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A STUDY ON THE DEVELOPMENT AND EVALUATION OF TAMARIND CANDY BY BLENDING WITH DIFFERENT SPICES

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ABSTRACT

The study was done to look into the physicochemical properties of tamarind candy developed from blending different spices. The proximate analysis and the analysis of the antioxidants reveal that the candy contains reasonable amounts, which is beneficial for health. The sample was produced and stored at ambient temperature (43 ^{0}C) for 20 days. The colour of the sample was visually observed, and it was found that there was no difference in colour during accelerated storage conditions. The reading for the water activity of fresh candy was 0.344, and the result indicates that water activity was decreased as per the prolonged storage of the sample. The overall acceptability of tamarind candy after 20 days of accelerated storage was good. As per the study, the accelerated ageing test calculation indicates that a product shelf life of more than three months at ambient temperature is acceptable.

Keywords: Tamarind pulp, Tamarind candy, the shelf life of candy, antioxidants, physicochemical analysis.

INTRODUCTION

The tamarind fruit pulp is widely employed in many Indian cuisines. *Tamarindus indica* belongs to the family Leguminosae. The native place of tamarind plants in Africa. The fruit's pulp is sweet-sour in taste, juicy and brown when ripened. Tamarind is additionally used as folk medicine because the fruit exhibits a laxative effect due to malic acid and tartaric acid; hence, it is suitable for constipation. Tamarind fruit paste has many culinary uses, such as flavouring curries, chutneys, and plenty of traditional drinks[1]. In many developed countries, tamarind pulp is employed extensively within the confectionery industry[2]. The tamarind fruit pulp is an excellent source of sugars, water-soluble vitamins, minerals and phenolic compounds as antioxidants[3]. Tamarind is also rich in ascorbic acid and citric acid. Tartaric acid endorses iron absorption in the bloodstream[4]. Tamarind is a rich source of minerals like iron, phosphorus, potassium, and calcium. The ripe fruit pulp contains minerals like potassium (62-570 mg/100 g) and phosphorus (86-190 mg/100 g)[5]. Vitamins like thiamin and niacin are high, but vitamins A and C are low[6]. Cumin is a significant spice that is commonly used in our kitchen. Its scientific name is *Cuminum cyminum,* which belongs to the family Umbelliferae. Tamarind was traditionally used as a spice cum herb from an ancient era for treating gastric-related diseases. Many Ayurvedic medicines accommodate cumin as a crucial ingredient. Cumin seeds contain petroselinic acid[7]. Black pepper is scientifically known as *Piper nigrum* and the family *Piperaceae*. Like many spices, it is also used as a seasoning agent and folk medicine. Six sorts of black pepper are there in line the various regions. Black and white peppers are most commonly found in our kitchen. One tablespoon (6 grams) of ground black pepper contains moderate amounts of vitamin K (13% of the daily value DV), iron (10% DV), and manganese (18% DV), with trace amounts of other essential nutrients, protein, and dietary fiber[8]. Piperine is under study for its potential to increase the absorption of selenium, vitamin B_{12}, beta-carotene, curcumin, and other compounds[9]. All age groups of consumers like confectionary products, and to bring the benefits together of the herbs mentioned above, candies are prepared by mixing all the herbs in measured quantities. However, the formulation must be standardized for its better quality, stability and shelf life.

Objectives

- Preparation of Tamarind candy
- Proximate analysis of raw tamarind and candy

MATERIALS AND METHOD

Tamarind, spices and salt are procured from the local market. The pulp was extracted overnight, soaking in water. The method of extraction is mentioned below (Figure 1). Literature was reviewed for the procedure of making candies. The flow diagram is given below for the preparation of tamarind candy using spices and salt (Figure 2).

Table 1: Recipe for candy making

Ingredients	Quantity
Tamarind pulp	25ml
Sugar	125g
Cumin powder	0.234g
Rock salt	0.447g
Black paper	0.125g
Glucose syrup	160ml

Extraction of pulp from tamarind

Tamarind was procured from the market and cleaned. It was soaked in water at a ratio of 1:2 overnight. To obtain pulp, it was correctly mashed and passed through the sieve.

