



Physico-chemical, Microbiological, and Sensory Evaluation of Mango Enriched Probiotic Yogurt

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Abstract

Yogurt is cultured dairy product produced by fermenting milk with or without added non-fat dry milk with *Lactobacillus bulgaricus* and *Streptococcus thermophilus* bacteria. Attempt was made to manufacture yoghurt with different concentrations of mango pulp 4, 6, 8, 10 and 12% respectively. After that physio-chemical and microbiological analysis of experimental sample T3 contain highly score average range 8.37 as compare to treated experimental sample T1, T2, T4, T5. In this study a major researched has been found after the 5th and 6th day of incubated sample with coliform colonies has been disappeared due to the highly metabolic activity of probiotic stains *Lactobacillus bulgaricus*, *Streptococcus thermophilus*, *Bifidobacterium*'s, *Lactobacillus acidophilus*. *L. rhamnosus*. Probiotics strains reached viable counts of 107 to 108 colony forming units (CFU)/mL in all treatments. Sensory panellists provided higher hedonic scores to T3 for flavour, colour and appearance, body and texture compared to T1, T4 and T5 but flavour and overall acceptability ratings amongst T1 to T5 were comparable.

Keywords: probiotic yogurt; probiotics strain; mango pulp; sensory evaluation.

Introduction

Yogurt is cultured dairy product produced by fermenting milk with or without added non-fat dry milk with *Lactobacillus bulgaricus* and *Streptococcus thermophilus* bacteria. It is usually containing 12-14 % total milk solids and has soft, easily crumbled and custard like consistency and a clean distinct flavor. The word yogurt is derived from Turkish word 'Yugurt' and is traditional food beverage in Balkan and Middle east. However, its vogue has also discovered and escalate in India too. Yogurt has unique nutritional attributes as they supply high quality proteins, also excellent source of calcium, phosphorus and potassium less zinc along with significant quantities of several vitamins. The carbohydrate content is easily absorbed even by lactose maldigestions. Presence of b-D-galactosidase activity in probiotic yogurts indicates its suitability for lactose-intolerant infants (Sarkar and Mishra 1998). Yogurt could be used for infant and adult feeding for its higher Ca/Na ratio as compared to RDA value in



USA. In 1878 to 1945, A Bulgarian medical student, Stamen Grigor was the first to discover *Bacillus bulgaricus* (LB) a lactic acid bacterium using in yogurt culture recently (McGee et al., 2014)

Yogurt is being manufacturing from milk with or without the addition of some natural derivatives of milk such as skim milk powder, caseinates or cream, whey concentrates with a gel formation structure that results from the coagulation of milk protein and degraded the lactose into lactic acid by specific species of bacterial cultures. Sometime, these bacterial strains must be “Viable and copious” at the time of consumption. The most commonly type of yogurt commercially available in market are set type yogurt and strained yogurt; though lately frozen yogurt and drinking yogurt have become quite popular. Set type of yogurt is fermented in retail customers and no further stirring or water take place after the fermentation manufacture process. Strained (Stirred and Greek style) yogurt is fermented in closed tank under sterile continuous mild stirring and after the completion of fermentation process a portion of the whey is removed. Due to the manufacturing process, the two different types develop a different texture; Set type yogurt develops a continuous gel texture, whereas strained yogurt displays a viscous creamy smooth texture.

Previous research stated that yogurt can be defined by the symbiosis of two strains of bacteria (*Streptococcus thermophiles* and *Lactobacillus bulgaricus*) in a sterile favourable environment at a very low temperature (36 – 42°C) for 3–8 h (Cirone et al., 2013). Both these bacterial strains must remain live and functionally active in final product with at least 10 million bacteria/g (Bodot et al., 2013). The manufacturing process to which pre-pasteurized skimmed milk is submitted, before it is turned into yogurt is responsible for change in protein, carbohydrate and lipid. When milk lactose is used as the fermentation substrate lactic acid and a series of another compound are formed and released, contributing to its aroma. After that its consequence of a decrease in pH and development of undesirable microorganism is delayed, phosphorous and calcium present in milk is converted into their soluble form, the majority of protein, now calcium free are better digested by probiotic by proteolytic and most commonly proteolytic enzyme, which increase its digestibility and over all bioavailability.

As per statista marker study in 2023, in the yogurt market volume is expected to amount to 21.72bn kg by 2028. The Yogurt market is expected to show a volume growth of 5.7% in 2024. The average volume per person in the Yogurt market is expected to amount to 11.9kg in 2023. The mango, or *Mangifera indica*, is a tropical fruit that is prized for its vivid hue, sweet taste and rich flavour. It is also thought to contain phytochemicals that have interesting biological qualities (Palafox-Carlos et al., 2012). The peel of mangos, which makes up around 20% of the fruit, has a lot of beneficial ingredients, such as enzymes, carotenoids, polyphenols, vitamin C, and E. Numerous biological attributes, such as anticancer, antioxidant, antibacterial, anti-inflammatory, cardiovascular, and hepatoprotective actions are demonstrated by these constituents (Jahurul et al., 2015; Quintana et al., 2021). Mango peel thus offers an alternative to the creation of new goods. Plain yogurt contains less phenolic compound and antioxidant compound though rarely health benefit of human health. In addition of functional compound like



carotenoid, fortified vitamin, VitE, flavonoids, polyphenols may highly functional food derivatives for health benefit (John and Singla, 2021).

