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Research Article

Comparison of Chemically Treated, Pasteurized, and Microwave-Treated (at Different Time Durations) Chia Seeds Added To Mango-Whey Beverage, during Different Storage Periods, for Physicochemical and Sensory Parameters

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Foods that are widely consumed and accepted have had a lot of ingredients added to them which may enhance consumer's health. Whey is a byproduct in the manufacture of cheese or curd and is widely used in different beverage formulations, due to its nutritional importance. In this context, a mango-based functional whey drink, added with omega-3-rich chia seeds, was developed. Different treatments of beverage were as follows: untreated control beverage (T_0), application of chemicals (T_1), pasteurization (T_2), and microwave heating (at 4 different time lengths: T_3, T_4, T_5, and T_6). Beverages were analyzed for physiochemical, microbiological, and sensorial changes, during the 90 days of storage. Chemical analysis of chia seeds before incorporation in beverages revealed a high nutritional profile of chia seeds, especially the presence of essential fatty acids. Significant variation in color parameters of the beverage was observed as a result of treatments and storage, with optimum values observed for T_3. A decrease in pH and an increase in acidity during storage for all treatments were evident, with the most significant results for microwave treatment for longer time periods. The total plate count of the beverage was the highest (2.36 ± 0.1 CFU/ml) for control, followed by chemically treated (2.17 ± 0.1 CFU/ml) and lowest in microwave-treated (0.98 ± 0.1 CFU/ml) at the start of the experiment, and this total plate count was found to be increasing in all treatments during storage. Total solids were increased and soluble solids were decreased during longer microwave treatments and also during storage of all treatments. The most acceptable treatment T_3 (85 ml whey, 2 g chia seed, and microwave heating for 30 sec) was further subjected to a storage study and it was observed that the sensory scores gradually decreased during the 90 days of storage. From the outcomes of the study, it was concluded that microwave treatment of the beverage for 30 sec, as compared to longer durations and chemical preservatives, was proved helpful for optimum quality retention of the formulated beverage, with minimum deteriorating effects. Thus, proper treatments of different beverage formulations, following the necessary protocols, could provide the consumers safe, healthy, and nutritious beverages.

1. Introduction

In the food sector, there is now considerably more emphasis on the production of food products with various bioactive components. The creation of these multifunctional meals satisfies the expectations of the targeted customers. Given the ubiquity of whey as the most palatable drink with nutritional components, the creation of such a drink with functional food ingredients appears to be fascinating [1]. Application of different novel processing technologies has been proven helpful in enhancing the functionality and nutritional contents of beverages [2].

Whey, a byproduct of the cheese-making process, is the primary component of whey drinks and has been employed for product development for the past 20 years. Whey provides several nutritional benefits since it contains a variety of bioactive substances [3]. After preparation, whey still contains lactose, minerals, water-soluble vitamins, and bioactive whey proteins [4]. Although whey has been utilized in the food sector for approximately twenty years, attention to its utilization has lately grown due to its strong nutritional and functional qualities [5]. Milk sugars, proteins, minerals, fat-soluble and water-soluble vitamins, polyphenols, and water, each component of whey have different medical advantages [6].

The phytochemistry of fruits and vegetables gives us the foundation for combining their constituent parts to create value-added products that are brimming with antioxidants and antimicrobial elements that prevent pathogen invasion and free radical production, respectively [7, 8]. Mango (*Mangifera indica* L.), a tropical fruit, has been well known among the most eaten fruits on the planet. It swiftly and significantly softens as the fruit gets closer to becoming fully ripe [9]. Mango is one of the greatest fruits, because of its alluring scent, delectable taste, superb flavor, and high nutritional content [10]. Mango juice, nectar, and squash are the three significant refreshments of mango fruit, which contain nutritional components [11], and thus, consumption of mango-based beverages could prove helpful in mediating the normal body functions.

Fruit components such as seeds, peels, pulp, concentrate, and juices serve as the foundation for creating food formulations that increase the nutritional content of finished goods [7, 12]. *Salvia hispanica* L. is the scientific name for chia, an herb plant that is a member of the *Lamiaceae* family. The health advantages of chia seeds to people are thought to be related to their greater levels of alpha-linolenic acid [13]. Some common food products such as cakes, pasta, biscuits, and bread which have been developed by using chia seeds can be found in the markets [14]. Stirred yogurt was prepared by using different doses of chia seeds [15]. Kowaleski et al. [16] used chia seeds and strawberries for the formation of yogurt. Chia seed extract in whey drink were found in the studies of Kwon et al. [17].

Large-scale foods and beverages frequently contain benzoates and sorbates as preservatives since they work as antibacterial and antifungal agents and have very low toxicity for mammals. They are universally acknowledged to be inherently free of carcinogenicity. The Joint FAO/WHO Expert Committee on Food Additives (JECFA) has investigated the safety of these compounds [18, 19]. However, ultrasonic and microwave processing have lately garnered significant potential in food applications as green processing technologies [20]. Microwaves are electromagnetic waves that range in frequency from 300 MHz to 1000 GHz. Microwaves heat up quickly and produce better results [21]. The use of microwaves has been shown to be beneficial in preserving the optimum amount of nutritious ingredients and eliminating any bacteria found in treated juices [22, 23]. Microwave processing could be applied as a good technique that utilizes less energy and time [24]. Although the reviewed literature provided a number of processing and preservation experiments, conducted on fruit juices and beverages, there is a lack of study about the whey beverage formulations added with chia seeds and mango pulp. This unique and novel recipe has been tested in the current study, which could enrich the market with a brand-new product, the consumers wish to take. Microwaved preservation of this beverage could minimize the food safety concerns of the food business operators. Keeping in view the valorization potential of whey, the nutrition and delicacy of mango, and the health-promoting potential of chia seeds, the present research project was designed to develop innovative and functional whey beverage formulations, enriched with mango pulp and chia seeds, and to investigate the different physicochemical, sensory, microbial, and storage parameters of beverage under different processing and preservation treatments.

2. Materials and Methods

2.1. Procurement of Raw Materials. Raw materials for beverage development were chia seeds and mangoes, which were purchased from the local market of district Sargodha, Pakistan. Other ingredients of the beverage such as sugar and guar gum were purchased from Imtiaz Super Market, Sargodha district, Pakistan, whereas two chemical preservatives used (sodium benzoate and potassium metabisulfite) were purchased from Sigma Chemical Store, Islamabad, Pakistan. Whey was collected from Fauji Foods Ltd., Bhalwal, Pakistan, and mango-whey beverage was prepared in laboratories of the Institute of Food Science and Nutrition, University of Sargodha. Before the onset of the experimental study, whey and mango pulp were handled under aseptic conditions to avoid any microbial contamination and spoilage.

2.2. Recipe Development and Treatment Plans. A recipe with different ingredients, of mango-whey beverage, was adopted from the earlier experiments of Alane et al. [25] and is presented in Table 1. Different treatments were T_0, T_1, T_2, T_3, T_4, T_5, and T_6, and these treatments, except control, were applied in chemical preservation, pasteurization, and different times of microwave heating as presented in Table 1. A microwave oven (MRO-AV200E, Hitachi, Japan) was used to microwave the beverages (200 ml in a 500 ml beaker) at 90°C and 400 W, as was reported earlier by Zia et al. [26]. The

Table 1: Treatment plan with recipe and preservation techniques.

Treatment	Whey (mL)	Chia seeds (g)	Sugar (g)	Mango pulp (g)	Guar gum (g)	Variables
T_0	85	2	8	5	0.1	N/A
T_1	85	2	8	5	0.1	Sodium benzoate 0.25 g and potassium metabisulfite 0.75 g
T_2	85	2	8	5	0.1	Pasteurization (85°C for 15 min)
T_3	85	2	8	5	0.1	Microwave heat (30 sec)
T_4	85	2	8	5	0.1	Microwave heat (60 sec)
T_5	85	2	8	5	0.1	Microwave heat (90 sec)
T_6	85	2	8	5	0.1	Microwave heat (120 sec)

abovementioned microwave treatment was employed to treat juices because of its special features, including thermostatic control, temperature resistance up to 120°C, a working temperature range of 25°C to 100°C with an accuracy of 0.3°C, and a 220 Volt/50 Hz electric supply. A double wall pasteurizer (Harvest Hi-Tech Equipment Pvt. Ltd, Coimbatore, India) was used for the pasteurization process, and it was heated to a maximum temperature of 85°C using water as the heating medium. The temperature probe was used to keep an eye on the beverage's temperature when it was moved to the pasteurizer once it had reached equilibrium. Pasteurization was performed by following the procedure of Prithviraj et al. [27]. By following the guidelines of Pandiselvam et al. [28], to find out the impact of different preservation techniques on the physicochemical and microbiological qualities during storage duration, aseptically packed beverage samples were kept at a refrigeration temperature of 5 ± 2°C for 3 months.

