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Development of novel herb supplemented soymilk fortified fruit based dairy yoghurt

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Abstract
Value added products based on combination of dairy & plant derived foods provide ample scope to satisfy rising demand for health products. The study was carried out to develop novel Herb Supplemented- Fruit based- Soy milk fortified- Yoghurt. Toned milk was used as base material, and was fortified with different concentrations of soy milk (10,20,30,40,50%), mango pulp (5,10,15,20%) &*Tulsi* herb (Holy Basil) leaf extract (0.5,1,1.5%). Based upon sensory evaluation the level of ingredients was selected. Yoghurt prepared using 10% soy milk 15% mango pulp & 1% Tulsi leaf extract gave best sensory score. The novel yoghurt samples were analysed for physicochemical properties viz. pH, titrable acidity, water activity, Total soluble solids, ash, carbohydrate, fat, antioxidant activity and protein and was compared with market available sample of mango fruit Yoghurt. The novel product found good acceptability. The protein content & antioxidant activity for novel yoghurt was found to be higher whereas fat, carbohydrate content was lower which can be attributed to soy milk

Keywords: Dairy, yoghurt, soy, tulsi, fruit yoghurt, sensory evaluation

Introduction
Increasing consumer awareness, changing lifestyle makes it vital to develop new products which can capture consumer's imagination, have greater convenience and provide for better nutrition. About 7% of the milk produced in India is used for the manufacture of fermented dairy products (Sarkar, 2008) [22]. In recent past demand for yogurt and milk based fermented products has also increased because of increasing health consciousness of consumer. Yogurt market in India grew at a CAGR of 28.9% during 2011-2015 (Tech Sci Research Report). One of the major challenges for dairy sector is to develop dairy foods that promote health and nutrition. Yoghurt contributes to beneficial microorganism that helps in improving gut health. The microbial ecology in the Gastro Intestinal Tract (GIT) influences many functions in our body such as digestion. All these aspects make yoghurt a good choice for the development of functional foods. In recent years, many different food ingredients, including green and black teas (Jaziri *et al.* 2009) [10], lentil flour (Zare *et al.* 2011) [26], nano-powdered chitosan (Seo *et al.* 2009) [23], dietary fiber (Staffolo *et al.* 2004) [24], and evening primrose oil (Lee *et.al.* 2006) [13], have been included in yogurt formulations to improve nutritional value.

India has a vast horticulture base with a wide range of fruit but post harvest losses of semi perishables and perishables in India are also amongst highest in the world, this call for creation of new avenues for the utilization of fruits in an effective manner. Incorporation of fruits in to fermented milk products would generate a great demand for processed fruits, which might help checking the post-harvest losses and the economic loss to the nation and would enhance the profitability of milk and fruit producer as well as processors. So there is great scope for value added products based on combination of dairy & plant food amidst rising demand for health products. Mango is king of fruits and national fruit of India grown all across tropical and subtropical parts of the country. It is processed to make pulp & thus reduce wastage. It is a rich source of antioxidants.

Soybeans, contain a high nutritional value, it is used for making soy milk, and is a popular food all over the world (Muller *et al*., 1998) [15]. Soy milk is commonly processed into yoghurt, tofu, aburage and yuba (Hammond and Jez, 2011) [7]. Soybean protein contain acceptable amount of essential amino acids which are recommended for daily intake as a balanced diet (Erdman and Fordyce, 1989) [4]. Soybean has been reported to impart several health benefits such as improvement in bone mineral density (Kreijkamp-Kaspers *et al*., 2004) [12], lowering of plasma cholesterol (Anthony *et al*., 1996) [1], prevention of cancer (Kennedy, 1998) [11], and provide protection against bowel and kidney disease (Friedman and Brandon, 2001) [6] because

of the presence of isoflavone, saponins, protein and peptide in soybean (Friedman and Brandon, 2001; Michelfelder, 2009; Xiao, 2008) [6, 14, 25]. Soy milk has 3-4% protein, 1.5-2.0% fat and 5-6% carbohydrates. Soymilk-based yogurt, namely, soy yogurt or sogurt, is produced by the fermentation of soymilk using lactic acid bacteria. Because of its beany flavour, insufficient acidity, hard and coarse texture (S.Y.Lee, 1990) [21], sogurt is not widely accepted by consumers. Great efforts have been made by several researchers to improve the flavour and textural properties of sogurt, for instance by calcium fortification (F. Yazici, 1997) [5], microware treatment (S. Bhattacharya, 2007) [20], ultra-high pressure homogenization (N.S. Cruz, 2009) [16], the addition of mango pulp (P. Kumar, 2003) [19] or inulin with raffinose and glucose (O.N. Donkor, 2007) [17].

