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QUALITY OF COW MILK PLAIN SET YOGHURT AS AFFECTED BY ULTRAFILTRATION PROCESS

Narayana NMNK[*1] and Gupta VK[2]
[1]Department of Animal Science, Faculty of Agriculture, University of Ruhuna, Mapalana, Kamburupitiya, Sri Lanka
[2]Division of Dairy Technology, National Dairy Research Institute, Karnal, 132001, Haryana, India

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ABSTRACT

The aim of the study was to investigate the effect of ultrafiltration (UF) process on quality of cow milk plain set yoghurt. Direct UF of cow skim milk and highly concentrated UF cow skim milk retentate addition were used to adjust the total solids (TS) of yoghurt milk, at two different UF concentration levels (UFCLs). Ultrafiltered (approximately to 1.5 and 2 fold) cow skim milk/ equivalent 5 fold UF skim milk retentate added cow skim milk were standardized to 3.3% fat and 13.8% TS. Yoghurts were prepared by inoculating with 2% yoghurt culture (*Lactobacillus delbrueckii* subsp. *bulgaricus: Streptococcus thermophilus*) and analyzed for chemical composition, spontaneous whey syneresis, water holding capacity, textural and sensory attributes. Protein, lactose and ash percentages of yoghurt prepared from direct UF milk were 5.27±0.04, 4.20±0.03 and 0.82±0.02, whereas, in yoghurt prepared from retentate added milk were 5.18±0.02, 4.28±0.03 and 0.84±0.01, respectively at 1.5 fold UFCL, which had optimum quality product. The values were not significantly different in yoghurt made by direct UF concentrated milk compared to retentate added milk. Further, it was observed that protein percentage increased and lactose content progressively decreased significantly (p<0.05) in yoghurt with increase in UF concentration/ UF retentate addition with similar TS in yoghurt milk. None of the quality parameters tested showed significant difference with UF process so that both procedures would be recommended at 1.5 fold UFCL to produce good quality yoghurt with enhanced protein content without addition of stabilizers.

Key words: UF process, UF retentate, UFCL, Spontaneous whey syneresis, Water holding capacity, Textural attributes, Sensory attributes

INTRODUCTION

Yoghurt is perhaps the most popular fermented dairy product as witnessed by its worldwide distribution. Horiuchi *et al*. (2009) reported that the global sales of yoghurt in year 2006 were approximately US$ 40 billion. According to a recent research conducted by Global Industry Analysts Inc., it was predicted that by 2015, global yoghurt consumption will reach 20.6 million tons, equaling US$ 67 billion in sales. Asia presents a huge opportunity due to the rising incidence of lifestyle-related health concerns, such as diabetes and obesity, brought on by rapid economic development and rising income levels (Anon, 2010). Yoghurt is retailed in one of the three physical states, namely set, stirred and fluid/drinking (Tamime and Robinson, 1999) according to the method of production and the physical structure of coagulum. The set yoghurt is produced by packaging the yoghurt mix into individual containers before fermentation. As a commercial product it is important that the set yoghurt has curd with sufficient hardness to stand up to the impact caused by shaking during transportation (Horiuchi *et al*., 2009). Nielsen (1975) suggested that the texture of set yogurt should be firm enough to remove it from the container with a spoon. According to Lewis and Dale (1994) set yoghurt should have a glossy surface appearance without excessive whey. Whey syneresis is a major defect of set-style yoghurt (Lucey, 2001). The formulation of yoghurt products with optimum consistency and stability to whey syneresis is of primary concern to the dairy industry (Biliaderis *et al*., 1992). Some of the methods adopted by manufacturers to address this problem include addition of skim milk powder, addition of natural or synthetic gums and stabilizers, enzymetic stimula-

*Corresponding author: nayananarayana1970@gmail.com

tion of protein interaction in milk, addition of texturing starters etc. However, some of these methods are of limited use with the increased consumer demand towards more „natural‟ product with no additives and stabilizers.

