



A STUDY ON MICROBIAL AND SENSORY PROPERTIES OF DIETETIC FLAVOURED MILK

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ABSTRACT

The present study aimed with the use of artificial sweetener (Aspartame) as substitute for sugar at 25, 50, 75 and 100 percent level in flavoured milk preparation. In microbial quality analysis of dietetic flavoured milk samples, the standard plate count (cfu/ ml) of dietetic flavoured milk samples were 78×10^{-4} , 63×10^{-4} , 51×10^{-4} , 23×10^{-4} and 21×10^{-4} for control treatments T1, T2, T3, and T4 respectively. Analysis of data showed significant difference ($p < 0.01$) between control and treatments indicating that addition of aspartame tablets and sugar at different level produce significant change in the standard plate count. There was no growth of Coliform in control and treatments T1, T2, T3 and T4 indicating that addition of aspartame tablets and sugar at various levels dose not promote the Coliform growth. Analysis of data with regard to yeast and mold count (cfu/ gm) showed no significant difference between control and treatments indicating that addition of aspartame tablets and sugar at different level produce significant change in the yeast and mold count due to minimal processing of dietetic flavored milk during preparation and contain other ingredients rather than sugars. The sensory qualities such as color, flavor, and taste of the control and treatment dietetic flavored milk revealed no significant change in the sensory score of the product up to 75 percent replacement of sugar with aspartame.

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INTRODUCTION

Flavored milk is becoming an integral part of market milk industry because it has good consumer acceptance as a refreshing and nourishing milk beverage. Recent years have witnessed a large increase in the market penetration of flavored milk into the total fluid milk market (Repate *et al.* 2010). The manufacture of flavored milk offer dietary advantage. The milk in its natural form is not considered suitable for many people, for example, those suffering from heart ailments. Infant and sick persons also cannot properly digest full fat milk. All these problems can be counteracted by manufacturing low fat flavored milk. Skim milk is a byproduct of dairy industry. It can be used in the manufacturing of flavoured milk because of its unique nutritional value. Flavoured milk is a sweetened milk beverage, usually made using low fat milk, sugar, permitted colorings and artificial or natural flavors. Flavoured milk, in the Indian market, has a projected annual growth rate of 27% (Baisya, 2005). The flavoured milk market in India is dominated by popular brands like Amul, Britannia and Nestle and the major regional brands include Saras. The advantage of

including flavored milk in the diets of children is that, despite its added sugar content, flavored milk has a nutrient profile similar to that of plain milk, and the benefits of flavored milk outweigh the detriments of added sugar. Flavored milk contains the same essential nutrients as plain milk, including protein, calcium, potassium, phosphorus, vitamin A, vitamin B12, iodine, and riboflavin. Nowadays, human beings are permanently in contact with different compounds biologically active (Stubbs *et al.* 1996). These are medicaments, plant growth re- gulators, active components of food additives as antioxidants, sweeteners and many other com- pounds. Escalating prevalence of worldwide obesity and its correlation to other chronic diseases has led to low calorie and sugar free products to move into the main stream of food market. Moreover, people choose low sugar for health reasons owing to prevailing diabetic conditions or motivated by calorie consciousness. Artificial sweeteners are the compounds that mimic the effect of sugar on tongue. They generally pass through the human body unmetabolized without possessing any calorie (Bhardwaj and Beniwal, 2009). In general, nowadays preference among the people to assume a high degree of confidence in wholesomeness and safety of foods and low calorific diet than those based on high energy value. The present study envisages the use of artificial sweetener

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(Aspartame) as substitute for sugar at different level in flavoured milk preparation.

