Effect of somatic cell count on dairy products: a review

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Abstract: Somatic cells are the most essential factors naturally present in milk, and somatic cell count (SCC) is used as an indicator of monitoring mastitis incidence in the herd and also to assess the quality of milk. In addition, SCC is frequently used to determine quality payments to dairy producers. The SCC is directly related to get maximum milk production from individual cow and a lower SCC indicates better animal health, as somatic cells originate only from inside the animal's udder. SCC monitoring is important because as the number of somatic cells increases, milk yield is likely to fall, primarily due to the damage to milk-producing tissue in the udder caused by mastitis pathogens and the toxins they produce, particularly when epithelial cells are lost. Keeping low SCC will allow good quality more raw milk and provide a better product to milk processors whether used as fluid milk or converted to milk based products. Somatic cells containing lipolytic and proteolytic enzymes lead to degrade major nutrients fats and proteins, respectively. Elevated SCC is related to udder inflammation, which leads to alter the normal microbial count and physicochemical parameters of milk, as well as the quality of heat treated fluid milk and milk based product. The objective of this review is to discuss on the SSC and endogenous enzymes released from somatic cells in raw milk as well as effect of somatic cells count and their endogenous enzymes in processed milk and milk based products.

Keywords: milk; mastitis; somatic cell; proteolysis; UHT

1. Introduction

Milk is accredited to be a high-value nutritional biological fluid composed of water, proteins, fat, sugar, minerals, etc. Other essential components prevailing naturally in raw milk are somatic cells (SCs), and the predominant cell type, in addition shed epithelial cells, in most species is leucocytes, comprising macrophages, polymorphonuclear neutrophils cells (PMNs), and lymphocytes (Boutinaud and Jammes, 2002). The amount of SCs, generally called somatic cell count (SCC), in milk is used as an imperative indicator of udder health since SCs are involved in protecting the mammary gland from infection as part of the innate immune system. SCs are recognized for one of the most important defense components of the mammary gland against disease or intramammary infections (Paape et al., 1979, 2002; Sharma et al. 2011). The four major cell types constituting SC, namely, macrophages, PMNs, lymphocytes, and epithelial cells. Macrophages are largely the predominant cell type in healthy cow milk. They can combat against bacterial invasion quickly by engulfing action. Moreover, macrophages take part in the specific immunity as do lymphocytes (Burvenich et al. 2003). PMNs can be enlisted and increase milk SCC when the infection continues. They can be present to a large extent in mastitic milk, even up to 92 % in bovine milk (Paape et al.,1979).When PMNs arrive at the site of infection, they phagocyte microorganisms and destroy them by using a combination of oxidative and non-oxidative mechanisms (Pham, 2006). Lymphocytes have a contributing factor in the specific immune system. They are the only cells able to distinguish the antigens through specific membrane receptors for invading pathogen.
Mammary epithelial cells are the cells that produce milk. They are shed from the mammary epithelium during lactation (Boutinaud and Jammes, 2002). The epithelial cells are the major defense line of the mammary glands, and they may play a part in the immunity of neonates in different species (Boutinaud and Jammes, 2002). The factors, animal species, milk production level, lactation stage, environmental and management practices influence the SSC in raw milk (Rupp et al., 2000). Keeping in mind the variable factors, SCC is still used as an important indicator of raw milk quality in ruminant (Hunt et al., 2013; Sharma et al., 2011). The dairy cattle udder is considered to be infected when SCC more than 2×10^5 cells per ml of raw milk, and when SCC more than 4×10^5 cells per ml of raw milk, it is deemed unfit for human consumption in the European Union (EU). The legal SCC threshold for milk acceptance in dairy industries varies in different countries, e.g., the values for bovine milk in Germany, Canada, and the USA are 1×10^5, 5×10^5, and 7.5×10^5 cells.mL^{-1}, respectively (Olechnowicz and Jaskowski, 2012; Schwarz et al., 2011).