Yoghurt texture can be improved by increasing the TS content in the milk base. This in turn increases viscoelastic properties and water holding capacity (WHC) proportionally (Sodini *et al.,* 2004) which leads to reduced whey syneresis. When considering a milk base for yoghurt with a determined TS content, the nature and relative proportions of the different proteins in the dry matter should be of significant importance for the texture of the final product (Modler *et al*., 1983). Savello and Dargan (1997) stated that the ideal system of protein enrichment involve increasing both casein and whey proteins. This can be achieved by use of UF technique. Ultrafiltration process offers several benefits to yoghurt formulations such as reduction of harshness caused by excessive acidity, reduction in lactose content, improvement of texture due to the increased protein content in base milk that minimize the need of stabilizer-like additives and production of a more "natural" product demanded by the current consumer.

Considerable amount of research has been conducted and reported on the use of UF for the production of cultured milk products including yoghurt. (Kosikowski, 1979; Abrahamsen and Holmen, 1980; Becker and Puhan, 1989; Biliaderis *et al*., 1992; Khorshid *et al*., 1992; Brazuelo *et al*., 1995; Savello and Dargan, 1997; Domagala and Kupiec, 2003; El-Khair, 2009). Some of the workers used direct UF milk and others used highly concentrated retentate to fortify and standardize yoghurt milk. However, no studies have been conducted to compare these two methods on compositional, physicochemical, sensory and textural properties of set yoghurt. Within this context, the current study was carried out to check whether the properties of yoghurt made employing these two procedures are distinctly different and make recommendations thereof.

MATERIALS AND METHODS

Materials

Raw cow skim milk and cream (50-55% fat) was obtained from Experimental Dairy of National Dairy Research Institute, Karnal, Haryana, India. As the starter culture, commercial yoghurt (Nestle‟) containing *Streptococcus thermophilus* and *Lactobacillus delbrueckii* subsp. *bulgaricus* was used. Pilot UF plant (Tech-Sep, France) with tubular module (channel diameter, 6 mm) having ZrO_2 membrane (membrane surface area, 1.68 m^2 and membrane molecular weight cut off, 50,000 Dalton) was used for UF of skim milk.

Methods

Cleaning of the UF plant and UF of cow skim milk

The UF plant was cleaned by flushing with water and running with hot alkali (0.8% NaOH) at 75°C for 15 min followed by hot water flushing for 20 min and circulating acid (0.3% HNO_3) at 80°C for 15 min with inlet and outlet pressures of 4.6 and 3.6 kg/cm^2, respectively on the retentate side. Cow skim milk was heated to 80°C, cooled to 55-60°C and transferred into the balance tank of UF plant and ultrafiltered at 50-55°C approximately to 1.5, 2.0 and 5 UFCLs.

Production of experimental yoghurts

Ultrafiltered cow skim milk/~5 fold UF retentate added cow skim milk were standardized to 3.3% fat and 13.8% total solids by adding calculated amount of cow milk cream in case of 1.5 UF fold/equivalent retentate added milk and by adding calculated amount of cream and water in case of 2 UF fold/equivalent retentate added milk. Resultant standardized milks were pre heated to 65-70°C, homogenized in a two-stage homogenizer (M/s Goma Engineers, Mumbai) at 2000 and 500 psi at 1st and 2nd stages, respectively, heat treated to 85°C/30 min in a thermostatically controlled water bath (NAVYUG, India), cooled immediately to 42°

C, inoculated with 2% yoghurt culture, mixed well, filled in to clean polystyrene cups, covered with lids and incubated at 42±1°C until desired titratable acidity (TA) of ≥0.8% lactic acid (LA) was achieved. Yoghurts were then immediately transferred to a refrigerator maintained at 4°C. All the trials were carried out in triplicates.