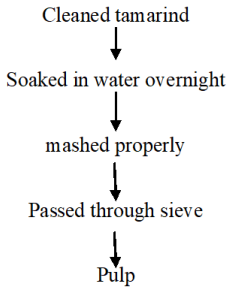

Figure 1: Extraction of pulp

Process of preparation of tamarind candy

Extracted pulp (25 ml) was poured into a heated pan. The sugar was added and cooked for 10 minutes at a constant temperature until the sugar caramelized. Then glucose syrup was added and cooked for 5 minutes. Then rock salt, cumin powder, and black pepper were added in the abovementioned quantity. Mixed thoroughly when the temperature reached 150 ^{0}C, the flame was turned off. The recipe was poured into molds. After cooling, the candy was packed.

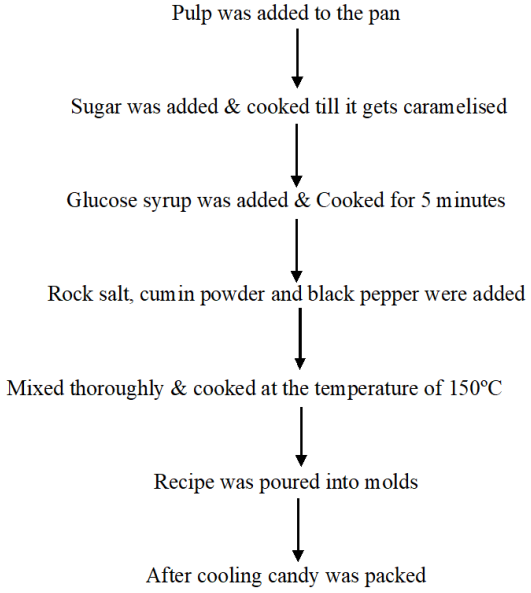

Figure 2: Candy Making

Analysis of the proximate composition of raw tamarind and tamarind candy

Moisture

The standard protocol estimated the moisture content. Approximately 3 gm of the sample was taken in a moisture dish. The sample was put in a hot air oven at 105 ^{0}C for 8 hours. The sample was then cooled at room temperature inside the desiccators, and the residue was weighed. The amount of weight loss is the moisture content. The experiment was performed triplicately, and the mean was recorded[10]. The amount of moisture was measured using the equation:

$$MC\ \% = ((M_2-M_1))/S\times 100 \qquad (1)$$

Where, M_1 =mass of dish with material before oven drying, M_2 =mass of dish with material after oven drying, S= Sample weight

Ash content

The ash percentage of tamarind was measured with the standard protocol. Briefly, 3 gm of the sample was placed into a crucible and heated using a gas flame until the complete charring occurred. After that, the crucible was put in a muffle furnace maintained at 550 ℃ for 8 hours[10]. Ash content was calculated from the inorganic residue as per given formula below:

$$Ash\% = \frac{M_2-M_1}{S}\times 100 \qquad (2)$$

Where, S = weight of the sample, M_1 = weight of empty crucible, M_2 = weight of crucible with ash

Fat content

The standard Soxhlet extraction procedure estimated fat content. Approximately 3 gm sample was kept in a thimble and placed inside the fat extraction assembly of the Soxhlet extractor. Then, approximately 250 ml of petroleum ether was allowed to reflux through the sample inside the Soxhlet machine for 24 hours. At the end of the extraction process, the thimble was detached from the apparatus, and most of the ether was distilled off by recovering it in the Soxhlet apparatus. Once the volume of ether in the Soxhlet flask had reached a small level, it was moved into a reweighed dry beaker[10]. Then, ether was allowed to evaporate by rotator evaporator and weighed.

The crude fat content was measured using the formula given below

$$\text{Fat \%} = \frac{M_2-M_1}{\text{Sample weight}}\times 100 \qquad (3)$$

Where, M_1= empty flask weight, M_2= Flask weight with extracted fat, S. Wt.= Sample weight is taken for analysis

Crude fiber

The crude fiber was estimated by standard protocol. The fat-free sample (3 gm). 200 ml of 1.25% H_2SO_4 was introduced in a conical flask and refluxed for 30 minutes. After that, the content inside the flask was filtered using a Buchner funnel and the residue that remained after filtration was rinsed twice with hot distilled water (50-100 ml). Then, the residue was moved back into the flask, and 1.25% NaOH (200 ml) was mixed to facilitate the digestion with alkali for about 30 minutes. After alkali digestion, the flask's content was recovered using ashless filter paper and rinsed with hot water to remove the alkali residue. The content obtained after washing was placed in a reweighed silica crucible and dried at 105 ℃ for three hours. The weight of the oven-dried crucible was noted, and then it was put in a muffle furnace at 550 ℃ for 3 hours. At last, the weight of the crucible with ash was recorded after cooling it in a desiccator[10]. The crude fiber was determined as the formula mentioned below.