2.3. Preparation of the Mango-Based Whey Beverage Added with Chia Seeds. For incorporation in mango-based whey beverage, first of all, chia seeds were soaked in water. After that, whey and mango pulp were mixed together in a juice blinder, and sugar was added to the mixture and ground again. At the end, chia seeds were added to the beverage. Then, the beverage was subjected to different microwave heat treatments, pasteurization, and chemical preservation. The beverage prepared was filled in bottles with labels and kept chilled at 5 ± 2°C. For the development of the formulated beverage, guidelines from the studies of Tanwar et al. [29] were taken and adjusted accordingly.

2.4. Analysis of Chia Seeds

2.4.1. Proximate Composition. Using the Association of Official Analytical Chemists' procedures, the crude fiber, moisture, ash, fat, nitrogen-free extract (NFE), and protein contents of chia seeds were calculated by following their respective methods of AOAC [30], with required modifications. The NFE percentage of chia seeds was analyzed by subtracting the percentage of crude fiber, moisture, fat, crude protein, and ash from 100, as indicated in the NFE value given as

$$\text{NFE}\ (\%) = 100 - (\text{moisture}\% + \text{ash}\% + \text{crudeprotein}\% + \text{crudefiber}\% + \text{crudefat}\%). \tag{1}$$

Before incorporation in mango-whey beverage, the proximate composition of chia seeds was determined, which is shown in Table 2. These results revealed that the percentage of fiber, fat, and protein was high in chia seeds. The high value of fat was attributed to the higher percentage of fatty acids in chia seeds, while the lower amount of moisture showed the lower perishability of chia seeds. These findings were found in line with those of Otondi et al. [31] Rodríguez Lara et al. [32], and Aamer et al. [33].

2.4.2. Fatty Acid Composition. The method used by Aamer et al. [33] was adjusted to ascertain the fatty acid composition of chia seeds. After being ground to 100 g, the chia seeds were extracted three times using 0.5 L of n-hexane at 60°C for four hours. A rotavapor apparatus was then used to mix and concentrate the extracted materials. By mixing 1 mL of Hex and 1 mL of a 2 N methanolic potassium hydroxide solution with 100 μL of the resultant oil, stirring for 15 seconds, and letting the mixture sit at room temperature for five minutes, fatty acid methyl esters were produced. The upper layer, which included methyl esters of fatty acids, was removed and kept at −20°C for additional examination of fatty acids. The fatty acid composition of chia seeds was investigated by gas chromatography (GC) (7820A, Agilent, Santa Clara, CA, USA), coupled with a FID and a Trace TR-FAME capillary column (i.d. 0.25 μm, 60 m × 0.25 mm, Thermo Fisher Scientific, Grand Island, NY, USA). The analysis condition was programmed as follows: the injector and FID temperatures were kept at 250°C and the initial oven temperature was held at 80°C for 3 min. It was then increased to 215°C (15°C/min) and finally up to 215°C (20 min hold time). Nitrogen was used as carrier gas at 1 mL/min, with a split ratio of 1 : 20. The chromatogram of the authentic fatty acids was used to characterize the fatty acids according to their retention times. The fatty acid composition was expressed as a percentage of the total fatty acids.

The results of the analysis of the fatty acid composition of chia seeds are shown in Table 3, from where it was revealed

Table 2: Proximate analysis of chia seeds.

Chemical parameters	Quantity (%)
Moisture	5.67 ± 0.08
Ash	3.98 ± 0.12
Fat	35.66 ± 0.09
Protein	25.01 ± 1.25
Fiber	26.92 ± 2.32
NFE	2.78 ± 0.05

Values are presented as means of triplicate analysis, along with standard deviations. NFE, nitrogen-free extract.

Table 3: Fatty acid composition of chia seeds.

Fatty acid compounds	Values (%)
Linolenic acid	51.15 ± 0.70
Linoleic acid	25.42 ± 0.30
Oleic acid	7.88 ± 0.40
Palmitic acid	7.63 ± 0.30
Mysteric acid	8.11 ± 0.19

Values are presented as means of triplicate analysis, along with standard deviations.

that the major unsaturated fatty acids that were present in chia seeds were linolenic acid, linoleic acid, and oleic acid. Some long-chain saturated fatty acids were also present in chia seeds, and these were palmitic and myristic acids. This fatty acid composition of chia seeds was also supported by earlier studies of Kulczynski et al. [34], Hernandez-Pérez et al. [35], and Aamer et al. [33].

2.5. Analysis of the Formulated Beverage

2.5.1. Color Analysis. Using a ColorQuest XE (HunterLab), color assessments of all the treatments of mango-whey beverages were carried out. The colorimeter has a 10° angle of view with specular reflection and a D65 illuminant. The values of color parameters, including a^*, b^*, and L^*, were established by following the procedures adopted by Siefarth et al. [36], with slight modifications. Briefly explaining, before sample measurement, the equipment was first standardized against white and black tiles. The color properties were then determined in triplicate, and the averages of the results were recorded.

2.5.2. pH Analysis. Using an electronic digital pH meter (Inolab pH 720, WTW 82362), the mango-whey beverage's pH was estimated by following the protocols adopted by Siefarth et al. [36]. First of all, the pH meter was calibrated with a buffer solution of pH 4 and 7. Then, an appropriate amount of beverage was taken as a sample and was put in the beaker. The electrodes of the pH meter were placed in the sample to check the pH, and all analyses were repeated in triplicate to find out the mean values.

2.5.3. Determination of Titratable Acidity. The titratable acidity of mango-whey beverage was determined by following the procedure used by Dong et al. [37], with slight changes. Briefly explaining, a sample of 9 ml of mango-whey beverage was placed in a titration flask and then phenolphthalein (2-3 drops) was added to it. This mixed solution was titrated against 0.1 N NaOH until it turned into a light pink color. The titratable acidity of mango-whey beverage was determined with the volume of NaOH used, and the below given formula was used to calculate the acidity %.

$$\% \text{ Titrable acidity} = \frac{0.009 \times \text{Vol. of NaOH used}}{\text{weight of the sample}} \times 100. \tag{2}$$

2.5.4. Determination of Total Solids. To determine the total solids of formulated beverages, a known amount of mango-whey beverage samples was placed in a crucible and relocated to a hot air oven for drying. The content of total solids in mango-whey beverage was determined by drying the samples to a constant weight at 105°C, overnight, using a heating oven by adopting the procedure given in AOAC [36], with slight modifications.

$$\text{Total solids}\,(\%) = \frac{\text{Weight of dried sample}\,(g)}{\text{Weight of sample}} \times 100. \tag{3}$$

2.5.5. Analysis of Viscosity. The samples of the mango-whey beverage were conditioned at 4°C before the viscosity was measured. The viscosity of the beverage samples was measured by using a Lamy Rheology Instruments' rotator-type viscometer. All mango-whey beverages with chia added had their apparent viscosity measured, with minor modifications, using the technique outlined by Cheong et al. [38]. Spindle number 5 was used to measure viscosity, and 50 rpm was the speed setting. The viscosity data were given in centipoises (cP).

2.5.6. Calculation of Brix. Since the lab refractometer had an upper number of 30°, the refractometer was first calibrated to 0° Brix using 1 ml of distilled water, and then, each sample (5 ml) was diluted with 10 ml of water to get a ratio of 1 : 2 before measuring the brix, and values were recorded by adopting the procedure used by Nduko et al. [39]. Briefly describing, with the help of a micropipette, a drop of the sample was placed on the detector of the refractometer and the value shown on the screen was noted. Calibrating the refractometer after each step, the same process was repeated thrice to find the mean value.

2.6. Total Plate Count. Thermo Fisher Scientific Inc.'s Oxoid™ plate count agar was used for the microbiological study of beverage samples. The inoculum was mixed into the plate using the pour plate technique, and the agar and inoculum were then united by gently rotating the plate. After that, incubation was performed at 37°C for 48 hours. The total plate count testing was employed for bacterial evaluation for the safe storage duration criteria; however, it was examined every hour for the next eight hours. Colony-forming units per milliliter (CFU/ml) would be the definition of total plate count, and mold and yeast count, and guidelines were taken from the experiment of Amelia et al. [40].

$$\text{Total Plate Count (TPC)} = \frac{\text{Average numbers of colonies} \times \text{dilution factor}}{\text{volume TPC was expressed as CFU/ml}}. \tag{4}$$

2.7. Sensory Evaluation of Different Treatments of Formulated Beverages. Sensory evaluation of all treatments of mango-whey beverage was first carried out before storage, to find out the most acceptable treatment, which was then further analyzed through a sensory panel of experts, at the end of each month, during the 90 days of storage. The Institute's assessors used a nine-point hedonic scale to evaluate the sensory qualities such as color, flavor, texture, and taste and overall acceptability. The sensory assessment was planned for 80 participants, as was earlier reported by Lawless and Heymann [41], as they suggested 70 to 100 expert people for the sensory evaluation test. Participants were given 150 ml of iced beverage in transparent cups during the test, which was conducted in separate booths. Scores were calculated by using a nine-point hedonic rating system. The means and standard deviations of the data were displayed.

