Karonda. Underutilized plants, in general, might provide a solution to the socio-economic problems of food and health insecurity. Since Karonda is an arid-region fruit that naturally thrives in various topographically diverse regions of Kashmir, it can be grown in a variety of soils at lower costs. The natives of the study area consume the fruits of wild Karonda plants without realizing how many nutrients they contain. Therefore, the current research study was designed to formulate a refreshing functional drink and assess the phytochemical and nutritional values of Karonda fruit juice.

2. MATERIALS AND METHODS

All analytical grade chemicals were purchased from Merck.

2.1. Collection of Samples. The fresh samples of Karonda fruit were randomly collected from the village area of Baila Chanair, Tehsil Hajira (latitude 33° 43′ 54″ N, longitude 73° 54′ 58″ E, and altitude of 1087 m), District Poonch, Azad Kashmir. The samples were then transferred to the laboratory of Food Science and Technology. A plant sample was also collected and deposited for formal analysis at the National Herbarium (Plants of Azad Jammu and Kashmir) and identified as *C. spinarum* L. (Figure 1) by Dr. Amir Sultan, Program Leader/Curator at the National Herbarium of Pakistan, National Agricultural Research Centre, Pakistan Agricultural Research Council, Islamabad, Pakistan, with the voucher Acc. no. RAW1102620. The samples of ripened fruits were harvested using the hand-picking method. When fully ripened, the fruits turn black. Due to their shorter shelf life, the picked fruits were immediately placed into a dry ice box. After

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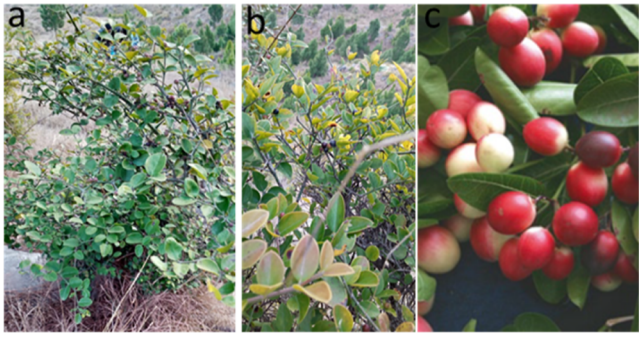

Figure 1. *C. spinarum* L. (a) Whole plant with ripened fruits from the selected site, (b) a branch with ripened fruits, and (c) fruits at various ripening stages (Source: Lim, 2012).[28]

sorting and cleaning, they were transferred to the refrigerator until they were utilized. The sampled fruits displayed colors ranging from pinky-white, pink, to black or reddish-purple, with total soluble solids (TSS) measured in °Brix ranging from 12 to 16.

2.2. Formulation of Juice. The collected Karonda fruit was washed and destemmed. Fruit juice was extracted using a juicer machine. The juice was then filtered through a muslin cloth. After filtration, the beverage was prepared using different concentrations of Karonda fruit pulp. The °Brix level of each pulp concentration was optimized by adding sugar content (Table 1). To preserve the juice, 0.1% sodium benzoate was

Table 1. Details of Different Karonda Fruit Beverage Formulations[a]

treatments	Karonda fruit-pulp (%)	°Brix
T1	20	12
T2	20	14
T3	20	16
T43	30	12
T5	30	14
T6	30	16
T7	40	12
T8	40	14
T9	40	16

[a]The effect of preservatives and concentration of preservatives were optimized after optimizing pulp concentration and °Brix.

added as a preservative. The Karonda fruit was stored at a refrigerated temperature of 4 °C for 8 weeks at each interval (0, 2, 4, 6, 8, and 10 weeks) for subsequent biochemical and sensory characterization. The optimization of fruit base concentration and °Brix was adjusted following the modified method of Nazareth et al.[29]

2.3. Physicochemical and Phytonutritional Characterization of Karonda Juice. *2.3.1. pH.* The pH of the juice samples was measured using a pH meter (Bench-top pH meter KDD002 AUXI Lab, Spain) in accordance with the AOAC method (981.12) guidelines.[30]

2.3.2. Titratable Acidity. Titratable acidity (TA) was measured following the standard method provided by the Association of Official Analytical Chemists (AOAC).[31] Five milliliters of fruit juice was mixed with 20 mL of purified water and filtered to obtain a clear extract. A 5 mL aliquot of the extract was titrated with 0.1 N NaOH, using a few drops of phenolphthalein as an indicator, until a light pink color appeared as the endpoint. The data were recorded for three replications, and the percent TA was calculated in terms of ascorbic acid using the following formula

$$\text{TA \%} = [(\text{mL of NaOH used} \times \text{normality of NaOH} \times \text{equiv weight of NaOH})/(\text{weight of sample} \times \text{volume of aliquot})] \times 100$$

2.3.3. Ascorbic Acid Content. The vitamin C content of the samples was measured using the titrimetric standard method described by AOAC, which involves the use of a dye solution of 2,6-dichlorophenol indophenol that is reduced to a neutral compound by ascorbic acid.[30] In this method, 5 mL of the juice sample was added to a 100 mL conical flask, followed by the addition of 5 mL of a 4% *meta*-phosphoric acid solution. The sample was then titrated with the 2,6-dichlorophenol indophenol dye until the endpoint was reached, indicated by a light pink color. The vitamin C content was calculated using the formula below

$$\text{ascorbic acid (mg/100 mL)} = F \times T \times 100/S \times D$$

where F = standardization factor = mL of ascorbic acid/mL of pigment used, T = mL of pigment used for the sample, S = mL of diluted sample taken for titration, and D = mL of the sample taken for dilution.

2.3.4. Total Phenolic Content. The total phenolic content of the product sample was estimated using the spectrophotometric method with the HALO DB-20 UV−vis double beam spectrophotometer by ATTEKNO SOLUSI Indonesia.[32] Initially, 2.5 mL of 10% Folin-Ciocalteu's Reagent and 2 mL of 7.5% sodium carbonate solution were mixed with a 0.5 mL sample. The reaction mixture was then incubated at 45 °C for 40 min, and the absorbance was measured at 765 nm using the HALO DB-20 UV−vis double beam spectrophotometer by ATTEKNO SOLUSI Indonesia. The average value of three readings was recorded as the mean total phenolic content. The phenolic content was expressed as milligrams of gallic acid equivalent per 100 mL of juice sample.

2.3.5. Total Flavonoids. The total flavonoid content of the product formulations was determined using the spectrophotometric method described by Chang and colleagues.[33] A sample weighing 0.1 g was mixed with 0.1 M potassium acetate, followed by the addition of 0.1 mL of aluminum chloride and 2.8 mL of distilled water. The mixture was then incubated at room temperature for 30 min. The absorbance of the reaction mixture against the blank was measured at a wavelength of 430 nm using the HALO DB-20 UV−vis double beam spectrophotometer by ATTEKNO SOLUSI Indonesia. The resulting values for total flavonoids were calculated using the quercetin curve and expressed as milligrams of gallic acid equivalents per milliliter (mg GAEs/mL).

2.3.6. Total Anthocyanin. The total anthocyanin content was measured using a spectrophotometer, specifically the HALO DB-20 UV−vis double beam spectrophotometer by ATTEKNO SOLUSI Indonesia. The pH differential method, as described by Golmohamadi et al.[34] with a few modifications, was employed.[27] Two dilutions of the juice sample were prepared using potassium chloride buffer (pH 1.0) (0.5 mL of sample extract and 3.5 mL of potassium chloride) and sodium acetate buffer (pH 4.5) (0.5 mL of sample extract and 3.5 mL of sodium acetate) against a blank. The mixtures were allowed to equilibrate for 15 min. The absorbance of each dilution was

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Table 2. Mean Values of All Tested Parameters for Nine Karonda Juice Formulations[a]

treatments	pulp (%)	°Brix	pH	titratable acidity (%)	vitamin C (mg/100 mL)	total phenolics (mg/100 mL)	total flavonoids (mg/mL)	total anthocyanins (mg/100 mL)	total antioxidants (%)	total sugars (%)
T1	20%	12	3.04^{e}	0.81^{abc}	22.26^{d}	19.86^{f}	0.64^{e}	31.59^{h}	48.88^{e}	13.17^{f}
T2	20%	14	3.21^{bcd}	0.86^{a}	18.84^{e}	27.27^{c}	0.68^{ed}	40.85^{g}	56.46^{d}	14.43^{c}
T3	20%	16	3.34^{a}	0.84^{ab}	23.70^{cd}	31.60^{a}	0.71^{b}	44.95^{f}	56.55^{d}	13.43^{f}
T4	30%	12	3.16^{d}	0.77^{bc}	23.78^{cd}	21.57^{c}	0.65^{de}	64.32^{e}	62.96^{c}	14.00^{e}
T5	30%	14	3.16^{d}	0.70^{d}	23.01^{cd}	25.38^{d}	0.66^{cde}	65.91^{e}	64.57^{b}	15.13^{d}
T6	30%	16	3.29^{abc}	0.75^{cd}	24.49^{bcd}	30.98^{ab}	0.68^{e}	68.75^{d}	65.25^{b}	17.45^{e}
T7	40%	12	3.32^{ab}	0.63^{e}	27.17^{b}	29.81^{b}	0.75^{a}	79.19^{e}	56.91^{d}	18.40^{b}
T8	40%	14	3.18^{cd}	0.52^{f}	25.52^{bc}	30.77^{ab}	0.76^{a}	100.70^{b}	65.31^{b}	18.92^{a}
T9	40%	16	3.37^{a}	0.55^{f}	31.48^{a}	32.16^{a}	0.77^{a}	110.23^{a}	67.71^{a}	13.17^{f}
ANOVA *P*-values	storage		0	0	0	0	0	0	0	0
	treatment		0	0	0	0	0	0	0	0
	treatment × storage		0	0.003	0	0	0	0	0	0

[a]Means for all parameters of different treatments with regard to storage intervals are shown in capital letters, while different letters indicate significant interactions ($P \leq 0.05$).

