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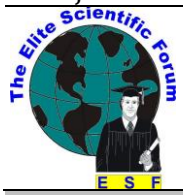

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RESEARCH ARTICLE

Physicochemical Quality and Sensory Attributes of Soy Milk Yogurt Blended with Buffalo Milk

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ABSTRACT

Soy milk and soy yogurt are not commonly used in Pakistan and consumers have no awareness about these products. Present study was designed for the development of soy yogurt using different concentrations of buffalo milk to decrease the beany taste of soy milk and soy yogurt. Properly cleaned soy beans were boiled at 100°C for 30 min. After dehulling cleaned with cold distilled water, grinded with water (1:3, w/v) and mixture was sieved. In manufacturing of soy yogurt, buffalo milk and soy milk were pasteurized for 30 minutes at 85°C with constant stirring to kill all pathogenic microorganisms, then cooled to 42°C and commercial starter culture was then used for the production of soy yogurt. Throughout storage study soy yogurt were assessed for its physico-chemical quality parameters (Protein, fat, acidity, pH, total solid, ash) and sensory evaluation during 21 days storage with seven days interval. All results obtained analyzed through two ways Analysis of Variance (ANOVA) using Mini Tab software. Highest score was awarded to T_0 with high values of fat% (4.48±0.01), Protein% (4.15±0.05), Total Solids% (14.49±0.03), Ash% (0.70±0.01), Acidity (0.91±0.01) and pH (4.52±0.01) and the results indicated significant effects ($P<0.05$) of storage on soy yogurt sensory attributes.

INTRODUCTION

Yogurt is considered as one of the earliest fermented products, which is liked and consumed all around the world. Yogurt is a fermented milk product produced by the action of lactic acid producing bacteria *Streptococcus thermophillus* and *Lactobacillus delbrueckii* ssp *bulgaricus* (Walstra et al., 1999). Soy bean milk is very nutritious and a healthy product as it contains no cholesterol, no lactose, low fat and high proteins. Soy milk is obtained from soybean. It plays an important role to fight against many diseases like hypercholesterolemia, cancers, menopause symptoms, helps in control osteoporosis, and heart diseases. Because of these qualities it can play a vital role in the field of medicine. Soy milk in many countries may be used as a substitute for cow milk. According to the local preferences it is sweetened and flavored. Soy-beverages are marketed in Taiwan, Hong Kong, Japan, Thailand & Malaysia (Rozina, 2012). By increasing the probiotics amount, use of fermented soymilk improves the intestinal tract function (Chang et al., 2005). Many people do not like particular beany flavor of soy milk. Due to the undesirable "beany" taste soy products have limitations in the Western culture as reported by Buono et al. (1990) and Favaro et al. (2001). Lactic acid reduces the "beany-flavor" through fermentation (Favaro et al., 2001). The people having low income accepted soy yogurt as alternative to the dairy milk yogurt at home level, as it is considered as a cheap raw material along with a good protein source. Soy milk and soy yogurt have been used as a drink and food due to its good compositional attributes, absence of cholesterol, higher protein contents, raw materials are easily accessible in markets and with the use of simple technologies they can be easily processed (Haenlein, 1996). In Pakistan buffalo milk is easily available and in excess quantity. In order to mask the beany taste/flavor of soy milk, present study has been designed to blend soy milk with buffalo milk to check an optimum level of buffalo milk to increase the acceptability and consumer demand of soy milk yogurt.

MATERIALS AND METHODS

The study has been carried out in the University of Veterinary & Animal Sciences, Department of Dairy Technology, (Ravi Campus), Pattoki. The starter culture namely (*Lactobacillus bulgaricus*) and (*Streptococcus thermophillus*) were obtained from Sacco-Clerici Italy (SAAF International, Dairy food pharma Lahore). Soy beans were procured from local market, Lahore, Pakistan. Fresh buffalo milk procured from Dairy Animals Training & Research Center, Pattoki. Whereas plastic cups from local market have been purchased for shelf life study.