2.8. Statistical Analysis. The current study's treatments were all administered a total of three times before a statistical analysis was conducted. The data were translated into means and standard deviations using a statistical analysis. The experimental data were processed using SPSS 22.0 (SPSS Inc., Chicago, IL, USA) and SigmaPlot 12.0 Statistical Analysis software (Systat Software, Inc., San Jose, CA, USA) to indicate the variables with statistically significant effects ($P < 0.05$). Every experiment was run in a different order, and the model was able to fit the data from the experiments. Following Steel and Torrie's [42] conventional techniques, an analysis of variance (ANOVA) was carried out.

3. Results and Discussion

3.1. Color Profile of the Mango-Whey Beverage

3.1.1. L^* Value. The L^* value indicates the darkness to the whiteness of any food material. The mean values of L^* for treatments and storage are shown in Table 4, and the values were found to be decreasing with increasing the time of microwave heat treatment for formulated beverages. A decrease in the values of L^* may be due to the Maillard reaction that might have occurred during the heat treatment [43]. In addition, polyphenols also polymerize during heat treatments, producing new compounds with dark colors, which are similar to Maillard reaction products [44]. Another reason could be the addition of chia seeds, which might have shifted the color of the beverage towards dark. The whiteness of beverages also decreased during the storage, as there was a decrease in the L^* value at the time of storage. The decrease in the whiteness during storage may be due to the changes in the composition of the drink [22]. So the treatments and storage time have a highly significant effect on the L^* value of the whey drink.

Comparing current experimental results with previous findings, it was observed that there was a decrease in the color of the formulated foods during storage and upon heat treatment. The study was conducted to evaluate the effects of radio frequency heating on the properties of stirred yogurt [36], and high frequency and prolonged treatment times were found involved in decreasing the L^* value of the yogurt. Results of the current study were also compared with the color values provided by another study of Buse et al. [45], which showed similar findings, witnessing the significant effect of different microwave heat durations on the colors of the treated beverages. A slight decrease in the L^* value in all treatments of chia seeds added to mango beverage was also reported by Aamer et al. [33], and these results agreed with the present ones.

3.1.2. a^* Value. The a^* value indicates the redness to the greenness of the food items, and the mean values of the a^* parameter of mango-based chia seeds-added whey beverage processed under different conditions are shown in Table 5. From the experimental results, it was observed that beverages having a short time of microwave exposure showed higher positive values of a^*, which were also significantly affected during the storage study, and were found in a decreasing mode, with the passage of storage time. This decrease in values of a^* may be attributed to the heat treatment by microwave and pasteurization, which might have produced the brown-colored compounds [22]. Experimental outcomes witnessed that a^* values were significantly affected by treatments and storage time. When the results of the current study were compared with previous findings of Siefarth et al. [36], a similar decreasing pattern of a^* parameter was observed during storage of yogurt treated with different radio frequencies and durations. Current experimental findings about color parameters affected by processing and storage conditions were also compared with the results of the study by Buse et al. [45], conducted on drinks added with powdered chia seeds and processed under different conditions. Gehlot et al.

Table 4: Mean value for L^* parameter of mango-whey beverage developed with different processing and preservation techniques.

Treatments	Storage days				Mean ± SE
	0	30	60	90	
T_0	79.10 ± 0.2^a	77.39 ± 0.4^b	75.02 ± 0.2^c	70.02 ± 0.1^e	77.38 ± 0.2^A
T_1	77.39 ± 0.4^b	75.61 ± 0.4^c	75.47 ± 0.3^c	71.21 ± 0.3^{de}	75.96 ± 0.3^B
T_2	75.38 ± 0.1^b	74.89 ± 0.2^b	73.15 ± 0.1^c	72.29 ± 0.1^{cd}	73.93 ± 0.1^C
T_3	73.16 ± 0.2^c	72.53 ± 0.2^{cd}	71.82 ± 0.1^{de}	72.13 ± 0.2^{cd}	72.41 ± 0.2^D
T_4	70.93 ± 0.2^e	68.86 ± 0.2^f	67.56 ± 0.1^g	63.34 ± 0.2^i	67.67 ± 0.2^E
T_5	64.82 ± 0.2^h	61.34 ± 0.1^j	59.21 ± 0.1^k	58.86 ± 0.2^k	61.06 ± 0.2^E
T_6	59.87 ± 0.6^k	58.72 ± 0.4^l	57.76 ± 0.3^m	56.67 ± 0.2^{mn}	58.87 ± 0.2^F
Mean ± SE	72.32 ± 0.2^A	70.65 ± 0.1^B	69.44 ± 0.2^C	68.40 ± 0.2^D	

*Each mean value within the same column, followed by different letters is highly significant at $P < 0.05$. T_0 = control; T_1 = 85 ml whey, 2 g chia seed, 0.25 g sodium benzoate, and 0.75 g potassium metabisulfite; T_2 = 85 ml whey, 2 g chia seed, and pasteurization; T_3 = 85 ml whey, 2 g chia seed, and microwave for 30 sec; T_4 = 85 ml whey, 2 g chia seed, and microwave for 60 sec; T_5 = 85 ml whey, 2 g chia seed, and microwave for 90 sec; T_6 = 85 ml whey, 2 g chia seed, and microwave for 120 sec.

Table 5: Mean values for a^* parameter of mango-whey beverage developed with different processing and preservation techniques.

Treatments	Storage days				Mean ± SE
	0	30	60	90	
T_0	2.31 ± 0.2^j	2.09 ± 0.1^{mn}	1.98 ± 0.1^n	1.87 ± 0.1^{no}	2.06 ± 0.1^G
T_1	2.54 ± 0.2^j	2.27 ± 0.1^k	2.19 ± 0.2^m	2.14 ± 0.1^{mn}	2.17 ± 0.2^F
T_2	2.60 ± 0.2^i	2.34 ± 0.2^j	2.30 ± 0.3^j	2.29 ± 0.2^k	2.38 ± 0.2^E
T_3	2.83 ± 0.1^{ghi}	2.80 ± 0.1^{ghi}	2.73 ± 0.2^{hi}	2.69 ± 0.1^{hi}	2.76 ± 0.1^D
T_4	3.37 ± 0.1^e	3.07 ± 0.1^f	2.99 ± 0.2^{fg}	2.91 ± 0.2^{fgh}	3.09 ± 0.2^C
T_5	4.14 ± 0.1^{bcd}	4.09 ± 0.1^{cd}	3.93 ± 0.2^d	3.51 ± 0.2^e	3.92 ± 0.2^B
T_6	4.67 ± 0.2^a	4.57 ± 0.3^a	4.36 ± 0.3^b	4.22 ± 0.3^{bc}	4.46 ± 0.3^A
Mean ± SE	3.51 ± 0.1^A	3.39 ± 0.2^B	3.24 ± 0.1^C	3.15 ± 0.2^C	

*Each mean value within the same column, followed by different letters is highly significant at $P < 0.05$. T_0 = control; T_1 = 85 ml whey, 2 g chia seed, 0.25 g sodium benzoate, and 0.75 g potassium metabisulfite; T_2 = 85 ml whey, 2 g chia seed, and pasteurization; T_3 = 85 ml whey, 2 g chia seed, and microwave for 30 sec; T_4 = 85 ml whey, 2 g chia seed, and microwave for 60 sec; T_5 = 85 ml whey, 2 g chia seed, and microwave for 90 sec; T_6 = 85 ml whey, 2 g chia seed, and microwave for 120 sec.

[13] reported that during the three months of storage, a decline in chlorophyll of chia seeds added to the mango drink was significant, and this may have been caused by the thermal breakdown of chlorophyll, which produced the yellow pigments known as pheophytins, as well as the oxidation and isomerization of carotenoids during heating.

3.1.3. b^* Value. The mean values of the b^* parameter of color analysis of formulated beverages are shown in Table 6. The present study's results showed that there was a decrease in values of b^* during storage of all treatments. Values of b^* shifted from yellowness to blueness, which showed that the yellowness of the whey drink was due to the composition of milk, which produced the colored components in the whey beverage. Visible changes in the color of whey beverage added with chia seeds were significantly dependent upon microwave heat treatment durations and storage days. For the values of b^*, all treatments had higher values than the control; however, a change in the time of microwave treatment had a significant effect on a^* and b^* values in all studied samples, and these results are in agreement with those of Aamer et al. [33].

Present findings were also paralleled with the color values of chia seeds-added drink developed in another similar study by Buse et al. [45], which showed a decrement of b^* value in drinks during storage. The consequences of the microwave heat treatments on the color values of beverages, observed in the current study were also strongly related with previous results presented by Siefarth et al. [36], as the b^* values were decreased during the storage of drinks.