Materials and Methods

Materials

The research work was conducted in the Animal Product Technology Lab & Food Science Technology Laboratory, at National Institute of Food Technology Entrepreneurship & Management (NIFTEM), Haryana. Mother Dairy brand market milk of Toned variant was used as base material for developing the novel product. Fresh, green Tulsi leaves (*Ocimum sanctum*) were collected from NIFTEM Campus, Sonepat. Soy milk was prepared in lab from soybean procured from local market. Amul Sagar brand Skim Milk Powder & sugar were also purchased from local market. Freeze dried culture of *Lactobacillus bulgaricus* and *Streptococcus thermophilus* were obtained from CHR Hansen, Gurgaon. Superior quality sweetened mango pulp was procured from USFDA FCE (Food Canning Establishment) registered Swad Alphonso Mango Pulp.

Soy milk preparation

Soymilk was prepared as per the method described by Ojha *et al*. (2015) [18]. Approximately 200 gm soybean split was prepared from cleaned beans by soaking in water for half an hour and then dried in sunlight for 2 days. The soybean split was prepared by breaking in stone miller. This soybean split was then cleaned. The purpose of soybean split making was to dehull the beans. The dehulled soybean split were soaked in slightly warm water (of 45 ºC) at room temperature for a period of 16 hours and grinded with water at a bean to water ratio 1:9. The resultant slurry was strained through 3 layered muslin cloth to remove soy okara. The digital refractometer was used to measure brix of soymilk.

Preparation of Novel Yogurt:

Novel yogurt was manufactured using toned market milk of Mother Dairy brand. In order to boost the SNF level of milk to 12.5 %, skim milk powder was used. This milk sample was replaced with 10%, 20%, 30%, 40% and 50% concentrations of soymilk of TS 9% and on basis of sensory evaluation the content of soy milk was optimized. The optimized soy milk replaced base was added with 5, 10, 15, 20% mango pulp to determine the optimum rate of mango pulp addition. Tulsi Leaf Extract (TLE) at the concentration of 0.5%, 1% and 1.5 % was added to the final mix. The mixes were homogenized at 2000 & 500 psi through homogenizer (make Goma Engg). The mixes were batch pasteurized at 90 ºC for 10 min and cooled to 42 ºC. The yogurt culture was added to the mix at 42 ºC. The Yogurt mixes were poured into polystyrene cups and incubated at 40-41 ºC to pH 4.5 before cooling to 4 °C. Samples were stored at 4 ºC until further usage. The preparation of novel Yoghurt is summarized in Figure 1.