Preparation of Soy Milk

Soy beans have been cleaned properly and boiled for 30 minutes at (100°C). De-hulling was performed with hand rubbing. The de-hulled soy beans were then washed with cold distilled water. Prepared soy beans were then grinded in blender with (13, w/v) soy beans to water ratio for 03 to 05 minutes. In sterilized muslin cloth, mixture was then sieved. After sieving soy milk boiled for (10) minutes (Masamba and Ali, 2013) to remove any microbial contaminations.

Preparation of Soy milk blended with buffalo milk

T1 and T4 were made with pure buffalo and soy milk respectively. While the blends of other treatments were made with different concentrations of soy milk (25%, 50% and 75%) and buffalo milk (75%, 50% and 25%) respectively, then refrigerated at (4°C) till further use.

Preparation of Soy yogurt

The soy milk, buffalo milk and blends warmed upto (43 to 45°C). Then sugar (2%) and gelatin (0.04%) were added to improve its acceptability profile and mixed well. Milk was again heated for 30 minutes at 90°C. After pasteurization, blends were cooled to 45°C and *thermophillic* culture was inoculated as per manufacturer's description. The cultured milk was then filled in (200 mL) plastic cups and all soy yogurt samples were incubated at 43°C for 04 to 06 hours as described by Masamba and Ali (2013). The Prepared samples of yogurt were then stored in refrigerator at (4°C).

Physico-chemical analysis

Fat, protein, TS, acidity, ash and pH of soy yogurt were analyzed by the methods as prescribed by (AOAC, 2000).

Sensory evaluation

Sensory evaluations were done at 0, 7, 14 and 21 days for the parameters of taste or flavor, color, appearance, and overall acceptability by panel of judges using nine point hedonic scale according to Peryam et al. (1952). All yogurt samples in transparent cups were labeled with 03 digits random code. The yogurt samples for sensory evaluation were given to the panelists and were asked to score them.

Microbiological examination

Soy yogurt samples were then examined for total plate count (TPC) according to (Marshall, 1992).

Sterilization of glassware and media

Total plate count media was prepared in distilled water and then autoclaved for 15 minutes at 121°C at 15 lbs pressure for sterilization purpose. The glassware's such as beakers, petri plates, pipettes were cleaned, and then sterilized at (180°C) for 2 to 3 hours using hot air oven.

Sample preparation

In test tubes phosphate buffer solution was prepared and poured (9ml) as Blank. In first blank (one gram) yogurt sample was taken then shaked test tube for 25 times and then transferred (1ml) in next test tube. Same procedure was continued for remaining test tubes.

Total plate count (TPC)

To calculate microbial count total plate count media was used for the soy yogurt samples. Each sample mixed very well and then next dilution was prepared. The sterilized petri-dishes inoculated with (1 ml) sample carefully obtained from prepared dilutions. The plate count agar was then transferred and mixed it properly. Incubation started for 48 hours at 32°C. Then the plates having colonies 20 to 200 were chosen for counting through Colony counter. The total number of bacterial colonies/ml were calculated by multiplying the dilution factor with number of colonies.

Statistical Analysis

Using two-way analysis of variance, the data collected was statistically analyzed and determine the treatment effect on storage (Steel et al. 1997) by using SAS 9.1 statistical software.

RESULTS AND DISCUSSION

Soy yogurt samples were investigated for physicochemical attributes and the parameters were (protein, fat, total solid, pH, acidity and ash) including sensory evaluation during storage study of (21 days) with 7 days interval. Soy yogurt samples prepared with different combinations of buffalo milk, were kept at refrigeration temperature for storage study. Results indicated that fat, ash contents and pH were continuously dropped while total solid, protein and acidity values showed constant increase in all the treatments.

Physico-chemical parameters

Fat % of soy yogurt blended with buffalo milk

The mean values of different treatments for fat of soy yogurt observed in (Table 1), all the treatments varied from each other. Highest values were recorded significantly in (T_0) 4.48% and the lowest value was noted 0.47 in (T_4). It was observed that fat contents were slightly decreased during 21 days of storage and statistically it was non-significant. Analysis of variance showed that the effects of storage on treatments were found non-significant and the interaction of treatments showed significant difference to each other. Present study results of soy yogurt are in line with the results of

Amanze and Amanze (2011), they reported for fat contents (1.3%). In present study results regarding fat contents were higher because pure buffalo milk was utilized for the blends of yogurt.