3.2. pH of the Mango-Whey Beverage. The mean values of pH for different treatments of whey beverage are presented in Table 7, where pH value relates to the concentration of hydrogen ions. The pH value of the mango-whey beverage decreased as a result of pasteurization and an increase in time of heat exposure by microwave. The decrease in the pH of the whey drink with the passage of storage time probably was due to the production of acids during the storage, as an increase in the percentage of acids may lead to an increase in the concentration of hydrogen ions as well as a decrease in pH. Second, evaporation of moisture during heat treatment and storage might also have resulted in the concentration of acids, causing a decrease in the pH of the beverages, as microwave and pasteurization resulted in a more significant decrease in pH, possibly due to the greater moisture loss [34]. Another opinion could also be included in reasoning the drop in pH, which may be attributed to the destruction of lactose owing to the production of lactic acid [22].

Table 6: Mean value for b^* parameter of whey beverage developed with different processing and preservation techniques.

Treatments	Storage days				Mean ± SE
	0	30	60	90	
T_0	14.56 ± 0.1^{ij}	13.59 ± 0.1^{k}	13.06 ± 0.1^{m}	12.95 ± 0.1^{m}	13.59 ± 0.1^{G}
T_1	16.67 ± 0.2^{j}	14.55 ± 0.3^{ij}	13.12 ± 0.1^{k}	12.98 ± 0.1^{m}	14.21 ± 0.2^{F}
T_2	17.51 ± 0.3^{j}	15.70 ± 0.4^{k}	15.68 ± 0.4^{k}	15.53 ± 0.3^{k}	16.11 ± 0.3^{E}
T_3	19.61 ± 0.1^{h}	18.70 ± 0.3^{I}	18.39 ± 0.2^{i}	17.71 ± 0.1^{j}	18.60 ± 0.2^{D}
T_4	21.20 ± 0.2^{f}	20.34 ± 0.3^{g}	20.11 ± 0.3^{gh}	19.86 ± 0.3^{gh}	20.38 ± 0.3^{C}
T_5	24.19 ± 0.2^{cd}	23.77 ± 0.2^{de}	23.48 ± 0.1^{e}	23.11 ± 0.2^{e}	23.64 ± 0.2^{B}
T_6	25.54 ± 0.3^{a}	25.47 ± 0.2^{ab}	24.85 ± 0.3^{bc}	24.66 ± 0.3^{c}	25.13 ± 0.3^{A}
Mean ± SE	21.61 ± 0.3^{A}	20.80 ± 0.3^{B}	20.50 ± 0.3^{B}	20.17 ± 0.2^{C}	

*Each mean value within the same column, followed by different letters is highly significant at $P < 0.05$. T_0 = control; T_1 = 85 ml whey, 2 g chia seed, 0.25 g sodium benzoate, and 0.75 g potassium metabisulfite; T_2 = 85 ml whey, 2 g chia seed, and pasteurization; T_3 = 85 ml whey, 2 g chia seed, and microwave for 30 sec; T_4 = 85 ml whey, 2 g chia seed, and microwave for 60 sec; T_5 = 85 ml whey, 2 g chia seed, and microwave for 90 sec; T_6 = 85 ml whey, 2 g chia seed, and microwave for 120 sec.

Table 7: Mean value for pH of whey beverage developed with different processing and preservation techniques.

Treatments	Storage days				Mean ± SE
	0	30	60	90	
T_0	4.62 ± 0.1^{a}	4.57 ± 0.2^{ab}	4.47 ± 0.2^{bc}	4.32 ± 0.2^{de}	4.49 ± 0.2^{A}
T_1	4.54 ± 0.3^{ab}	4.43 ± 0.3^{bcd}	4.34 ± 0.2^{cde}	4.22 ± 0.3^{e}	4.38 ± 0.3^{B}
T_2	4.48 ± 0.2^{abc}	4.38 ± 0.2^{cd}	4.22 ± 0.1^{e}	3.96 ± 0.2 fg	4.26 ± 0.2^{C}
T_3	3.97 ± 0.2^{fg}	3.83 ± 0.3^{ghi}	3.77 ± 0.1^{hij}	3.68 ± 0.2^{j}	3.91 ± 0.2^{D}
T_4	3.89 ± 0.4^{fgh}	4.03 ± 0.4^{f}	3.96 ± 0.5^{fg}	3.74 ± 0.3^{ij}	3.81 ± 0.4^{E}
T_5	3.67 ± 0.2^{i}	3.59 ± 0.2^{j}	3.49 ± 0.3^{k}	3.39 ± 0.2^{l}	3.59 ± 0.2^{F}
T_6	3.01 ± 0.1^{k}	2.58 ± 0.2^{l}	2.51 ± 0.1^{i}	2.45 ± 0.1^{m}	2.65 ± 0.1^{G}
Mean ± SE	4.30 ± 0.3^{A}	4.25 ± 0.3^{A}	4.15 ± 0.2^{B}	3.98 ± 0.3^{C}	

*Each mean value within the same column, followed by different letters is highly significant at $P < 0.05$. T_0 = control; T_1 = 85 ml whey, 2 g chia seed, 0.25 g sodium benzoate, and 0.75 g potassium metabisulfite; T_2 = 85 ml whey, 2 g chia seed, and pasteurization; T_3 = 85 ml whey, 2 g chia seed, and microwave for 30 sec; T_4 = 85 ml whey, 2 g chia seed, and microwave for 60 sec; T_5 = 85 ml whey, 2 g chia seed, and microwave for 90 sec; T_6 = 85 ml whey, 2 g chia seed, and microwave for 120 sec.

Results of the previous similar findings from the experiments of Siefarth et al. [36] can be correlated with the current study's results of pH, as the pH of stirred yogurt was decreased with the increase in radio wave frequency and storage days. Another previous research on yogurt showed a similar decrease in pH, recorded with the increase in storage time and heat treatment [46]. Results of a latest study from Pinky et al. (2023) were also in line with the current findings, suggesting the use of novel processing techniques as useful for drinks processing.

Similar research was conducted by Adulvitayakorn et al. [47], when they evaluated microwave and conventional thermal processing methods for processing sugarcane juice. They found that the sugarcane juice processed at 700 W using microwave technology had a minor pH reduction. In addition, they connected the temperature variation of the juice samples being treated with the pH variation, as in current experiments, longer microwave treatments might have raised the temperature of the beverage, resulting in loss of moisture. Yikmis [48] extracted the juice from red and yellow watermelons and subjected them to various processing methods in order to evaluate the physicochemical characteristics of both the processed and raw juices. Juice samples that were left unprocessed, pasteurized, and ultrasonic treated at shorter times did not differ in pH from one another, and this contradiction of results could be justified by considering the ultrasound as a nonthermal processing method, which did not rise the temperature of the beverage to such extent as the microwave could raise. Malik et al. [49] looked into the effects of microwave treatments and storage on the pH of lemon cordial. They found that, when treated for 120 seconds, the pH of the cordial increased nonsignificantly when compared to untreated samples, but that, over the course of a 90-day storage period, the pH of the cordial gradually decreased, which they associated with the treatment's decrease in organic acid content.

3.3. Titratable Acidity of the Mango-Whey Beverage. The mean values of titratable acidity of chia seeds added to mango-whey beverage are shown in Table 8. Titratable acidity is directly related to the pH and acid concentration within the food material. The titratable acidity of the beverage increased with the increase in microwave heating time, as well as the storage days. Increased acidity of beverages, due to heat treatments and longer storage durations, could develop a sour taste in the beverages. Prolonged storage time resulted in the production of acids due to enzymatic and nonenzymatic chemical reactions, which was probably due to the presence of acid-producing bacteria. These bacterial colonies might have utilized the sugars and other components present in chia-based whey drinks, which may have led to the higher production of acids. This phenomenon resulted in an increase in acidity, as well as a drop in pH. The acids of

Table 8: Mean value for titratable acidity (%) of whey drink developed with different processing and preservation techniques.

Treatments	Storage days				Mean ± SE
	0	30	60	90	
T_0	0.78 ± 0.1^{r}	0.81 ± 0.1^{q}	0.89 ± 0.2^{p}	0.91 ± 0.2^{n}	0.84 ± 0.1^{G}
T_1	0.87 ± 0.2^{op}	0.92 ± 0.1^{n}	0.98 ± 0.2^{lm}	0.87 ± 0.2^{p}	0.91 ± 0.2^{F}
T_2	0.92 ± 0.4^{no}	0.95 ± 0.3^{mn}	1.02 ± 0.3^{l}	1.09 ± 0.2^{k}	0.99 ± 0.3^{E}
T_3	1.26 ± 0.3j	1.32 ± 0.2^{hi}	1.34 ± 0.2^{ghi}	1.40 ± 0.2^{de}	1.33 ± 0.2^{D}
T_4	1.30 ± 0.1^{ij}	1.35 ± 0.2^{fgh}	1.37 ± 0.1^{efg}	1.39 ± 0.2^{def}	1.35 ± 0.1^{C}
T_5	1.43 ± 0.2^{d}	1.49 ± 0.2^{c}	1.57 ± 0.3^{b}	1.63 ± 0.1^{a}	1.53 ± 0.2^{B}
T_6	1.79 ± 0.2^{def}	1.77 ± 0.1^{efg}	1.75 ± 0.1^{fgh}	1.62 ± 0.1^{hi}	1.76 ± 0.1^{A}
Mean ± SE	1.15 ± 0.3^{D}	1.21 ± 0.3^{C}	1.25 ± 0.2^{B}	1.27 ± 0.2^{A}	

*Each mean value within the same column, followed by different letters is highly significant at $P < 0.05$. T_0 = control; T_1 = 85 ml whey, 2 g chia seed, 0.25 g sodium benzoate, and 0.75 g potassium metabisulfite; T_2 = 85 ml whey, 2 g chia seed, and pasteurization; T_3 = 85 ml whey, 2 g chia seed, and microwave for 30 sec; T_4 = 85 ml whey, 2 g chia seed, and microwave for 60 sec; T_5 = 85 ml whey, 2 g chia seed, and microwave for 90 sec; T_6 = 85 ml whey, 2 g chia seed, and microwave for 120 sec.

the beverage were also increased due to the evaporation of moisture and concentration of whey drink with an increase in time of microwave heating [34, 34].

