SOMATIC CELL COUNT AS AN INDICATOR OF UDDER HEALTH STATUS UNDER MODERN DAIRY PRODUCTION: A REVIEW

A. SHARIF AND G. MUHAMMAD[1]

Livestock and Dairy Development Department, Punjab, Lahore;
[1]Department of Clinical Medicine and Surgery, University of Agriculture, Faisalabad, Pakistan

INTRODUCTION

Mastitis, an important disease of dairy animals, influences the quality and quantity of milk. Penetration of pathogenic microorganisms in the teat canal irritates and invades the delicate mammary tissue, causing an inflammatory response and consequent changes occur in the milk. Degree of these changes depends on the infecting agent and the inflammatory response. Mastitis, the inflammation of the udder regardless of the cause, is the most costly disease of dairy cattle and results in severe economic losses from reduced milk production, treatment cost, increased labor, withheld milk following treatment and premature culling. Among infectious agents, bacterial pathogens are major threat to mammary gland. These microorganisms are often contagious, widely distributed in the environment of dairy animals and thus increase prevalence rate of intra-mammary infections (IMI).

Field surveys of major livestock diseases in Pakistan have ranked mastitis as number one disease of dairy animals (Candy *et al.,* 1983). The disease in Pakistan is mostly contagious in nature (Ali *et al*., 2008). It affects the milk quality and production of cow along with changes in the composition of milk, and the extent to which various compositional changes occur depends on the inflammatory response (Kitchen, 1981; Korhonen and Kaartinen, 1995). Main changes in the udder include; leakage of ions, proteins and enzymes from the blood into the milk due to an increased permeability, invasion of phagocytising cells into the milk compartment, and a decrease in the synthetic capacity of the gland, resulting in decreased concentration of certain milk constituents (Korhonen and Kaartinen, 1995). The affected quarter may also produce substances related to the inflammatory reaction such as acute phase proteins (Eckersall *et al.,* 2001).

Sub-clinical mastitis is important due to the fact that it is 15 to 40 times more prevalent than the clinical form, is of long duration, difficult to detect, adversely affects milk quality and production of dairy animals and constitutes a reservoir of microorganisms that can affect other animals within the herd due to its contagious nature (Schultz *et al.,* 1978). Losses due to mastitis may even be higher in developing countries because standard mastitis control and prevention practices (e.g. pre and post milking antiseptic teat dipping and dry period antibiotic therapy) recommended by National Mastitis Council (NMC) of USA are not being carried out in these countries (Nickerson, 1994). Milk available to our human population is contaminated with pus due to high incidence of mastitis in our dairy animals (Allore, 1993; Khan and Muhammad, 2005), with high occurrence of pathogenic microbes in dairy buffaloes (Ahmad, 2001). The gold standard to measure inflammation is the cytological examination i.e. milk somatic cell count (SCC) and other methods are compared with SCC (Hamann, 2002). The diagnosis of mastitis according to the International Dairy Federation (IDF, 1971) recommendations is based on the SCC and microbiological status of the quarter. This paper reviews the importance of detection of sub-clinical phase of mastitis based on SCC and deleterious effect of high SCC on milk production and composition.

SOMATIC CELL COUNT IN MASTITIC MILK

An inflammatory response is initiated when bacteria enter the mammary gland. Somatic cell counts and bacteriological examination indicate the status of mammary gland as SCC in milk increases during intra-mammary infection (Harmon, 1994). The SCC of milk is widely used to monitor udder health and the milk quality. Elevated SCC primarily consists of leucocytes, which include macrophages, lymphocytes and neutrophils. During inflammation, major increase in SCC is because of the influx of neutrophils into milk and at that time over 90% of the cells may be polymorphonuclear leukocytes. Higher the SCC, greater is the risk of raw milk contamination with pathogens and antibiotic residues (Jones, 2006).

