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Augmentation of Rheological Properties of *Dahi*

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Abstract

Dahi is the most popular indigenous milk product due to its functional properties. Market survey of *dahi* revealed diversity in its chemical and microbiological quality due to adoption of different manufacturing technology. Recently the inclination of consumers towards healthful foods resulted in a renewed interest in *dahi*. Process standardization for its production is emerging to obtain a consistent product with improved rheological properties besides enhanced functional properties to project it as a functional food in the world market. In the present investigation modifications for firm body (type of milk, pre-treatment of milk, manipulation of starter cultures, modulation of incubation temperature), manipulation of starter cultures for pleasant flavour and technology for higher shelf-life have been delineated.

Key words: *Dahi; Rheological properties; Shelf-life*

Introduction

Fermentation is the oldest method of milk preservation. Dahi is considered the oldest Indian fermented milk product and is the western equivalent to yoghurt. Health consciousness coupled with enhanced health care cost has led consumer's inclination towards healthful foods, which eventually resulted in coining the term "functional foods" (Sarkar, 2019). Functional foods are foods, which besides having nutritional properties possess certain health benefit extending properties. According to Functional Food Center, functional food may be defined as natural or processed foods that contains known or unknown biologically-active compounds, the foods, in defined, effective, and non-toxic amounts, provide a clinically proven and documented health benefit for the prevention, management or treatment of chronic disease (Martirosyan and Singh, 2015).

Dietetic significance of *dahi* is well documented (Yadav *et al*., 2008, Vijayendra and Gupta, 2012, Mohania *et al*., 2013) and consumer's inclination towards healthful foods has resulted in renewed interest in *dahi*. Functional properties of *dahi* (Sarkar *et al*., 2011) and probiotic- supplemented food have projected them as a functional food in the current era of self-care and complementary medicine (Sarkar, 2013). Introduction of probiotic cultures and inclusion of diverse food additives (fruit juices, herbs, spices) has been suggested for enhancing the functional properties of traditional dahi (Sarkar, 2016 a, Sarkar and Sur, 2017) to project dahi into the global market as a functional food (Sarkar et al., 2011). Murugan et al. (2013) reported remarkable demand for dahi in international market owing to its high nutritional value and therapeutically benefits.

Several market survey reports indicate diversity in physico-chemical, microbial, nutritional and therapeutic properties of dahi (Sarkar et al., 1996 a, b, Sarkar, 2008, Shekh et al., 2009, Dey et al., 2011, Chanda et al., 2013, Islam et al., 2017, Patel et al., 2018). Numerous attempt have been made to enhance the functional properties of dahi with the incorporation of plant (oats, rice and almond) milk (Rai et al., 2018), flax lignan (Paul et al., 2016), inclusion of probiotics (Shandilya et al., 2016), co-encapsulation of probiotics with prebiotics (Vivek et al., 2013) and inclusion of microencapsulated flaxseed oil microcapsules (Goyal et al., 2016). Relatively limited efforts have been taken to improve the rheological properties of traditional dahi. In the present article, an attempt has been made to review diverse research findings to suggest technological modifications that can be adopted for optimizing dahi production with augmented rheological properties with enhanced shelf-life to project dahi as a functional food in the world market.

Modifications For Firm Body

Type of milk

Numerous research on modulation of milk composition have been done to obtain dahi with desirable rheological properties. Attempt to manufacture dahi from milk from different breeds such as cow, buffalo, goat or their admixtures have been reported. Acidification of milk under the influence of starter cultures is important to obtain dahi with desirable rheological properties and extent of acidification is influenced by the buffering capacity of the milk, which is influenced by the total solids content of milk (Gastaldi et al., 1997, Varghese and Mishra, 2008). Whey expressed after the growth of a mixed culture of Streptococcus thermophilus and Lactobacillus delbrueckii subsp. bulgaricus in milk is capable of inhibiting a wide range of undesirable as well as pathogenic microorganisms. Highest inhibitory activity were noted in buffalo milk, followed by in cow milk and goat milk (Singh and Kaul, 1982), indicating better growth of yoghurt starters in buffalo milk than in cow milk or goat milk. Attempts were also made to investigate rate of acidification in mixed milk by yoghurt cultures. Highest rate of acid development by yoghurt cultures were noted in sterilized goat milk, followed by in mixed goat milk and cow milk or cow milk (Bozanic et al., 1998). In general, the citrate content in goat's milk is rather low when compared with cow's milk and, as a consequence, such milk may not be suitable for diacetyl production by mesophilic lactococci alone (Abrahamsen and Rysstad, 1991). However, low levels of acetaldehyde in goat's yoghurts have been attributed to the relatively high concentration of glycine in the milk; glycine can inhibit the enzyme involved in the conversion of threonine to acetaldehyde and glycine (Abrahamsen and Rysstad, 1991). Yoghurt obtained from goat milk have a loose body due to lower as1-casein content and high casein micelle dispersion coupled with the low buffering capacity of goat milk, resulting in over-acidification (Martín-Diana et al., 2003). Problem of low firmness and high whey separation in goat milk yoghurt could be overcome by rigorous monitoring of the heat-treatment, incubation temperature and concentrations of fat, protein and total solids (Lucey, 2002, Abbasi et al., 2009, Espírito-Santo et al., 2013).