$$\text{Crude fiber (\%)} = \frac{W_1-W_2}{W}\times 100 \quad (4)$$

Where, W = sample weight taken, W1= sample + crucible after drying, W2 = sample + crucible weight after ashing

Carbohydrate percentage

The carbohydrate content of tamarind was calculated using the difference method as mentioned in the standard protocol (AOAC).

$$\text{Carbohydrate (\%)} = [100-(\text{moisture} + \text{ash} + \text{fat} + \text{protein})] \quad (5)$$

Protein content

Protein was measured by the Kjeldahl method with Kjeldahl digestion (Buchi-Kjeldahl digester kn-449) and titration unit (Kjel Flex k-360). About 1 gm of sample along with digestion mixture of catalyst (K_2SO_4:$CuSO_4.5H_2O$ in ratio 4:1) and 20 ml of conc. H_2SO_4 was taken in the Kjeldahl digestion tube. The sample was placed at 389 ^{0}C for 4 hours unit the solution became green or colourless. A blank sample was used comprising only a catalyst mixture of K_2SO_4. $CuSO_4$ and H_2SO_4. After digestion, the sample was allowed to cool at 40-50 ^{0}C, followed by stem distillation for 3 minutes using 40% NaOH (90 ml). Then, released ammonia was retained in 50 ml of 4% boric acid solution comprising 1-2 drops of mixed indicator (0.1% methyl red + 1% bromocresol green in 2:1). The obtained solution of boric acid was titrated using 0.1 N estimated by using the formula given below[10]

$$\% \text{ Protein} = N \times \text{conversion factor}$$

Total phenolic content and antioxidants analysis

Sample preparation (extraction)

The tamarind sample of 10 gm was combined with 60% methanol (100 ml). The extraction was done using a shaking incubator at 36 ℃ at 150 rpm for 12 hours. Later, the extra methanol was evaporated by using a rotator evaporator. The extract was stored at -20 ℃ to estimate TPC and antioxidant capacity.

Total Phenolic Content

TPC was measured using the procedure Folin–Ciocalteu (fc). The sample extract (10 mg) was added to the Fc reagent (500 µl) and then combined with 6 ml distilled water. After mixing the content of the sample for 1 minute, 10 ml of distilled water was used to make the volume and incubated for 2 hours. Readings were taken at 750 nm using a UV-visible spectrophotometer[11]. The data is given as milligram gallic acid equivalent (mgGAE/gm db.).

Total Flavonoid Content

The total flavonoid content of ethanol extract was evaluated with the standard method. In a test tube, 10 mg of extract was taken, and 1ml methanol was added. Then 200 µl of 10% aluminium chloride solution was added into the same test tube, followed by 200 µl of 1M potassium acetate. At last, 5.6 ml of distilled water was mixed with the reaction mixture. The reaction mixture was then incubated for 30 minutes at room temperature to complete the reaction. Then, the absorbance of the solution was measured at 415 nm using a spectrophotometer against a blank. Methanol was used as a blank[11]. The flavonoid compounds in tamarind extract were expressed in mg/g quercetin equivalent (QE).

Antioxidant potential using diphenyl-1-picrylhydrazl [DPPH]

DPPH inhibition percentage was measured as per protocol. The DPPH reagent (3.9 ml) was combined with sample extract (0.1 ml), and the resulting solution was incubated in the dark for 30 minutes. Then, sample absorbance was observed at 517 nm against methanol as blank. A control sample containing 3.9 ml of 0.1 mM DPPH solution and 0.1 ml of methanol was also prepared, and measurements for the same sample were taken immediately at 517 nm. The scavenging activity of the DPPH radical was examined by using the formula as follows:

$$\% \text{ of Inhibition} = (\text{absorbance for control} - \text{sample absorbance}/\text{absorbance for control}) \times 100 \quad (6)$$

Antioxidant potential as 2,2 azino-bis-3-ethyl-benzothiazoline-6-sulphonic acid (ABTS)

ABTS was carried out as per the procedure. ABTS cation radical was made by reacting 2.45 mM potassium persulfate and 7 mM ABTS (1:1) in water and stored in dark conditions for 12-16 hours before analysis. The ABTS cation was mixed with methanol (1:60) to obtain an absorbance reading of 0.70±0.02. Then, 2 ml of ABTS was mixed with 20 µl of tamarind extract, and absorbance values were taken after 6 minutes at 734 nm wavelength range[12]. The result of the ABTS assay was displayed as mMTE/100g db.