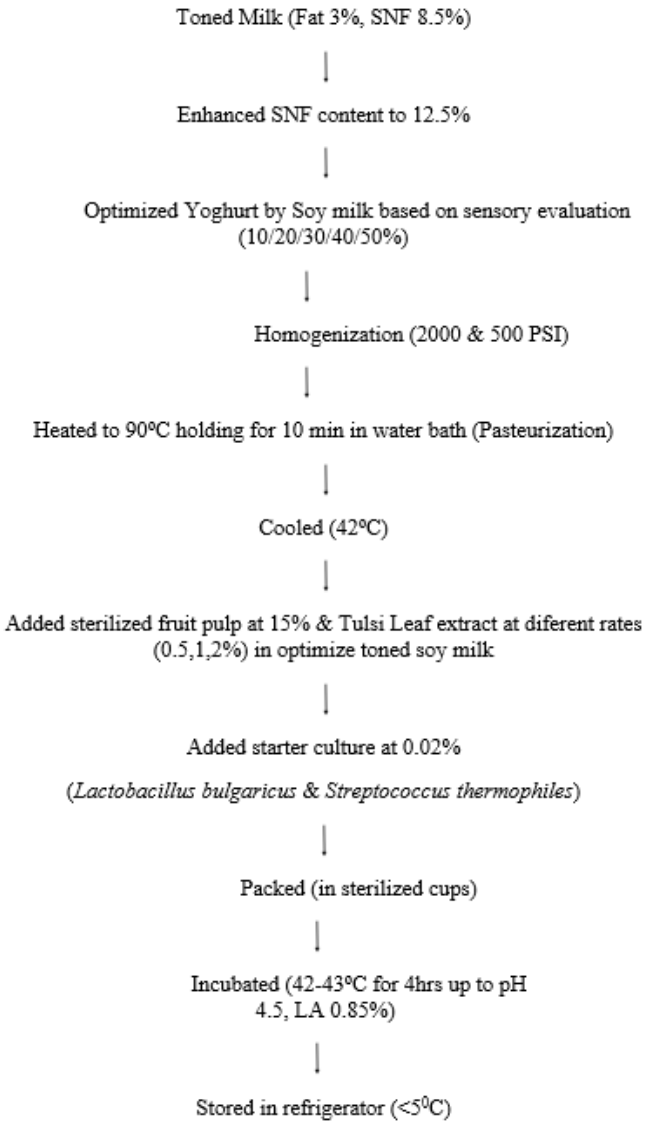

Fig 1: Flow diagram for the preparation of Novel Fruit Yoghurt

Methods of Analysis

The pH content was determined through digital pH meter - make LABINDIA pH analyzer supplied by Lab India Analytical Instruments Pvt. Ltd. Mumbai, India. The total soluble solids content was determined through digital refractrometer Model RX-7000i, Make ATAGO CO. LTD, Japan. The water activity of yoghurt samples was determined through digital water activity analyzer (Pre Aqua Lab water activity analyser, Decagon Devices Inc, USA). The Titrable Acidity of yoghurt sample (expressed as percent lactic acid) was determined as per the procedure laid in IS 1166 – 1973. The protein content of yoghurt was determined using micro-Kjheldhal method. The carbohydrate content was calculated by subtracting sum of Moisture, Protein, Fat and Ash from 100. Fat was analyzed using Rose Gottleib Method as per AOAC (2000) [2]. The color of samples was measured using Hunter Lab Colorimeter (Konica Minolta CR-400), in terms of Hunter (redness, ranging -60 to +60 indicating green to red). It measures the absorbance of different wavelengths of light in a solution. The colorimeter was calibrated against a standard instrument white tile (L= 95.750, a= - 0.375, b= 2.359). Samples were taken in poly bag to a level at which the measurement area nod of colorimeter CR-400, which is 1 cm

in diameter, would be immersed. Then by clicking the measurement button values of a*, b*, and ΔE were measured.
Antioxidant activity of novel yoghurt was determined by a method based on the ability of the antioxidant to scavenge the 1, 1-diphenyl-2-picrylhydrazyl (DPPH) cation radical, as described by Brand-Williams *et al*. (1995) [3].
Sensory evaluation was performed to determine the acceptability of the Novel yoghurt, to standardize the rate of incorporation of soy milk & TLE for preparing yoghurt with high acceptance. Responses were taken from a semi- trained panel of 25 Scholars and faculty members from NIFTEM. Their opinions were recorded for taste, flavor, appearance, texture, and overall acceptability on nine-point Hedonic scale.

Statistical Analysis: All experiments were carried out in triplicate (n=3). Data were analyzed by ANOVA - SPSS (16.0) version.

Results and Discussion

Yoghurt is a good carrier for quality nutrients and provides immense scope for incorporation of plant derived food proteins from soy milk thereby further augmenting the functionality of quality milk products.

Selection of level of Soy milk incorporation for Novel Yoghurt

Soy milk is rich source of protein. Incorporation of soy milk is a good alternative for value addition in novel dairy products, but the problem with increased level is its beany flavour. The sensory scores revealed that up to 10% level it gave good sensory perception. The results are summarized in Table -1.