Different attempts were made to fortify or supplement milk for enhancing it total solid content with the objective of producing dahi with desirable rheological properties. In an experiment, whey-soya milk (5:1) was finally diluted to 9:1, 8:1, 7:1 and 6:1 with whey and skim milk powder was added to have the final total solid concentration of 16, 18 and 20 %. Dahi prepared from whey-soya milk having whey to soybean ratio of 5:1 and 20 % total solids was found to be most acceptable in terms of production time (5 h) and sensory evaluation scores (7.7 on 9 point hedonic scale). Whey-soya milk dahi was comparable to milk dahi and could be stored up to 10 days (Rathi et al., 2015). Attempt was made to produce dahi from milk incorporated with spray-dried iron-whey protein concentrate (Fe-WPC) conjugate and ferrous sulfate (FeSO4). Dahi obtained from Fe-WPC conjugate fortified milk (15-25 mg/L) had better sensory, textural and physical attributes as compared with those obtained from FeSO4 fortified milk and control dahi (Gandhi et al., 2022).

Raju and Pal (2014) suggested fortification of milk with fiber (inulin and soy fiber) at a level of 1.5 % during the manufacture of misti dahi, though their overall acceptability were significantly lower than control dahi but above the minimum acceptable score. Fortified low fat functional dahi prepared with the supplementation of skimmed buffalo milk with 0.5% β-glucan had significantly less whey separation, higher viscosity, better instrumental color values and sensory scores in comparison to control dahi and other dahi prepared with different levels (0.25, 0.75 and 1.0%) of β-glucan (Bhaskar et al., 2017). In a another research, Singh et al. (2012) fortify stirred dahi with Strawberry polyphenol extract (0.5 mg /mL) and the resultant product had higher scores than control dahi for flavour, consistency, appearance and overall acceptability throughout the storage of 14 days at 7-8°C. Dahi was prepared from by mixing cow milk with coconut milk showed increasing trends in the values for pH, Total Solids but reverse trend in acidity, protein and carbohydrate (Matin et al., 2020).

Pre-treatment of milk

Heating of milk is an important processing variable for the preparation of yogurt and it influences the physical properties and microstructure of yogurt (Lee and Lucey, 2010). When the pH of preheated milk decreases during fermentation, denatured whey proteins aggregate and extensive cross-linking between whey proteins and caseins occurs (Lee and Lucey, 2004). During the manufacture of fermented milk products, milk is heat treated with the objective of destroying the microorganisms, to increase shelf life, to improve texture and ultimately to improve the quality of final products (Anwer et al., 2013). Shekhar et al. (2013) recommended boiling of milk without holding period in contrast to heating at 63°C/30 min to produce best quality dahi with least syneresis (6.8 vs 7.2 ml). Heat-treatment of milk at 90°C/30 min should be recommended due to a firm curd formation with elongated micelle of size about 235nm with protein matrix consisted of a long micellar chain over 70°C, which resulted in a soft product with an open structure and spherical shaped casein of size about 300 nm (Tomar and Prasad, 1989, Turambekar and Kulkarni, 1991). Heat-treatment of milk at 90^{0}C or 95^{0}C were reported to be equally good for obtaining fermented milk products with less syenersis, more viscosity, better texture profile and overall acceptability (Anwer et al., 2013).