T1= 20% fruit Karonda pulp +12 °Brix T4= 30% fruit Karonda pulp +12 °Brix T7= 40% fruit Karonda pulp +12 °Brix

T2= 20% fruit Karonda pulp +14 °Brix T5= 30% fruit Karonda pulp +14 °Brix T8= 40% fruit Karonda pulp +14 °Brix

T3= 20% fruit Karonda pulp +16 °Brix T6= 30% fruit Karonda pulp +16 °Brix T9= 40% fruit Karonda pulp +16 °Brix

measured at 515 and 700 nm using the HALO DB-20 UV−vis double beam Spectrophotometer. The anthocyanin pigment content was calculated as milligrams of cyanidin-3-glucoside per 100 mL utilizing an extinction coefficient of 29,600 and a molecular weight of 449.2.

2.3.7. Total Sugars. The reducing and non-reducing sugar contents were estimated in the juice formulations using the titrimetric method of AOAC (2006).[30] The reducing and non-reducing sugar content was estimated from the juice formulations following the AOAC-reported method (2006; method no. 939.03). The reduction in sugar content was evaluated by the appearance of a brick-red color, which indicated the endpoint. Afterward, methylene blue was added to confirm whether the brick-red color changed to blue or not. The percent of total sugars was obtained by adding both reducing and non-reducing sugars.

2.3.8. Antioxidant Activity. Antioxidant activity was evaluated using the DPPH method as reported by Brand-Williams et al.[35] For this, an extract of different concentrations (1000 μg/mg) was prepared by adding 0.5 mL of the juice sample, followed by the addition of 1 mL of freshly prepared DPPH (0.25 mm) and 1 mL of ethanol. Each sample was thoroughly mixed and kept in the dark for 30 min at room temperature. Subsequently, the sample was assessed for DPPH radical scavenging activity using a spectrophotometer (HALO DB-20 UV−vis double beam spectrophotometer by ATTE-KNO SOLUSI Indonesia) at 517 nm. The antioxidant activity was calculated using the following formula

$$\text{DPPH scavenging activity } \% = (A_0 - /A_0) \times 100$$

where A_0 is the absorbance of the control and A_S is the absorbance of the sample.

2.3.9. Sensory Evaluation. Organoleptic testing was conducted by a panel of five judges selected from the Food Science and Technology department. Before beginning the evaluation, the judges received training on the coding method and rating of sensory characteristics. Sensory evaluation of nine juice treatments for color, flavor, taste, and overall acceptability was performed to determine the differences between formulations. The evaluation was assessed using a 9-point hedonic scale, ranging from extremely liked (9) to extremely disliked (1), as described by Purewal et al.[36] The judges were served with the juice formulations three times, and three responses were collected from each person. The data was then analyzed statistically.

3. STATISTICAL ANALYSIS

The data were obtained as the mean of three replications and subjected to analysis of variance (ANOVA) at an α level of 0.05 for all studied parameters. The means were statistically separated using Tukey's range test, with the software package Statistics 8.1.1 being employed to determine the significant differences among the means. The normality of distributions and homogeneity of variances were checked before running the Tukey HSD test. To observe the association among the different analyzed parameters, the mean values of each treatment for physicochemical attributes, functional attributes, and sensory attributes were compiled. Pearson's linear correlation analysis was then conducted, and a heat map analysis was generated using R, following the method previously reported by Jobil et al.[37]

4. RESULTS AND DISCUSSION

4.1. pH. pH is a quantitative measure of the acidity or basicity of aqueous or other liquid solutions. It serves as a controlling factor in the development of flavor, acts as a preservative, and determines the stability of certain bioactive compounds. A highly significant effect of pH was observed among the treatments and storage intervals.

An increase in pH was observed in treatments with higher concentrations of Karonda pulp and soluble solid content in the beverage sample, as shown in Table 2. The lowest pH value (3.04) was found in T1 (20% fruit Karonda pulp + 12 °Brix), while the highest pH value (3.37) was observed in T9 (40%

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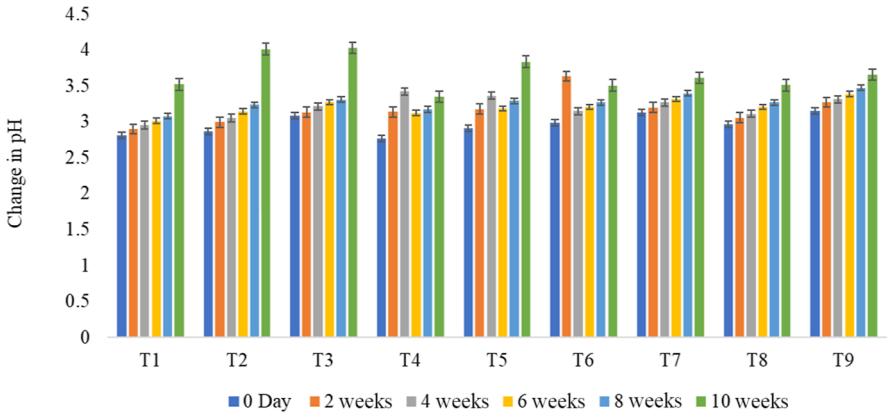

Figure 2. Effect of storage intervals on the pH of the Karonda juice formulations.

fruit Karonda pulp + 16 °Brix). The mean results related to storage time are presented in Figure 2. The graph depicts an increasing trend in pH for all samples as the storage period advanced. The pH values ranged from 2.96 to 3.66 after 10 weeks of storage. The lowest pH value of 2.96 was recorded on day zero of the storage period, while the highest pH value of 3.66 was observed on the final day of the storage interval. Overall, an increasing trend in pH was observed with prolonged storage across all treatments.

The increase in pH may be attributed to the acid hydrolysis of certain polysaccharides into disaccharides, such as starch into sucrose, fructose, glucose, etc. This acid hydrolysis of polysaccharides into mono- and disaccharides, resulting in increased sweetness and decreased sourness, could be a cause for the pH increase during the extended storage of Karonda juice. These findings align with a previous study by Jan and Masih,[38] which reported a similar trend in guava juice. Another study by Rehman et al.[39] also described a comparable trend in kinnow juice.

4.2. Titratable Acidity. Total TA (TTA) is a measure of the amount of acid or acids present in a food sample. Acidity is an important characteristic of beverages as it reflects the freshness of juices. Excessively low acidity can lead to a loss of freshness, while high acidity can result in a sharp and unpleasant taste, making the beverage difficult to consume.

After analyzing the TA in fruit juice, the obtained results were statistically analyzed using ANOVA, which revealed a highly significant effect among treatments and storage intervals. However, the interaction effect was only found to be significant. The highest acidity value (0.86) was observed in T2, while the lowest value (0.52) was noted in T8, as shown in Table 2. The beverage samples exhibited a consistent decrease in acidity with an increasing concentration of Karonda pulp. This observation is in line with a previous report by Jeppsson and Johansson,[40] which indicated that Karonda fruit is more basic in nature.

The percentage of TA during the storage period, as depicted in Figure 3, showed a declining trend. The highest acidity value (0.95) was observed on day zero, while the lowest value (0.46) was recorded after 10 weeks of storage. Analogous observations were reported by Divate et al.[41] The gradual decrease in total acidity during storage is attributed to the interaction between the organic components of the juice at room temperature and the conversion of acids into sugars and salts by the action of invertase enzymes.[42] A similar decreasing trend in TA was also observed in kinnow juice by Singh et al.[43]

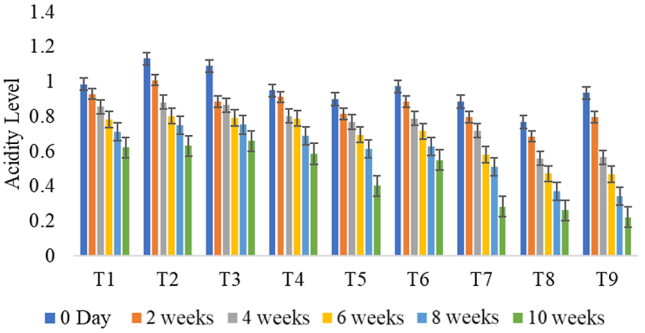

Figure 3. Effect of different storage intervals on the acidity level of nine Karonda juice formulations.

Additionally, another study by Pareek et al.[44] supported our findings, reporting a decreasing trend of acidity in mandarin juice during storage.

4.3. Ascorbic Acid. Ascorbic acid, commonly known as vitamin C, is often marketed and consumed as a dietary supplement. It is a water-soluble vitamin found in citrus fruits as well as various other fruits and vegetables. Vitamin C plays a crucial role in maintaining healthy connective tissue and is believed to function as an antioxidant. Severe deficiency of vitamin C can lead to scurvy. Ascorbic acid is a potent reducing and antioxidant agent that aids in fighting bacterial infections, detoxifying reactions, and collagen formation in fibrous tissues, teeth, bones, connective tissue, skin, and capillaries. Vitamin C has the potential to reduce LDL cholesterol levels, protect against tumor cells by scavenging free radicals, and neutralize the effects of nitrates. It also strengthens the immune system, alleviates cataracts, and reduces symptoms of the common cold.[45]

Our results indicate that storage, treatments, and their interactions had a highly significant effect on the ascorbic acid content of the different samples. For individual mean comparisons, significant results were subjected to Tukey's HSD test at a significance level of ($P \leq 0.05$).