Protein% of soy yogurt blended with buffalo milk

The protein contents of soy yogurt of different treatments are shown in (Table 1). The Protein contents slightly decreased and were statistically non-significant during storage of (21 days) but the different treatments showed significant difference to each other. Decline in protein contents in present study may be due to the proteins breakdown by lactic acid during storage of 21 days. The results are very close to the results of Amanze and Amanze (2011) they reported in soy yogurt 2.02% protein and 2.7% in soy/cow yogurt; in present research the results are higher due to the blends with buffalo milk. Whereas throughout storage period significant decrease in protein contents was observed, due to the culture and residual coagulants remain viable and this assisting the hydrolysis of proteins. A relatively higher free moisture content presented during the storage period favors the hydrolysis and hydration of the proteins and this probably due to the reason for reduced protein contents throughout storage.

Total Solids % of soy yogurt blended with buffalo milk

The mean value regarding total solids of soy yogurt in different treatments are shown in (Table 2). Significantly highest total solid contents noted in (T_0) and lowest value was recorded in (T_4). Different treatments showed significant difference to each other. Present results are significant with the finding of Amanze and Amanze (2011) they reported 9 to 10%. In present results decline in the total solid contents during storage study was due to the action of lactose fermenting bacteria and conversion of lactose into the lactic acid.

Ash% of soy yogurt blended with buffalo milk

Ash contents of soy yogurt in different treatments are provided in (Table 2). It indicated that entire treatments varied significantly from each other. In (T_4) lowest values 0.44 were recorded. The ash contents approximately remained the same during the storage period of 21 days and were not influenced by the time period. Effects of storage on ash% were non-significant and the interaction of treatments and storage was found non-significant. Ash contents in food stuffs depicted inorganic residues which are residual after organic matter has been burnt away.

Acidity % of soy yogurt blended with buffalo milk

The acidity was increased during the (21 days) storage period in all treatments. The maximum increase (1.07%) in acidity (Table 3) was noted in (T_0) and minimum was observed in (T_4) due to the growth of psychrophilic bacteria at (4°C) there may be increase in acidity% by converting lactose into the lactic acid. In present study storage effect on treatments was highly significant and interaction was also found significant among treatments and storage.

Table 1: Mean values for Fat and Protein of soy yoghurt blended with buffalo milk

Treatment	Fat %				Protein %			
	Day 0	Day 7	Day 14	Day 21	Day 0	Day 7	Day 14	Day 21
T_0	4.48 ± 0.01^{a}	4.48 ± 0.03^{a}	4.48 ± 0.05^{a}	4.47 ± 0.12^{a}	4.15 ± 0.05^{a}	4.13 ± 0.01^{a}	4.15 ± 0.08^{a}	4.12 ± 0.02^{a}
T_1	4.18 ± 0.13^{a}	4.19 ± 0.04^{a}	4.19 ± 0.09^{a}	4.18 ± 0.33^{a}	3.75 ± 0.03^{b}	3.74 ± 0.01^{b}	3.74 ± 0.06^{b}	3.73 ± 0.06^{b}
T_2	3.10 ± 0.14^{b}	3.11 ± 0.01^{b}	3.11 ± 0.01^{b}	3.11 ± 0.15^{b}	3.56 ± 0.07^{c}	3.54 ± 0.02^{c}	3.54 ± 0.04^{c}	3.53 ± 0.04^{c}
T_3	1.79 ± 0.17^{c}	1.79 ± 0.02^{c}	1.79 ± 0.12^{c}	1.78 ± 0.05^{c}	3.18 ± 0.11^{d}	3.17 ± 0.03^{d}	3.17 ± 0.01^{d}	3.16 ± 0.05^{d}
T4	0.49 ± 0.33^{d}	0.48 ± 0.09^{d}	0.48 ± 0.04^{d}	0.47 ± 0.19^{d}	2.81 ± 0.06^{e}	2.78 ± 0.04^{ef}	2.78 ± 0.00^{ef}	2.76 ± 0.02^{f}

*Rows and Columns with the same letter having non-significant difference.