Milk intended for dahi manufacture could be concentrated either by addition of sweet cream butter milk, condensed cow skim milk or with the adoption of vacuum concentration or by membrane processing such as Ultrafiltration process and Reverse osmosis and should not have a total solid content of more than 14% (Roy Chowdhury and Bhattacharyya, 2014). Concentration of milk intended for dahi manufacture employing ultrafiltration is more promising in contrast to the addition of skimmed milk powder. Marafon et al. (2011) had delineated distinct rheology properties of fermented milks manufactured from milk concentrated by different methods. Addition of skimmed milk powder result in a product with comparable viscosity but greater syneresis in comparison to ultrafiltration. Adoption of ultrafiltration as a method of milk concentration may be advantageous due to greater retention of milk properties as it does not have any additional heat treatment, whereas powdered caseinates or skimmed milk are subjected to heat during its manufacture. Ultrafiltration proved to be superior to skimmed milk powder addition due to increased whey retention and better viscosity in goat milk fermented products (Moreno-Montoro et al., 2018). These differences may be attributable to casein alteration by the spray-drying process or to the increase of all milk compounds in the same proportion with the addition of powdered skimmed milk, whereas ultrafiltration of milk mainly concentrates caseins, which are responsible for formation of the yoghurt coagula (Domagała, 2012, Moreno-Montoro et al., 2015). Higher rate of acid development was noted in dahi prepared from concentrated ultrafiltered cow milk in contrast to those obtained from normal cow milk, higher being observer at higher concentration (Khurana et al., 2014). Dahi prepared from ultrafiltered milk and milk with added ultrafiltrate retentate had greater firmness, stickiness, work of shear, work of adhesion and sensory scores and lower whey syneresis than those made from milk with added skim milk powder (Meena et al., 2015).

Pushkala and Srividya (2011) encountered significantly lower whey syneresis in dahi obtained with substitution of skimmed milk powder by 0.15% Aloe gel even after storage at 10°C for 7 days. Dahi obtained employing a combination of 10% sugar with 2% culture scored highest for body and consistency and an elevation in sugar level to 14% induced lowering of scores (Akter et al., 2010). Natural yoghurt obtained from 15-70% fresh cow milk and 85-30% soya milk had enhanced protein content and exhibited a protein–efficiency ratio of 2.60 (Karmas and Bachmann, 1991). Substitution of 25% buffalo milk with soymilk is suggested during the production of misti dahi employing Lactocoocus lactis subsp. lactis culture (Singh et al., 2015). In a recent study, Pratap et al. (2018) concluded that dahi manufactured with 30% replacement of toned milk with soy milk is most acceptable in terms of organoleptic properties as well as physico-chemical properties. Islam et al. (2016) recommended use of 5% carrot juice with skim milk for preparation of dahi with acceptable taste, flavour, body and consistency with lower bacterial count. In another study. Sarker et al. (2018) suggested incorporating 15% carrot juice whole milk to obtain dahi with highest organoleptic properties.

Manipulation of starter cultures

Good quality fermented milk could be produced from a milk without enhancing its total solid content with the adoption of exopolysaccharide (EPS) producing starter cultures. Advantages proclaimed with the application of EPS producing cultures are requirement of low levels of chemical additives (De Vuyst et al., 2001, Jolly et al., 2002), better textural attributes at low total solid content (Wacher-Rodarte et al., 1993, De Vuyst et al., 2001) and improved sensory properties (Folkenberg et al., 2006).

The rheological properties of the polysaccharides depend on the monomeric composition, the number of side chains, the chain length and the charge (neutral or anionic) of the polysaccharides, as well as the anomeric configuration of the monosaccharides and the sequence in which they are arranged (Nath and Malik, 2016). Exopolysaccharides of microbial origin are long chain, high-molecular-mass water-soluble polymers which may be ionic or non-ionic and have potential applications in food industries as texturizers, viscosifiers, emulsifiers and synerisis-lowering agents due to their pseudoplastic rheological behaviour and water-binding capacity (Cerning, 1990). Vijayendra et al. (2008) reported EPS produced by non-ropy strain of Leuconostoc sp. CFR 2181, isolated from dahi consisted mainly of 91% glucose with minor quantities of 1.8% rhamnose and 1.8% arabinose. Most common EPS producers are Lactobacillus casei, Lactobacillus acidophilus, Lactobacillus brevis, Lactobacillus curvatus, Lactobacillus delbrueckii subsp. bulgaricus, Lactobacillus helveticus, Lactobacillus rhamnosus, Lactobacillus plantarum, Lactobacillus johnsonii etc.