The results presented in Table 2 show that the amount of ascorbic acid increases with the increasing concentration of Karonda pulp in the beverage sample. The highest value of ascorbic acid was observed in T9 (31.48%), while the lowest value was noted in T2 (18.84%). The reason behind the rise in ascorbic acid with Karonda pulp is that Karonda fruit is a rich source of ascorbic acid, as supported by the findings of Siyum and Meresa.[68] The impact of storage intervals on the vitamin C

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content of the beverages is shown in Figure 4. There was a gradual decrease in ascorbic acid content in Karonda juice

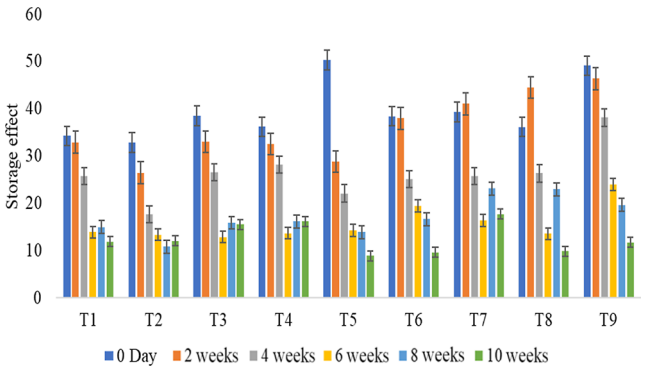

Figure 4. Effect of storage on the vitamin C content of nine Karonda juice formulations.

from 39.44 to 12.56 mg. The maximum value of ascorbic acid was recorded on day zero (39.44%), and the lowest value (12.56%) was recorded on the last day of storage. Since vitamin C is water-soluble and sensitive to heat, light, and oxygen, it undergoes oxidation, resulting in the irreversible conversion of ascorbic acid to dehydroascorbic acid. This oxidation process occurs due to the action of oxidase enzymes and is further accelerated during storage. Another reason for the decreasing trend in ascorbic acid is enzymatic and non-enzymatic reactions, as well as the formation of complexes such as hydroxyl-methyl-furfural compounds, which degrade ascorbic acid to dehydroascorbic acid, furfural, and/or carbolic acid. A similar decreasing trend in ascorbic acid content was reported by Kabasakalis et al.[46] in fruit juices. Our findings are also consistent with a previous study by Pareek et al.,[44] which reported a decrease in vitamin C content in mandarin juice during storage.

4.4. Total Phenolic Content. Phenolic compounds are important constituents found in plants and possess redox properties that contribute to antioxidant activity. Fruits serve as a rich source of various bioactive compounds, including phenolics and phenolic acids. These bioactive compounds play a crucial role in determining the freshness and quality characteristics of fruit juices, whether in their fresh or processed forms. From a human physiological perspective, phenolic compounds have significant roles in defense reactions, such as exhibiting anti-aging, anti-inflammatory, antioxidant, and anti-proliferative activities.

The results presented in Table 2 demonstrate a significant increase in the total phenolic content with increasing concentrations of Karonda pulp and SSC in the beverage samples. The highest value was recorded in T9 (32.16), while the lowest value was recorded in T1 (19.86). The observed increase in total phenolic content in the beverage samples can be attributed to the higher content of Karonda pulp. This finding aligns with the research by Khuanekkaphan et al.,[49] which establishes Karonda as a significant source of phenolic acid, explaining its contribution to the elevation of total phenolic compounds.

As depicted in Table 3, there is a significant increase in phenolic content during the first six weeks of storage, followed by a significant decrease during the last four weeks of storage. The lowest value of phenolic content was observed at day zero (16.07), while the highest value was observed after six weeks (38.67). A similar trend was also reported by Reque et al.[47] The increase in total phenolic content over the extended storage period can be attributed to enzymatic hydrolysis or the biodegradation of previously unextractable bound phenolic compounds. The total phenolic content is influenced by various factors, such as environmental conditions, light exposure, and temperature. Consequently, the reduction in total phenolic content after a long storage period is a result of these factors, as observed by Choi et al.[48]

4.5. Total Flavonoids. Flavonoids are a class of polyphenolic secondary metabolites commonly found in plants that are commonly consumed by human beings for dietary purposes. These compounds offer several medicinal benefits, including anticancer, anti-inflammatory, and antiviral proper-

Table 3. Effect of Storage Intervals on the Total Phenolic and Flavonoid Content of Nine Karanda Juice Formulations[a]

	total phenolics (mg/100 mL)						total flavonoids (mg/mL)					
treatments	0 day	2 weeks	4 weeks	6 weeks	8 weeks	10 weeks	0 day	2 weeks	4 weeks	6 weeks	8 weeks	10 weeks
T1	$20.89 \pm 1.41^{p-V}$	$17.00 \pm 0.62^{u-z}$	$22.16 \pm 1.11^{n-t}$	$24.90 \pm 1.35^{m-p}$	$23.95 \pm 1.35^{m-p}$	10.27 ± 0.56^{a}	$0.73 \pm 0.01^{e-k}$	$0.70 \pm 0.05^{g-o}$	$0.69 \pm 0.00^{g-p}$	$0.63 \pm 0.01^{m-f}$	$0.62 \pm 0.01^{n-f}$	0.50 ± 0.02^{t}
T2	12.89 ± 0.39^{za}	$18.90 \pm 0.69^{q-w}$	$38.65 \pm 1.35^{e-g}$	$42.03 \pm 0.65^{c-e}$	41.33 ± 1.89^{a}	10.39 ± 1.89^{a}	$0.79 \pm 0.05^{d-f}$	$0.69 \pm 0.02^{g-p}$	$0.68 \pm 0.01^{h-p}$	$0.65 \pm 0.01^{j-q}$	$0.64 \pm 0.01^{l-r}$	$0.62 \pm 0.02^{o-s}$
T3	$22.58 \pm 1.70^{n-s}$	$21.63 \pm 3.55^{o-u}$	$42.82 \pm 1.24^{c-e}$	50.97 ± 1.24^{a}	33.76 ± 1.86^{hi}	$17.85 \pm 0.31^{s-y}$	0.86 ± 0.01^{cd}	$0.73 \pm 0.02^{g-p}$	$0.79 \pm 0.05^{d-f}$	$0.71 \pm 0.00^{f-l}$	$0.65 \pm 0.01^{j-r}$	0.54 ± 0.02^{st}
T4	$13.42 \pm 1.52^{y-a}$	$13.89 \pm 1.01^{x-a}$	$22.71 \pm 1.13^{n-r}$	$26.72 \pm 1.31^{k-n}$	$34.00 \pm 1.17^{g-i}$	$18.68 \pm 0.30^{r-x}$	$0.66 \pm 0.00^{i-q}$	$0.68 \pm 0.02^{h-p}$	$0.67 \pm 0.01^{i-p}$	$0.69 \pm 0.01^{g-p}$	$0.58 \pm 0.00^{g-t}$	$0.64 \pm 0.02^{l-r}$
T5	12.59 ± 1.86^{za}	$13.16 \pm 2.20^{y-a}$	$31.94 \pm 0.75^{h-j}$	$29.20 \pm 0.84^{i-l}$	$39.22 \pm 1.65^{d-f}$	$26.17 \pm 1.62^{k-o}$	$0.74 \pm 0.01^{e-i}$	$0.70 \pm 0.01^{g-n}$	$0.62 \pm 0.00^{n-r}$	$0.63 \pm 0.00^{l-r}$	$0.70 \pm 0.00^{g-n}$	$0.57 \pm 0.01^{f-t}$
T6	10.03 ± 0.15^{a}	$23.66 \pm 2.37^{n-q}$	$36.21 \pm 1.18^{f-h}$	51.14 ± 0.48^{a}	$36.15 \pm 1.50^{f-h}$	$28.72 \pm 1.99^{i-m}$	$0.61 \pm 0.01^{p-s}$	$0.69 \pm 0.00^{g-p}$	$0.73 \pm 0.05^{e-k}$	$0.67 \pm 0.01^{i-p}$	$0.73 \pm 0.01^{e-k}$	$0.65 \pm 0.02^{k-t}$
T7	$21.86 \pm 1.30^{o-t}$	$24.69 \pm 2.430^{l-p}$	44.90 ± 0.66^{bc}	$30.05 \pm 0.67^{i-k}$	$40.79 \pm 1.01^{c-f}$	$16.49 \pm 2.07^{v-z}$	0.95 ± 0.03^{ab}	$0.66 \pm 0.05^{i-q}$	$0.66 \pm 0.01^{i-q}$	$0.67 \pm 0.01^{i-p}$	$0.66 \pm 0.01^{i-q}$	0.91 ± 0.02^{bc}
T8	$14.23 \pm 1.47^{w-a}$	$15.48 \pm 3.39^{w-z}$	44.02 ± 0.96^{cd}	$43.45 \pm 1.39^{c-e}$	$41.16 \pm 1.58^{c-e}$	$26.30 \pm 1.69^{k-e}$	0.89 ± 0.05^{bc}	$0.71 \pm 0.05^{f-m}$	$0.77 \pm 0.6^{e-g}$	$0.70 \pm 0.00^{g-n}$	0.80 ± 0.05^{de}	$0.68 \pm 0.01^{h-p}$
T9	$16.77 \pm 0.50^{v-z}$	$17.73 \pm 2.03^{t-y}$	49.16 ± 0.41^{ab}	49.59 ± 0.44^{ab}	49.87 ± 1.99^{a}	9.84 ± 1.80^{a}	$1.03 \pm 0 \pm 06^{a}$	$9.65 \pm 0.04^{k-r}$	$0.68 \pm 0.01^{h-p}$	$0.76 \pm 0.00^{e-h}$	$0.77 \pm 0.01^{e-g}$	$0.74 \pm 0.02^{e-j}$

[a]Numbers represent the means of three replications of each treatment at different storage intervals. The standard deviation of each interval reading has been mentioned. The superscripts letters showing interactions and non-significant interactions are shown by the same letters, while different letters indicate significant interactions ($P \leq 0.05$).

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ties. Flavonoids have also been found to exhibit neuroprotective and cardioprotective effects.

The analysis of variance for the total flavonoid content of different beverage samples provided evidence of a significant difference among various treatments, storage intervals, and their interactions. To compare the means, the results were subjected to the Tukey's HSD test at a significance level of ($P \leq 0.05$).