In the trials of Gehlot et al. [13], the acidity of the mango-mint RTS drink versions prepared without and with 2% chia seeds increased significantly over the course of three months of storage. Results regarding titratable acidity of a similar previous study reported by the authors in [46] were in agreement with the current findings. Another previous finding showed similar results, as an increase in acidity was recorded with the increase in storage time and time of heat treatment of the drink. Another study conducted on the manufacturing and storage of stirred yogurt, processed with different heat treatments, showed that titratable acidity was increased with the increase in heating time and storage days [37]. Moussa et al. [15] reported that the postacidification effect of yogurt was decreased with the addition of chia seeds and the minimum value of postacidification was shown by the treatment having 3% chia seeds, and these results could be correlated with current experiments in a positive way that chia seeds might have contributed towards maintaining the acidity and pH of the formulated beverage.

Findings of Yikmis [48], upon different treatments including microwave and pasteurization of juices, presented nonsignificant findings about the effects of pasteurization and sonication at various times on titratable acidity. As sonication has been known as a nonthermal technique for food processing, that is why the temperature of the juices might not have been raised, due to which variation in titratable acidity and pH was nonsignificant. Similar reports were also present in the findings of Yikmis [50]. Adulvitayakorn et al. [47] discovered a considerable rise in the titratable acidity of sugarcane juice processed at 700 W. This effect on titratable acidity was shown to be connected with loss of temperature, which is in line with the current findings. Their titratable acidity was estimated by comparing juice samples that were thermosonicated and microwave-treated with sugarcane juice that had been heat-treated traditionally.

Malik et al. [49] looked into how storage and microwave treatments affected the titratable acidity of lemon cordial and found that it gradually decreased over time. They postulated that longer microwave treatments and higher temperatures may have destroyed fermenting microorganisms, which may have led to a decrease in the generation of organic acids. Titratable acidity reduces as a result of the acid's use in the hydrolysis of polysaccharides, which turns nonreducing sugars into reducing sugars [51]. Titratable acidity is one of the important parameters defining the taste, shelf life, and microbial deterioration of fruit juices; therefore, the impact of processing conditions on titratable acidity and optimizing the process protocols for maintaining the pH and acidity of juices is very crucial.

3.4. Total Plate Count of the Mango-Whey Beverage. Statistical analysis of the results obtained has shown that microwave heat treatment significantly influenced the total plate count of mango-whey beverages, as microwave treatment caused a significant reduction in total plate count, with increased exposure. The mean values of the total plate count of the whey mango drink are provided in Table 9. The highest value was found for T_1 and the lowest for T_6 at 90 days of refrigerated storage. As the duration of microwave heating was increased, a significant decrease in the total plate count of the beverage was noticed, whereas control presented the highest total plate count, as there was no treatment applied for the preservation of the beverage. Pasteurization also exhibited a relatively higher total plate count of the beverage than the microwave, indicating that the microwave is an effective approach for juice treatment. Results of the total plate count analysis of current experiments were supported by the findings of Abdul Alim et al. [52], where they reported that the total plate count of the mulberry-whey beverage was found to increase during storage, unless proper heat treatment was applied. Microbes require a specific water activity and moisture content for their growth, and as the temperature increases, it increases the solid content of the beverage, thus reducing the pH and moisture contents, which ultimately results in the lowering of the microbial count [53]. When the time duration of microwave heating was increased in current experiments, a significant decrease in the total plate count of the beverage was evident. Jacob et al. [43] reported that the microbial load of untreated juices significantly increases, both during atmospheric or cold storage.

Table 9: Mean value for total plate count ($\log_{10}$ CFU/ml) of whey beverage developed with different processing and preservation techniques.

Treatments	Storage days 0	30	60	90	Mean ± SE
T_0	2.36 ± 0.1^{b}	2.39 ± 0.2^{b}	2.43 ± 0.2^{ab}	2.48 ± 0.2^{a}	2.41 ± 0.2^{A}
T_1	2.17 ± 0.1^{cde}	2.18 ± 0.1^{cd}	2.21 ± 0.1^{c}	2.25 ± 0.1^{c}	2.20 ± 0.1^{B}
T_2	1.77 ± 0.2^{h}	1.86 ± 0.2^{g}	1.88 ± 0.2^{g}	1.93 ± 0.2^{g}	1.86 ± 0.2^{C}
T_3	1.48 ± 0.1^{j}	1.63 ± 0.2^{i}	1.69 ± 0.1^{hi}	1.73 ± 0.1^{h}	1.63 ± 0.1^{D}
T_4	1.38 ± 0.2^{dfg}	1.35 ± 0.1^{def}	1.32 ± 0.2^{fgh}	1.30 ± 0.1^{hi}	1.37 ± 0.2^{E}
T_5	1.04 ± 0.1^{f}	1.05 ± 0.1^{f}	1.09 ± 0.1^{ef}	1.18 ± 0.2^{def}	1.07 ± 0.1^{F}
T_6	0.98 ± 0.1^{g}	0.85 ± 0.1^{h}	0.78 ± 0.1^{i}	0.71 ± 0.1^{j}	0.83 ± 0.1^{G}
Mean ± SE	1.96 ± 0.1^{C}	2.02 ± 0.2^{B}	2.16 ± 0.2^{AB}	3.29 ± 0.1^{A}	

*Each mean value within the same column, followed by different letters is highly significant at $P < 0.05$. T_0 = control; T_1 = 85 ml whey, 2 g chia seed, 0.25 g sodium benzoate, and 0.75 g potassium metabisulfite; T_2 = 85 ml whey, 2 g chia seed, and pasteurization; T_3 = 85 ml whey, 2 g chia seed, and microwave for 30 sec; T_4 = 85 ml whey, 2 g chia seed, and microwave for 60 sec; T_5 = 85 ml whey, 2 g chia seed, and microwave for 90 sec; T_6 = 85 ml whey, 2 g chia seed, and microwave for 120 sec.

Storage duration also had a significant influence on the total plate count of whey drink, as with the increase in the storage period, the growth of microbes was significantly increased in all treatments. Similarly, treatment during storage also had a significant influence on the total plate count of the drink. With the increase in storage period, the microbial count also increased due to variations in pH, acidity, and moisture contents of the beverage, as well as increased enzymatic activity also increased the microbial count of whey drinks [54]. Moussa et al. [15] conducted a study in which stirred yogurt was prepared by using different doses of chia seeds and found that the bacteria present in yogurt have maximum viability as chia seeds added to yogurt exhibited maximum antioxidant and antimicrobial activities. The addition of chia seeds in mango-whey beverages might have provided the favorable environment for the growth of microorganisms, but at the same time microwave heat treatment caused a significant reduction in the growth and survival of microorganisms.

Kowaleski et al. [16] used chia seeds and strawberries for the formation of yogurt, and the acceptability of yogurt was up to 70 percent for the formulation, having 12 percent strawberry and 6 percent chia seeds. There were 107 colony-forming units of lactic acid bacteria during storage. Moreover, 106 colony-forming units of *Bifidobacteria* were also produced during storage, and these microorganisms could be considered as probiotics. Similarly in another study, the addition of chia seed extracts increased the growth of lactic acid bacteria and fermentation rate of set-type yogurt, as was reported by Kwon et al. [17]. Gupta et al. [55] calculated complete bacterial counts and yeast and mold counts of whey-based dairy drinks, which were expanded from 3.14 to 6.48 log CFU/ml and 1.14 to 2.10 log CFU/ml, respectively, during refrigeration storage. A similar fashion study by Moussa et al. [56] revealed that storage of juices without any treatments results in an increase in microbial counts as in the guava-whey drink, whose complete counts were high going from 1120 to 2500 CFU/ml, and mold and yeast counts changed between 0.0 and 18 CFU/ml, during storage. Thus, microwave heat treatment used in comparison to untreated and pasteurized samples, in current experiments, was capable of controlling the microbial growth of the beverage samples, the basic agents responsible for food safety concerns.