Mastitis is characterized by an increased number of inflammatory cells, and SCC in milk is used as an indirect measure of the degree of the udder health. The single most important factor affecting SCC in milk is mammary gland infection and all other factors such as age, stage of lactation, weather are of minor importance (Eberhart *et al.,* 1979; Reneau, 1986). Kitchen (1981) commented that use of SCC to diagnose udder disease was the first widely used screening procedure and even today it has retained its position as the most reliable and specific test for mastitis diagnosis. The somatic cells present in the milk of a healthy cow are mainly macrophages (66-88%); in addition, there are

neutrophils, and epithelial and mononuclear cells (Ostensoon *et al.,* 1988; Sandholm 1995). The proportion of neutrophils is only 1-11% in a healthy quarter but increases upto 90% in a quarter with IMI (Sandholm, 1995). The proportion of neutrophils as the percentage of the SCC has been proposed as a mastitis indicator (Hamann and Kromker 1997). Jafri (1981) carried out total and differential leukocyte count in the milk of Holstein cows in Pakistan and indicated higher leukocyte count in udder inflammation than normal. Instead of total SCC, differential cell counting may provide more information about the health status of a quarter (Ostensoon *et al.,* 1988), but this approach has not been widely adopted in practical conditions. Milk SCC has been used extensively as an indicator of IMI since nineteen-sixties. SCC has been included as a component of the definition of mastitis and the original limit for SCC of a healthy quarter was 500,000 cells/ml (IDF, 1971). The threshold of levels for SCC is based on a population mean plus two times standard deviation for one measurement of the foremilk from an individual quarter. The definition is a guide for diagnosis, even though 50% of truly infected quarters could at any time have a cell count less than the break-point of 500,000 cells/ml (IDF, 1971).

The inflammation of the mammary gland can be characterized by an increase in SCC. This trait is used as an indicator of udder health for management and selection purposes (Rodriguez *et al.,* 2000). SCC values higher than 283,000 cell/ml indicate the presence of mastitis (Guidry, 1985; Reneau, 1986). It has also been pointed out that SCC is always compared with bacteriology, and these tests can never agree completely. Recently, a SCC limit of 100,000 cells/ml is suggested for a healthy quarter (Hillerton, 1999) and SCC for the composite milk from a cow should not exceed 100,000 cells/ml (Kromker *et al.,* 2001).

The composition of milk is markedly influenced by the health status of the udder. Inflammatory process or mastitis generally leads to an increase in SCC in milk which has been associated with changes in milk components and properties (Auldist and Hubble, 1998). In most developed countries milk quality is defined by the SCC and bacterial count ("standard plate count" or SPC) in pre-pasteurized bulk tank milk. Somatic cells are composed of white blood cells (WBC) and occasional sloughed epithelial cells. Most cells found in normal bovine milk are WBC (macrophages) that function as early warning signals when bacteria invade the udder. The SCC of a cow that is not infected with mastitis is usually less than 200,000 cells/ml and many cows maintain SCC values of less than 100,000 cells/ml. When mastitis causing bacteria invade the udder, the macrophages present in the udder signal the cow's immune system to send neutrophils to the udder to engulf and destroy the bacteria. More than 90% of SCC in infected gland is composed of neutrophils (Miller and Paape, 1985) and SCC greater than 200,000 cells/ml is almost always caused by mastitis (NMC, 1996; Jones, 1986). SCC in sub-clinically affected mastitis Jersey cows varied from 21000 to 330000 cells/ml in Pakistan (Ahmad *et al.,* 1988).