Low fat dahi obtained employing EPS producing cultures of L. Lactis subsp. lactis PM23, S. thermophilus ST and L. lactis NCDC 191 found to be more acceptable in terms of body, texture and flavour in contrast to dahi made with EPS non-producing culture NCDC 167 (Behare et al., 2009). Isolated strain of Streptococcus thermophilus IG16 was capable of producing both capsular and ropy polysaccharides (211 mg/L) comprising of rhamnose and galactose (5.3:1) and its application resulted in less syneresis in curd (Behare et al., 2010). Lactobacillus fermentum V10 was capable of producing 247.37±0.76 mg/L polysaccharides and its inclusion in low fat dahi exhibited optimum acid production, lesser whey separation, higher viscosity, increased adhesiveness and stickiness whereas decreased firmness and work of shear as compared to control dahi made employing L. delbrueckii subsp. bulgaricus NCDC 285 or L. delbrueckii subsp. bulgaricus 09 cultures with or without addition of exopolysaccharides (Behare et al., 2013). Microstructural studies showed that dahi made with EPS-producing strains had relatively compact linear structure with more open structure and pores with discontinuous casein matrix than the controlled dahi (Praveen, 2000).

Modulation of incubation temperature

Fermentation occurs in three distinct and noticeable phases. Diverse chemical and biochemical reactions that occur during the lactic acidification process (Lucey, 2004) result in slight decrease in pH at the initially phase, followed by a rapid decline in pH values at second phase and the third phase is characterized by a tendency to stabilization, with little variation in the pH levels (Brabandere and Baerdemaeker, 1999, Jeanson et al., 2009). A portion of lactic acid combines with calcium to form calcium lactate. The casein devoid of calcium is coagulated at isoelectric point and the fermentation is accompanied by gelling of protein and appearance of clear whey on the surface of the product.

Faster fermentation at a higher incubation temperature may be preferred for industrial production but it can lead to several defects like an increase in whey separation (Lee and Lucey 2003, 2004, Purwandari et al., 2007), a decrease in gel firmness, viscosity and smoothness, poor sensory properties (Tamime and Robinson, 2007, Wu et al., 2009) and a weaker protein network with a coarser microstructure (Lee and Lucey 2004). Contraction of the protein strands due to rapid acidification and increased hydrophobic interactions at higher fermentation temperature resulted in a weaker network containing thinner protein strands and thereby higher syneresis (Lee and Lucey, 2004). Lower fermentation temperature may be advantageous over higher fermentation temperature. Cryo-scanning electron microscopy and confocal laser scanning microscopy of buffalo yoghurt denoted a compact microstructure with better viability of probiotic L. acidophilus La-5 at lower (37°C) fermentation temperature but firmer body with a more porous microstructure and a higher degree of syneresis at higher (43°C) culturing temperature (Nguyen et al., 2014).

Manipulation of Starter Cultures for Pleasant Flavour

Metabolic activities of starter cultures differ with the type of milk resulting in diverse fermented products with distinct chemical composition and different volatile compounds (Guler and Gursoy-Balci, 2011). An enhancement in volatile acidity (2.9 vs. 2.6 ml/50g) with the inclusion of L. acidophilus R and Leuconostoc dextranicum with LF-40 were reported (Sarkar et al., 1996 a), however a decline in volatile acidity (2.4 to 1.7 ml/50g) were encountered with the inclusion of nisin @ 25 RU/ml to the dahi after 12h of incubation (Sarkar and Misra, 2002). Vijayendra and Gupta (2014) noted higher volatile acidity production by dahi cultures in buffalo milk (32.5 ml/50 g) than in cow milk (29.2 ml/50 g) but reduced (16 ml/50 g) with the conjugated growth with probiotic cultures (Lactobacillus acidophilus and Bifidobacterium bifidum), irrespective of the type of milk. Similarly, production of volatile acidity and acetaldehyde by yoghurt cultures enhanced significantly with the inclusion of probiotic cultures in both types of milk.