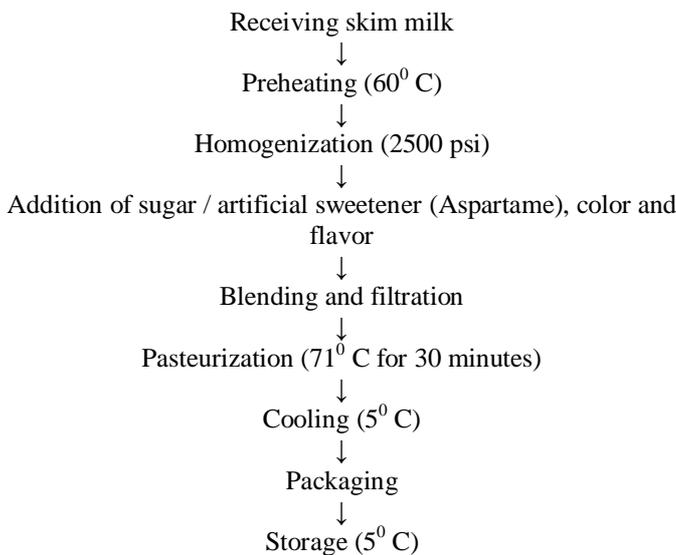
MATERIALS AND METHODS

Fresh skim milk collected from SNP dairy plant, Nagari, Madurai. Artificial sweeteners, (Aspartame), Sugar Free Natura, Cadila Health Care Ltd., Ahmadabad, India. Sugar purchased from local market and artificial flavour (Badam), OROR flavours and chemicals (P) Ltd, Madurai. The total bacterial count (SPC), Coliform count and yeast and mould counts were carried out by Plate count agar, Violet Red bile Agar and Potato Dextrose agar respectively were obtained as dehydrated media from M/s. Hi-Media Laboratories, Mumbai.

Methods of manufacture

The measured quantity of skim milk was taken. The milk was filtered through muslin cloth. Milk was preheated heated to 60°C and homogenized at 2500 psi. The milk is pasteurized at 75°C for 30 minutes and then it was allow to cool to 40°C after cooling, sugar/ Aspartame, color and flavors were added in milk. Milk was filled in bottle and capped then milk was stored in refrigerator. (De, 1980)

Flow diagram for preparation of Flavoured milk



Treatment Details

| Trial | Skim milk (lts) | Sugar(g) | Aspartame Tab (Nos) | Flavor Percentage | Color (ml) |
|-------------|-----------------|----------|---------------------|-------------------|------------|
| T0- Control | 1 | 80 | 0 | 0.5 | 0.5 |
| T1 | 1 | 60 | 4 | 0.5 | 0.5 |
| T2 | 1 | 40 | 8 | 0.5 | 0.5 |
| T3 | 1 | 20 | 12 | 0.5 | 0.5 |
| T4 | 1 | 0 | 16 | 0.5 | 0.5 |

Microbial Analysis

All the samples were subjected to microbiological analysis for Standard Plate Count (SPC), Coliform count and Yeast and Mould Count (YMC).

Preparation of diluents

Normal saline was used for serial dilution of samples. One ml of flavoured milk sample was transferred aseptically into 9 ml of sterile saline and serial dilutions made up to 10⁻⁹ dilution factor. One ml of appropriate dilution (10⁻⁶ to 10⁻⁹) was used for enumeration of bacteria and 10⁻¹ dilutions for Coliform count and 10⁻² yeast and mould count.

Standard Plate Count

Preparation of media

The standard plate count of the samples were analyzed as per the methods followed in IS: 5402 (1969). Plate count agar was used for this purpose. 1 ml of sample of appropriate dilution (10⁻⁴ to 10⁻⁶) was transferred in duplicate of sterile Petridis. Approximately 15-20 ml of the melted agar medium was then poured at 30 to 40 °C and mixed thoroughly by rotating the Petridis. After solidification invert the plates and incubate at 37°C ± 0.5 °C for 24 - 48 hours. The colonies were enumerated with colony counter.

Coliform Count (CFC)

The presence of Coliform was estimated as per the procedure described in BIS: 10. SP: (Part XI)-1981.

Yeast and Mould Count (YMC)

Yeast and mould counts were determined using Potato Dextrose Agar medium as per the procedure of IS: 5403 (1969)

Sensory Evaluation

The flavoured milk prepared under different treatment combinations were suspected to sensory evaluation by a panel of judges for colour and appearance, flavour consistency and mouth feel. The scoring was done on 9 point hedonic scale as described by (Gupta, 1976).

The scores ranged between 1 and 9, depending upon the liking. The scores were,

| Characters | Score |
|----------------------------|-------|
| Extremely liked | 9 |
| Liked very much | 8 |
| Liked moderately | 7 |
| Liked slightly | 6 |
| Neither liked nor disliked | 5 |
| Disliked slightly | 4 |
| Disliked moderately | 3 |
| Disliked very much | 2 |
| Extremely disliked | 1 |

Statistical Analysis

The data were tabulated and subjected to statistical analysis by means of analysis of variance (ANOVA) as per the procedure given by Snedecor and Cochran (1994).

RESULTS AND DISCUSSION

Prevention of food adulteration (PFA) rules, 1956 specifies microbial supplies for pathogens such as *E. coli*, *Staphylococcus aureus*, *Listeria monocytogenes* in foods frequently involved in food-borne diseases. According to standards given by PFA these microorganisms must be absent in one gram of milk and milk products (flavoured also). Hence we checked if the microorganisms were present or absent in one ml of the dietetic flavoured milk representing whether there was contamination or not.