3.5. Total Solid Content of the Mango-Whey Beverage. The mean values of total solid contents of different treatments of mango-whey beverage, during different storage days, are presented in Table 10. Analysis of results has shown that microwave heat treatment had significant influences on the total solid content of whey drink, and as the microwave time was increased, the rise in total solids was more significant. Similarly, the storage period also had a significant influence on the total solid content of the whey drink, and as the storage days increased, total solids were decreased for all treatments. The highest value of total solid content was found for T_6 and the lowest for T_0. Increased temperature as a result of pasteurization and microwave treatment for longer periods caused an increase in the total solid content of the whey drink by evaporating the moisture content. The results of the total solid content analysis of current experiments were in line with the findings of Panghal et al. [57], where papaya-based whey drink was developed and total solid contents were decreased during storage.

Total solid contents and moisture showed an inversely proportional relationship with each other. Reduction in the moisture content as a result of microwave treatments for longer time periods reduced the total solid content of whey beverages, whereas, during the storage period, viscosity decreased due to the increase in syneresis and enzymatic activity, which decreased the solid content of the whey beverage [58]. The results of total solids of mango-whey beverages were in accordance with the research outcomes of Tanwar et al. [35], in which the total solids of mango-whey beverages were decreased during storage. As total solids of microwave-treated beverages were decreased during storage in the current study, these results exhibited a close resemblance to those of Sattar et al. [59].

Yikmis [48] observed nonsignificant results for total solids of the juice when comparing raw melon juice with pasteurized and ultrasonicated juice, offering comparable findings as have been reported in the current study. Similar results were also seen in the studies conducted by Bora et al. [60], who found that shorter durations of sonication and microwave had no discernible impact on the solid components of banana juice. Similar findings were also reported by Yikmis, [50] during a comparison of thermal, nonthermal, and chemical treatment of juices with untreated juice.

Table 10: Mean value for total solid content of whey beverage developed with different processing and preservation techniques.

Treatments	Storage days 0	30	60	90	Mean ± SE
T_0	12.11 ± 0.1^{k}	11.15 ± 0.1^{l}	10.65 ± 0.3^{m}	9.21 ± 0.1^{n}	10.78 ± 0.1^{G}
T_1	14.42 ± 0.1^{j}	13.24 ± 0.2^{k}	12.55 ± 0.2^{k}	11.73 ± 0.2^{l}	12.98 ± 0.2^{F}
T_2	18.39 ± 0.3^{f}	17.58 ± 0.1^{g}	16.25 ± 0.2^{hi}	15.63 ± 0.2^{i}	16.96 ± 0.2^{E}
T_3	19.65 ± 0.1^{de}	18.58 ± 0.1^{f}	17.59 ± 0.2^{g}	16.70 ± 0.1^{h}	18.13 ± 0.1^{D}
T_4	21.53 ± 0.2^{b}	20.34 ± 0.3^{cd}	19.55 ± 0.4^{e}	17.45 ± 0.2^{f}	19.35 ± 0.3^{C}
T_5	23.0 ± 0.2^{b}	20.37 ± 0.2^{c}	20.14 ± 0.3^{fg}	18.88 ± 0.2^{i}	19.97 ± 0.2^{B}
T_6	25.21 ± 0.1^{a}	22.23 ± 0.2^{c}	21.19 ± 0.1^{d}	$19,12 \pm 0.1^{i}$	20.12 ± 0.1^{A}
Mean ± SE	19.40 ± 0.2^{A}	18.02 ± 0.1^{B}	16.81 ± 0.3^{C}	15.68 ± 0.2^{D}	

*Each mean value within the same column, followed by different letters is highly significant at $P < 0.05$. T_0 = control; T_1 = 85 ml whey, 2 g chia seed, 0.25 g sodium benzoate, and 0.75 g potassium metabisulfite; T_2 = 85 ml whey, 2 g chia seed, and pasteurization; T_3 = 85 ml whey, 2 g chia seed, and microwave for 30 sec; T_4 = 85 ml whey, 2 g chia seed, and microwave for 60 sec; T_5 = 85 ml whey, 2 g chia seed, and microwave for 90 sec; T_6 = 85 ml whey, 2 g chia seed, and microwave for 120 sec.

3.6. Total Soluble Solids (Brix) of the Mango-Whey Beverage. Brix represents the sugar-to-acid ratio of food commodities. Perishable food commodities possess a higher content of water, and as vitamins, sugars, and amino acids are soluble in water, this high water content causes an increase in the sugar-to-acid ratio in food commodities. The values of the brix of different treatments during different storage days are presented in Table 11. Statistical analysis has shown that microwave heat treatments have significantly influenced the brix of whey drinks. The highest value was found for T_0 and the lowest for T_6, at the start of the study, and during storage, the brix of all treatments was found in decreasing mode. Pasteurization and prolonged microwave time decreased the brix of whey beverage. Similarly, the storage period also had a significant influence on the brix of the whey drink, and as the storage period increased, the acidity level increased, and it decreased the brix of the whey beverage. Results of brix of current analyses were in the range of findings of Kaur et al. [61], where fruit juice-added whey beverage exhibited a decrease in brix during storage. The beverage's lengthy storage caused the monosaccharide and other sugars to break down, which resulted in a decrease in brix. This decrease may also be due to the conversion of insoluble polysaccharides into reducing sugars. The level of reducing sugar might also have been increased due to the acid hydrolysis of sugars, which may have resulted in the breakdown of disaccharides into monosaccharides [13]. According to Nadeem et al. [62], total soluble solids (TSS) were decreased in carrot and grape juice, when sonication treatment was applied. It is possible that the fermentation of carbohydrates into ethyl alcohol, carbon dioxide, and water led to a decrease in TSS during storage. Findings of Pandiselvam et al. [63] and Pandiselvam et al. [22] were also in line with the current ones, where microwave processing of coconut juice was carried out in a similar fashion study.

When compared to individual microwave, sonication, and untreated treatments, the synergy of ultrasonics and microwaves, as reported by Zia et al. [36], showed no change in the TSS of the sugarcane juice samples. This was likely because there was a lower temperature rise than with current microwave processing for longer periods of time. In melon juice processing, Liu et al. [64] evaluated ultrasonic and ultrahigh temperature treatments, along with a control group. The results showed that there was no significant difference in the juice's TSS. These differences in results were possibly due to a higher power and longer time durations of microwave used in previous studies, when compared with the current one. Malik et al. [49] detected a significant decrease in the TSS of juice over storage periods in both the control and microwave-treated samples, which provided supportive data. They connected the rise in internal temperature of the treated samples, which caused the water to evaporate, with the reduction in TSS brought about by microwave treatment. Juices' TSS decreases during storage as a result of yeast and lactic acid bacteria converting carbohydrates into their corresponding acids and alcohols [36].

3.7. Viscosity of the Mango-Whey Beverage. The viscosity of dairy commodities is directly proportional to lactic acid production. When the synthesis of lactic acid is enhanced, it increases the protein content that produces a viscous gel structure in the dairy product. The analysis of the viscosity values of all treatments of mango-whey beverage is presented in Table 12. The highest value was observed for T_6 and the lowest for T_0. The microwave heating time had significantly influenced the viscosity of the whey drink. Similarly, the storage period had a significant effect on the viscosity of whey beverage, and as the storage period increased, it reduced the viscosity of whey drink, by increasing the enzymatic activity, microbial reactions, and other chemical reactions. Another reason might be the rise of temperature, as it results in an increase in solid content and a decrease in moisture content of the beverage, which ultimately increases the viscosity [34]. Heating increases the viscosity of whey drink due to the interaction of phenolic content and protein that produced the gel-like structure, which improves the texture of whey drink and reduces the syneresis [65]. The addition of chia seed extracts increased the water-holding capacity, syneresis, and viscosity of set-type yogurt, as reported by Kwon et al. [17], just as chia seeds-added mango-whey beverage showed high values of viscosity. In order to study the changes that occurred in physicochemical properties and functional components, Sattar et al. [59] chosen to pasteurize peach-based functional beverage for 10 min at 90°C, microwave for 1.5 min at 850 W, and sonicate for

Table 11: Mean value for Brix of whey beverage developed with different processing and preservation techniques.