In order to fermented foods, yogurt production has increased by over 4% between 1995 and 2009 (USDA-ERS, 2013; Batmanglij, 2007) and corresponding yogurt intake has increased in the past decade (Nielsen and Ogden, 2015). Yogurt contains higher amount of Protein, VitB12, VitB2, potassium, calcium, magnesium and zinc than milk (USDA Food Data Central Database. 2018). Meta- analytes have shown that yogurt may help with lower risk of type 2 diabetes. They help in decreased the risk of cardiovascular disease (Danone Nutricia, 2013) and metabolic syndrome (PREDEMED Investigator, 2015). Yogurt consumption has also been shown to be associated with a lower body mass index (BMI), lower body weight/weight gain, smaller waist circumferences (MC), and lower body fat (Mozaffarian et al., 2018 and Wang et al., 2014). The present investigation was undertaken to assess the physical, microbiological and quality properties to develop the mango flavoured probiotic yogurt from different concentration of mango pulp.

Materials and Methods

2.1 Collection of Ingredients.

2.1.1 Procurement of milk sample

Proximate fresh raw mixed milk was collected from the mother dairy Etawah (U.P.) plant of Ghatanpur source. The milk was cooled immediately after receiving and used without prolonged storage. During this study all of the instrument's media, buffers & chemical reagents used in this research will be listed in the Appendix. " Sigma-Aldrich (St. Louis, MO, USA), Hi Media laboratory Pvt Ltd. (Mumbai, India), Merck Pvt. Ltd. (Mumbai, India)

& Biovision Inc. were among the manufacturers having delivered the media and reagents (AR or reagent grade) (California, USA), Laminar Air Flow make Optic technology Delhi, Sampling Bottle make Duran, Auto pipette make Borosil with Tips, Agar Media make Hi media, Dilution Blank (9 ml or 99 ml), Test Tube / Culture tube make Borosil, Durham's Tube, Petri Dish, Petri Dish Container make HI media, Hot Air Oven make Optics technology Delhi, Microscope, Autoclave EIC India Pvt. Gujrat, Water Bath make Optics technology, Vertex shaker make Optics technology, BOD Incubator make Optics technology, Colony counter make Optics technology, pH Meter make Thermo model no Orion star A214 / pH Strips, Media Making Utensil make Duran's and Refrigerator make Whirlpool.



2.1.2 Procurement of raw material

Spray dried skim milk powder (Strainer tested) manufactured by Mother dairy fruit and vegetables Pvt. Ltd. was purchased from local market. Cane sugar was purchased from the local market. Mango pulp purchased from Safal Mother dairy brand. Plastic cups of 200 ml. capacity were purchased from the local market.

2.1.3 Procurement of cultures

Freeze dried culture of Lactobacillus culture (Lactobacillus rhamnosus NCDC 610. Lactobacillus fermentum), Lactobacillus bulgaricus and Streptococcus thermophilus NCDC074 (1:1), Bifidobacterium and Lactobacillus acidophilus were obtained from National Collection of Dairy cultures, Dairy Microbiology Division, National Dairy Research Institute, Karnal and DVStarter Culture (YFL903) from CHR HANSEN Denmark.

2.2 Maintenance of Cultures

Normal yogurt cultures were maintained in sterilized skim milk and in litmus milk tubes. Sterilization was done at 15 psi for 15 min in an autoclave. The cultures were propagated by transferring a loop full of culture to the sterilized tube aseptically. These tubes were incubated at 37°C in an incubator. Prior to inoculation for preparation of product at least three transfers were made. Finally, the cultures were inoculated in 200 ml. sterilized milk at a rate of 1.0% to be used for the preparation of yogurt. After development of desired acidity, the mother culture was stored in refrigerated cabinet. Secondary cultures prior to use the culture were sub-cultured at 37 °C for 15-18 h in skim milk/MRS/M17 broth tubes.

2.2.1 Purity of cultures

Every time before the study, the cultures were evaluated for purity by Gram's staining, Negative staining and catalase test.

2.2.2 Gram staining

A thin smear was prepared with the help of inoculating loop by taking a drop of the cultures on the clean, grease free slide. The slide was air dried and fixed by heating with the low flame. The smear was stained with crystal violet for 1 min. Gram's iodine was covered on the slide for 30 sec and then rinsed with water. The smear was decolorized by treatment with decolourizer i.e., 95% alcohol. Final step is done with counter stain i.e., safranin (0.5%) for 1 min followed by rinsing with water. The stained smear was air dried and observed under oil immersion lenses.

2.2.3 Negative staining



The negative staining was performed by taking one drop each of bacterial suspension and Nigrosine stain on the clean, grease free slide. The smear was uniformly prepared, air dried and observed under oil immersion lens.

2.2.4 Catalase test

Aseptically added 2 ml of starter culture and equal volumes of 10% H₂O₂ in a test tube with proper mixing and observed for the formation of effervescence.

2.3 Physical examination of raw milk sample.

As per physical analysis of raw mixed milk sample test parameter like odour, taste, flavour, colour and consistency checked by FSSAI test manual. Organoleptic testing allows for quick separation of low-quality milk at the receiving platform. A strong sense of sight, aroma, order, taste and colour is required of the milk grader (IS 1479 (Part II) – 1997). Sensory evaluation test of all collected sample performed as per BIS method evaluation (IS 1479 (Part II) – 1997).