Use of soya milk for the manufacture of dahi is reported but its acceptability is lower due to its weak body and unpleasant flavour. Akabanda et al. (2010) suggested fermentation of soymilk to overcome the drawbacks and to make it more acceptable like other cultured dairy products. Dahi obtained from admixture of cow and coconut milk had lower sensory scores in contrast to normal dahi. Use of flavoring agents is suggested for improvement in its sensorial acceptability (Matin et al., 2020).

Technology for Higher Shelf-Life

The major problem associated with dahi is rapid deterioration of its quality and reduction of its shelf life due to frequent microbial contamination. Bio-preservatives such as nisin, naturally elaborated by Lactococcus lactis subsp. lactis can be added to fermented milk products to restrict the growth of starter cultures to avoid post-acidification. Mitra et al. (2010) reported production of nisin by L. lactis W8 during dahi manufacture'', which exhibited antibacterial activity against spoilage and pathogenic bacteria. Due to least post-acidification (0.69% lactic acid after 120h of storage at 15 ± 1°C) and highest volatile acid production (2.3 ml 0.1 N NaOH/50 mg curd after 6 h of incubation at 42 ± 1°C), incorporation of nisin (25 RU/ml) was recommended for the manufacture of stirred yoghurt containing nisin (Sarkar, 2016 b).

Attempt have been made to obtain spray dried dahi blended with different concentrations of maltodextrin (5 to12.5%) employing an inlet temperature of 120-140oC and outlet temperature of 70oC. Inclusion of 7.5% maltodextrin in dahi is recommended to obtain spray dried dahi owing to highest survival rate of lactobacilli (0.62±.4%) (Kothakota et al., 2014) due to encapsulating effect of maltodextrin (Oldenhof et al., 2005). Effect of addition of potato mash on the shelf-life of dahi revealed a decline in shelf-life resulting from enhanced fermentation due to added potato mash (Amin et al., 2012). Glusac et al. (2015) reported less post acidification in yoghurt due to addition of honey and whey protein concentrate during 21 days of storage at 4±1°C.

Shelf-life of dahi can enhanced with starter manipulation. As per FSSAI specification dahi should have an acidity of 0.70-0.90 % (BIS, 1980). After 72 h of storage of curd obtained by culturing with Brevibacillus brevis MMB 12 had lowest acidity (0.64%) and highest viable count (4.4x1010 cfu/ml) in contrast to those obtained employing Lb. delbrueckii subsp. bulgaricus (1.26% and 0.50x1010 cfu/ml) or 1:1 mixed Brevibacillus brevis MMB 12 and Lb. delbrueckii subsp. bulgaricus (0.72% and 0.38x1010 cfu/ml) indicated that Brevibacillusbrevis MMB 12 does not have the ability to ferment lactose. Application of such starter cultures would be beneficial to avoid post acidification (Kiran et al., 2012).

Ultrasonication is an emerging non-thermal processing technique used in food and dairy industry to improve the quality properties. Ultrasound has been shown to affect dairy protein denaturation (Villamiel and De Jongm 2000) and induced lowering of fermentation time (4-5h vs. 6-7h) during the manufacture of dahi from milk sonicated at different power level (96 to 498 W) and pulsations (30-70) for 1- 2h in contrast to normal method where milk is heated to 90ºC to and cooled to 37ºC for starter inoculation. Higher firmness (125.71 ± 0.07g vs. 95.05 ± 0.02g and an improvement shelf life of the curd by 2 days due to sonication treatment were noted (Banuu Priya et al., 2015).

Shelf-life of dahi can also be extended employing a mild heat treatment process named as thermization. Thermization is done at a temperature ranging between 50°C/30 min to 70°C/14 s), which is below pasteurization temperature (Sarkar et al., 1992 a, b, Behare and Prajapati 2007). Thermization of cultured milk product restrict over-acidification and development of off flavours by limiting the growth of contaminating flora.

Conclusion

Dahi, an Indian fermented milk product may be categorized as functional food owing to its nutritional and therapeutic significance. Rheological properties of traditional dahi could be enhanced by adopting modifications for firm body (type of milk, pre-treatment of milk, manipulation of starter cultures, modulation of incubation temperature), manipulation of starter cultures for pleasant flavor. Technology modulation for higher shelf-life of dahi is also suggested. Production of dahi with improved rheological properties and higher shelf-life may extend market reach and project dahi as functional food in global market.

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