Treatments	Storage days 0	30	60	90	Mean ± SE
T_0	10.81 ± 0.1^{a}	10.35 ± 0.2^{b}	10.12 ± 0.1^{c}	9.25 ± 0.1^{d}	10.44 ± 0.1^{A}
T_1	10.23 ± 0.2^{b}	10.18 ± 0.2^{b}	10.01 ± 0.3^{b}	9.10 ± 0.2^{b}	10.17 ± 0.2^{B}
T_2	9.74 ± 0.1^{c}	9.34 ± 0.1^{d}	9.26 ± 0.1^{de}	9.08 ± 0.2^{de}	9.35 ± 0.1^{C}
T_3	8.94 ± 0.2^{ef}	8.71 ± 0.2^{fg}	8.45 ± 0.3^{g}	8.10 ± 0.2^{h}	8.55 ± 0.3^{D}
T_4	7.60 ± 0.2^{i}	7.52 ± 0.3^{i}	7.47 ± 0.1^{i}	7.06 ± 0.3^{j}	7.41 ± 0.3^{E}
T_5	6.90 ± 0.1^{j}	6.45 ± 0.2^{k}	6.21 ± 0.2^{l}	6.01 ± 0.2^{m}	6.45 ± 0.2^{F}
T_6	6.51 ± 0.2^{k}	6.34 ± 0.2^{j}	6.02 ± 0.2^{m}	5.98 ± 0.2^{n}	6.21 ± 0.2^{G}
Mean ± SE	9.46 ± 0.1^{A}	9.22 ± 0.2^{B}	9.14 ± 0.1^{B}	8.92 ± 0.2^{C}	

*Each mean value within the same column, followed by different letters is highly significant at $P < 0.05$. T_0 = control; T_1 = 85 ml whey, 2 g chia seed, 0.25 g sodium benzoate, and 0.75 g potassium metabisulfite; T_2 = 85 ml whey, 2 g chia seed, and pasteurization; T_3 = 85 ml whey, 2 g chia seed, and microwave for 30 sec; T_4 = 85 ml whey, 2 g chia seed, and microwave for 60 sec; T_5 = 85 ml whey, 2 g chia seed, and microwave for 90 sec; T_6 = 85 ml whey, 2 g chia seed, and microwave for 120 sec.

Table 12: Mean value for viscosity (cP) of whey beverage developed with different processing and preservation techniques.

Treatments	Storage days 0	30	60	90	Mean ± SE
T_0	0.87 ± 0.2^{j}	0.81 ± 0.2^{ij}	$0.79 \pm \pm 0.1^{k}$	$0.71 \pm \pm 0.2^{i}$	0.79 ± 0.1^{G}
T_1	0.96 ± 0.1^{i}	0.85 ± 0.1^{j}	0.72 ± 0.2^{k}	0.68 ± 0.2^{k}	0.80 ± 0.1^{F}
T_2	1.16 ± 0.1^{f}	1.05 ± 0.1^{g}	0.98 ± 0.2^{hi}	0.87 ± 0.1^{j}	1.01 ± 0.2^{E}
T_3	1.28 ± 0.2^{d}	1.27 ± 0.1de	1.16 ± 0.1^{f}	1.02 ± 0.2^{gh}	1.18 ± 0.2^{D}
T_4	1.37 ± 0.2^{bc}	1.26 ± 0.2^{de}	1.13 ± 0.1^{f}	1.05 ± 0.2^{g}	1.20 ± 0.2^{C}
T_5	1.45 ± 0.1^{b}	1.38 ± 0.1^{d}	1.23 ± 0.2^{e}	1.17 ± 0.2^{e}	1.35 ± 0.2^{B}
T_6	1.51 ± 0.1^{a}	1.43 ± 0.2^{c}	1.38 ± 0.1^{d}	1.38 ± 0.1^{e}	1.49 ± 0.1^{A}
Mean ± SE	1.24 ± 0.1^{A}	1.16 ± 0.1^{B}	1.06 ± 0.1^{C}	0.97 ± 0.2^{D}	

*Each mean value within the same column, followed by different letters is highly significant at $P < 0.05$. T_0 = control; T_1 = 85 ml whey, 2 g chia seed, 0.25 g sodium benzoate, and 0.75 g potassium metabisulfite; T_2 = 85 ml whey, 2 g chia seed, and pasteurization; T_3 = 85 ml whey, 2 g chia seed, and microwave for 30 sec; T_4 = 85 ml whey, 2 g chia seed, and microwave for 60 sec; T_5 = 85 ml whey, 2 g chia seed, and microwave for 90 sec; T_6 = 85 ml whey, 2 g chia seed, and microwave for 120 sec.

90 min at 20 kHz of frequency. They then stored the beverage in the refrigerator for up to 30 days. TSS almost always remained stable, even in samples that have undergone microwave and ultrasound treatment, whereas the cloud values of all processed juice samples decreased with storage time.

To evaluate the effects of chia seed in ice cream stabilization, Feizi et al. [66] replaced a commercial stabilizer with chia seed gum, and the analysis of ice cream including fat globule size distribution, the viscosity of the ice cream mix, destabilized fat index, overrun, hardness of ice cream, and meltdown rate was performed, with conclusions that the ice cream was highly viscous due to chia seeds' gum. Thus, the chia seeds-added mango-whey beverage developed in the current study was an ideal recipe for food lovers, because the trio of chia seeds, mangoes, and whey contributed positively in developing a beverage with optimum quality.

3.8. Sensory Evaluation of the Mango-Whey Beverage. Sensory evaluation of the product was based on the five parameters, which included color, texture, taste, flavor, and overall acceptability. Statistical analysis of the sensory evaluation scores of all treatments of whey drink is presented in Table 13. The highest scores for all parameters were observed for T_3 and the lowest for T_6. The color profile of the whey drink was significantly influenced by the microwave heat treatment duration. The color of dairy products is one of the significant factors that influence the consumer acceptance. As the microwave duration was increased, it increased the yellowness of the whey drink, reducing the lightness, due to which consumers might have not preferred treatments with longer microwave times. The heating of the drink results in the caramelization that imparts a dark color to the drink. The flavor is a combination of taste and aroma, and the evaluation of flavor and taste was based on the taste buds such as bitter, sweet, umami, sour, and salty. Results indicated that T_3 obtained higher consumer acceptance for taste and flavor. As the temperature was increased, it increased solid contents, by decreasing the moisture and pH, which resulted in the bitterness and reduced the acceptance level of consumers. The appearance of dairy commodities has been found to be directly proportional to the lactic acid production. When the synthesis of lactic acid is enhanced, it results in increased protein content, which produces a viscous gel structure in dairy products. The texture profile of the whey drink was significantly influenced by the addition of chia seeds. Increased microwave heating duration increased the viscosity of the whey drink due to the interaction of the phenolic content and protein that produced a gel-like structure, which improves the texture of the whey drink and reduces the syneresis [58]. For overall acceptability, the

Table 13: Mean value for sensory evaluation of whey beverage developed with different processing and preservation techniques.

Treatment	Color	Flavor	Texture	Taste	Overall acceptability
T_0	7.92 ± 0.2^{b}	7.50 ± 0.3^{c}	6.98 ± 0.2^{d}	6.5 ± 0.2^{d}	7.12 ± 0.3^{c}
T_1	7.14 ± 0.2^{c}	7.43 ± 0.3^{ab}	7.60 ± 0.3^{b}	7.60 ± 0.1^{b}	7.14 ± 0.3^{b}
T_2	7.13 ± 0.3^{c}	7.39 ± 0.4^{ab}	7.54 ± 0.4^{b}	7.11 ± 0.4^{bc}	6.51 ± 0.4^{d}
T_3	7.65 ± 0.2^{a}	7.76 ± 0.2^{a}	7.70 ± 0.2^{a}	7.70 ± 0.2^{a}	7.41 ± 0.3^{a}
T_4	7.21 ± 0.3^{c}	7.26 ± 0.4^{ab}	7.08 ± 0.2^{b}	7.03 ± 0.3^{bc}	5.99 ± 0.3^{e}
T_5	6.26 ± 0.2^{d}	7.04 ± 0.1^{b}	5.63 ± 0.2^{c}	6.69 ± 0.2^{c}	5.89 ± 0.2^{e}
T_6	6.12 ± 0.2^{f}	6.08 ± 0.2^{e}	5.41 ± 0.2^{e}	5.32 ± 0.1^{d}	5.54 ± 0.2^{f}

*Each mean value within the same column, followed by different letters is highly significant at $P < 0.05$. T_0 = control; T_1 = 85 ml whey, 2 g chia seed, 0.25 g sodium benzoate, and 0.75 g potassium metabisulfite; T_2 = 85 ml whey, 2 g chia seed, and pasteurization; T_3 = 85 ml whey, 2 g chia seed, and microwave for 30 sec; T_4 = 85 ml whey, 2 g chia seed, and microwave for 60 sec; T_5 = 85 ml whey, 2 g chia seed, and microwave for 90 sec; T_6 = 85 ml whey, 2 g chia seed, and microwave for 120 sec.

highest score was observed for T_3 and the lowest for T_6. Microwave heat treatment durations significantly influenced the overall acceptability of whey drink.

Shrestha and Dahal [67] developed a drink by utilizing natural whey with magnificent wholesome characteristics and blended flavors, alongside banana juice and the necessary measure of sugar. A huge variety in taste, color, flavor, and overall acceptability was seen by differing the organization of whey and banana juice. In a similar study, Yonis et al. [68] uncovered that the drink developed with 75% whey and 25% guava juice scored the greatest for practically all sensorial quality credits such as the taste, color, flavor, texture, and overall acceptability. An ice cream formulation was evaluated by Feizi et al. [66], and a sensory evaluation by a focused group showed that the ice cream was highly acceptable as it had a cream texture, smooth texture, and hardness was desirable without any crystallization.