FRAP

The reducing power of tamarind extract was determined according to standard procedure. 10mg/ml methanol concentration of tamarind was mixed with 2.5 ml of phosphate buffer [0.2M, pH 6.6] and 2.5 ml of potassium ferricyanide solution (1%) after incubation in the water bath at 50 ℃ for 30 minutes. 2.5 ml of 10% trichloroacetic acid was added to the mixture and centrifuged at 3000 gm for 10 minutes to stop the reaction. The supernatant (2.5 ml) was mixed with 2.5 ml distilled water and 0.1% $FeCl_3$ (0.5 ml), and at last, the absorbance was measured at 700 nm using a spectrophotometer. Higher absorption of reaction suggests a necessary reducing power. Trolox was used as positive control[13].

Water activity

The water activity of the tamarind sample was evaluated with a hand-operated portable digital water activity meter (Aqualab, USA). Tamarind sample was filled 70% in a plastic moisture-free cup provided along with the instrument and placed into the sample holder until the instrument sensor detected the water activity value. Water activity was estimated from the average of triplicate readings.

Titratable acidity

The ascorbic acid content in tamarind was determined with the N/10 NaOH visual titration method. A 5 gm sample was weighed and mixed with 100 ml of distilled water, appropriately stirred and then filtered using muslin cloth. Filtered juice was made up of 100 ml of distilled water. N/10 NaOH was filled in the burette, then 10 ml of volume was taken from the filtered juice. 2 ml of phenolphthalein indicator was added and titrated against N/10 NaOH filled in the burette till the colour changed to pink. A lower meniscus reading was noted[14]. The experiment was performed in triplicates, and the mean was measured using eq.

$$\text{Acidity} = \frac{\text{Titration volume} - \text{Eq.wt of acid} \times \text{Normality} \times 100}{\text{Weight of sample} \times 1000} \quad (7)$$

Vitamin C

The ascorbic acid content in tamarind was determined with the 2,6 dichlorophenol Indophenol (DCPIP) visual titration method. The DCPIP dye as an indicator, which is blue in an alkaline solution and red in an acidic solution, is reduced by ascorbic acid to a colourless form. About 10ml of 3% metaphosphoric acid extract of tamarind (filtrate) was titrated with DCPIP dye to the pink colour endpoint (persist 15 sec). The ascorbic acid content was calculated using the dye factor, determined by titration of standard ascorbic acid solution (dissolve ascorbic acid in 3% HPO3) with DCPIP dye[14]. Ascorbic acid expressed as mg/100 ml of tamarind was calculated using the following eq.

Dye Factor = 0.5/titer value

$$\text{Ascorbic acid (mg/100 ml)} = \frac{Titration\ volume \times Dye\ factor \times Volume\ madeup \times 100}{Aliquot \times volume\ of\ juice} \quad (8)$$

Reducing sugar

Tamarind sample 10 mg weighed accurately. A sample was mixed with 10ml methanol. 100 µl Aliquot of the sample, mixed with DNS reagent (3 ml). After that, heat the solution for 5 minutes at 100 ℃. 1 ml Rochelle salt and 2.9 ml distilled water

were added in the same test tube. After the sample cooled down, absorbance was measured at 540 nm, and a reading was noted[15].

Microbial examination of tamarind candy

i) Total bacterial count

The dilution plate method was followed to estimate bacterial populations in different samples (IS 4502). 9 ml of sterile saline water was mixed thoroughly with 1 gm of the sample; from this, 1 ml of the sample was transferred through a sterile pipette to a tube with a screw cap containing 9 ml sterile saline water, which gave a dilution of 10^2. Similarly, serial dilutions were made. 1 ml of sterile serially diluted sample was placed in the sterile petri dish to which cooled plate count agar medium was added, mixed thoroughly with the suspension, and then set to set after that incubated at 28±2 ^{0}C for 48 hours. Individual colonies were counted and multiplied[16].

ii) Yeast and moulds

The dilution plate method was followed for yeast and mould content (IS 4503). For estimating the fungal population, yeast extract dextrose-chloramphenicol Agar medium was used[17].