As depicted in Table 2, there is a significant increase in the total flavonoid content with increasing concentrations of Karonda pulp and SSC in the beverage samples. The highest value was recorded in T9 (0.77), while the lowest value was recorded in T1 (0.64). This increase in total flavonoid content by increasing the Karonda pulp and TSS concentration in beverage samples indicates that Karonda and soluble solids serve as good sources of flavonoids, as reported by Khuanekkaphan et al.[49]

The results regarding the impact of storage intervals on total flavonoids are presented in Table 3. It is evident from the table that there is a significant decrease in total flavonoids with the progression of storage. The significantly highest value of flavonoids was observed at the beginning of the experiment on day zero (0.81), while the significantly lowest content of total flavonoids was observed at the end of the storage period (0.65). The decrease in flavonoid content during storage could be attributed to a decrease in enzyme activity, which leads to the formation of phenolic compounds. Similar decreasing trends in flavonoid content were observed in cranberry juice by Borges et al.[50] and in raspberry juice by Bradish et al.[51]

4.6. Total Anthocyanin Content. Anthocyanins, also known as anthocyans, are water-soluble vacuolar pigments that can exhibit colors ranging from red, purple, and blue, to black. Berries, such as blueberries, raspberries, and others, are rich sources of anthocyanins, which give them their red, blue, purple, or black color. Anthocyanins extracted from edible plants have potential pharmaceutical applications.

The results regarding the impact of treatments on total anthocyanin content are presented in Table 2. Our study demonstrated that the anthocyanin content increased with increasing concentrations of Karonda pulp and soluble solids. The highest value of anthocyanin was observed in T9 (110.23), while the lowest value was observed in T1 (31.5). The increase in anthocyanin content by increasing the concentration of Karonda pulp in the beverage samples is due to the abundant amount of anthocyanins present in Karonda fruit.[18]

There is a significant decrease in total anthocyanin content as storage progresses, as shown in Figure 5. The significantly highest value of anthocyanin was observed at the beginning of the experiment on day zero (101.02), while the significantly lowest content of total anthocyanin was observed at the end of the storage period (29.43). The decreasing trend in anthocyanin content during prolonged storage was also reported by Kaur et al.[52] in strawberry pulp, attributed to the conversion of anthocyanins into insoluble brown pigments during storage, which supports our findings. Anthocyanin is sensitive to light, oxygen, temperature changes, pH changes, and enzymes. Strong negative correlations were observed between anthocyanin concentrations and the levels of haze, polymeric, and brown color development during storage. Therefore, the change in pigment concentration during storage is a result of anthocyanin degradation. A similar decreasing

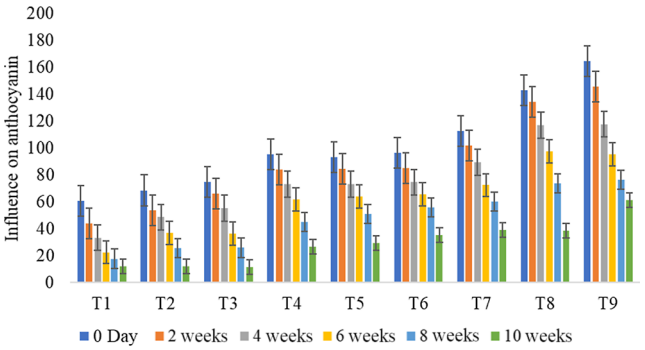

Figure 5. Influence of storage intervals on the anthocyanin content (mg/100 mL) of nine juice formulations.

trend in anthocyanin content was observed in strawberries and grape juice by Garzon et al.[53]

4.7. Total Sugars. The general term for soluble carbohydrates with a sweet flavor that is commonly found in food is sugar. Sugars can take various forms and are derived from multiple sources. Unlike non-reducing carbohydrates, which undergo partial oxidation by oxidizing agents, reducing sugars have the ability to reduce other compounds.

The analysis of variance conducted for total sugar content among the treatments and storage intervals revealed highly significant differences among the treatments, storage durations, and their interactions. To compare the mean values, the Tukey HSD test was applied ($P \leq 0.05$).

The results regarding the impact of different treatments are presented in Table 2. The table demonstrates that total sugar content increases with increasing concentrations of Karonda pulp and soluble solids (TSS). The highest total sugar content was observed in T9 (18.92%), which was significantly higher than the other treatments. The lowest total sugar content was observed in T1 (11.30%). The increasing trend of total sugar with higher levels of soluble solids and Karonda pulp concentration is independent of the source of sugars, similar to that of reducing and non-reducing sugars. This is because sugar is added to maintain different levels of soluble solids in the juice, and additionally, Karonda pulp contains complex sugars in the form of pectin. These results align with the findings of Gonçalves et al.[54] in strawberry pulp, where they reported significant nutrient content after 6 months of storage. Karonda, like strawberry, is a berry fruit with a higher total solids content, including pectin and other complex carbohydrates, which are not negatively affected by prolonged storage. Our findings are also consistent with the reported study of Bal et al.[55] in guava pulp. The increase in total sugar may be attributed to the solubilization of pulp constituents, the hydrolysis of polysaccharides such as pectin and starch, and the breakdown of complex carbohydrate polymers.

The data pertaining to different storage intervals and total sugar content are presented in Figure 6. The total sugar content increases from 12.90 to 16.89%. The significantly lowest value of total sugar was observed at the beginning of the experiment (12.90%), while the significantly highest content of total sugar (16.89%) was observed at the end of the storage period. The increase in total sugar can be attributed to the breakdown and solubilization of polysaccharides (pectin and starch) and insoluble carbohydrate polymers into simple sugars as the storage period progresses. Another factor contributing to the increase in total sugar is the loss of moisture over time.

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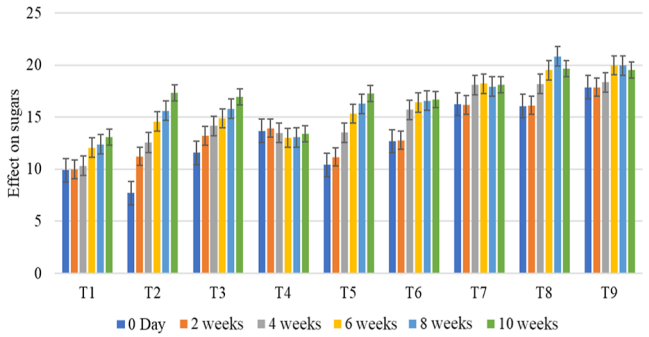

Figure 6. Influence of storage intervals on total sugars (%) of the nine Karonda juice formulations.

Similarly, Garg et al.[56] reported an increasing trend of total sugar in gooseberry juice, and Shahnawaz et al.[57] observed the same trend in blended mango and sea buckthorn pulp.

4.8. Antioxidant Activity. Antioxidants are compounds that prevent oxidation, a chemical reaction that can lead to the production of free radicals, potentially triggering further chain processes such as polymerization. Ascorbic acid, flavonoids, and phenols are antioxidants that halt these chain reactions. The two main sources of these antioxidants are industrial chemicals typically added to halt the oxidation process and plant-based antioxidants, renowned for their health benefits.

The treatments and storage intervals revealed a highly significant correlation among treatment, storage, and interactions. For individual comparisons, the means underwent the Tukey HSD test at $P \leq 0.05$ (Figure 7).

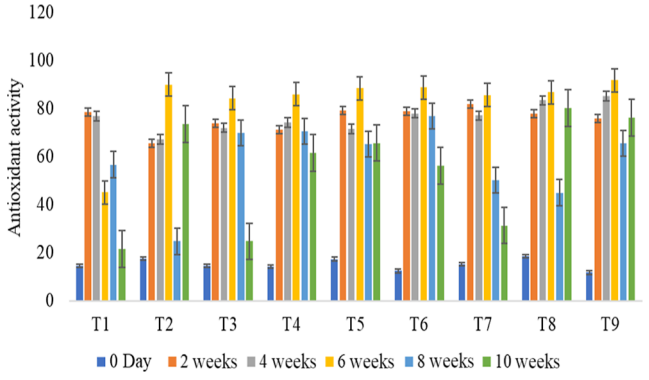

Figure 7. Effect of storage intervals on antioxidant activity (%) of the nine juice formulations.

The impact of different treatments, as shown in Table 2, demonstrated an increase in antioxidant activity with increasing concentrations of Karonda pulp and soluble solids (TSS). The highest antioxidant activity was observed in T9 (67.71%), significantly surpassing the other treatments. The lowest antioxidant activity was observed in T1 (48.88%). The enhancement in scavenging activity is attributed to the presence of various antioxidants in Karonda, including ascorbic acid, flavonoids, alkaloids, ursolic acid, cortisone, and triterpenoids. By increasing the concentration of Karonda, the scavenging activity of antioxidants increased. Similar results were also observed by Sajjabut et al.[58] in *C. carandas*.

The data regarding different storage intervals and antioxidant activity are presented in Figure 6. The table shows both an increasing and decreasing trend in antioxidant values. The antioxidant value increased from T1 to T6 (15.17−82.98) after 6 weeks of storage, while the antioxidant value exhibited a declining trend from T8 to T9 (58.28−54.58). A similar trend was observed with phenolics, as phenolics are key components for antioxidant activity in plants, and their increase or decrease is interdependent.[59] Total phenolics and anthocyanins are correlated with each other. Therefore, like phenols and anthocyanins, antioxidant activity also exhibits both increasing and decreasing trends. Another reason for the observed trends is the possible oxidation of bioactive compounds during storage. The similar decreasing trend of bioactive molecules during prolonged storage of kinnow−amla beverages, as reported by Purewal et al.,[36] supports our findings. They also reported an increase in antioxidant activity, as assessed by the DPPH method, during the storage of control fruits.

4.9. Sensory Evaluation. Food products undergo critical examination through the utilization of human senses in food sensory testing. Experienced testers evaluate characteristics such as appearance, texture, odor, and taste to assess the quality of a product and identify areas for improvement. In the case of Karonda fruit juice beverages, sensory attributes including overall acceptability, color, taste, and flavor were evaluated. A panel of 10 trained judges assessed the Karonda fruit juice samples using a hedonic scale ranging from extremely like to extremely dislike, with 9 points. Organoleptic parameters were evaluated at storage intervals of 0, 2, 6, 8, and 10 weeks.