Table 1: Mean & Standard Error of different level of soy milk on sensory properties of Yoghurt

Variable	Color & appearance	Body & Texture	Flavor	Overall Acceptability
T0	8.1333^{a}±.03333	8.0000±.05774	8.00±.000	8.00±.000
T1	8.1333^{b}±.03333	8.0333±.12019	8.00±.000	8.00±.000
T2	7.9667±.06667	7.8000±.05774	8.00±.000	8.00±.000
T3	7.3000±.05774	7.6000±.05774	8.00±.000	7.67±.333
T4	7.2333±.03333	6.7333±.12019	7.33±0.333	6.00±.000
T5	6.6667±.08819	6.3000±05774	6.00±.000	6.00±.000
	7.5722±.13402	7.4111±.16268	7.56±.185	7.28±.226

T0=Toned milk, T1=10% soy milk, T2=20%, T3=30%, T4=40%, T5=50% soy milk. Data are presented as mean ± Standard deviation (n=25) and a, b, c and d represents the significant difference within column (p<0.05)

Selection of level of Fruit pulp incorporation for Novel Yoghurt

The results reported in table-2, about sensory evaluation of the product revealed that the best product is manufactured by using 15 % Mango pulp. It can be seen that there was no adverse impact of addition of mango pulp even upto15% on colour and appearance of the product. Addition of mango pulp slightly weakened the body and texture of the product. It is obviously because of the reason that with addition of pulp the protein particularly the casein decreases which is responsible for the firmness of the curd. However, with 15% mango pulp the body and texture was liked very much by sensory panel. The sensory score for flavour increased with increase in level of mango fruit pulp up to 15% level where after it decreased at 20% level of mango pulp incorporation. The increase in flavour score may be due to the fact that mango is a popular fruit and we are accustomed to its taste. Decrease in score at 20% pulp level may be because of higher sweetness in product. Corresponding to the flavour the overall acceptability was highest with 15% mango pulp product. Jayalalitha *et al.* (2015) also obtained similar results in their study. They added 5, 10 and 15% mango pulp to the 30% soy milk fortified milk and found product with 15 % mango pulp to give best overall acceptability.

Table 2: Mean & Standard Error of different level of mango fruit pulp on sensory properties of Yoghurt

Variable	Colour & Appearance	Body & Texture	Flavour	Overall Acceptability
T1	8.13^{b}±.033	8.03^{c}±0.12	8.20^{bc}±.058	8.20^{b}±.058
T11	8.20^{b}±.057	7.97^{bc}±0.12	8.03^{ab}±0.12	7.93^{a}±.088
T12	7.97^{b}±.066	7.73^{bc}±0.07	8.20^{bc}±.058	8.03^{ab}±.067
T13	8.07^{b}±.088	7.60^{b}±0.15	8.30^{c}±.058	8.40^{c}±.058
T14	7.33^{a}±.088	6.73^{a}±0.12	7.80^{a}±.058	7.87^{a}±.033

T1- 0% fruit 10% soy milk 90% Toned milk, T11=5%, T12=10%, T13=15%, T14=20% Mango pulp incorporation. Data are presented as mean ± Standard deviation (n=25) and a, b, c, ab, and bc represents the significant difference within column (p<0.05)

Selection of level of green *Tulsi* Leaf Extract (TLE) for incorporation in Novel Yoghurt

The results reported in table-3, about sensory evaluation of the product having 0.5%, 1% and 1.5% TLE with 10% soymilk replacement and 15% mango pulp, revealed that the product with 0.5% and 1% TLE were at par with the product having no TLE, while the product having 1.5% TLE was graded significantly low. This may happen because at 1.5% level, it may suppress the pleasant smell of mango flavour vis a vis provide a sort of ayurvedic taste. The results reported in table 4, compares the antioxidant activity of different treatment with respect to standard and blank. The results revealed that anti-oxidant activity was found to be highest in T13 which signifies the importance of TLE.

On the basis of results obtained so far, a standardized Herb Supplement Fruit Yoghurt (HSFY) was made with 10% soymilk, 90% toned milk, 15% Mango Pulp and 1% TLE and the same was compared with Mango based fruit yoghurt procured from Market for Physicochemical analysis.