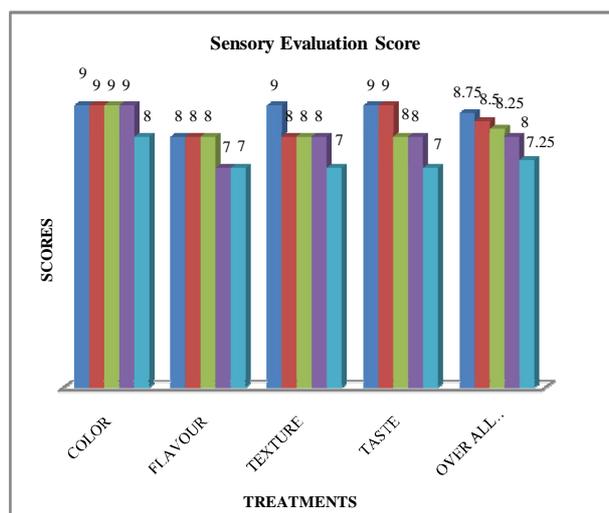
The standard plate count (cfu/ ml) of dietetic flavoured milk samples were 78×10⁻⁴, 63×10⁻⁴, 51×10⁻⁴, 23×10⁻⁴ and 21×10⁻⁴ for control treatments T1, T2, T3, and T4 respectively. The results of the samples were ranged from 21×10⁻⁴ to 78×10⁻⁴. Analysis of data showed significant difference (p<0.01) between control and treatments indicating that addition of aspartame tablets and sugar at different level

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produce significant change in the standard plate count due to minimal processing of sugars during preparation and presence of other ingredients rather than sugars.

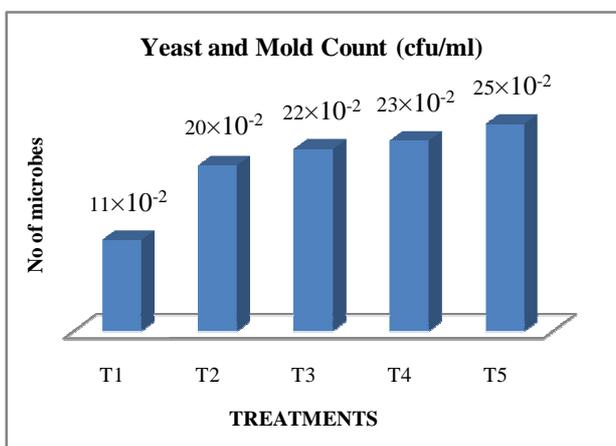
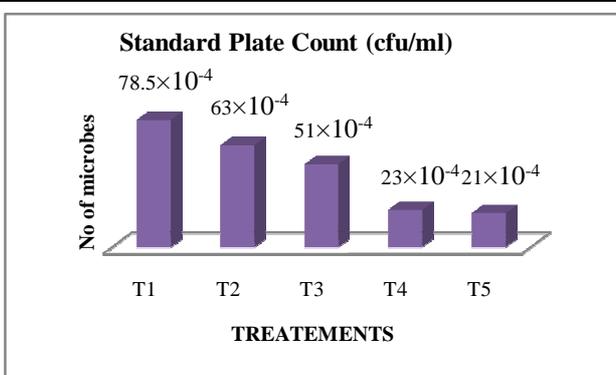
Table Microbial and Sensory analysis of flavoured milk

| Sl. No | Parameter | Values | |
|--------|---|----------|------------------------------|
| 1 | Standard Plate Count (cfu/ml) | RANGE | 21.0 – 78.5×10 ⁻⁴ |
| | | SED | 10.5789 |
| | | CD (.05) | 22.5485 |
| | | CD (.01) | 31.1735 |
| | | CV % | 31.63 |
| 2 | Yeast and Mould Count (cfu/ml) | RANGE | 11 – 25×10 ⁻² |
| | | SED | 4.1990 |
| | | CD (.05) | 8.9501 |
| | | CD (.01) | 12.3736 |
| | | CV % | 29.40 |
| 3 | Sensory Evaluation Score- Color | RANGE | 8.0 - 9.0 |
| | | SED | 2.4087 |
| | | CD (.05) | 4.8574 |
| | | CD (.01) | 5.8503 |
| | | CV % | 17.68 |
| 4 | Sensory Evaluation Score- Flavor | RANGE | 7.0-8.0 |
| | | SED | 3.4503 |
| | | CD (.05) | 13.5784 |
| | | CD (.01) | 34.4378 |
| | | CV % | 19.54 |
| 5 | Sensory Evaluation Score- Texture | RANGE | 7.0-9.0 |
| | | SED | 10.4865 |
| | | CD (.05) | 22.6387 |
| | | CD (.01) | 30.4397 |
| | | CV % | 29.46 |
| 6 | Sensory Evaluation Score- Taste | RANGE | 7.0-9.0 |
| | | SED | 6.4379 |
| | | CD (.05) | 22.6548 |
| | | CD (.01) | 31.4377 |
| | | CV % | 31.54 |
| 7 | Sensory Evaluation Score- Overall Acceptability | RANGE | 7.25-8.75 |
| | | SED | 18.4367 |
| | | CD (.05) | 29.3097 |
| | | CD (.01) | 46.7679 |
| | CV % | 18.64 | |