4.9.1. Color. The visual perceptual characteristic resulting from the interaction between the spectrum of light and the photoreceptor cells in the eyes is referred to as color in American English or colour in British English. Objects or materials are assigned color groups and physical color standards based on their physical characteristics, such as light absorption, reflection, or emission spectra. The physical characteristics of a product, such as its absorption and emission spectra, generally influence its light emission or reflection. Evaluating the organoleptic properties of color is crucial to ensuring customer satisfaction as it is the most significant attribute in terms of consumer appeal. Visual cues influence more than 90% of purchase decisions, with 85% of consumers identifying color as the primary factor in their purchase choices.[60]

The treatments and storage intervals exhibited a highly significant correlation, while there was a significant effect among the interactions. For mean comparisons, mean values were subjected to Tukey's HSD test at a significance level of ($P \leq 0.05$).

The results for color in relation to the treatments are presented in Table 4. The graph clearly shows that the color of juice samples increased with an increasing concentration of Karonda pulp. The highest value was recorded in T9 (8.511), while the lowest value was recorded in T2 (4.58). The increase in visual perception in the juice from T1 to T9, resulting from an increase in Karonda pulp concentration, is attributed to the presence of various pigments, including anthocyanins, flavonoids, and tannins, which contribute to color development in Karonda juice. Nguyen et al.[59] also observed and confirmed that polyphenol oxidase is primarily responsible for color expression in Karonda.

A study on sensory characteristics revealed a decrease in the color of the beverage from 7.07 to 4.69% during the storage period. The highest value of 7.07% was observed at zero day of storage, while the lowest value of 4.69% was observed at 90

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Table 4. Sensory Characterization of Nine Karonda Juice Formulations on a Scale of 0−9[a]

parameters	storage time	T1	T2	T3	T4	T5	T6	T7	T8	T9
color	0 day	6.8333 ± 0.153^{b-j}	6.1667 ± 0.351^{e-m}	6.9333 ± 0.153^{a-i}	7.3333 ± 0.500^{a-h}	6.6667 ± 0.577^{c-l}	6.8333 ± 0.764^{b-j}	6.3333 ± 0.577^{d-m}	7.6667 ± 0.577^{a-f}	8.9333 ± 0.115^{a}
	2 weeks	6.3333 ± 0.153^{d-m}	5.7000 ± 0.400^{f-n}	6.7667 ± 0.153^{b-j}	6.5333 ± 0.819^{c-l}	6.3667 ± 0.586^{d-m}	6.5667 ± 0.777^{c-l}	6.1000 ± 0.608^{e-n}	7.4667 ± 0.493^{a-g}	8.8667 ± 0.153^{a}
	4 weeks	4.8667 ± 0.208^{j-p}	5.4667 ± 0.351^{g-o}	6.1000 ± 0.200^{e-n}	6.0667 ± 0.874^{e-n}	6.0333 ± 0.551^{e-n}	6.2333 ± 0.862^{d-m}	6.8000 ± 0.608^{f-n}	7.2667 ± 0.493^{a-h}	8.7000 ± 0.200^{a-b}
	6 weeks	4.1333 ± 0.231^{n-q}	4.3667 ± 0.321^{m-q}	5.5667 ± 0.208^{j-n}	6.0333 ± 0.950^{e-n}	5.6667 ± 0.577^{f-n}	5.8667 ± 0.907^{f-n}	5.4333 ± 0.666^{h-o}	7.0333 ± 0.551^{a-i}	8.4333 ± 0.208^{a-c}
	8 weeks	3.4000 ± 0.361^{p-q}	3.3000 ± 0.500^{pq}	5.1667 ± 0.153^{a-d}	5.5333 ± 0.950^{g-n}	5.2333 ± 0.643^{i-p}	5.5333 ± 0.929^{g-n}	5.1667 ± 0.723^{i-p}	6.7333 ± 0.643^{b-k}	8.2000 ± 0.200^{a-d}
	10 weeks	2.4000 ± 0.693^{q}	2.5000 ± 0.500^{q}	3.5000 ± 0.866^{o-q}	4.6667 ± 1.155^{l-p}	4.7333 ± 0.643^{k-p}	5.1667 ± 1.041^{i-p}	4.8333 ± 0.839^{j-p}	6.5000 ± 0.700^{c-l}	7.9333 ± 0.115^{a-e}
flavor	0 day	7.1000 ± 0.200^{a-c}	6.9667 ± 0.153^{a-c}	7.0000 ± 0.200^{a-c}	6.3333 ± 1.528^{a-e}	6.5000 ± 1.323^{a-e}	7.6667 ± 0.577^{a}	6.5333 ± 0.500^{a-e}	7.6667 ± 0.577^{a}	8.1667 ± 1.041^{a}
	2 weeks	6.7667 ± 0.321^{a-d}	6.6667 ± 0.208^{a-d}	6.7333 ± 0.153^{a-d}	6.2433 ± 1.544^{a-e}	6.4333 ± 1.332^{a-e}	7.5433 ± 0.561^{ab}	6.4667 ± 0.751^{a-e}	7.6767 ± 0.686^{a}	7.9100 ± 1.055^{a}
	4 weeks	5.7333 ± 0.321^{a-f}	6.7667 ± 0.252^{a-d}	6.7667 ± 0.252^{a-d}	6.4233 ± 1.349^{a-e}	6.2000 ± 1.308^{a-e}	7.5567 ± 0.751^{ab}	6.2333 ± 0.651^{a-e}	7.6567 ± 0.486^{a}	7.8000 ± 1.015^{a}
	6 weeks	4.5667 ± 0.513^{b-g}	5.4000 ± 0.361^{a-f}	6.1000 ± 0.200^{a-e}	6.0567 ± 1.566^{a-e}	6.1000 ± 1.442^{a-e}	7.4567 ± 0.483^{ab}	6.0000 ± 0.721^{a-e}	7.4233 ± 0.541^{ab}	7.7433 ± 1.161^{ab}
	8 weeks	3.5000 ± 0.500^{e-g}	4.3667 ± 0.351^{c-g}	5.5000 ± 0.200^{a-f}	5.7433 ± 1.514^{a-f}	5.9000 ± 1.442^{a-f}	7.1767 ± 0.501^{a-c}	6.0333 ± 0.902^{a-e}	7.1767 ± 0.781^{a-c}	7.5000 ± 1.114^{ab}
	10 weeks	2.2333 ± 0.751^{g}	2.9000 ± 0.361^{f-g}	3.8333 ± 0.802^{d-g}	5.4000 ± 1.442^{a-f}	5.5667 ± 1.570^{a-f}	6.9333 ± 0.404^{a-c}	5.8000 ± 0.854^{a-f}	7.0333 ± 0.451^{a-c}	7.1667 ± 1.002^{a-c}
taste	0 day	7.2000 ± 0.200^{a-o}	6.9667 ± 0.153^{a-a}	7.0000 ± 0.200^{a-c}	6.3333 ± 1.528^{a-e}	6.5000 ± 1.323^{a-e}	7.6667 ± 0.577^{a}	6.5000 ± 0.500^{a-e}	7.6667 ± 0.577^{a}	8.1667 ± 1.041^{a}
	2 weeks	6.9000 ± 0.200^{a-o}	6.6667 ± 0.208^{a-d}	6.7333 ± 0.153^{a-d}	6.2433 ± 1.544^{a-e}	6.4333 ± 1.332^{a-e}	7.5433 ± 0.561^{ab}	6.4667 ± 0.751^{a-e}	7.6767 ± 0.686^{a}	7.9100 ± 1.055^{a}
	4 weeks	5.7333 ± 0.321^{a-f}	5.9000 ± 0.200^{a-f}	6.7667 ± 0.252^{a-d}	6.4233 ± 1.349^{a-e}	6.2000 ± 1.308^{a-e}	7.5567 ± 0.751^{ab}	6.2333 ± 0.651^{a-e}	7.6567 ± 0.486^{a}	7.8000 ± 1.015^{a}
	6 weeks	4.5667 ± 0.513^{b-g}	5.4000 ± 0.361^{a-f}	6.1000 ± 0.200^{a-e}	6.0567 ± 1.566^{a-e}	6.1000 ± 1.442^{a-e}	7.4567 ± 0.483^{ab}	6.0000 ± 0.721^{a-e}	7.4233 ± 0.541^{ab}	7.7433 ± 1.161^{ab}
	8 weeks	3.5000 ± 0.500^{e-g}	4.3667 ± 0.351^{c-g}	5.5000 ± 0.200^{a-f}	5.7433 ± 1.514^{a-f}	5.9000 ± 1.442^{a-f}	7.1767 ± 0.501^{a-c}	6.0333 ± 0.902^{a-e}	7.1767 ± 0.781^{a-c}	7.5000 ± 1.114^{ab}
	10 weeks	2.2333 ± 0.751^{g}	2.9000 ± 0.361^{f-g}	3.8333 ± 0.802^{d-g}	5.4000 ± 1.442^{a-f}	5.5667 ± 1.570^{a-f}	6.9333 ± 0.404^{a-c}	5.8000 ± 0.854^{a-f}	7.0333 ± 0.451^{a-c}	7.1667 ± 1.002^{a-c}
overall acceptability	0 day	7.2000 ± 0.200^{a-o}	7.0333 ± 0.153^{a-o}	7.7333 ± 0.252^{a-m}	6.4667 ± 0.577^{f-p}	7.0000 ± 1.000^{a-o}	8.1667 ± 0.289^{a-i}	8.5000 ± 0.866^{a-f}	8.0000 ± 0.000^{a-k}	9.0000 ± 0.000^{a}
	2 weeks	6.9000 ± 0.200^{a-o}	6.5333 ± 0.153^{e-o}	7.5000 ± 0.361^{a-m}	6.2667 ± 0.814^{g-p}	6.8100 ± 1.115^{c-o}	8.2333 ± 0.451^{a-h}	8.2833 ± 0.858^{a-g}	7.9767 ± 0.204^{a-k}	8.9267 ± 0.064^{ab}
	4 weeks	6.3000 ± 0.200^{g-p}	6.0333 ± 0.153^{j-q}	6.8333 ± 0.208^{b-o}	6.1000 ± 0.700^{i-p}	6.7233 ± 1.135^{d-o}	8.0667 ± 0.153^{a-j}	7.7667 ± 0.833^{a-m}	7.9900 ± 0.115^{a-k}	8.8667 ± 0.153^{a-c}
	6 weeks	5.1000 ± 0.173^{o-q}	5.2667 ± 0.252^{n-q}	7.3000 ± 0.058^{a-n}	5.9000 ± 0.781^{k-q}	6.5567 ± 1.186^{e-o}	7.8167 ± 0.161^{a-l}	7.7000 ± 0.964^{a-m}	7.7233 ± 0.040^{a-m}	8.7667 ± 0.058^{a-d}
	8 weeks	4.4000 ± 0.321^{p-r}	3.9333 ± 0.603^{o-r}	6.9333 ± 0.252^{a-o}	5.8000 ± 0.964^{l-q}	6.2767 ± 1.165^{g-p}	7.5667 ± 0.208^{a-m}	7.4500 ± 0.926^{a-m}	7.5767 ± 0.197^{a-m}	8.6000 ± 0.173^{a-e}
	10 weeks	2.5000 ± 1.323^{rs}	1.6667 ± 0.577^{s}	6.7333 ± 0.231^{d-o}	5.6667 ± 0.907^{m-q}	6.1333 ± 1.102^{h-p}	7.3000 ± 0.361^{a-n}	7.1667 ± 0.990^{a-o}	7.1667 ± 0.289^{a-o}	8.3333 ± 0.231^{a-g}