EFFECTS OF HIGH SCC

Milk production

Mastitis, caused by a variety of pathogenic microbes, is characterized by tissue changes leading to progressive change to the secretory apparatus and resulting in loss of milk production. Koldeweij *et al.* (1999) found linear relationship between production loss and Log_{10}(SCC). The established association between milk production and SCC has been increasingly used to estimate production loss due to mastitis (Barlett *et al.,* 1990). Jones (1986) suggested that SCC of 0.6 to 1 million cells/ml were associated with an 8 to 12% reduction in herd milk production. The greatest loss associated with high SCC is reduced milk yield. Additional losses are associated with changes in milk quality and composition. Increased cost also results from treatment, discarded milk and premature culling. The possibilities of drug residues in milk are also increased. In Nili-Ravi buffaloes, mastitis shortens lactation period of each animal by 57 days on an average and reduces 438 Kg of milk per lactation (Candy *et al.,* 1983). Jones (1986) reported that lowering the maximum available SCC is beneficial for milk producers and processors. Lower SCC results in higher milk yield and better milk quality. It is indicated that 6% of quarters could be infected when the bulk tank milk has a SCC of 200,000 cells/mL. This count is often considered as the measuring point for mastitis. By comparison, it has been estimated that a herd with a bulk tank SCC of 500,000cells/mL would have 16% infected quarters with 6% reduction in milk yield (NMC, 1996). When a herd had a bulk tank SCC of one million, 32% of quarters would be infected and loss in milk yield was estimated at 18%. Production loss calculated was percent of production expected at SCC of 200,000 cells/mL (NMC, 1996). Earlier studies at Washington State University compared mastitis control practices used by herds with low and high SCC (Hutton *et al.*, 1990). The herds with below 283,000 SCC also produced more milk per cow, with few cows infected with coagulase positive staphylococcus (*S. aureus*) and environmental streptococci. They reported that 25% of the cows in high SCC herds were infected with contagious mastitis pathogens. Zitny *et al.* (1995) studied the correlations of SCC with changes in milk production and composition of cows and found significant correlations between SCC and yield of milk, milk fat, milk protein and lactose. For cows producing milk with a SCC of <400000/ml of milk, the average milk yield was 3653 Kg/lactation, compared with 3289

Kg/lactation for cows with SCC of >400000/ml. Yields of milk fat, protein and lactose per lactation for cows with SCC of <400000/ml were 150 (4.11%), 123 (3.36%) and 171Kg (4.68%), respectively. For cows with SCC of >400000/ml, these figures were 135 (4.10%), 113 (3.43%) and 153Kg (4.64%), respectively. Kukovics *et al.* (1996) studied phenotypic correlations between SCC and milk components and found a negative correlation between SCC and daily milk yield and lactose content, and a positive correlation between milk fat, milk protein content and pH, although these correlations varied according to lactation and year.

Milk composition

Microbial infections change milk composition and render milk less suitable for consumption and processing. Research has shown conclusively that elevated SCC (>200,000 cells/mL) had a significant impact on the udder. Mastitis causes injury to milk secretory cells in the mammary gland which interferes with the synthesis of lactose, fat and protein (Schallibaum, 2001). It also affects the milk yield. There are changes in the permeability of membranes, which lead to increased leakage of blood components into the udder and changes in milk composition. Milk with elevated SCC has been referred to as mastitic milk. Sharif *et al.* (2007) estimated effect of severity of mastitis on milk lactose content in normal and sub-clinically affected dairy buffaloes and observed decreasing value of milk lactose with the severity of mastitis. Increase in SCC is correlated with a greater potential for proteolysis and consequently, with increased proteolysis indexes (Schaar and Funke, 1986; Auldist *et al.,* 1996). Mastitic milk undergoes more proteolytic activity than normal milk (Urech *et al.,* 1999). Haenlein *et al.* (1973) reported a significant decrease in casein content when either Holstein or Guernsey milk exceeded SCC of 500,000; depression was greater above one million SCC. SCC above 500,000 has been associated with poor quality cheese because of increased rennet to cutting time and lower curd firmness. The mastitis or elevated SCC is associated with a decrease in lactose, α-lactalbumin, and fat in milk because of reduced synthetic activity of mammary tissue (Harmon, 1994). In buffaloes, SCC and milk lactose are negatively correlated with each other and measuring lactose can reveal a change in SCC of buffalo milk in comparison with normal (Sharif *et al.,* 2007). The reduced lactose concentration is one of the very important factors for impaired acidification properties of milk with elevated SCC after adding starter cultures (Schallibaum, 2001). Delayed acidification favours the growth and survival of human pathogens that may be present in the raw material and impair the hygienic safety of the end product. With higher SCC, the concentrations of serum albumin and immunoglobulin increase which reduce heat stability of mastitis milk, causing coagulation, or flocculation during pasteurization, UHT treatment, etc. Pasteurized milk made from milk with over 500,000 SCC gave lower grade scores after storage. Also there is a decrease in calcium absorption from blood into milk and calcium acts as co-factor responsible for impaired coagulation characteristics of mastitis milk. Reichmuth (1975) showed graphically that as SCC exceeded 150,000, the concentrations of sodium, chloride, and whey nitrogen increase, whereas milk yield, potassium, and lactose decrease. Everson (1980) observed that milk with SCC of 700,000 and greater was rancid. Rancid and lipolyzed flavors are related to the breakdown of fats to various short-chain fatty acids. Gudding (1982) found that *S. aureus* infections caused higher free fatty acid concentration of milk. Milk with low SCC is justified economically. Benefits to the processor include higher cheese yield, better cheese quality, and higher hygienic safety of the product (Schallibaum, 2001). With decreasing bulk milk SCC, fat and lactose contents increased, with little effect on protein content (Schukken *et al.,* 1992). Fernandes *et al.* (2004) investigated the relationship between SCC and composition (total solids, fat, protein and lactose content) of milk from individual Holstein cows and indicated that SCC of individual cow's milk significantly correlated with a decrease in milk constituents only under conditions of average SCC in bulked milk above 1,000,000 cells/mL. Significantly reduced lactose content of milk in inverse proportion to the number of leukocytes has been observed in buffaloes in Pakistan (Qureshi and Ahmad, 1980) and in cows in India (Chandra, 1992). Negative correlation between lactose and chloride content has also been established in sub-clinically affected cows with high SCC (Ahmad *et al.,* 1988).