2. Enzymes of somatic cell
SCs are an imperative source of endogenous proteins, including enzymes. A large range of enzymes are released into milk after the lysis of SCs, and among them, lipases (e.g., lipoprotein lipase), oxidases (e.g., catalase and lactoperoxidase), glycosidases (e.g., lysozyme), and proteases including elastase, collagenase and cathepsins B, C, D and G, which contribute to hydrolysis of casein (Le Roux et al., 2003; O’Brien et al., 2004). In addition, plasmin is the principle proteolytic enzyme in milk from both healthy udders and udders with elevated SCC (Leitner et al., 2006). Plasmin is a heat-stable alkaline serine proteinase which exists in milk as a component of a complex system, including its zymogen (plasminogen), plasminogen activators (PAs) and inhibitors of both plasmin and PAs (Kelly et al., 2006). Level of active plasmin in milk is influenced by on the activator-inhibitor balance, with the balance in mastitic milk being in favor of activation (Chen et al., 2003). A list of SCs enzymes their activity in milk and milk products has been enlisted in Table 1.

<table>
<thead>
<tr>
<th>Somatic cells</th>
<th>Enzymes</th>
<th>Activities</th>
<th>Reference</th>
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<tbody>
<tr>
<td>Macrophage</td>
<td>Cathepsin-B</td>
<td>Protease</td>
<td>Guha and Padh, 2008</td>
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<td></td>
<td>Cathepsin-D</td>
<td></td>
<td>Dimen et al., 1988; Guha and Padh, 2008</td>
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<td>Cathepsin-H</td>
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<td>Guha and Padh, 2008</td>
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<td>Cathepsin-L</td>
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<td>Guha and Padh, 2008</td>
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<td></td>
<td>Cathepsin-G</td>
<td></td>
<td>Campbell et al., 1989; Considine et al., 2002</td>
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<td></td>
<td>Cathepsin-S</td>
<td></td>
<td>Guha and Padh, 2008</td>
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<td></td>
<td>Elastase</td>
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<td>Campbell et al., 1989; Prin-Mathieu et al., 2002</td>
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<td></td>
<td>Lipoprotein lipase</td>
<td></td>
<td>Azzara and Dimick, 1985a</td>
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<td></td>
<td>Collagenase</td>
<td></td>
<td>Prin-Mathieu et al., 2002.</td>
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<td></td>
<td>Myeloperoxidase</td>
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<td>Considine et al., 2000</td>
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<tr>
<td>PMNs</td>
<td>Cathepsin-B</td>
<td>Protease</td>
<td>Travis and Friz 1991, Magboul et al., 2001</td>
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<td></td>
<td>Cathepsin-C</td>
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<td>Travis and Friz, 1991</td>
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<td>Cathepsin-D</td>
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<td>Cathepsin-L</td>
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<td>Travis and Friz, 1991</td>
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<td>Cathepsin-G</td>
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<td>Baggionlini et al., 1978; Considine et al., 2002</td>
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<td>Considine et al., 2000</td>
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<td></td>
<td>Elastase</td>
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<td>Baggionlini et al.,1978; Travis and Fritz, 1991; Dubin et al., 1994; Considine et al., 2000; Prin-Mathieu et al., 2002.</td>
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<td></td>
<td>Lipoprotein lipase</td>
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<td>Azzara and Dimick, 1985b</td>
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<td>Collagenase</td>
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<td>Verdi and Barbano, 1991; Prin-Mathieu et al., 2002.</td>
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<td>Myeloperoxidase</td>
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<td>Mukherjee et al., 2004.</td>
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<td>Lymphocytes</td>
<td>Elastase</td>
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<td>Prin-Mathieu et al., 2002.</td>
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<td>Epithelial cells</td>
<td>Cathepsin-B</td>
<td>Protease</td>
<td>Lah et al., 1996; Guha and Padh, 2008</td>
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<td></td>
<td>Cathepsin-D</td>
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<td>Lah et al., 1996; Seol et al., 2006; Guha and Padh, 2008</td>
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<td>Unknown cell</td>
<td>Cathepsin-L</td>
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<td>Lah et al., 1996</td>
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<td>Cathepsin-K</td>
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<td>Moatsou, 2010</td>
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<td></td>
<td>Catalase</td>
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<td>Kitchen, 1976</td>
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Presently, there are dairy industries that determine milk quality on SCC figures with the intention of obtaining products with hygienic, sanitary, dietetic, nutritional, gustative and gastronomic quality (Boyazoglu and Morand- Fehr, 2001). However, high quality dairy products can only be produced from good quality milk. Quality milk should be able to tolerate technological treatment and be converted into products that fulfill the anticipations of consumers, in terms of nutritional, hygienic and sensory aspects (Ribeiro and Ribeiro, 2010). To this end, it is very essential to understand the effect of SCC on different dairy processes and product. The main effects of SCC in dairy products include lower yield in cheeses (Politis and Ng-Kwai-Hang, 1988), increased lipolysis in yogurt (Fernandes et al., 2007) and more pronounced proteolysis in pasteurized milk (Ma et al., 2000). Considering the potential effect of somatic cell count this review focuses on the state of our knowledge of SCs in the dairy field and on their effective role in dairy processes and products.