2.3 Chemical examination of raw milk sample.

Fat% of raw standardized mix milk was estimated by Gerber method as per IS:1224 (58) Part-I. Solid not fat is determined by using Richmand's formula as per IS: 1479(Part II). Acidity of the raw standardized mix milk in term **of percent Lactic acid was determined as per IS: 1479 (Part – I). All adulterant test is done by FSSAI test manual.**

2.4 Microbiological examination of raw milk sample.

Before processing the raw milk sample for microbiological analysis all materials sterilized by dry heat in a hot air oven at 170°C for 2 h and wet sterilization by autoclaving at 15 psi (121°C) for 20 min. After sterilization all materials were stored in hygienic condition. Collected samples were prepared for testing as per reference method (IS 11546: 2012 (Reaffirmed, 2018)). During testing of raw milk samples approximate dilution factor to be used range from Coliform (10⁻², 10⁻³), Aerobic plate count (10⁻⁴, 10⁻⁵, 10⁻⁶), Spore count (10⁻¹, 10⁻², 10⁻³) and Psychrophiles count (10⁻², 10⁻³). Coliform count is analysed by as per BIS standard method (IS: 5401:2018). Aerobic plate count of raw mixed milk samples was determined as per BIS standard method (IS: 5402:2018). After that Counted the colonies grown in petri plates between 30 - 300 cfu/ml in each plate. After that test result was interpreted by the calculation below mention.



$$\text{Calculation: } N = \frac{\Sigma C}{(N1+0.1N2) D} \text{ or } \frac{\Sigma C}{2.2 * d} \quad (1)$$

ΣC is the sum of colonies counted on all the dishes retained. N1 is the no. of dishes retained in the first dilution. N2 is the number of dishes retained in the second dilution. D is the dilution factor corresponding to first dilution. Coliform count of raw mixed milk samples was determined as per BIS standard method (IS: 5401-2012). Test result was interpreted by the calculation mention in para 2.5. Spore count of raw mixed milk samples was determined as per reference and BIS standard method (Kent et al., 2016; IS 1479-2003). Test result was interpreted by the calculation mention in para 2.5. Somatic cell counted by (DeLaval SCC Analyzer). Take 1 ml of milk in a beaker and mix properly. Charge the cassette with the help of piston approximately 0.1 ml of milk suck by the capillary action inside the cassette (No.-92865880). Milk passed inside the micron filter in cassette and somatic cell trapped inside there. Cassette placed inside the DeLaval SCC analyzer. Run the start button and wait for five minutes. Spectra of light is fall on the micron filter and absorb the intensity of light is directly proportional to the somatic cell concentration”.

$$\text{Calculation: Total no of reading on display} \times 1000 = \text{SCC/ml} \quad \text{-----} \quad (2)$$

2.5Preparation of yogurt, Mango probiotic yogurt

Firstly, standardized the good quality previously selected mixed milk based on the physio-chemical and microbiological test parameter as per FSSAI specification.

2.5.1 Preparation of sugar syrup.

Sugar syrup was prepared by mixing with the ration of 13% and sterilize at 80 °C. After that cool at 45 °C and filtered with SS Stainer. After that added in the final standardized batch as the same ratio.

2.5.2 Basic mix.

Selected raw mixed milk based on the parameter of physio- chemically as well as microbiologically. After that standardized the raw mixed milk based on the recipe (Fat %, SNF %, Sugar %, and additives) of yogurt as per FSSAI, BIS and MDTL specification. The desired composition of the yogurt mix was maintained by calculating the quantity of fat and SNF in the mixed raw milk from Ghatanpur sources and made up the fat % and SNF % up to Fat 4.7% and SNF11.30 % respectively by the addition of calculated amount of spray dried skim milk powder, using Pearson square method.Semi-finished goods (SFG) batch are standardized based on the all added ingredient as per recipe. After that calculate the finally calculation as per 1000 kg. As per 1000 kg of SFG standardized batch fat %, Total Solid (TS) and Protein part vary in finished good (FG) test. FG testing of all parameter should be done after preparation of yogurt.



2.5.3 Preparation of Yogurt.

Fresh whole mixed milk was procured from the NGC Ghatanpur (UP) standardized to fat 4.7% and SNF 11.30 % by adding skim milk. The standardized milk was heated to $92 \pm 2^\circ\text{C}$ for 6-10 min and cooled to 42°C . For preparation of yogurt, culture strains *Lactobacillus bulgaricus* and *Streptococcus thermophilus* (NCDC 074) or culture (YFL 903) was inoculated at 2 % (v/v) and incubated at 42°C till the acidity is measured 0.70-0.75% (figure no 1). The probiotic yogurt was prepared by inoculating culture strains *Bifidobacterium* and *Lactobacillus acidophilus* (DVS CHR HANSEN DENMARK) 2 % (v/v) and incubated at 42°C till the acidity is measured 0.70-0.75%.

2.5.4 Process flowchart of manufacturing of set mango probiotic yogurt

The manufacturing process for mango enriched probiotic yogurt was shown in figure no 2.

2.5.5 Control Sample

The control sample having 4.7% fat and 11.30% solid not fat was prepared by addition of calculated amount of mixed milk, and spray dried skim milk powder. The mixture was then heated to 80°C and maintain the level 13% in final product. The temperature was then raised to 92°C and held for a period of 15 min. it was then cooled to $40-42^\circ\text{C}$ and inoculated with normal yogurt culture at the rate of 2% using a ratio of 1:1 the bulk inoculated mixture was then divided into two equal part and one part is transferred into 100 ml polystyrene cups, caps, capped and incubated at $40-42^\circ\text{C}$ until desired body was obtained.