Milk quality

Milk is an important part of the diet of human beings. The nutritional requirement of the human body is fulfilled by consuming about a quart of quality milk containing vitamin A, ascorbic acid, thiamine and calories of energy needed daily by an average individual (Bilal and Ahmad, 2004). SCC is a useful index for detection of sub-clinical mastitis and milk quality. The SCC of normal milk is less than 200,000 cells/mL, higher SCC is considered as abnormality in milk and indicates udder infections. High SCC causes a rise in whey protein and a decrease in casein, resulting in a considerable lower cheese yields. Shorter shelf life and adverse milk flavor are other consequences of high SCC.

Ma *et al.* (2000) looked at the relationship between high SCC and quality of pasteurized fluid milk and created high SCC and clinical mastitis by infusing *Str. agalactiae* into two quarters of eight cows that had low SCC (less than 58,000). Milk samples were

commingled to achieve a SCC close to 750,000 and were then pasteurized at 165°F for 34 seconds. This milk was compared to pre-infected milk. At day 1, the high SCC milk smelled different, was less sweet, and lacked freshness aroma. Milk protein and fat breakdown occurred and acid degree value (rancid milk) increased. Between 14 and 21 days of storage, the high SCC milk had a greater rancid aroma and taste and also a bitter and astringent taste, followed by a bitter after taste. Milk fat continued to breakdown during cold storage, indicating that the responsible enzymes survived pasteurization. After some of the milk from these mastitis infected cows was processed, it contained high standard plate counts, psychrotropic bacteria and coliform counts. They concluded that the changes in milk, associated with development of mastitis, caused a deterioration of flavor quality and shelf life of milk after pasteurization. They suggested fluid milk processors to use milk with low SCC when seeking to extend shelf life and quality beyond 14 days of storage.

Significant decrease in milk main constituent i.e. lactose is observed in buffaloes in relation to the severity of mastitis (Sharif *et al.,* 2007). Casein, the major milk protein of high nutritional value, declines and low quality whey proteins increase which adversely affect the quality of dairy products such as cheese. Serum albumin, immunoglobulins, transferrin and other serum proteins pass into milk because of increased vascular permeability (Haenlein *et al.,* 1973). Milk lactose, an important disaccharide present in milk, is formed by the mammary gland from glucose or glycogen. Dietary or blood sugar level does not alter the lactose content of milk but the gland with clinical mastitis shows a decrease in lactose and increase in chloride contents (Schalm *et al.,* 1971). Severity of mastitis also decreased the food value of milk in terms of reduced protein and fat contents (Ullah *et al.,* 2005). In Holstein cows, total leucocyts and neutrophils in milk had a highly significant positive correlation with total whey proteins, lact-albumins and gamma globulins (Jafri, 1981).