Physico-chemical analysis

A pH meter (PHAN LABINDIA Model, Labtek Eng. Pvt. Ltd. India) was used for the determination of pH of yoghurt. The TA of yoghurt was determined using procedure recommended in BIS (1981a). Fat content of skim milk, UF retentates, cream and standardized milks was determined by Gerber method as described in BIS (1981a). Protein of yoghurt was determined by semi-micro kjeldhal method using Kjeltec digestion and distillation equipment (2300, Kjeltec Analyzer, FOSS). Lactose was determined by Lane-Enyon method described in BIS (1981b). Ash and TS contents were determined by methods described in BIS (1981b) and in ISO (1989), respectively.

Spontaneous whey syneresis (SWS)

Siphon method described by Amatayakul *et al*. (2006) was used with slight modifications to determine the SWS. A cup of yogurt (100 ml) was tilted immediately after removing from the refrigerator at an angle of 45° to collect the surface whey. Collected whey was siphoned out with a graduated syringe to which a needle was attached. The siphoning was performed within 10 s to avoid forced leakage of whey from the curd. The value was taken directly as the percentage of SWS.

Water Holding Capacity

The WHC was measured by a centrifuge method according to Supavititpatana *et al*. (2009). Within 12 h of the production of yogurt, a 10 g sample was centrifuged at 2,000 g for 60 min at 10±1°C. The supernatant was removed within less than 10 min and the wet weight of the pellet was recorded. The WHC was expressed as follows.

$$\text{WHC (\%)} = \frac{\text{pellet weight}}{\text{Sample weight}} \text{x}100$$

Textural attributes

Texture analysis was carried out according to the method given by Kumar and Mishra (2003) with slight modifications, using a TA-XT2i Texture analyser (M/s Stable Micro Systems, UK) fitted with a 25 kg load cell and was calibrated with a 5 kg standard dead weight prior to use. For determining the textural attributes, the pasteurized and cooled standardized milk was filled up to 80 ml in 100 ml pre-sterilized glass beaker and incubation was carried out. Experiments were carried out by compression tests that generated plot of force (N) versus time (s). A 25 mm perplex cylindrical probe was used to measure texture of yoghurt samples at a temperature of 10±0.5°C performing four repetitions. During analysis the samples were compressed up to 20 mm of their original depth. The speed of the probe was 0.5 mm/s during the compression and 2 mm/s during pre-test and relaxation. From the resulting force-time curves, firmness, stickiness, work of shear and work of adhesion were calculated using the Texture Expert Exceed software (version 2.55) supplied by the manufacturer along with the instrument.

Sensory evaluation

On the basis of desirable attributes for good quality yoghurt, the 100 point score card suggested by Ranganadham and Gupta (1987) was used. Yoghurts were served at 10±1°C to 7 trained judges.

Statistical analysis

Factorial arrangement of treatments (2x2) in a complete randomized design was used. The

GLM procedure of SAS (version 9.2) was used to analyse the data. Critical difference (CD) was calculated according to the method described by Rangaswamy (1995). Mean±SE (Standard Error) was calculated using MS-Excel software (version 2007) wherever required.

RESULTS AND DISCUSSION

Effect of method of fortification of milk using ultrafiltration process and UFCL on chemical composition, physicochemical parameters, textural and sensory attributes of cow milk plain set yoghurt is presented in Table 1.

Chemical composition of yoghurt

Total solids, fat, and titratable acidity were maintained approximately same in all the samples of yoghurt. Even though it was observed that protein content was higher, while lactose and ash contents were lower in yoghurt prepared with direct UF concentrated milk compared to yoghurt prepared with retentate added milk, none of the constituents showed statistically significant difference with the UF process. Slightly lower amounts of lactose and ash content in yoghurt prepared with direct UF concentrated milk than UF retentate added milk might be due to their removal during direct UF concentration process. Further, to standardize the yoghurt milk with highly concentrated retentate, skimmed cow milk was used, which might add additional lactose and ash to the yoghurt prepared.