3. Effect of somatic cell on pasteurized milk
SCC has an influence on the casein fractions of pasteurized milk, especially low-fat products. The most important consequence of the changes in casein fractions of pasteurized milk during storage is the enzymatic hydrolysis of casein associated with SCC. Certain significant sensory modifications in milk is found mainly the bitter taste due to peptides release, which were shown to be originated mostly from αs1- and β-casein (Lemieux and Simard, 1991). Compared with pasteurized low SCC milk, pasteurized high SCC milk was more susceptible to lipolysis during refrigerated storage. Milk lipases survived pasteurization. Shipe and Senyk (1981) reported that sufficient lipase activity remains after minimum pasteurization (72°C for 16 sec) and can induce rancidity during cold storage of pasteurized milk. They suggested that more severe treatment (79°C for 20 sec) was needed to completely inactivate lipoprotein lipases. Rancidity has been associated with elevated levels of FFA in milk (Shipe et al., 1980). A sensory threshold of about 1.0 meq/100 g fat has been previously reported (Bodyfelt et al., 1988; Case et al., 1985). Rancid off flavor has also been described as having a soapy, bitter, unclean taste and lingering aftertaste (Bodyfelt et al., 1988, Shipe et al., 1980). Bitterness has been associated with high levels of short-chain fatty acids (Bodyfelt et al., 1988, Shipe et al., 1980). Milk rancidity has been described as a major quality defect associated with market milk (Bandler, 1982).

During refrigerated storage, more extensive Casein Nitrogen (CN) degradation occurred in the high SCC milk than in low SCC milk. For high SCC milk samples, compared with d-1 level, CN/TP decreased by an average of 4.04% after 21 d of cold storage. Actual levels of CN degradation were probably even higher because the Kjeldahl method for nitrogen analysis may underestimate the extent of proteolysis (Driessen and van der Waals 1978). Decreased CN/TP during cold storage of milk, especially in high SCC milk, indicated that significant levels of proteolytic activity remained after pasteurization. High SCC raw milk has been shown to have high concentrations of plasmin (Driessen and Waals, 1978), plasminogen and proteases of somatic cell origin (Driessen and van der Waals 1978). Plasmin is heat stable with large percentages surviving minimum pasteurization (72°C for 15 sec) (Politis et al., 1989). Even after UHT treatment, 30 to 40% of plasmin activity can still remain (Kitchen et al., 1970). Other researchers have indicated that extensive proteolysis in milk can result in the accumulation of small in milk can result in the accumulation of small hydrophobic peptides, causing bitterness and astringency. The high SCC milk had significantly higher scores for bitter and astringent flavors at 21 d than low SCC milks. Although bitterness is rarely a problem associated with pasteurized milk, it can be a problem when levels of heat-resistant plasmin are high, as is the case during mastitis (Kitchen, 1981). Proteolysis wills a more significant problem in extended shelf life of refrigerated fluid milks.

4. Effect of somatic cell on cheese
The production of dairy products from milk with elevated SCC has been characterized by reduced product yield, reduced yield efficiency, increased losses in the production of cheese (e.g. whey) and reduced product quality (Auldist et al., 1996; O’Brien et al., 2004). Authors agree that as SCC increases fat in whey increases, moisture in cheese increases and protein in cheese decreases. Protein and fat recovery were found to considerably reduce as milk SCs increased. The recoveries of fat and protein in cheese are a more consistent method of evaluating the effects of SCC on cheese yield than comparing definite and adjusted cheese yields recommended by Lucey and Kelly (1994). They indicated that the reduction in the recoveries of fat and protein in cheese with elevated SCC may be due to impaired rennet coagulation and cheese making properties or increased proteolysis and lipolysis in high SCC milk.