The development of innovative thermal and nonthermal food processing techniques has made it feasible to produce food products that are safer, healthier, and have acceptable sensory qualities. According to Ozkan et al. [69], the use of nonthermal processing was proven to be an efficient strategy in maintaining maximum antioxidant chemicals and acceptable sensory qualities in natural fruits and their juices. The length of the beverage's microwave treatment and storage time had a significant impact on its sensory qualities [63].

The use of microwave heat treatments for 30 sec was proved effective in maintaining the optimum quality of the formulated beverage; that is why consumers preferred this treatment over untreated and pasteurized beverage samples. The most acceptable treatment T_3 (85 ml whey, 2 g chia seed, and microwave heat treatment for 30 sec) was further subjected to a storage study, to find out the influence of storage on the sensory aspects and consumer acceptance level.

3.9. Storage Study of the Most Acceptable Treatment. Whey beverage supplemented with 2 g chia seed that had a microwave treatment for 30 sec (T_3) was most acceptable for consumers in terms of taste, color, flavor, texture, and overall acceptability. The three-month storage period had a significant influence on the sensory aspects of T_3. The means of sensory scores are shown in Table 14. An increase in the storage period significantly decreased the acceptance level of consumers, as was evident from the decreased scores obtained during increased storage days. As the storage period increased, the enzymatic activity, chemical reaction, and microbial growth also increased, which would possibly have influenced the taste, color, and flavor of whey beverages. From the storage study results, it was found that formulated beverage got good scores during the 60 days of storage, but a further increase in storage time caused a reduction in consumer preferences, so the chia seeds-added mango-whey beverage microwaved for 30 sec was recommended for consumption during the 60 days of refrigerated storage.

Gupta et al. [55] conducted experiments for the exploration of microbial and physicochemical properties of cocoa and whey protein-advanced utilitarian dairy drink, that was exposed to storage at a refrigeration temperature of 4 ± 1°C, and these findings proposed that the drink could be stored for up to 18 days at 4 ± 1°C. Experimental investigation of Shrestha and Dahal [67] showed that refreshments arranged with 85% fluid whey and 15% banana juice could be stored for 30 days under refrigerated conditions, without the expansion of additives. Incorporation of chia in the form of gel was performed to avoid the competition of starch and ground chia, by Zettel et al. [70], and it was found that the volume yield of bread loaf as well as the stability of dough during fermentation was increased by the addition of chia gel, providing the high consumer acceptance attributed to chia seeds, just as our product got good preferences. During the three months of storage, a significant decline in the sensory scores for the mango-mint drink with chia seeds added was observed in the areas of color and appearance, taste, mouthfeel, flavour, and overall acceptability [13]. Whey beverages developed in the current study were initially pleasing in color and appearance with high sensory scores, but changes were evident as three months of storage progressed. It might be because mango pulp and chia seeds initially had attractive colors during storage, but those colors started to fade as ascorbic acid, carotenoids, and total phenols degraded with time. The Maillard process, which occurs when sugars and amino acids are mixed in an acidic environment, may be the cause of the beverage colors deteriorating after storage.

Table 14: Mean values for sensory evaluation of most acceptable treatment of whey beverage (T_3).

Treatment T_3	Storage days			
	0	30	60	90
Color	7.90 ± 0.1^a	7.37 ± 0.2^b	6.26 ± 0.2^c	4.78 ± 0.2^d
Taste	7.48 ± 0.2^a	6.37 ± 0.3^b	5.40 ± 0.3^c	2.95 ± 0.3^d
Flavor	7.91 ± 0.1^a	6.99 ± 0.2^{ab}	6.15 ± 0.2^b	3.48 ± 0.2^c
Texture	7.34 ± 0.4^a	6.55 ± 0.2^b	5.95 ± 0.3^b	4.28 ± 0.4^c
Overall acceptability	7.92 ± 0.3^a	7.24 ± 0.4^b	4.74 ± 0.3^c	3.62 ± 0.3^d

*Each mean value within the same column, followed by different letters is highly significant at $P < 0.05$. T_3 = 85 ml whey, 2 g chia seed, and microwave for 30 sec.

Tangerine juice samples were processed and preserved using microwave treatments by Demirok and Yikmis [71], who also produced comparable findings for storage trials. In a related study, the antioxidant capacity of untreated and pasteurized juices reported a decreasing trend during storage, following a first-order kinetics [72], supporting the findings of the current work regarding the decrement of nutritional potential of blend juices during storage. Correlating these experiments with the current one, it could be argued that either untreated, or pasteurized and microwaved, all treatments of beverages start deteriorating after a limited storage duration under controlled refrigerated conditions. Microwave and ultrasonic as green processing technologies work chemically (free radicals) and mechanically (cavitation and shock waves). Juices have higher antioxidant capacities and retain more bioactive compounds during storage under controlled circumstances, therefore processing them using ultrasound and microwaves separately or in combination yields better results [73].

4. Conclusion

In the current experiments, a novel beverage formulation containing whey, mango pulp, and chia seeds was developed, which had undergone different preservation techniques, to study their effect on chemical, physical, microbial, and sensory attributes. The chemical composition of chia seeds revealed the high nutritional profile of chia seeds, before incorporation into beverages. The color of chia seeds added to mango-whey beverages was significantly affected by pasteurization and microwave treatments. There was a significant increase in the titratable acidity of the beverage with the increase in time of storage and microwave heating, whereas the pH value significantly decreased. Microwave heat treatment significantly influenced the total plate count of whey beverages, as the highest value of total plate count was found for T_1 and lowest for T_6. Storage duration also had a significant influence on the total plate count of beverages as with the increase in the storage period, the growth of microbes was significantly increased, but as the microwave treatment time was increased the total plate count decreased. An increase in the microwave heating caused an increase in total solids. On the other hand, brix was decreased, both for increased microwave time and storage. Sensory analysis showed that the best results were shown by the treatment, which had 2 g of chia seeds, and was applied 30 sec of microwave heat treatment (T_3). The conclusive result of the sensory evaluation of T_3 at different days of storage showed that good scores were found at 0 and 30 days of storage, scores at 60 days of storage were also acceptable, but were highly decreased at 90 days. In the end, it could be concluded that microwave treatment for 30 sec, of formulated beverage, provided the best results to obtain the optimum parameters for physicochemical, microbial, and sensory characteristics during storage. The food processing industry can employ the microwaved mango-whey beverage with chia seeds added as an effective and premium functional food.

5. Recommendations

Fruit extracts and juices can be used in functional beverages in a variety of ways because fruits are valuable dietary sources and are recognized for being rich in phenolic and antioxidant components. These beverages are becoming more and more popular worldwide. Consumers are becoming more conscious of the nutritional content and possible health advantages of food. Whey is a byproduct of cheese production that can be used to make useful food items. Chia seed is a rich source of omega-3 fatty acids and is recommended for patients suffering from cardiovascular disease and for intended consumers for fiber intake. Mango-whey beverage enriched with chia seeds is recommended for the sportsmen and adults as a potent source of energy and valuable nutrients. This beverage has been also recommended for the school-going children suffering from protein deficiency and is particularly significant for athletes and bodybuilders for mass gain. Further phytochemical and antioxidant studies are required on these formulated beverages processed and preserved under different conditions for different time periods, in order to commercialize these value-added functional food formulations.

Data Availability

The data used to support the findings of the study are available from the corresponding author upon request.

Additional Points

Highlights. (i) Mango-based whey beverage, added with chia seeds, is an exotic and nutritious drink. (ii) Comparison of different processing and preservation techniques for beverage formulation. (iii) Significant changes in pH, acidity, brix, viscosity, phenolics and sensory parameters during the 90 days of storage. (iv) Microwave treatment had a significant influence on the physiochemical, nutritional and sensory profiles of beverages. (v) Microwave treatment for 30 sec provided the best results as compared to other treatments.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

Authors' Contributions

Ashiq Hussain conceptualized the study. Mansoor Hassan and Farzana Siddique curated the data and performed the formal analysis. Haya Fatima acquired the funding. Amer Ali Mahdi and Sameh A. Korma investigated the study. Syeda Ayesha Batool developed the methodology. Mansoor Hassan administered the project. Saima Noreen collected the resources. Faiza Iftikhar Gorsi developed the software. Farzana Siddique and Shazia Yaqub supervised the study. Tahira Siddique and Amer Ali Mahdi validated the study. Samina Kauser and Sameh A. Korma visualized the study. Ashiq Hussain wrote the original draft of the manuscript. Ashiq Hussain, Amer Ali Mahdi, and Sameh A. Korma wrote, reviewed, and edited the manuscript. All authors contributed equally in the preparation of this manuscript and gave their consent for publication.

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