2.5.7 Experimental Sample

The different Mango pulp concentration level 4% to 8% within the experiment samples were adjusted by the addition of the pulp. The different concentration level of Mango pulp was blended into the second part of the bulk inoculated mixture were then further processed similar to control samples.

2.5.8 Storage

After the incubated period, the cups were removed from incubator and cooled in ice bath to $8-9^\circ\text{C}$.

2.6 Physical test of prepared yogurt.

2.6.1 Wheying off

To determine the rate of wheying off in sample the method suggested by Chawla and Balachandram, (2006) was used with slight modification 50 ml of inoculated sample were taken in the cups and the same treatment, from incubation to storage, was loosened from the sides of the cup and emptied



into a glass funnel with a water filter No.1.the funnel was placed on the graduated glass cylinder. The quality of whey collection after 15 min. was noted down. This was measured as the wheying off (ml)of the samples.

2.7 Chemical analysis of yogurt and mango probiotic yogurt.

The titratable acidity of yogurt samples, expressed as percent lactic acid was determined as per the procedure laid down in IS: 1166- 1973.After that calculated the desire value of acidity.

Calculation.

$$\text{Titratable acidity (Lactic Acidity)} = \frac{9 \text{ VN}}{W} \quad (3)$$

V= Volume of 0.1 N NAOH used in titration; N= Normality of NAOH; W= Weight of sample.

The pH of yogurt sample was determined using the digital pH meter (EUTECH, model pH 700, India).Fat % of yogurt samples was estimated by Gerber method as per (IS 1479-2003).Solid not fat in the yogurt samples done by using Richmand's formula as per (IS 10083-1982).

Calculation

$$\text{SNF\%} = \text{Total Solid\%} - \text{Fat\%} \quad (4)$$

Total solid in yogurt were determined by gravimetrically, the procedure laid down in the FSSAI manual& IDF, (2005).

2.8 Microbiological analysis of yogurt and mango probiotic yogurt.

Different dilution factor was used for analysis of probiotic yogurt like 10^{-1} , 10^{-2} for Coliform, 10^{-7} , 10^{-8} for *Lactobacillus bulgaricus* and *Streptococcus thermophilus*, 10^{-1} , 10^{-2} , 10^{-3} for *Bifidobacterium's* and *Lactobacillus acidophilus* and 10^{-6} , 10^{-7} for *Lactobacillus rhamnosus*.

2.8.2Determination of coliform count: (IS:5401-2012).

Coliform cfu/gm in yogurt is analysed by BIS standard method (IS: 5401-2012). After that test result calculated by below mention calculation.

$$\text{Calculation:} \quad N = \frac{\sum C}{(N1+0.1N2) D} \quad \text{or} \quad \frac{\sum C}{2.2 * d} \quad (5)$$



ΣC is the sum of colonies counted on all the dishes retained; N1 is the no. of dishes retained in the first dilution; N2 is the no of dishes retained in the second dilution; D is the dilution factor corresponding to first dilution.

2.8.3 Determination of *Streptococcus Thermophilus* count: (IS: 5401-2012).

Transfer 10 ml of well-mixed milk sample to 90 ml diluent (phosphate buffer solution) & mix well to make first dilution. If required make second dilution by transferring 1 ml of first dilution into 9 ml diluent. Arrange 2 Petri plates for each sample mark them with sample no. and date. Transfer 1 ml in each plate respective dilution of sample to be tested. Before pouring the media in plate adjusted the pH 7.1. Add 15 to 20 ml of M017 (Hi-media code – M1036) agar, previously melted and cooled to about 45°C. Mix the contents thoroughly by rotating the plates and allow the agar to solidify. Invert plates and incubate the plates at 44 ± 1°C for 48h. After incubation count the colonies.

Calculation:
$$N = \frac{\Sigma C}{(N1 + 0.1N2) * D} \quad \text{or} \quad \frac{\Sigma C}{2.2 * d} \quad (6)$$

ΣC is the sum of colonies counted on all the dishes retained; N1 is the no. of dishes retained in the first dilution; N2 is the no of dishes retained in the second dilution; D is the dilution factor corresponding to first dilution.

2.8.4 Determination of *Lactobacillus acidophilus* count: (IS: 5401-2012).

Transfer 10 ml of well-mixed milk sample to 90 ml diluent (phosphate buffer solution) & mix well to make first dilution. If required make second dilution by transferring 1 ml of first dilution into 9 ml diluent. Arrange 2 Petri plates for each sample mark them with sample no. and date. Transfer 1 ml in each plate respective dilution of sample to be tested. Before pouring the media in plate adjusted the pH 5.5. Add 15 to 20 ml of MRS (Hi-media code – M641) agar, previously melted and cooled to about 45°C. Mix the contents thoroughly by rotating the plates and allow the agar to solidify. Invert plates and incubate the plates at 37 ± 1°C for 48 h. After incubation count the colonies.

Calculation:
$$N = \frac{\Sigma C}{(N1 + 0.1N2) * D} \quad \text{or} \quad \frac{\Sigma C}{2.2 * d} \quad (7)$$



ΣC is the sum of colonies counted on all the dishes retained; N_1 is the no. of dishes retained in the first dilution; N_2 is the no of dishes retained in the second dilution; D is the dilution factor corresponding to first dilution.