RESULTS

Table 2: Proximate analysis result of raw tamarind

Parameters	Number of trails			Statistical reading	
	T_1	T_2	T_3	Mean	S. D
Moisture % (w/w)	26.298	23.216	27.458	25.658	2.192
Ash % (w/w)	5.021	3.275	4.689	4.328	0.927
Fat % (w/w)	7.731	6.487	6.061	6.760	0.867
Crude fiber	12.150	14.056	14.056	12.168	1.878
Water activity (a_w)	0.568	0.565	0.547	0.560	0.0113
Titratable acidity	7.350	8.200	15.750	10.433	4.623
Vitamin C	22.200	22.200	22.200	22.200	0

Table 3: Proximate analysis result of tamarind candy

Parameters	Number of trails			Statistical reading	
	C_1	C_2	C_3	Mean	S. D
Moisture %(w/w)	1.549	1.763	1.534	1.616	0.128
Ash %(w/w)	2.760	2.835	2.771	2.789	0.040
Fat %(w/w)	1.050	0.699	0.434	0.728	0.308
Crude fiber	1.053	1.112	0.750	0.973	0.192
Protein % w/w	--	--	--	0.60	--
Carbohydrates	--	--	--	94.267	--
Water activity (a_w)	0.481	0.508	0529	0.506	0.024
Vitamin C	39.600	37.740	26.64	34.660	7.007
Titratable acidity (% Sorbic acid)	1.050	1.350	1.35	1.200	0.150

Table 4: Phytochemical content and antioxidant analysis results of tamarind

Parameters	Number of trails			Statistical reading	
	T_1	T_2	T_3	Mean	S. D
TPC	37.536	38.412	37.243	37.732	0.610
TFC	1.008	1.0018	1.008	1.011	0.005
DPPH	11.198	11.222	11.257	11.226	0.029
ABTS	91.965	91.965	91.989	91.973	0.013
FRAP	76.120	76.096	76.226	76.147	0.064
Reducing sugar	0.724	0.665	0.665	0.685	0.03

Table 5: Phytochemical content and antioxidant analysis results of tamarind candy

Parameters	Number of trails			Statistical reading	
	C_1	C_2	C_3	Mean	S. D
TPC	44.574	45.747	45.454	45.259	0.619
TFC	0.566	0.576	0.566	0.569	0.005
DPPH	29.655	29.665	29.702	29.672	0.024
ABTS	72.285	72.285	72.265	72.277	0.013
FRAP	91.648	91.648	91.648	91.648	0
Reducing sugar	0.5811	0.5811	0.5803	0.580	0.004

Table 6: Phytochemical content and antioxidant analysis results of raw tamarind and tamarind candy

	TPC	TFC	DPPH	ABTS	FRAP	Reducing Sugar
Raw Tamarind	37.732±0.610	1.011±0.005	11.226±0.029	91.973±0.013	76.147±0.064	0.685±0.03
Tamarind Candy	45.259±0.619	0.569±0.005	29.672±0.024	72.277±0.013	91.648±0	0.580±0.004

Table 2 highlights the proximate analysis of raw tamarind. The moisture % of raw tamarind was 25.658%. Ash percent was 4.328%. The fat percentage was 6.760%. The crude fiber was 12.168. The water activity of raw tamarind was 0.560. Titratable acidity was 10.433. The vitamin C content of raw tamarind was 22.200. Table 3 highlights the proximate analysis of tamarind candy. The moisture percentage of tamarind candy was 1.616%. Ash percentage was 1.616%. The fat percentage of tamarind candy was 0.728%. The crude fiber for tamarind candy was 0.973. Protein percentage was 0.60. Carbohydrates for tamarind candy was 94.267%. Water activity for tamarind candy was observed at 0.506. The titratable acidity of tamarind candy was 1.200. The vitamin C content of tamarind candy was 34.660.

Data for the antioxidant analysis of raw tamarind is summarised in Table 4. Data shows the presence of phenols and flavonoids in the extract, and the amounts are 37.732 ± 0.610 mg gallic acid/g and 1.011 ± 0.005 mg quercetin/ g, respectively. In Tables 4 and 5, the results of antioxidant activity assays are 11.226±0.029, 91.973±0.013, 76.147±91.648 for tamarind and 29.672±0.024, 72.277±0.013, 91.648±0 for tamarind candy DPPH, ABTS and FRAP respectively. Table 4 and 5 highlights the result of reduced sugar of raw tamarind was 0.685±0.03, and reduced sugar of tamarind candy was 0.580±0.004. Here, the reduced sugar for raw tamarind is more than in tamarind candy. It may be because of the cooking at higher temperatures.