[a]Mean values of different treatments and storage intervals are shown in capital letters, non-significant interactions are shown by the same letters, while different letters indicate significant interactions ($P \leq 0.05$). Where,

T1= 20% fruit Karonda pulp	+12 °Brix	T4= 30% fruit Karonda pulp	+12 °Brix	T7= 40% fruit Karonda pulp	+12 °Brix
T2= 20% fruit Karonda pulp	+14 °Brix	T5= 30% fruit Karonda pulp	+14 °Brix	T8= 40% fruit Karonda pulp	+14 °Brix
T3= 20% fruit Karonda pulp	+16 °Brix	T6= 30% fruit Karonda pulp	+16 °Brix	T9= 40% fruit Karonda pulp	+16 °Brix

days of storage. The table indicates that the color of the juice samples deteriorates as the storage period progresses. This decline is attributed to the degradation of pigments, enzymatic browning, and oxidative loss caused by the presence of oxygen during storage. Song et al.[61] observed a similar decreasing trend in color during the storage of blueberry juice. A similar decline in the color score during prolonged storage was also observed in kinnow and amla juice by Purewal et al.[36] in orange juice.

4.9.2. Flavor. The sensory impressions experienced while eating and drinking are referred to as flavors. These impressions are created by the chemical perceptions of taste and smell. In order to perceive flavor as a whole, the evaluation of several chemical irritants by the "trigeminal senses" in the mouth and throat is crucial. The addition of natural and artificial flavoring molecules can alter the taste of food, helping to maintain a pleasurable and authentic flavor without any undesirable notes.

The results for flavor in relation to the treatments are presented in Table 4. It is evident from the table that the flavor of the juice samples increased with an increasing concentration of Karonda pulp and TSS. The highest value was recorded in

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T9 (7.71), while the lowest value was recorded in T1 (4.98). This increase in the flavor of the juice samples from T1 to T9 can be attributed to the presence of various nutraceutical components in Karonda fruit. Therefore, increasing the pulp concentration also enhances the flavor of the juice, leading to the higher score given by the judges for T9.

A decreasing trend in flavor was observed with the progression of storage. The lowest score (5.20) was observed on the final day of the experiment, while the highest score (7.10) was reported on the first day of storage. This loss of flavor is attributed to the degradation and conversion of flavoring substances, such as octanal, ethyl butanoate, decanal, and β-myrcene, into complex chemicals over time. Bhuiyan et al.[62] observed a similar decreasing trend in flavor in functional beverages. Similarly, Cheng et al.[63] found a decreasing trend in flavor in mandarin juice.

4.9.3. Taste. The sensory system responsible for the perception of flavor is the sense of taste. When a substance in the mouth chemically combines with taste receptor cells on taste buds, primarily located on the tongue, it generates or stimulates the sensation of taste. The taste of a food component is determined by the activation of nerves.

The results of the treatments in relation to taste are presented in Table 4. It is evident from the table that the taste of the juice samples increased with increasing concentrations of Karonda pulp and soluble solids (SS). The highest value was recorded in T9 (7.70), while the lowest value was recorded in T1 (5.09), which contained 40% Karonda pulp and 16 Brix. This increase in taste from T1 to T9 can be attributed to the presence of nutraceutical components in Karonda fruit. Therefore, as the pulp concentration increases, the taste of the juice also improves, leading to the higher score given by the panel of judges for T9.

The taste of the juice declined as the storage time increased. During the storage period, the highest score (7.23) was recorded on the first day of the experiment, while the lowest score (5.21) was recorded on the last day. This decline in taste can be attributed to the presence of oxygen in the air, which breaks down the sugar in Karonda juice and negatively affects its taste. Another factor is the presence of tannin in Karonda juice, which also contributes to a decrease in taste score over time. Similar decreasing trends in taste were observed in fruit juices by Rocha and Bolini[64] and Samborska et al.[65] Additionally, previous studies by Purewal et al.[36] also demonstrated a declining trend in the taste of kinnow and amla juice over storage intervals, which aligns with our investigation.

4.9.4. Overall Acceptability. Consumer beliefs and expectations have been shown to significantly impact the overall acceptability of food based on its sensory characteristics, and these influences directly affect consumer behavior, including consumption. The treatments and storage intervals demonstrated a highly significant correlation among them. The results were then subjected to the Tukey's HSD test at a significance level of ($P \leq 0.05$) for the mean comparison.

The comparative results indicated that the highest overall acceptability score (8.74) was observed for T9, followed by T8 (7.73), while the lowest score (5.07) was recorded for T2. The overall acceptability of the treatments increased with increasing concentrations of Karonda pulp and TSS values (Figure 8). The beverage sample with 40% Karonda pulp and 16 °Brix yielded the most favorable results in terms of acceptability. T9 received the highest score and was deemed more acceptable by the judges.

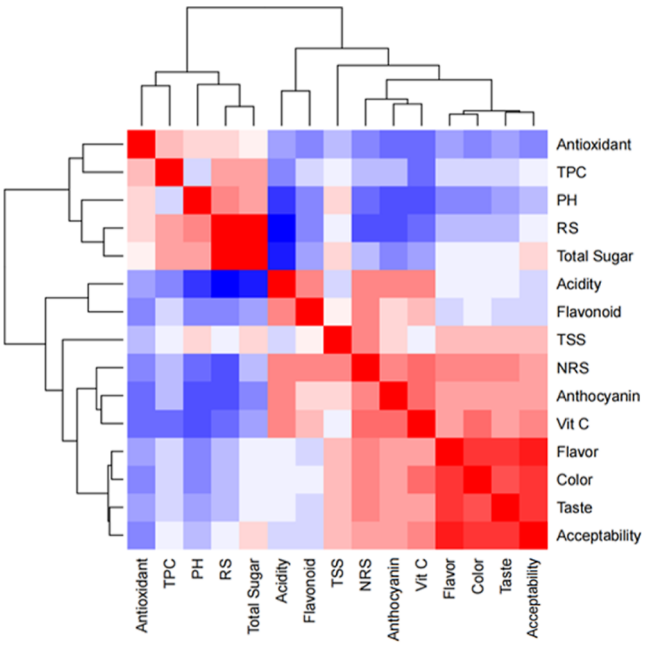

Figure 8. Heat map analysis of parameters studied for nine Karonda juice formulations.

The scores for overall acceptability given by the judges, as shown in Table 4, exhibited a decreasing trend with increasing storage intervals. The highest score (7.67) was observed on the first day of storage, while the lowest score (5.85) was recorded on the last day of the experiment.

Over time, the decline in color, flavor, taste, and overall acceptability of Karonda juice can be attributed to the presence of tannin and the loss of flavoring substances such as octanal, ethyl butanoate, decanal, and myrcene. These substances may be lost due to breakdown and conversion into complex chemicals. Bhuiyan et al.[62] observed a similar decreasing trend in the overall acceptability of functional beverages. Likewise, Samborska et al.[65] observed a decreasing trend in berry fruit juice. The decline in overall acceptability of Karonda juice with increasing storage duration is also consistent with the findings of Purewal et al.,[36] whose results demonstrated a diminishing trend in the overall acceptability of kinnow and amla juice over storage intervals of approximately 10 weeks.

4.10. Heat Map and Correlation Analysis. Heat map analysis in food provides a visual representation of the distribution and intensity of chemicals, which helps in identifying trends, changes, and hotspots and enhances understanding of the composition and quality of food samples. The cluster heat map (Figure 8) revealed that antioxidants, TPC, pH, RS, and total sugar were grouped together but showed weak or no association. However, these traits exhibited a strong positive association with acidity, flavonoid, NRS, and vitamin C. Similarly, the remaining traits were clustered together, with acidity showing a strong positive association with pH, RS, and total sugar. Flavor, color, taste, and acceptability demonstrated positive associations with antioxidants, TPC, pH, and RS, but showed negative associations with TSS, NRS, anthocyanin, and vitamin C.