Elevations of SCs in milk have a parallel relationship with the moisture content of cheese, stated by Barbano et al. (1991), Auldist et al. (1996), Vianna et al. (2008). Conversely, this conclusion is not undisputed across the literature with Cooney et al. (2000) and O’Brien et al. (2004) finding no significant correlation between SCC and cheese moisture content which were also counted in the meta-analysis. Increases in cheese moisture with
elevated SCC may be triggered by a slow, weak congealing due largely to altered milk protein composition, mineral disproportion and an increased milk pH (Auldist, 2000). As milk SCS increased the protein content of cheese significantly decreased which is asserted by Cooney et al. (2000) and Andrea et al. (2007). Vianna et al. (2008) found no significant difference in the protein content of Prato cheese produced with high (>700,000 cells/mL) and low (<200,000 cells/mL) SCC milk. The reduction in protein recovery can clarify fully this decline in cheese protein with elevated SCC. Auldist (2000) attributes this as being largely due to reduction in casein as a percentage of total protein, since it is commonly casein that is adjusted into the curd, while the whey is expelled during syneresis. In addition it could be argued that the increased moisture content of cheese as SCC increased could adversely influence the milk solid not fat (SNF) content of the cheese. Cooney et al. (2000) affirmed that as SCC increased the fat content in cheese increased (P<0.05). On the other hand, Rogers and Mitchell (1994) identified that as milk SCC increased cheese fat decreased, explained by increased fat losses to the whey. Very sporadic studies found a significant relationship between milk SCC and the fat content of cheese.

Author’s largely reach in an agreement that there is a detrimental effect on the organoleptic properties of cheese occur when the levels of SCC increase (Barbano et al. 1991; Rogers and Mitchell, 1994; Popescu and Angel, 2009). Auldist and Hubble (1998) found that negative effects on the organoleptic properties of cheese were testified for milk with SCC as low as 100,000 cells/ml. Grandison and Ford (1986) concluded that even a small increase in SCC can negatively impact cheese processing and Seynk et al. (1985) recommended cheese manufacturers to keep SCC <200,000 cells/ml. The available literature highlights the significance of conserving low bulk milk somatic cell count (BMSCC) for high quality cheese production. The cheese yield will decrease by up to 1-4%, depending on the level of somatic cells. In the figure above, the loss in cheese is 318g per 100 kg milk when the somatic cell count increases from 240,000 to 640,000, equal to 3.26%.

5. Effect of somatic cell on ultra-high temperature (UHT) product
The UHT milk usually has a shelf-life of four months and no modifications in the milk components should occur during this period. In particular, the changes in the proteins of UHT milk during storage are of major concern to the dairy industry (Recio et al., 1996). However, there is little information available on the influence of SCC on the casein fractions of UHT milk, except for a report by Auldist et al. (1996) who found that UHT milk produced from raw milk with a high SCC tended to gel faster than low-SCC milk. Gelation of UHT milk during storage (age gelation) is a major factor limiting its shelf-life. The gel which forms in UHT milk is a three-dimensional protein matrix formed by the whey proteins, particularly β-lactoglobulin, interacting with casein, chiefly κ-casein, of the casein micelle. The major proteinaceous linkages which develop during the heat treatment result in formation of b-lactoglobulin—κ-casein complexes (βk-complexes) (Elfgam and Wheeock, 1978). The following factors which influence the quality of milk have an effect on the gelation behavior of UHT milk.

The proteolysis has been attributed to both the natural milk proteinase and proteases produced by psychrotrophic bacterial contaminants of raw milk. The proteolytic activity associated with SCC in fluid milks has been extensively studied (Senyk et al., 1985; Verdi et al., 1987; Saeman et al., 1988; Ballou et al., 1995). Grufferty and Fox (1988) reported that proteolysis of casein caused by milk proteinase is responsible for gelation of UHT milk during storage. Approximately 80% of milk total nitrogen from bovine milk is in casein. Bovine casein can be classified into four types of protein with different properties: αs1-, αs2-, β- and κ-casein, which account for 38%, 10%, 34% and 15% of total casein, respectively (Fox et al., 2000).}

associated with SCC (Saeman et al., 1988; Ballou et al., 1995), which leads to the corresponding reduction of αs1- and β-casein fractions in milk (Randolph et al., 1974; Verdi et al., 1987). As some SCC-related proteases are heat-resistant (Prado et al., 2006), such effects are expected to occur in ultra-high-temperature (UHT) milk during storage. Proteolysis of UHT milk during storage at room temperature decreases its shelf life and might cause undesirable precipitation or gelation (Topçu et al., 2006). The enzymatic hydrolysis of casein, associated with SCC, may also lead to detrimental sensory changes in milk, mainly a bitter taste due to the release of bitter peptides (Prado et al., 2006).