Table 3: Mean & Standard Error of different level of *Tulsi* Leaf Extract on sensory properties of Yoghurt

Variable	Colour & Appearance	Body & Texture	Flavour	Overall Acceptability
T1	8.20^{b}±.033	7.90^{ab}±0.12	8.10^{bc}±.058	7.90^{ac}±.058
T11	8.10^{b}±.057	7.85^{ab}±0.12	7.90^{bc}±0.12	7.80^{ac}±.088
T12	8.05^{b}±.066	7.75^{ab}±0.07	7.80^{bc}±.058	7.75^{ac}±.067
T13	7.05^{a}±.088	6.10^{aa}±0.15	6.30^{c}±.058	6.10^{c}±.058

T1- 15% fruit 10% soy milk 90% Toned milk and 0% TLE, T11=0.5%, T12=1.0%, T13=1.5%, TLF incorporation. Data are presented as mean ± Standard deviation (n=25) and a, b, c, ab, ac, and bc represents the significant difference within column (p<0.05)

Table 4: Comparison of Anti-oxidant activity of Treatments with varying concentration of TLE

Sample Name		Absorbance (at 517 nm)	% Inhibition
Ascorbic 0.2		0.0949	78.43
Ascorbic	0.4	0.04	90.91
Ascorbic	0.6	0.019	95.68
Ascorbic	0.8	0.015	96.59
T1		0.32	21.70
	T11	0.24	45.4545
Treatments with TLE	T12	0.23	47.7272
	T13	0.21	51.1515
Blank		0.44	-

Physicochemical & colour Properties of Novel Yoghurt

The results mentioned in Table-5 and 6 revealed the physiochemical comparison and color comparison between HSFY and Market sample. Soy milk fortification increases the level of protein whereas fat content gets lowered. The pH and acidity of Yoghurt is influenced by addition of fruit pulp and was slightly more acidic as also observed by Osman & Razig (2010). There is not much influence on water activity as compared to market sample. The carbohydrate content & total soluble solid content of the novel yoghurt sample was found to be lower than market sample this can be attributed to less sugar incorporation or the source of fruit pulp.

Conclusion

The prepared novel yoghurt sample was found to have higher antioxidant activity because of inclusion of Tulsi leaf extract and mango pulp. Antioxidants are helpful in preventing oxidative stress related consequences. Herbs like mint, basil, coriander, ginger, beet root have been tried in Yoghurt and other dairy products like sandesh and have found to have positive effect Chakarborty *et al*. (2017).

Table 5: Comparative Physicochemical attributes of novel HSFY with Market samples (Mango Flavor Yoghurt)

Parameter	HSFY	Market Sample
pH	4.39	4.42
a_w	0.9769	0.9778
TSS (^{0}Brix)	25.13	26.1
TA (%LA)	1.18	1.17
Protein (%)	4.14	3.61
Fat (%)	4.03	4.30
Carbohydrate	16.46	17.53
Moisture (%)	74.70	73.8
Ash	0.67	0.66
Antiradical Activity(%DPPH Scavenging)	47.72	15.90

Table 6: Comparative colour score for Market sample & HSFY

Sample	Market Sample	HSFY
L*	84.43±0.02^{a}	81.106±0.548^{aa}
a*	- 6.035±0.01^{b}	-7.34±0.55^{bb}
b*	35±0.52^{c}	37.74±1.15^{cc}

Data are presented as mean ± Standard deviation (n=3) and a, b, c, aa, bb, and cc represents the significant difference within row (p<0.05)