The correlation matrix (Figure 9) indicated a strong positive correlation between pH, RS, total sugars, and TPC, but these

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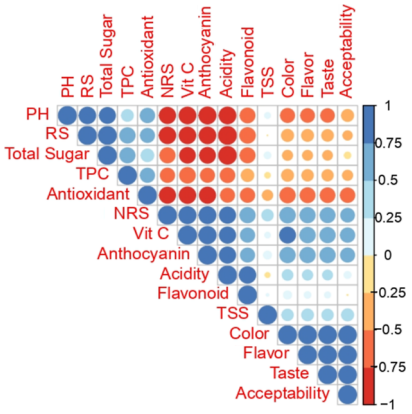

Figure 9. Correlation analysis of all studied parameters.

traits exhibited a strong negative correlation with NRS, vitamin C, anthocyanin, acidity, and flavonoid. Except for a few, pH and antioxidants showed a strong negative relationship with the rest of the traits. Color, flavor, taste, and acceptability also showed weak or no correlation with pH, RS, total sugars, TPC, and antioxidants. The use of heat maps for quality assessment and optimization of juice generation from fruit samples has been previously well reported.[37,66,67] In a previous study, Zhang et al.[67] generated a heat map through Pearson correlation analysis to comprehensively assess fruit quality and identify aroma-active compounds in green pepper. Their research findings elucidated the primary texture-related attributes of pepper and identified the key aroma-active compounds, thereby offering valuable insights for improving pepper quality through breeding programs and establishing consumer guidelines.

5. CONCLUSIONS

Underutilized fruits have untapped potential to address the issue of food insecurity and benefit the global population. *C. spinarum* L., an undervalued berry fruit found in Azad Jammu and Kashmir, is currently being wasted due to a lack of awareness among the local community. The presented investigation aims to highlight the potential of this fruit and promote the development of functional food products. Among the various beverage formulations tested, the one with a 40% fruit base and a Brix value of 16° was found to be the most suitable in terms of functional and quality attributes, as well as consumer preference. This beverage is a rich source of phytonutrients, including anthocyanin, phenolics, and antioxidants, which are essential for human health and provide protection against various diseases. Therefore, this new prototype formulation holds the potential for commercialization, offering a profitable business opportunity while providing the community with a nutritious and refreshing functional food product. The research supports the production of valuable food products using the nutrient-rich Karonda fruit, thereby reducing the risk of nutritional deficiencies. Furthermore, it provides guidance for the development of an appropriate breeding program to enhance the current nutritional profile of the wild Karonda ecotypes used in the preparation of the first functional beverage.

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Notes

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REFERENCES

(1) Corbo, M. R.; Bevilacqua, A.; Petruzzi, L.; Casanova, F. P.; Sinigaglia, M. Functional beverages: The emerging side of Functional Foods. *Compr. Rev. Food Sci. Food Saf.* **2014**, *13*, 1192−1206.

(2) Cong, L.; Bremer, P.; Mirosa, M. Functional beverages in selected countries of Asia Pacific Region: A Review. *Beverages* **2020**, *6*, 21.

(3) Siró, I.; Kápolna, E.; Kápolna, B.; Lugasi, A. Functional Food. product development, marketing and Consumer Acceptance—a review. *Appetite* **2008**, *51*, 456−467.

(4) Liu, X.; Zhao, Y.; Mu, J.; Zhang, J.; Zhang, A. Determination of geographical origin of concentrated apple juice through analysis of stable isotopic and mineral elemental fingerprints: Preliminary results. *J. Sci. Food Agric.* **2021**, *101*, 3795−3803.

(5) Zapata, M.; Chaparro, D.; Rojano, B.; Alzate, A.; Restrepo, L.; Maldonado, M. Effect of storage time on physicochemical, sensorial, and antioxidant characteristics, and composition of Mango (cv. azúcar) juice. *Emir. J. Food Agric.* **2017**, *29*, 367.

(6) Granato, D.; Barba, F. J.; Bursać Kovačević, D.; Lorenzo, J. M.; Cruz, A. G.; Putnik, P. Functional Foods: Product Development, technological trends, efficacy testing, and safety. *Annu. Rev. Food Sci. Technol.* **2020**, *11*, 93−118.

(7) Sun-Waterhouse, D. The development of fruit-based functional foods targeting the health and wellness market: A Review. *Int. J. Food Sci. Technol.* **2011**, *46*, 899−920.

(8) Nazir, M.; Arif, S.; Khan, R. S.; Nazir, W.; Khalid, N.; Maqsood, S. Opportunities and challenges for functional and medicinal beverages: Current and future trends. *Trends Food Sci. Technol.* **2019**, *88*, 513−526.

(9) Arora, R. K. *Diversity in underutilized plant species: an Asia-Pacific perspective*; Bioversity International, 2014.

(10) Abanoz, Y. Y.; Okcu, Z. Biochemical content of cherry laurel (Prunus laurocerasus L.) fruits with edible coatings based on caseinat, Semperfresh and lecithin. *Turk. J. Agric. For.* **2022**, *46*, 908−918.

(11) Dawadi, P.; Shrestha, R.; Mishra, S.; Bista, S.; Raut, J. K.; Joshi, T. P.; Bhatt, L. R. Nutritional value and antioxidant properties of Viburnum mullaha Buch.-Ham. Ex D. Don fruit from central Nepal. *Turk. J. Agric. For.* **2022**, *46*, 781−789.

(12) Devmurari, V.; Shivanand, P.; Goyani, M.; Vaghani, S.; Jivani, N. A review: Carissa congesta: phytochemical constituents, traditional use and pharmacological properties. *Pharmacogn. Rev.* **2009**, *3*, 375.

(13) Taş, A.; Gundogdu, M. Physiological characterization of wild cornelian cherry genotypes in terms of phenolic compounds, organic acids and antioxidants. *Genet. Resour. Crop Evol.* **2023**, *10*, DOI: 10.1007/s10722-023-01578-9.

(14) Gemechu, B.; Dagmawit, A.; Venkataramana, K. A Review of the Medicinal and Antimicrobial Properties of *Carissa spinarum* L. *Am. J. Biomed. Res.* **2020**, *8*, 54−58.

(15) Fatima, A.; Singh, P. P.; Agarwal, P.; Irchhaiya, R.; Alok, S.; Verma, A. Treatment of various diseases by Carissa spinarum L.: a promising shrub. *Int. J. Pharm. Sci. Res.* **2013**, *4*, 2489−2495.

(16) Mundaragi, A.; Thangadurai, D. Process optimization, physicochemical characterization and antioxidant potential of novel wine from an underutilized fruit *Carissa spinarum* L. (Apocynaceae). *J. Food Sci. Technol.* **2017**, *38*, 428−433.

(17) Sarkar, R.; Kundu, A.; Banerjee, K.; Saha, S. Anthocyanin composition and potential bioactivity of Karonda (Carissa Carandas L.) fruit: An Indian source of biocolorant. *LWT—Food Sci.* **2018**, *93*, 673−678.

(18) Meghwal, P. R.; Singh, S. K.; Singh, A.; Pathak, R. Characterization of karonda (Carissa Carandas) accessions under Arid Region. *J. Appl. Hortic.* **2014**, *16*, 157−160.

(19) Wiart, C. Medicinal plants classified in the family Apocynaceae. In *Medicinal Plants of Asia and the Pacific*; CRC Press: Boca Raton, Florida, 2006.

(20) Das, S. C.; Prakash, J.; Deb, A. K.; Biswas, T. Medicinal value of underutilized fruits in Hilly Tripura. *Acta Hortic.* **2013**, 135−141.

(21) Doshi, G.; Chaskar, P. K.; View, M. Antiinflammatory and antibacterial potential of *Carissa carandas* and *Carissa spinarum* nanoemulsions. *Int. J. Pharm. Sci. Res.* **2017**, *8*, 5137−5145.

(22) Patel, S. Food, pharmaceutical and industrial potential of *Carissa* genus: An overview. *Rev. Environ. Sci. Biotechnol.* **2013**, *12*, 201−208.

(23) Kaunda, J. S.; Zhang, Y. J. The Genus Carissa: An Ethnopharmacological, Phytochemical and Pharmacological Review. *Nat. Prod. Bioprospect.* **2017**, *7*, 181−199.

(24) Singh, S.; Bajpai, M.; Mishra, P. *Carissa carandas* L. − phyto-pharmacological review. *J. Pharm. Pharmacol.* **2020**, *72*, 1694−1714.

(25) Wani, R. A.; Prasad, V. M.; Hakeem, S. A.; Sheema, S.; Angchuk, S.; Dixit, A. Shelf life of Karonda jams (*Carissa carandas* L.) under ambient temperature. *Afr. J. Agric. Res.* **2013**, *8*, 2447−2449.

(26) Navya, D. V.; Kukanoor, L.; Gorabal, K.; Bhat, A.; Jalawadi, S. Studies on organoleptic qualities and colour (L* a* b*) values of Karonda (*Carissa carandas* L.) blended squash during storage. *J. Pharmacogn. Phytochem.* **2020**, *9*, 994−997.

(27) Chauhan, A.; Tanwar, B.; Intelli. Influence of Processing on Physicochemical, Nutritional and Phytochemical Composition of Ficus carica (Fig) Fruit. *Res. J. Pharm., Biol. Chem. Sci.* **2015**, *6*, 1474−1489.

(28) Lim, T. K. Carissa spinarum. *Edible Medicinal and Non-Medicinal Plants*; Springer: Dordrecht, 2012.

(29) Nazareth, M. S.; Shreelakshmi, S. V.; Shetty, N. P. Identification and characterization of polyphenols from Carissa spinarum fruit and evaluation of their antioxidant and anti-quorum sensing activity. *Curr. Microbiol.* **2021**, *78*, 1277−1285.

(30) Association of Official Analytical Chemists. *Official Methods of Analysis*; The Association of Official Analytical Chemists. Inc., 18th ed.: Arlington, USA, 2006.

(31) Association of Official Analytical Chemists. *Official Method of Analysis*; The Association of Official Analytical Chemistry Washington, 14th ed.: Arlington, 1990; Vol. *1*, pp 88-91.

(32) Singleton, V. L.; Orthofer, R.; Lamuela-Raventos, R. M. Analysis of Total Phenols and Other Oxidation Substrates and Antioxidants by Means of Folin-Ciocalteu Reagent. *Methods Enzymol.* **1999**, *299*, 152−178.