Table 2: Mean values for Total Solids and Ash of soy yoghurt blended with buffalo milk

Treatment	Total Solid %				Ash %			
	Day 0	Day 7	Day 14	Day 21	Day 0	Day 7	Day 14	Day 21
T_0	14.49 ± 0.03^{a}	14.46 ± 0.33^{ab}	14.45 ± 0.03^{ab}	14.44 ± 0.14^{ab}	0.70 ± 0.01^{a}	0.68 ± 0.05^{ab}	0.65 ± 0.03^{bc}	0.63 ± 0.03^{dc}
T_1	14.30 ± 0.01^{ab}	14.28 ± 0.15^{ab}	14.26 ± 0.22^{b}	14.24 ± 0.13^{b}	0.69 ± 0.03^{a}	0.67 ± 0.02^{ab}	0.63 ± 0.02^{dc}	0.61 ± 0.02^{de}
T_2	13.12 ± 0.11^{c}	13.06 ± 0.11^{c}	13.04 ± 0.20^{c}	13.03 ± 0.17^{c}	0.62 ± 0.05^{dc}	0.60 ± 0.01^{de}	0.58 ± 0.05^{ef}	0.55 ± 0.02^{fg}
T_3	12.06 ± 0.01^{d}	12.03 ± 0.15^{d}	12.02 ± 0.17^{d}	12.00 ± 0.16^{d}	0.56 ± 0.03^{fg}	0.53 ± 0.01^{hg}	0.51 ± 0.01^{hi}	0.48 ± 0.03^{ji}
T4	11.69 ± 0.09^{e}	11.67 ± 0.09^{e}	11.66 ± 0.15^{e}	11.64 ± 0.12^{e}	0.51 ± 0.02^{hi}	0.49 ± 0.02^{ji}	0.46 ± 0.02^{jk}	0.44 ± 0.01^{k}

*Rows and Columns with the same letter having non-significant difference.

Table 3: Mean values for Acidity and pH of soy yoghurt blended with buffalo milk

Treatment	Acidity%				pH			
	Day 0	Day 7	Day 14	Day 21	Day 0	Day 7	Day 14	Day 21
T_0	0.91 ± 0.01^{dc}	1.00 ± 0.03^{ab}	1.04 ± 0.01^{a}	1.07 ± 0.00^{a}	4.52 ± 0.01^{a}	4.44 ± 0.02^{bc}	4.30 ± 0.07^{f}	4.22 ± 0.02^{g}
T_1	0.90 ± 0.01^{dce}	1.01 ± 0.05^{a}	1.02 ± 0.07^{a}	1.05 ± 0.02^{a}	4.51 ± 0.01^{a}	4.40 ± 0.04^{d}	4.33 ± 0.02^{ef}	4.12 ± 0.01^{i}
T_2	0.79 ± 0.03^{f}	0.83 ± 0.03^{dfe}	0.88 ± 0.01^{dce}	0.92 ± 0.02^{bc}	4.52 ± 0.04^{a}	4.41 ± 0.04^{cd}	4.31 ± 0.02^{f}	4.11 ± 0.02^{i}
T_3	0.69 ± 0.05^{hi}	0.72 ± 0.04^{gh}	0.79 ± 0.04^{gf}	0.82 ± 0.02^{fe}	4.51 ± 0.02^{a}	4.45 ± 0.03^{b}	4.35 ± 0.03^{e}	4.16 ± 0.05^{h}
T4	0.59 ± 0.01^{j}	0.63 ± 0.05^{ji}	0.67 ± 0.02^{jhi}	0.71 ± 0.01^{ghi}	4.52 ± 0.02^{a}	4.43 ± 0.01^{bcd}	4.32 ± 0.05^{ef}	4.19 ± 0.01^{gh}

*Rows and Columns with the same letter having non-significant difference.