PHYSIOLOGIAL FACTORS AFFECTING SCC

Stage of lactation

The secretion of somatic cells in milk is influenced by the number and stage of lactation, and management practices (Harmon, 1994). Milk from uninfected quarters displays change in SCC as number of lactations or days in milk increase. SCC of milk from uninfected quarters rise from 83,000 at 35 days postpartum to 160,000 by day 285. Lactation stage affects the SCC as after parturition SCC is high and then decreases to the normal level within 4-5 days after calving (Barkema *et al.,* 1999). Towards the end of lactation period, SCC again increases slightly (Brolund, 1985; Miller and Paape, 1988). SCC decreases to a low level within three days in healthy quarters but remains high in the infected ones; thus SCC can be used in early post-partum to detect new IMIs (Barkema *et al.,* 1999). Since SCC in uninfected cows is high at freshening, lowest from peak to mid-lactation, and highest at drying off, a plot of monthly SCC inversely correlates to lactation curve (Reneau, 1986). A modest rise in the SCC of the uninfected quarters at the end of lactation is in fact a dilution effect (Harmon (1994). Schepers *et al.* (1997) described that the logarithm of SCC was high at the beginning of the lactation, dropped to a minimum between 40 and 80 days postpartum, and then steadily increased until the end of lactation. In the work of Rodriguez *et al.* (2000), the SCC decreased to a nadir at about 60 day of lactation and then increased, although not in a monotonic mode, without regaining the initial level. SCC increases in older cattle and/or at the end of lactation due to increased prevalence of infection and permanent glandular damage from previous infections (Barlett *et al.,* 1990). In buffalo milk, number and stage of lactation did not affect the somatic cells and lower average SCC such as 136000/ml in July-August, 10800/ml in May-June and 76000/ml in December-January is reported in Indian Murrah buffaloes (Singh and Ludri, 2001).

Milking frequency

Milking frequency also affects milk SCC. A shift from two times a day to three times a day milking decreases bulk milk SCC and the proportion of high SCC cows (Hogeveen *et al.,* 2001), while very short milking intervals (4 h and less) increase SCC (Hamann, 2001). Long milking intervals with automatic milking systems (AMS) increase bulk milk SCC (Pettersson *et al.,* 2002) and this is suggested to be due to the increased number of IMI and rise in the milk SCC of individual cows with the longest milking intervals. Kelly *el al.* (1998) investigated the effects of reducing the frequency of milking of cows in lactation on milk SCC, polymorphonuclear leukocyte (PMN) count, chemical composition and proteolytic activity in Holstein Friesian cows. Milk lactose levels were significantly decreased and pH, lactalbumin levels, plasmin and plasminogen activities significantly increased by reducing milking frequency.

According to Kukovics *et al.* (1996), SCC were higher in afternoon than in morning, and also increased with age, year and lactation number. Significant differences were observed between breeds. Schukken *et al.* (1992) conducted a study to evaluate the Bulk Milk Somatic Cell Count (BMSCC) control program and observed the effect of the program time on milk composition and milk quality. The data from approximately 9500 farms in Ontario, Canada, was analyzed. The SCC data showed a seasonal pattern. The expected lowest mean SCC occurred in April and expected highest mean SCC occurred in October

Percentage of fat and lactose increased significantly with decreasing bulk SCC.

Conclusions

Understanding the relationship between the production of high quality milk and SCC due to mastitis in dairy herds is fundamental for the profitability of the dairy business. 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. Optimum production and maximum daily yield of milk can only be achieved if mastitis is prevented at herd level by adopting guidelines of Mastitis Control Program. The intra-mammary infections among dairy animals persist for longer periods of time, associated with elevated SCC, and affect milk production in dairy animals. The disease involves interplay between management practice and infectious agents. High prevalence of mastitis can be controlled by routine screening tests. Problem of mastitis is encountered from both contagious and environmental pathogens. The successful control of mastitis relies much on antibiotic treatments, which is a challenge in food animals, particularly use of broad spectrum, multi-component products and use of prophylactic treatment. Curative therapy with antibiotics remains only moderately effective and depends on the stage at which disease is treated. The implementations of research conducted on mastitis control through better management would result in flourishing of prosperous and stable dairy industry especially in developing countries. Managers of dairy herds should cull animals having recurrent mastitis, this will decrease treatment costs, avoid recurrent use of antibiotics and overcome problem of resistance and drug residues in milk. Proper milking techniques, improved sanitation, effective use of teat dipping and dry period therapy and improvement in management are needed to reduce SCC by reducing the spread of new infections.

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