(33) Chang, C.-C.; Yang, M.-H.; Wen, H.-M.; Chern, J.-C. Estimation of total flavonoid content in propolis by two complementary colometric methods. *J. Food Drug Anal.* **2020**, *10*, 3.

(34) Golmohamadi, A.; Möller, G.; Powers, J.; Nindo, C. Effect of ultrasound frequency on antioxidant activity, total phenolic and anthocyanin content of red raspberry puree. *Ultrason. Sonochem.* **2013**, *20*, 1316−1323.

(35) Brand-Williams, W.; Cuvelier, M. E.; Berset, C. Use of a free radical method to evaluate antioxidant activity. *LWT—Food Sci. Technol.* **1995**, *28*, 25−30.

(36) Purewal, S. S.; Kamboj, R.; Sandhu, K. S.; Kaur, P.; Sharma, K.; Kaur, M.; Salar, R. K.; Punia, S.; Siroha, A. K. Unraveling the effect of storage duration on antioxidant properties, physicochemical and sensorial parameters of ready to serve Kinnow-Amla beverages. *Appl. Food Res.* **2022**, *2*, 100057.

(37) Jobil, A. J.; Parameshwari, S.; Husain, F. M.; Alomar, S. Y.; Ahmad, N.; Albalawi, F.; Alam, P. Scientifically formulated avocado fruit juice: Phytochemical analysis, Assessment of its antioxidant potential and consumer perception. *Molecules* **2021**, *26*, 7424.

(38) Jan, A.; Masih, Er. D. Development and quality evaluation of pineapple juice blend with carrot and orange juice. *Int. j. sci. res. publ.* **2012**, *2*, 1−8.

(39) Rehman, M. A.; Khan, M. R.; Sharif, M. K.; Ahmad, S.; Shah, F. U. H. Study on the storage stability of fruit juice concentrates. *Pak. J. Food Sci.* **2014**, *24*, 101−107.

(40) Jeppsson, N.; Johansson, R. Changes in fruit quality in black chokeberry (*aronia melanocarpa*) during maturation. *J. Hortic. Sci. Biotechnol.* **2000**, *75*, 340−345.

(41) Divate, S. M.; Savale, B. G.; Patil, N. B.; Relekar, P. P. Study of preparation and standardization of Karonda (*Carissa congesta* L.) crush. *Pharma. Innov* **2020**, *9*, 176−179.

https://doi.org/10.1021/acsomega.3c03386
ACS Omega 2023, 8, 32643−32655

(42) Singh, S.; Godara, R. K.; Saini, R. S.; Sharma, J. Standardization of processing technology for Baell blended Bael (Aegele marmelos) ready-to-serve beverages. *Haryana J. Hortic. Sci.* **2005**, *34*, 263.
(43) Singh, S. V.; Jain, R. K.; Gupta, A. K. Changes in quality of debittered kinnow juice during storage. *J. Food Sci. Technol.* **2009**, *46*, 598−600.
(44) Pareek, S.; Paliwal, R.; Mukherjee, S. Effect of juice extraction methods and processing temperature-time on juice quality of Nagpur Mandarin (citrus reticulata blanco) during storage. *J. Food Sci. Technol.* **2010**, *48*, 197−203.
(45) Pehlivan, F. E. *Vitamin C: An Antioxidant Agent*. ISBN987-953-51-3422; IntechOpen, 2017. 69660.
(46) Kabasakalis, V. Ascorbic acid content of commercial fruit juices and its rate of loss upon storage. *Food Chem.* **2000**, *70*, 325−328.
(47) Reque, P. M. R. S.; Steffens, R. S.; Jablonski, A.; Flôres, S. H.; Rios, A. d. O.; de Jong, E. V. Cold storage of blueberry (Vaccinium spp.) fruits and juice: Anthocyanin stability and antioxidant activity. *J Food Compost Anal.* **2014**, *33*, 111−116.
(48) Choi, M.-Y.; Chai, C.; Park, J. H.; Lim, J.; Lee, J.; Kwon, S. W. Effects of storage period and heat treatment on phenolic compound composition in dried citrus peels (Chenpi) and discrimination of Chenpi with different storage periods through targeted metabolomic study using HPLC-dad analysis. *J. Pharm. Biomed. Anal.* **2011**, *54*, 638−645.
(49) Khuanekkaphan, M.; Khobjai, W.; Noysang, C.; Wisidsri, N.; Thungmungmee, S. Bioactivities of Karanda (*Carissa Carandas* Linn.) fruit extracts for novel cosmeceutical applications. *J. Adv. Pharm. Technol. Res.* **2021**, *12*, 162.
(50) Borges, G.; Degeneve, A.; Mullen, W.; Crozier, A. Identification of flavonoid and phenolic antioxidants in black currants, blueberries, raspberries, Red Currants, and cranberries. *J. Agric. Food Chem.* **2009**, *58*, 3901−3909.
(51) Bradish, C. M.; Perkins-Veazie, P.; Fernandez, G. E.; Xie, G.; Jia, W. Comparison of flavonoid composition of Red Raspberries (*Rubus idaeus L.*) grown in the Southern United States. *J. Agric. Food Chem.* **2011**, *60*, 5779−5786.
(52) Kaur, M.; Poonam, A.; Mudasir, Y. Retention of Colour and Phytochemicals in strawberry pulp in response to the pulping method, chemical treatments, pasteurization, and storage period. *Int. J. Agric. Sci.* **2018**, *10*, 6283−6288.
(53) Garzon, G. A.; Wrolstad, R. E. Comparison of the stability of Pelargonidin-based anthocyanins in strawberry juice and concentrate. *J. Food Sci.* **2002**, *67*, 1288−1299.
(54) Gonçalves, G. A.; Resende, N. S.; Carvalho, E. E.; Resende, J. V.; Vilas Boas, E. V. Effect of pasteurisation and freezing method on bioactive compounds and antioxidant activity of Strawberry Pulp. *Int. J. Food Sci. Nutr.* **2017**, *68*, 682−694.
(55) Bal, L. M.; Ahmad, T.; Senapati, A. K.; Pandit, P. S. Evaluation of quality attributes during storage of guava nectar cv. Lalit from different pulp and TSS ratio. *J. Food Process. Technol.* **2014**, *5*, 329−334.
(56) Garg, V.; Barwal, V. S.; Sarera, S. Preparation and evaluation of vitamin C enriched fruit drink. *J. Food Sci. Technol.* **2008**, *45*, 524−526.
(57) Shahnawaz, M.; Wali, A.; Durrani, Y.; Ayub, M.; Muhammad, A.; Khan, R.; Khan, H. Refrigerated storage studies of mango seabuckthorn blended pulp with selected preservative and ginger extract. *Int. J. Food Nutr. Sci.* **2012**, *1*, 01−07.
(58) Sajjabut, S.; Pewlong, W.; Eamsiri, J.; Chookaew, S. Antioxidant potential of Carissa carandas seed comparing to the dietary supplement products. *Food Appl. Biosci. J.* **2018**, *6*, 95−104.
(59) Nguyen, Q. D.; Vu, T.-D.; Nguyen, T. T.; Phan, T.-K.; Pham, H.-T.; Nguyen, P. T. Microencapsulation of karonda (Carissa carandas L.) anthocyanin extracts: Effects of drying conditions on antioxidant contents and activities of spray-dried powder. *Pigm. Resin Technol.* **2022**, DOI: 10.1108/PRT-12-2021-0141.
(60) García-Madariaga, J.; Blasco López, M. F.; Burgos, I. M.; Virto, N. R. Do isolated packaging variables influence consumers' attention and preferences? *Physiol. Behav.* **2019**, *200*, 96−103.
(61) Song, H.-N.; Ji, S.-A.; Park, H.-R.; Kim, H.-H.; Hogstrand, C. Impact of various factors on color stability of fresh blueberry juice during storage. *Prev. Nutr. Food Sci.* **2018**, *23*, 46−51.
(62) Bhuiyan, M. H. R.; Shams-Ud-Din, M.; Islam, M. N. Development of functional beverage based on taste preference. *J. environ. sci nat. resour.* **2012**, *5*, 83−87.
(63) Cheng, Y.; Li, G.; Wu, H.; Liang, G.; Wang, H. Flavor deterioration of Mandarin juice during storage by MDGC-MS/O and GC-MS/PFPD. *LWT–Food Sci. Technol.* **2022**, *159*, 113132.
(64) Rocha, I. F.; Bolini, H. M. Passion fruit juice with different sweeteners: Sensory profile by descriptive analysis and acceptance. *Food Sci. Nutr.* **2015**, *3*, 129−139.
(65) Samborska, K.; Eliasson, L.; Marzec, A.; Kowalska, J.; Piotrowski, D.; Lenart, A.; Kowalska, H. The effect of adding berry fruit juice concentrates and by-product extract to sugar solution on osmotic dehydration and sensory properties of apples. *J. Food Sci. Technol.* **2019**, *56*, 1927−1938.
(66) Kalaimurugan, D.; Sivasankar, P.; Manikandan, E.; Durairaj, K.; Lavanya, K.; Vasudhevan, P.; Lakshmanan, R.; Venkatesan, S. Spatial determination of soil variables using GIS method and their influence on microbial communities in the Eastern Ghats region. *Trop. Ecol.* **2019**, *60*, 16−29.
(67) Zhang, J.; Wang, C.; Wang, J.; Yang, Y.; Han, K.; Bakpa, E. P.; Li, J.; Lyu, J.; Yu, J.; Xie, J. Comprehensive fruit quality assessment and identification of aroma-active compounds in green pepper (Capsicum annuum L.). *Front. Nutr.* **2023**, *9*, 1027605.
(68) Siyum, Z. H.; Meresa, T. A. Physicochemical properties and nutritional values of *carissa spinarum* L. "agam" fruit. *Int. J. Fruit Sci.* **2021**, *21*, 826−834.

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