It was observed that protein content increased and lactose content progressively decreased significantly (p<0.05) in yoghurt with the increase in UF concentration/UF retentate addition (even though water is added in the case of 2 fold direct/retentate added milk to maintain similar TS level in all the treatments). Ash content was also observed to be increased but was not statistically significant with the increase in UF concentration/UF retentate addition.

Table 1: Comparison of method of fortification of yoghurt milk using ultrafiltration process and UFCL on quality aspects of cow milk plain set yoghurt

Characteristic	1.5 fold		2 fold + water	
	Direct	Retentate	Direct	Retentate
Compositional aspects				
TS (%) Fat	13.60^{a} ±0.02	13.61^{a} ±0.01	13.62^{a} ±0.03	13.57^{a} ±0.03
(%) Protein	3.31^{a} ±0.01	3.30^{a} ±0.00	3.32^{a} ±0.02	3.31^{a} ±0.01
(%) Lactose	5.27^{a} ±0.04	5.18^{a} ±0.02	5.58^{b} ±0.02	5.49^{b} ±0.07
(%)	4.20^{a} ±0.03	4.28^{a} ±0.03	3.88^{b} ±0.07	3.91^{b} ±0.04
Ash (%)	0.82^{a} ±0.02	0.84^{a} ±0.01	0.85^{a} ±0.01	0.87^{a} ±0.02
Physicochemical parameters				
TA (% LA)	0.86^{a}±0.00	0.86^{a} ±0.01	0.86^{a} ±0.01	0.86^{a}±0.01
pH	4.54^{a} ±0.00	4.54^{a} ±0.01	4.56^{b} ±0.00	4.56^{b}±0.00
WHC (%)	63.49^{a}	63.15^{a}	63.06^{a}	62.48^{a}
Textural attributes				
Firmness (N)	1.88^{a}	1.85^{a}	1.94^{b}	2.09^{b}
Stickiness (N)	-0.41^{a}	-0.4^{a}	-0.46^{b}	-0.44^{b}
WoS (N.s.)	54.47^{a}	56.66^{a}	58.52^{b}	60.45^{b}
WoA (N.s.)	-2.09^{a}	-2.09^{a}	-2.42^{b}	-2.46^{b}
Sensory attributes				
Flavour	41.24^{a}	41.90^{a}	40.62^{b}	40.29^{b}
Body & texture	28.05^{a}	27.57^{a}	27.83^{a}	27.62^{a}
Acidity	8.93^{a}	9.02^{a}	8.79^{b}	8.50^{b}
Colour & appearance	8.98^{a}	9.02^{a}	9.19^{a}	9.00^{a}
Overall Acceptability	92.19^{a}	92.50^{a}	91.43^{b}	90.40^{b}

Mean of 3 trials
$^{a, b}$ Means with different superscripts within each row differ significantly (p<0.05)

Physicochemical parameters

pH was observed to be 4.54 in the yoghurt made using direct as well as 5 fold retentate added milk at 1.5 fold UF concentration level. However, at higher UFCL, pH of yoghurt was observed to be significantly (p<0.05) higher than the pH of yoghurt made from milk concentrated to 1.5 fold. This might be due to the buffering effect of UF milk at higher folds (Premaratne and Cousin, 1991; El-Gazzar and Marth, 1991). Nevertheless, significant differences were not observed in the pH of yoghurts made by using direct and retentate addition method.

Whey syneresis was not observed in any of the yoghurts made. El-Khair (2009) reported that the yoghurts made with added UF skim milk retentate displayed minimal free whey, whereas, the control yoghurt made from added skim milk powder was criticized for whey separation. Further, the author mentioned that the UF skim milk retentate served as a stabilizer in yoghurts to improve texture and reduce whey separation. Water holding capacity of yoghurt was observed to be unaffected either with the method of fortification of yoghurt milk using UF process or with UFCL. The reason to have non-significant effect of WHC with UFCL might be due to the added extra water to similarize TS content at higher UFCL even though, protein is higher in higher than lower UFCL.