There was no growth of coliform in control and treatments T1, T2, T3 and T4 indicating that addition of aspartame tablets and sugar at various level dose not promote the growth of coliform and the samples were analyzed for the count immediately after preparation may be the reason for the absence of growth. Analysis of data indicating that addition of replacement of aspartame and sugar at any level does not promote the growth of Coliform bacteria. The results were in accordance with the report of Prevention of Food Adulteration (PFA) rules, the standards given by Prevention of Food Adulteration Rules, (1956) that *E. coli* and *S. aureus* should be absent in 1 gram of milk and milk products. The yeast and mold count (cfu/ ml) of dietetic flavoured milk samples were 11×10⁻², 20×10⁻², 22×10⁻², 23×10⁻² and 25×10⁻² for control treatments T1, T2, T3, and T4 respectively. The results of the samples were ranged from 11×10⁻² to 25×10⁻². Analysis of data showed significant difference between control and treatments indicating that addition of aspartame tablets and sugar at different level produce significant change in the yeast and mold count due to minimal processing of dietetic flavoured milk during preparation and presence other ingredients along with sugars. The result obtained was similar to the report of Sailaja Palthur *et al.* (2014).

The flavoured milk prepared under different treatment combinations were suspected to sensory evaluation by a panel of judges for color and appearance, flavor consistency and mouth feel. The scoring was done on 9 point hedonic scale as described by (Gupta 1976). Analysis of the data with regard to flavor score of dietetic flavoured milk revealed no significant difference between control and treatments indicating that addition of aspartame tablets and sugar different level produce a marked change in the flavor of the dietetic flavoured milk. The mean with regard to flavor scores of dietetic flavoured milk samples were 8, 8, 8, 7 and 7 for control treatments T1, T2, T3 and T4 respectively. The results of the samples were ranged from 7 to 8 per cent. But the flavor score of dietetic flavoured milk samples with 25 and 50 percent replacement of sugar with aspartame tablets had similar scores as that of control. Charanjiv Singh *et al.* (2005) reported that types of milk and the variety did not influence the organoleptic scores of flavoured milk beverage to a significant extent.



Sensory Evaluation of Dietetic Flavoured Milk

The results of textural characters of dietetic flavoured milk samples were ranged from 7 to 9. Highly Significant difference in the body and textural scores of dietetic flavoured milk samples was observed. The data with regard to T4 had very low score for texture indicating that 100 percent replacement of sugar with aspartame tablets produce loose body and varying degree of precipitation of flavoured milk noticed resulting in lower score value of the product.

The data with regard to color scores of dietetic flavoured milk samples were 9, 9, 9, 9, and 8 for control and treatments T1, T2, T3, and T4 respectively. The results of the samples were ranged from 8 to 9. Analysis of the data revealed significant difference in the score of color scores of dietetic flavoured milk samples was observed among all the samples. Difference in the color and appearance score of dietetic flavoured milk samples might be due to wide variation in raw material, amount of sugar added, reaction between sugar and amino acid produce maillard browning, method (direct) of preparation and duration of heat treatment used for manufacture of dietetic flavoured milk. The score for taste characters of dietetic flavoured milk samples were 9, 9, 8, 8 and 6, for control treatments T1, T2, T3, and T4 respectively. The results of the samples were ranged from 6 to 9. Highly Significant difference in the taste scores of dietetic flavoured milk samples was observed. The data with regard to T4 had very low score for taste indicating that 100 percent replacement of aspartame tablets produce unacceptable taste resulting in lower score value.

The result of the present investigation was in close agreement with the report of Jothylingam and Pugazhenth (2013) revealed the sensory evaluation, dietetic herbal flavoured milk up to 75 % replacement of sugar with aspartame (TA3) and 100 % replacement of sugar with sucralose (TS4) were concluded as the ideal, as they attained maximum score for overall acceptability. There was no difference in over all acceptability among the control and different dietetic herbal flavoured milk with artificial sweeteners due to storage effect. The marketing of dietetic flavored milk mainly depend upon the sensory attributes of the product. The sensory qualities such as color, flavor, and taste of the control and treatment dietetic flavored milk were evaluated by a panel of five judges. The analysis of the data revealed no significant change in the sensory score of the product up to 75 percent replacement of sugar with aspartame.

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