2.8.5 Determination of *Bifidobacterium* count: (IS: 5401-2012).

Transfer 10 ml of well-mixed milk sample to 90 ml diluent (phosphate buffer solution) & mix well to make first dilution. If required make second dilution by transferring 1 ml of first dilution into 9 ml diluent. Arrange 2 Petri plates for each sample mark them with sample no. and date. Transfer 1 ml in each plate respective dilution of sample to be tested. Before pouring the media in plate adjusted the pH 6.5. After that add the solution A (Dicloxacillin solution-10 ml), solution B (Lithium chloride solution – 5 ml) and solution C (Cysteine hydrochloride solution – 980 ml) in MRS media and mix it properly by swirling action. Add 15 to 20 ml of MRS (Himedia code – M641) agar, previously melted and cooled to about 45°C. Mix the contents thoroughly by rotating the plates and allow the agar to solidify. Invert plates and incubate the plates at 37 ±1°C for 48 h. After incubation count the colonies.

$$\text{Calculation: } N = \frac{\Sigma C}{(N_1 + 0.1N_2) D} \quad \text{or} \quad \frac{\Sigma C}{2.2 * d} \quad (8)$$

ΣC is the sum of colonies counted on all the dishes retained; N_1 is the no. of dishes retained in the first dilution; N_2 is the no of dishes retained in the second dilution; D is the dilution factor corresponding to first dilution.

2.8.6 Determination of *Lactobacillus acidophilus* count: (IS: 5401-2012).

Transfer 10 ml of well-mixed milk sample to 90 ml diluent (phosphate buffer solution) & mix well to make first dilution. If required make second dilution by transferring 1 ml of first dilution into 9 ml diluent. Arrange 2 Petri plates for each sample mark them with sample no. and date. Transfer 1 ml in each plate respective dilution of sample to be tested. Before pouring the media in plate adjusted the pH 6.5. Add 15 to 20 ml of MRS (Himedia code – M641) agar, previously melted and cooled to about 45°C. Mix the contents thoroughly by rotating the plates and allow the agar to solidify. Invert plates and incubate the plates at 37 ±1°C for 48 h. After incubation count the colonies.

$$\text{Calculation: } N = \frac{\Sigma C}{(N_1 + 0.1N_2) D} \quad \text{or} \quad \frac{\Sigma C}{2.2 * d} \quad (9)$$



ΣC is the sum of colonies counted on all the dishes retained; N1 is the no. of dishes retained in the first dilution; N2 is the no of dishes retained in the second dilution; D is the dilution factor corresponding to first dilution.

2.9 Sensory evolution of control samples of yogurt & probiotic mango yogurt (Ranganathan and Gupta, 2017).

To assure the flavour, the body and texture, colour and appearance of the product, the samples were subjected to sensory evaluation by adopting a 9-point Hedonic scale by expert committee members of sensory panel.

2.103.16 Statistical analysis.

The analysed samples were expressed as mean \pm standard deviation. Furthermore, statistical significance was tested by employing analysis of variance (ANOVA) and data comparison between means by Tukey's test pair wise comparison. For computation of data, software application programmers' life Microsoft Word, Excel and Prism Graph Pad (Prism version 10 for windows) were used. The data obtained were analysed statistically by the analysis of variance technique and critical difference.

Table -1 Basic analysis of Raw Standardized Milk (n= 30 days)

S. No	pH	Acidity%	Moisture%	Fat%	SNF%	TSS%	Protein%	Ash%
L-1	6.65 \pm 0.03	0.116 \pm 0.003	85.93 \pm 0.17	5.30 \pm 0.10	8.77 \pm 0.09	14.07 \pm 0.17	36.55 \pm 0.30	0.61 \pm 0.03
L-2	6.66 \pm 0.02	0.116 \pm 0.003	85.45 \pm 0.14	5.89 \pm 0.08	8.67 \pm 0.07	14.55 \pm 0.14	36.48 \pm 0.34	0.62 \pm 0.03
L-3	6.66 \pm 0.01	0.121 \pm 0.005	84.17 \pm 0.22	6.59 \pm 0.16	9.23 \pm 0.10	15.82 \pm 0.22	36.67 \pm 0.43	0.62 \pm 0.03
L-4	6.66 \pm 0.01	0.113 \pm 0.005	86.43 \pm 0.23	5.09 \pm 0.16	8.47 \pm 0.09	13.56 \pm 0.22	36.45 \pm 0.27	0.63 \pm 0.04
L-5	6.66 \pm 0.02	0.116 \pm 0.005	86.01 \pm 0.15	5.37 \pm 0.11	8.63 \pm 0.10	13.99 \pm 0.16	36.58 \pm 0.44	0.67 \pm 0.05
L-6	6.66 \pm 0.02	0.115 \pm 0.004	85.25 \pm 0.58	5.81 \pm 0.38	8.91 \pm 0.25	14.75 \pm 0.59	36.56 \pm 0.32	0.63 \pm 0.04
L-7	6.62 \pm 0.04	0.117 \pm 0.002	85.06 \pm 0.37	5.99 \pm 0.26	8.98 \pm 0.07	14.97 \pm 0.31	36.61 \pm 0.57	0.64 \pm 0.05
L-8	6.66 \pm 0.04	0.113 \pm 0.005	85.29 \pm 0.39	5.84 \pm 0.29	8.88 \pm 0.12	14.71 \pm 0.39	36.78 \pm 0.35	0.73 \pm 0.03
L-9	6.66 \pm 0.02	0.115 \pm 0.004	85.25 \pm 0.58	5.89 \pm 0.08	8.67 \pm 0.07	14.55 \pm 0.14	36.48 \pm 0.34	0.62 \pm 0.03
L-10	6.66 \pm 0.02	0.116 \pm 0.005	86.01 \pm 0.15	5.37 \pm 0.11	8.63 \pm 0.10	13.99 \pm 0.16	36.58 \pm 0.44	0.67 \pm 0.05