Textural attributes

All the textural attributes namely firmness (peak force obtained during the penetration of the probe), stickiness (negative peak force obtained during the withdrawal of the probe), work of shear (WoS) (i.e. area under the penetration cycle) and work of adhesion (WoA) (i.e. area under the withdrawal cycle) of yoghurt were observed to be non significant with the method of fortification of yoghurt milk. However, all the textural attributes of yoghurt were observed to be significantly (p<0.05) increased with increasing UFCL, either with direct method or with retentate addition method. This is because of the higher protein level in yoghurt made from 2 UF fold compared to 1.5 UF fold which increase protein matrix density and hence textural attributes.

Sensory attributes

Highest flavour score of 41.90 out of maximum possible 45 was obtained by yoghurts made from 5 fold UF skim milk retentate added milk to maintain same milk solids as in 1.5 fold UF concentrated milk, whereas, lowest score was obtained by yoghurts made from 5 fold UF skim milk retentate added milk to maintain same milk solids as in 2 fold UF concentrated milk. Flavour score of yoghurt was observed to be non significant with method of UF of milk. However, it was observed that the flavor score decreased significantly (p<0.05) with increasing UFCL. Body and texture score was highest in yoghurts made from milk concentrated to 1.5 UF fold by direct method. The range was observed to be from 27.57 to 28.05 out of maximum possible 30, and no differences were observed either with the method of fortification of milk using UF process or with the UFCL. Total solids in yoghurts were maintained nearly similar value (13.8%) by adding calculated amount of water to higher fold. However, protein was higher in 2 fold direct/ retentate added yoghurt milk than 1.5 fold. Proteins play a significant role in body and texture improvement of yoghurt. However, body and texture can be improved up to some extent by increasing the protein content of yoghurt milk (Abrahamsen and Holmen, 1980). This might be the reason to have non-significant scores for body and texture between UFCLs since increase of protein is slight in yoghurt made from 2 fold UF concentrated milk compared to 1.5 UF fold. Further, when protein is higher, slightly rubbery texture was noted in yoghurts made reducing the body and texture scores.

Acidity, colour & appearance as well as overall acceptability scores of yoghurts were also not significantly different with the method of UF. Acidity score ranged from 8.5 in yoghurts made from retentate added 2 fold UF concentrated milk to 9.02 in retentate added 1.5 fold UF concentrated milk. Acidity score was significantly (p<0.05) lower in yoghurts made by 2 compared to 1.5 fold UF concentrated milk (Table 1). Even though incubation of yoghurts stopped nearly at same acidity level, slightly higher pH value was observed in higher UF fold. This may be because of the buffering capacity of UF milk due to higher protein content even though, nearly similar TS were maintained in each treatment. This may be the reason to have lower scores for acidity at higher UF fold. Further, this was reflected in flavor and overall acceptability scores. Colour & appearance and Overall acceptability scores ranged from 8.98-9.19 and 90.4-92.5 respectively. Overall acceptability score was significantly (p<0.05) higher in yoghurt made from 1.5 compared to 2 UF fold. However, none of the parameters were significantly different with yoghurts made from direct UF concentrated milk or retentate added milk.

According to the results of the sensory evaluation, it was clear that, use of direct UF concentrated milk or 5 fold UF skim milk retentate added milk for the production of plain yoghurt, did not create any significant difference to the sensory scores of yoghurt. Further, yoghurts made at 1.5 fold UFCL obtained significantly (p<0.05) higher sensory scores than at 2 fold UFCL with similar TS.

CONCLUSION

Either the use of direct UF concentrated milk or 5 fold UF skim milk retentate added milk for the production of plain set yoghurt did not affect compositional, physicochemical, textural and sensory attributes of the product significantly. Further, use of 1.5 fold UFCL was better than 2 fold UFCL in terms of sensory attributes of the product. Therefore, any feasible method at 1.5 fold UFCL can be recommended for the production of good quality plain set yoghurt without use of stabilizers and with lesser amount of TS.

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