Table 4: Total plate count of soy yoghurt blended with buffalo milk

Treatments	Storage Days			
	Day 0	Day 07	Day 14	Day 21
T_0	$7.6x10^6 \pm 0.33^j$	$1x10^7 \pm 0.07^h$	$8.5x10^6 \pm 0.05^i$	$3.2x10^6 \pm 0.04^m$
T_1	$1.1x10^7 \pm 0.23^g$	$8.0x10^8 \pm 0.26^a$	$7.1x10^7 \pm 0.09^c$	$4.1x10^6 \pm 0.33^l$
T_2	$1.2x10^7 \pm 0.13^f$	$5.5x10^7 \pm 0.09^d$	$6.7x10^6 \pm 0.21^k$	$1.9x10^6 \pm 0.01^n$
T_3	$1.2x10^7 \pm 0.17^f$	$3.8x10^8 \pm 0.33^b$	$3.9x10^7 \pm 0.35^e$	$3.0x10^6 \pm 0.15^n$
T_4	$1.1x10^6 \pm 0.15^o$	$6.7x10^6 \pm 0.23^k$	$1.2x10^6 \pm 0.08^o$	$3.0x10^6 \pm 0.30^n$

*Rows and Columns with the same letter having non-significant difference.

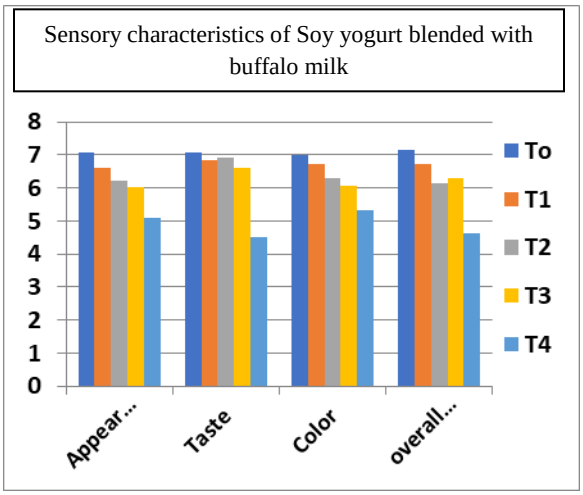

Fig. 1: Treatment effect on Soy yogurt sensory characteristics.

pH of soy yogurt blended with buffalo milk

The mean value for pH of soy yogurt blended with buffalo milk presented in (Table 3). Results showed that all treatments varied significantly from each other. In (T_0) and (T_2) the highest pH value was noted i.e. 4.52 and significantly lowest value recorded 4.11 in (T_2). The pH trend declined during the (21 days) storage period. Results of present study are in line with findings of Manhal and Kamal (2010). It was observed that during storage the pH, acetaldehyde and whey separation decreased whereas lactic acid was increased.

Microbiological Examination

Microbiological examination of soy yogurt blended with buffalo milk total plate count was conducted using Plate Count Agar during storage on (0, 7, 14 and 21 days). Soy yogurt blended with buffalo milk was evaluated for total plate count. The highest microbial count observed in (T_0). The results of total plate count are given in (Table 4). All the treatments showed significant results to each other and the storage effect also showed significant difference with treatments. Microbial count was reduced due to the increase in acidity% but still it was within acceptable range reported by Ali et al (2013).

Sensory Evaluation

Overall acceptability is depending on various quality characteristics like taste, flavor, appearance, color and texture (Fig. 1) revealed that in all treatments there was decline in overall acceptability. Results of overall acceptability with treatments, days and interaction were found (P=0.000) statistically highly significant. The highest score was awarded to (T_0), there were significant effects of storage observed on sensory attributes of the soy yogurt.

It is concluded that soy yogurt can be adopted as a substitute to the dairy milk yogurt because it has several therapeutic values and it is a good protein source. The "beany" flavour of soy milk and soy yogurt can be abridged and acceptability could be improved by the addition of 50% buffalo milk. The overall acceptability of soy milk and soy yogurt can also be increased with the addition of sugar, honey and other sweeteners.

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Author's contribution

SI designed the experimental study and wrote the paper. AH conducted the trial. IJ, MJ, IA, AI and MA helped in trial, statistical analysis and manuscript write up. All authors read and approved the final manuscript.

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