Note- L1- SAHAJ, L2- JONIHAN, L3- MAALV MAHILA, L4- LALGANJ, L5- DIBIYAPUR, L6- KANNAUJ, L7-
JANAHABAD, L8- GHATANPUR, L9- LAKSHYA FOOD, L10- VERKA

Table -2 Quality statistical analysis of Raw Standardized Milk (n= 30 days)

Sample no	RM	BR	Sodium (ppm)
L-1	29.02±0.27	41.55±0.39	434±8.18
L-2	28.88±0.20	41.60±0.41	437±8.38
L-3	29.05±0.36	41.63±0.47	558±10.26
L-4	28.76±0.20	41.47±0.25	451±9.26
L-5	28.81±0.24	41.50±0.37	442±8.56
L-6	28.85±0.28	41.60±0.28	447±11.08
L-7	29.07±0.18	41.38±0.35	440±5.69
L-8	29.36±0.59	41.31±0.50	432±18.02
L-9	28.76±0.20	41.60±0.41	597±8.38
L-10	28.88±0.20	41.60±0.28	587±11.08

Note- L1- SAHAJ, L2- JONIHAN, L3- MAALV MAHILA, L4- LALGANJ, L5- DIBIYAPUR, L6- KANNAUJ, L7-
JANAHABAD, L8- GHATANPUR, L9- LAKSHYA FOOD, L10- VERKA



Table- 3 Quantitative Statistical analysis of adulteration test of Raw Standardized Milk (n= 30 days)

Sample no	Melamine (ppb)	Aflatoxin M1 (ppb)
L-1	60.17±5.17	0.29±0.08
L-2	60.17±4.82	0.27±0.08
L-3	60.83±5.43	0.26±0.09
L-4	62.50±8.28	0.29±0.08
L-5	59.50±6.74	0.28±0.07
L-6	60.17±5.17	0.28±0.09
L-7	59.50±6.75	0.32±0.08
L-8	58.17±4.82	0.17±0.05
L-9	90.83±9.43	1.26±0.19
L-10	80.83±7.43	0.96±0.12

Note- L1- SAHAJ, L2- JONIHAN, L3- MAALV MAHILA, L4- LALGANJ, L5- DIBIYAPUR, L6- KANNAUJ, L7- JANAHAABAD, L8- GHATANPUR, L9- LAKSHYA FOOD, L10- VERKA



Table- 4 Microbiological Statistical analysis of Raw Standardized Milk (n= 60 days)

Sample no	MBRT (Minute)	Coliform (cfu/ml)	APC (cfu/ml)	Somatic Cell
L-1	97.17±9.89	565±226	146233±17947	112997±18249
L-2	78.9±6.32	2128±160	2248833±166669	325129±34590
L-3	49.83±10.58	2282±258	4201666±142322	3343516±22751
L-4	58.92±7.25	2980±338	3150000±115220	329096±29546
L-5	71.83±9.28	4181±125	3176666±160136	235354±24207
L-6	60.5±10.64	5939±187	3213333±165014	235096±22017
L-7	60.5±11.81	4695±132	3195000±195355	235354±23500
L-8	100.75±9.78	148±18	122000±16370	100903±15713
L-9	55.92±5.25	2980±338	4150000±115220	429096±29546
L-10	50.83±9.28	5081±115	4176666±160136	435354±24207

Note- L1- SAHAJ, L2- JONIHAN, L3- MAALV MAHILA, L4- LALGANJ, L5- DIBIYAPUR, L6- KANNAUJ, L7- JANAHAABAD, L8- GHATANPUR, L9- LAKSHYA FOOD, L10- VERKA



Table- 4a Microbiological statistical analysis of Raw Standardized Milk (n= 60 days)

Sample no	MScfu/ml	TScfu/ml	Psychotropiccfu/ml
L-1	33±20	3±1	29375±11100
L-2	57±26	3±1	40200±13058
L-3	54±35	2±1	46433±17872
L-4	64±31	3±1	36117±12626
L-5	40±25	3±2	40783±12590
L-6	50±18	3±1	38750±9593
L-7	40±26	2±1	40417±16844
L-8	22±8	2±1	2016±1896
L-9	90±25	20±2	66433±17872
L-10	74±31	15±2	56117±12626

Note- L1- SAHAJ, L2- JONIHAN, L3- MAALV MAHILA, L4- LALGANJ, L5- DIBIYAPUR, L6- KANNAUJ, L7- JANAHAABAD, L8- GHATANPUR, L9- LAKSHYA FOOD, L10- VERKA



Table- 5 Sensory Evaluation of Raw Standardized Milk by 9-point hydronic scale (n= 60 days)

Sample no	Score
L-1	7±0.68
L-2	6±0.70
L-3	6±0.79
L-4	6±0.81
L-5	6±0.34
L-6	6±0.68
L-7	6±0.66
L-8	8±0.58
L-9	5±0.90
L-10	5±0.99