Mastitic milk (i.e. with high somatic cell count, SCC) subjected to UHT treatment is more susceptible to gelation than normal milk (Swartling, 1968). This has been attributed to increased proteolytic activity resulting from an elevated level of plasmin (Bastian and Brown, 1996; Saeman et al., 1988). Auldist et al. (1996) showed that, during storage at 20°C, UHT-treated high-SCC milk showed more proteolysis than low-SCC milk, but the level of proteolysis did not correlate with the tendency of the milks to gel. Stage of lactation was a much more significant factor than SCC; however, at any particular stage of lactation, the milks with the highest SCC gelled fastest. Kelly and Foley (1997) reported that (indirectly) UHT-treated high- and low-SCC milks had very little residual plasmin activity and a low tendency to gel. However, with added plasminogen both high- and low-SCC milks showed gelation, which was more pronounced in high-SCC milk. They concluded that low levels of plasmin, arising from activation of plasminogen by an elevated level of plasminogen activator in the high-SCC milks, were responsible for this enhanced gelation.

6. Effect of somatic cell count on fermented product

Probiotic fermented dairy products, mainly yoghurt, has been consumed with a long history of safe use (Maragkoudakis et al., 2006). The term probiotic is defined as live microorganisms that when administered in adequate amounts confer a health benefits on the host (Vasiljevic and Shah, 2008). Probiotic bacteria as well as yoghurt bacteria have proteolytic activity. Proteinases and peptidases constitute the primary enzymes in lactic acid bacteria for proteolysis in milk protein as a source of amino acids and nitrogen (Donkor et al., 2006). Previous studies have shown that the proteolytic activity of S. thermophilus, L. delbrueckii spp. L. bulgaricus, and L. acidophilus was much higher than that of Bifidobacterium spp. (Shihata and Shah, 2000). The possible effect of different SC levels in milk on the nitrogen components and proteolysis index of yoghurt milk and resulting probiotic set yoghurt (Bavarian et al., 2010). Proteolysis of casein leads to decrease in the relative proportion of caseins (αs1 and β-casein) with simultaneous clear increased levels of γ-caseins and protease peptones (Leitner et al., 2006). Proteolysis in processed milk has been measured by monitoring changes in nitrogen levels such as decreases in casein nitrogen (CN) or increases in non-protein nitrogen (NPN). These changes have been linked to changes in functionality, such as micro-structural changes (e.g. casein flocculation) and increases in viscosity in UHT milk (Chen et al., 2003).

The contribution of the activities of certain enzymes to the quality of certain dairy products has been the subject of considerable researches, for example, increased plasmin activity is accompanied by increasing clotting time, loss of moisture in cheese, reduced curd stability and yield (Kelly & Fox, 2006). Reduced amount of TN in yoghurt milk samples by elevation of SC levels was due to the reduction in the synthesis and secretion ability of mammary tissue as a result of mastitis (Lee et al., 1991; Ogola et al., 2007). Decreasing trend in CN concentration is likely related to activation of plasmin system resulting in partial CN degradation (Leitner et al., 2006). Most previous studies have concerned cathaecin C and G collagenase and elastase activities in blood during the inflammatory response, which exhibit proteolytic activity towards bovine casein (Haddadi et al., 2006).

7. Conclusions

Previously various attempts were taken to study the effect of SCs but a few facts have been carried out to highlight the effective role of SCs on dairy products without the other parallel factors in high-SCC in milk. This review would provide a thorough idea on the SC count and composition and, therefore, their endogenous enzyme profiles in milk. Such a thumbprint would support to depict more exactly the SC impact on technological properties and ultimate quality of dairy products besides the primary microbiological and nutritional features of milk. By considering the literature data, we have pointed out in this review that SCs can have a desirable role, in relations of hastening of proteolysis in the cheese-ripening process and improvement of cheese sensory quality, increased lipolysis in yogurt and more pronounced proteolysis in pasteurized milk. Dairy industry is a large and dynamic segment of the agricultural economy of many nations and in current situation this industry is the backbone of developing countries. So this literature will certainly help to enhance the
knowledge on SCC and their effect in dairy processes and products so that a good and profitable dairy business could establish for the advancement of different developing countries.

Conflict of interest
None to declare.

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