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Conflict of Interest

The author(s) declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

References

1. Anthony MS, Clarkson TB, Hughes CL, Morgan Jr TM, Burke GL. Soybean isoflavones improve cardiovascular risk factors without affecting the reproductive system of peripubertal rhesus monkeys. J Nutr. 1996; 126:43-50.
2. AOAC, 17th edn. Official method 905.02 Fat in milk, 2000
3. Brand-Williams, Wendy, Cuvelier ME, Berset CLWT. Use of free radical method to evaluate antioxidant activity. LWT-Food Sci Technol. LWT - Food Science and Technology. 1995; 28:25-30.
4. Erdman JW Jr, Fordyce EJ. Soy products and the human diet. Am. J Clin. Nutr. 1989; 49:725-737.
5. Yazici F, Alvarez VB, Hansen PMT. Fermentation and properties of calcium-fortified soy milk yogurt, J Food Sci. 1997; 62:457-461.
6. Friedman M, Brandon DL. Nutritional and health benefits of soy proteins. J Agric. Food Chem. 2001; 49:1069-1086.
7. Hammond BG, Jez JM. Impact of food processing on the safety assessment for proteins introduced into biotechnology-derived soybean and corn crops. Food Chem. Toxicol. 2011; 49:711-721.
8. "Indian Dairy Products Market By Product Type, Competition, Forecast & Opportunities, 2011 – 2021'' report by Tech-Sci Research.
9. IS 1166 – 1973, Specification for condensed milk, BIS, New Delhi.
10. Jaziri I, Ben Slama M, Mhadhbi H, Urdaci MC, Hamdi M. Effect of green and black teas (*Camellia sinensis* L.) on the characteristic microflora of yogurt during fermentation and refrigerated storage. Food Chem. 2009; 112:614-620.

11. Kennedy AR. The bowman-birk inhibitor from soybeans as an anticarcinogenic agent. Am. J Clin. Nutr. 1998; 68:1406S-1412S.
12. Kreijkamp-Kaspers S, Kok L, Grobbee DE, EH de Haan, Aleman A, Lampe JW *et al.* Effect of soy protein containing isoflavones on cognitive function, bone mineral density and plasma lipids in postmenopausal women: A randomized controlled trial. J Am. Med. Assoc. 2004; 292:65-74.
13. Lee SJ, Hwang JH, Lee S, Ahn J, Kwak HS. Property changes and cholesterol-lowering effect in evening primrose oil-enriched and cholesterol-reduced yogurt. Int J Dairy Technol. 2006; 60:22-30.
14. Michelfelder AJ. Soy: A complete source of protein. Am. Fam. Physician 2009; 79:43-47.
15. Muller U, Weber W, Hoffmann A, Franke S, Lange R, Vieths S. Commercial soybean lecithins: A source of hidden allergens? Zeitschrift Lebensmitteluntersuchung Und-Forschung A. 1998; 207:341-351.
16. Cruz NS, Capellas M, Jaramillo DP, Trujillo AJ, Guamis B, Ferragut V. Soymilk treated by ultra-high-pressure homogenization: Acid coagulation properties and characteristics of a soy-yogurt product, Food Hydrocolloids 2009; 23:490-496.
17. Donkor ON, Henriksson A, Vasiljevic T, Shah NP. Rheological properties and sensory characteristics of set-type soy yogurt, J Agric. Food Chem. 2007; 55:9868-9876.
18. Ojha A, Bhojak N, Sharma A, Joshi S. Indirect Micellar Spectrophotometric determination of lead in various soybean based products using green chemistry. Asian J of Dairy & Food Research. 2005; 34(2):336-340.
19. Kumar P, Mishra HN. Effect of mango pulp and soymilk fortification on the texture profile of set yoghurt made from buffalo milk, J Text. Stud. 2003; 34:249-269
20. Bhattacharya S, Jena R. Gelling behavior of defatted soybean flour dispersions due to microwave treatment: Textural, oscillatory, microstructural and sensory properties, J Food Eng. 2007; 78:1305-1314.
21. Lee SY, Morr CV, Seo A. Comparison of milk-based and soymilk-based yogurt, J Food Sci. 1990; 55:532-536.
22. Sarkar S. Innovations in Indian Fermented Milk Products-A Review, Food Biotechnology 2008; 22(1):78-97.
23. Seo MH, Lee SY, Chang YH, Kwak HS. Physicochemical, microbial, and sensory properties of yogurt supplemented with nanopowdered chitosan during storage. J Dairy Sci. 2009; 92:5907-5916.
24. Staffolo MD, Bertola N, Martino M, Bevilacqua A. Influence of dietary fiber addition on sensory and rheological properties of yogurt. Int. Dairy J. 2004; 14:263-268.
25. Xiao CW. Health effects of soy protein and isoflavones in humans. J Nutr. 2008; 138:1244S-1249S.
26. Zare F, Boye JI, Orsat V, Champagne C, Simpson BK. Microbial, physical and sensory properties of yogurt supplemented with lentil flour. Food Res Int. 2011; 44:2482-2488.