Note- L1- SAHAJ, L2- JONIHAN, L3- MAALV MAHILA, L4- LALGANJ, L5- DIBIYAPUR, L6- KANNAUJ, L7- JANAHAABAD, L8- GHATANPUR, L9- LAKSHYA FOOD, L10- VERKA



3.0 Result and Discussion

Yogurt's quality is strongly influenced by the kind and calibre of the raw materials used in its production. While solids not fat content, regulate the product's physicochemical properties, compositional factors like fat content mostly determine the body, texture and flavour of the final product. The trend for the future is to add fruit pulp to the mix to enhance the dairy products' organoleptic quality. In addition to processing parameters, quality is crucial to standardizing the compositional parameters in order to acquire during this study. In the context of the aforementioned, the following is explored under. Different treatment' control and experimental yogurt's fat %, TS%, acidity %, wheying off, pH, flavour, colour and appearance, body and texture have all been examined. The results of the analysis shown in tables 1 to 9 for further statistical analysis of these data using the mean and standard deviation evaluation methods. Yogurt made from raw standardized blend milk that had 6% sugar and without mango pulp was supplied as T₀. T₁ was a raw standardized blend milk yogurt that has been sweetened with 6% sugar and 4% mango pulp. T₂ was served as raw standardized blend milk yogurt with 6% sugar and 6% mango pulp. T₃ was served as raw standardized blend milk yogurt with 6% sugar and 8% mango pulp. T₅ was offered as yogurt made from raw standardized blend milk with 6% sugar and 12% mango pulp.

3.1 Physico-chemical evaluation

The result shown the control samples (T₀) had an average fat % 4.53 and range of 4.41 to 4.67, experimental samples (T₁) had an average fat % 4.39 and range of 4.31 to 4.48, experimental samples (T₂) had an average fat% 4.28 and range of 4.22 to 4.36, experimental samples (T₃) had an average fat% 4.21 and range of 4.17 to 4.25, experimental samples (T₄) had an average fat% 4.15 and range of 4.09 to 4.19 and experimental samples (T₅) had an average fat% 4.01 and range of 3.96 to 4.09 respectively in table no (2). Total solid % in control sample (T₀) had an average of total solid % 18.88 and range of 18.28 to 19.27, experimental samples (T₁) had an average total solid % 19.79 and range of 19.06 to 20.99, experimental samples (T₂) had an average total solid % 20.51 and range of 19.21 to 21.31, experimental samples (T₃) had an average total solid % 23.18 and range of 23.09 to 23.21,



experimental samples (T₄) had an average total solid % 26.38 and range of 25.83 to 26.98 and experimental samples (T₅) had an average total solid % 28.45 and range of 27.97 to 28.78 respectively in table no (2). Total acidity in control yogurt (T₀) is typically 0.72 and ranges from 0.70 to 0.75, whereas the acidity of experimental yogurt (T₁) is typically 0.71 and ranges from 0.72 to 0.78. In the experimental yogurt, (T₂) is regularly 0.71 had a range of 0.70 to 0.75, (T₄) had an average of 0.81 and a range of 0.71 to 0.92, and (T₅) had an average of 0.88 as well as a range of 0.83 to 0.95. Above study shown that the acidity percentage of sample no T₃, T₄ and T₅ is suddenly increased after the six days of investigation respectively. It was evident from data in table no2 that the average pH of control yogurt (T₀) is 4.83 and a range of 4.50 to 5.30, while in the experimental yogurt (T₁) had an average of 4.68 and a range of 4.32 to 5.02. In the experimental yogurt (T₂) had an average of 4.65 and a range of 4.50 to 4.89 and in (T₃) had an average of 4.61 and a range of 4.50 to 4.82 and in (T₄) had an average of 4.54 and a range of 4.50 to 4.62 and in (T₅) had an average of 4.09 and a range of 4.0 to 4.15. Previous study on analysis of yogurt shown that the average compositional value of fat, SNF, acidity and pH were approximately 3.0 to 4.5, 8.5 to 9.50, 18.75 to 24.50 and 5.21 to 4.29 maximums respectively suggested by (Helal et al., 2018).

The result shown the control samples (T₀) had an average of wheying off method sample volume is 4.93 and range of 4.29 to 5.77, experimental samples (T₁) had an average of wheying off method sample volume is 5.09 and range of 4.68 to 5.50, experimental samples (T₂) had an average of wheying off method sample volume is 4.96 and range of 4.58 to 5.31, experimental samples (T₃) had an average wheying off method sample volume is 4.47 and range of 3.54 to 4.98, experimental samples (T₄) had an average wheying off method sample volume is 4.12 and range of 3.54 to 4.53 and experimental samples (T₅) had an average wheying off method sample volume is 4.75 and range of 3.49 to 5.71 respectively in table no (3).