Chart 1 shows the growth of total plate count in the sample. The total plate count content in fresh tamarind candy was less than 10, and on the 5th, 10th, 15th, and 20th days, the total plate count was 718, 1827, 3000 and 4636 cfu/g, respectively. Table 7 indicates the growth of yeast and mould in the given sample is insignificant; it's almost the same for the duration of storage.

Table 8 reflects the effect of storage time on sensory attributes and water activity of the tamarind candy sample. The colour of the sample was visually observed, and it was found that there was no difference in colour during accelerated storage conditions. Characteristics odour of the sample was absorbed during the storage period of the sample. The water activity of fresh candy was 0.344, and the result indicates that the water activity was decreased as per the long storage of the sample. The least water activity of candy was observed on the 20th day of storage, which was 0.287.

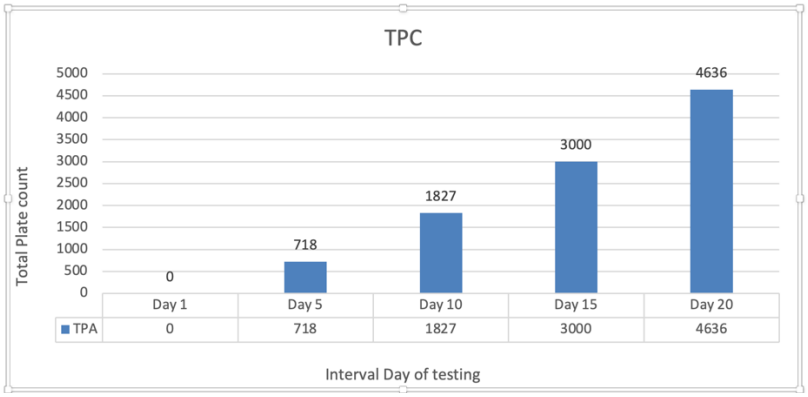

Chart 1: Total plate count in tamarind candy

Table 7: TPC and Yeast & mould count in tamarind candy

Testing Parameter	UOM	Test Method	Test Result Day-1	Test Result Day-5	Test Result Day-10	Test Result Day-15	Test Result Day-20
Total Plate Count	CFU/gm	IS 5402	<10	718	1827	3000	4636
Yeast & Mould	CFU/gm	IS 5403	<10	<10	<10	<10	<10

Table 8: The effect of storage time on the sensory attribute of tamarind candy

Testing parameter	Testing method	Observation				
		Day 1	Day 5	Day 10	Day 15	Day 20
Odour	Organoleptic	Characteristics	Characteristics	Characteristics	Characteristics	Characteristics
Colour	Visual	Dark brown colour	Dark brown colour	Dark brown colour	Dark brown colour	Dark brown colour
Water activity at 26 ℃	Water activity meter	0.3445	0.3284	0.3060	0.2974	0.2875

DISCUSSION

More than two methods should be used to determine the antioxidant capacity of food. It is important to determine by a mixture of different antioxidants with different action mechanisms, among which synergistic interactions. In the present work, we selected three oxidant systems that measured colour disappearance with free radical DPPH, ABTS, and FRAP. These assays are based on the scavenging of radicals (DPPH, ABTS, and FRAP), which convert it to a colourless product. The degree of this discolouration affects the amount of DPPH, ABTS, and FRAP that has been scavenged. Microbial and sensory analysis of tamarind candy was done using the accelerated shelf-life method, in which the product was stored under elevated stress conditions (such as humidity, pH, and temperature). This research stored a sample of candy at 48 ℃, and analyses were done at regular 5-day intervals for 20 days. The overall acceptability of tamarind candy after 20 days of accelerated storage condition was good, and as per the calculation of the accelerated ageing test, it indicates that at ambient temperature, the shelf-life of the sample is more than three months.

CONCLUSION

The results of the physicochemical analysis showed that the tamarind candy contains low moisture percentage and less water activity, preventing the growth of food spoilage bacteria. The resulting high amounts of vitamin C and tamarind candy's antioxidant values will help acquire the minerals and health benefits associated with nutritional deficiency. The tamarind candy is stored without any deterioration in sensory attributes and microbial count and is acceptable for up to three months of storage. Hence, it can be said that blending with different spices makes the candy not only palatable but also good from a nutritional and health points of view.

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