3.2 Microbiological evaluation of yogurt.

The microbiological test result of yogurt and probiotic yogurt of control and experimental samples shown in table no 5. The average of coliform count was 0.55 cfu/ ml and range from 0 to 4 as the same average of LB count was 1.51×10^8 cfu/ml and range from 0.01×10^8 to 4.7×10^8 and average range of ST count was 1.67×10^8 cfu/ ml and range from 0.01 to 5.2 in yogurt samples. In Probiotic yogurt the bacterial strain is different, due to the activity of beneficial strain maintained the gut pH of small intestine. The average of coliform count was 1.44 cfu/ ml and range from 0 to 8 cfu/ml as the same average of BB count was 1.57×10^7 and range from 0.5×10^7 to 4.5×10^7 and average range of LA count was 1.78×10^8 cfu/ ml and range from 0.3 to 3.5. Both the same sample were analyzed up to self-life of the products like 28 days. During the startup of the study coliform was found up to 4 to 5 days after the six day of study coliform found absent both the sample. This study may be concluded that the beneficial bacterial strain like LB, ST, BB and LA are more abundant on the coliform strain and digested by enzymatic activity. This study shown that the count of LB, ST, BB and LA decreased slowly after the 8 to 9 days of incubation. After 13



days of incubation the counts of beneficial strain LB, ST, BB and LA decreased fast. The potency of bacterial strain should be less after 14 days of incubation. In this study there is no significant difference found. In previous study by (Lororia and Martin, 2017; Moon and Reinbold, 2014) reported that the one third of viable count of ST, LB, BB and LA decreased due to the long incubation period of yogurt sample. The motility and mobility rates if the viable count had been in slow motion after the seven days of incubation. These test result score of control and experimental yogurt samples mention respectively in figure no 2.

3.3 Sensory Evaluation

The data shown in table no 4, the average offflavour in control yogurt (T₀) is typically 6.78 and ranges from 5.96 to 8.1, whereas the average offflavour of experimental yogurt (T₁) is typically 7.33 and ranges from 6.52 to 8.10. In the experimental sample, (T₂) average was regularly 8.33 had a range of 7.70 to 8.65, (T₃) had an average of 8.47 and a range of 8.10 to 8.85, (T₄) had an average of 7.68 as well as a range of 7.15 to 8.33 and (T₅) had the average range of 6.11 and range from 5.88 to 6.45. Body and texture data shown in table no 4 that the average of control yogurt (T₀) is 7.88 and a range of 6.45 to 8.56, While in the experimental yogurt (T₁) had an average of 7.92 and an range of 7.12 to 8.45, (T₂) had an average of 8.37 and a range of 7.96 to 8.84, (T₃) had an average of 8.37 and a range of 8.10 to 8.90, (T₄) had an average of 7.54 and a range of 7.10 to 7.98 and in (T₅) had an average of 6.83 and a range of 6.10 to 7.66. of yogurt and probiotic yogurt. Colour and Appearance data shown in table 4, the average of experimental yogurt (T₁) had an average flavour rating of 7.42 and a flavour rating range of 6.42 to 8.21. This contrasts with the control yogurt (T₀) had an average flavour rating of 6.51 and flavour rating ranged from 5.45 to 7.28. In the experimental yogurt, (T₂) had an average score of 7.56 and a range of 6.14 to 8.28, (T₃) had an average score of 8.01 and a range of 6.0 to 8.64, (T₄) had an average score of 8.30 and a range of 7.95 to 8.67 and (T₅) had an average score of 8.31 and a range of 7.10 to 8.95. Previous study on analysis of probiotic yogurt shown that sensory evaluation of yogurt based on appearance, flavour and texture average of control (T₁) was 7.33 and treated sample (T₂, T₃ and T₄) were 6.45, 5.80 and 7.42) maximums respectively reported by (Fatima and Hekmat, 2020)

Conclusion

On the basis of the aforementioned data, it can be stated that mango pulp can be successfully used to make fruit yogurts. It processes all the attributes of a high-quality fruit yogurt. Mango pulp when added in yogurt made from standardized raw mix milk, enhances its flavour, body, texture, colour and appearance compared to yogurt made from natural standardized raw mix milk. Mango probiotic yogurt carrier properties when inoculated with probiotic

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stains *Lactobacillus bulgaricus*, *Streptococcus thermophilus*, *Bifidobacterium's* and *Lactobacillus acidophilus*. which attained and maintained mean viable counts of 10^8 CFU/mL in all mango pulp based treated thereby exceeding the minimum requirement of 10^6 CFU/mL for therapeutic effects. Mango probiotic yogurt has a higher total solid concentration as per physico-chemical and microbiological investigations. This characteristic improves how the body and texture of the batter are formed. Mango probiotic yogurt has less wheying off. This characteristic aids in the gel development of mango pulp probiotic yogurt. The microbiological analysis data shown that the probiotic strain is most abundant on coliform strain. During microbiological analysis of yogurt, the 1st day 6 to 7 cfu/gm of coliform count seem. But during the self-life study of yogurt up to 28 days the coliform colonies disappear after the 6th days of tested sample of yogurt. This may be concluded that the viable count of probiotic strain digested the coliform colonies due to the high potency power of probiotic strain and also help in the high self-life of product. In addition of 8% mango pulp in yogurt has strong qualities, acid flavour, smooth texture and softness. Increased mango pulp addition had a negative impact on the yogurt's flavour as well as its body and texture. In order to achieve the best results, 8% of mango pulp was advised to be added. After sensory evaluation the most of customer likely the 8% added mango pulp in the yogurt. The novel probiotic stains *Lactobacillus bulgaricus*, *Streptococcus thermophilus*, *Bifidobacterium's* and *Lactobacillus acidophilus* helping in relief the urogenital infection and solve the gastrointestinal problem in patients. Yogurt is a good supplemented functional food diet with beneficial bacterial viable strains which help in lactose intolerance peoples.

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