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Influence of Season on Total and Differential Bulk Cow Milk Somatic Cell Counts in Dromedary (*Camelus dromedaries*) and Bovine Milk

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Abstract: The influence of season on Somatic Cell Counts (SCCs), differential leukocytes (Macrophage (MAC), Lymphocyte (LYM) and Poly Morphonuclear Neutrophils (PMNs)) in camel's and cow's milk was compared. Determining the Somatic Cell Count (SCC) in the cow's milk samples, we established that during the Winter season the somatic cell count was significantly higher in comparison to the dry season ($p<0.05$). However, our results did not determine significant effect of season on SCC in dromedary milk. In wet season, macrophages were the predominant cell type in camel's milk and PMN in cow's milk. In camel's milk, MAC were the dominant cell type at the wet season and tended to maintain high levels as season changed. In cow, PMN numbers were higher in wet season and declined gradually with advanced season while LYM number increased in dry season ($p<0.05$). Our results suggest that camels are more resistant to season variation in comparison to cows.

Key words: Somatic cells, leukocyte populations, camel's milk, season, dry, suggest

INTRODUCTION

Dromedary milk (*Camelus dromedaries*) can servive and produce considerable amount of milk in recurrent and prolonged hot and dry environment (Bekele *et al*., 2011). Thus, camel milk is considered one the most valuable food sources of nomadic people in arid and semi-arid areas and had been consumed for centuries due to its nutritional values and medicinal properties (Kenzhebulat *et al*., 2000; El Zubeir and Nour, 2006; Farah *et al*., 2007; Lorenzen *et al.,* 2011). However, most camel milk is produced in traditional farming or pastoral systems by hand milking that cannot provide consistent quantity and quality of raw milk for urban markets (Abeiderrahmane, 2005). The Somatic Cell Count (SCC) in bovine milk is an indicator of udder health and milk quality and can be related to the cellular immune response after an inflammatory stimulus (Leitner *et al*., 2000a, b, 2006, 2008; Ostensson *et al*., 1988; Silanikove *et al*., 2006; Vangroenweghe *et al.,* 2002).

All milk variety contains a certain level of somatic cells. These are Neutrophils (PMN cells), Lymphocytes (LYM) and Macrophages (MAC). The increase in SCC results from a transfer of white blood cells (immune cells) from the blood to the mammary gland (Sladek *et al*., 2006; Verdi and Barbano, 1988). The increase in SCC in milk from infected animals is accompanied by a change in the leukocytes (Krrchen, 1981; Paape and Tucker, 1966). A typical Intra Mammary Infection (IMI) with coliforms is associated with a dramatic increase of the PMN level up to 90%. Thus, the number of PMNs may be a more useful indicator in the evaluation of udder health than the SCC (Vangroenweghe *et al*., 2002). In addition to IMI, stage of lactation, season, milk yield and number of lactations also influence SCC (Brolund, 1985; Harmon, 1994; Kennedy *et al*., 1982; Verdi and Barbano, 1991; Silanikove *et al*., 2010) Somatic cell counts have been determined in camel milk in the past but mainly from samples of individual animals to diagnose clinical or subclinical mastitis (Abdurahman, 1995, 1996; Chaffer *et al*., 2000; Guliye *et al*., 2002). To our knowledge, the interaction between season and cellular counting has not yet been fully considered in camel species. Therefore, the purpose of the present study was to examine variation in SCC and their subtypes in camel's milk between seasons and to compare them with cow's milk in similar situation.

MATERIALS AND METHODS

Milk sampling: The study was carried out using individual milk samples from 36 dromedary animals (*Camelus dromedarius*) of Maghrabi breed from the South and the Center of Tunisia and from 52 cows (Tunisian Holstein from one herd in the region of Sfax). The dromedaries were fed throughout the year exclusively

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by grazing. In the case of cows, there is no grazing area and the herd's nutrition is based on roughages and concentrates (aboveground system). For both species, individual samples during early morning milking were collected into sterile bottles between May 2008 and March 2009 in 2 different days. The first few streams of milk from each quarter were discarded. Bovine samples were obtained by automated milking systems but dromedary samples were obtained by manual milking.

Cow and dromedary milk samples were collected and transferred immediately to the laboratory without preservative. Upon arrival each individual milk sample (500 mL) was divided in two portions into sterile bottles. The first sub-sample was stored at 4°C until SCC analyses. The second was stored at -18°C until the rest of analyses (protein fractions, casein fractions) were performed.

Somatic cell count: Somatic cells were counted using a Fossomatic 5000 (Foss Electric, Hillerod, Denmark) according to International Dairy Federation Standard (IDF, 1995).

Differential somatic cell counts: Differential SCC was determined microscopically in smears stained with May-Grunwald and Giemsa reagents. In addition from each milk sample, two preparations were used for differential cell staining and determination of the cell type (Gargouri *et al*., 2008; adapted for cow's milk). In each slide, the SCC was enumerated in 16 subsections. The working factor was 393.1 in all cases and the average values were used for statistical analyses. The working factor was obtained taking into account the field diameter (0.18 mm), the observed total area (100 mm^2) and the milk volume (0.01 mL).

Statistics: Statistical evaluations were performed using SPSS Software (Version 13). Firstly, the frequency distributions for SCC were skewed and thus log10 transformation was applied prior to the analyses. Data were arranged according to two seasons; dry season (Aug-Oct) wet (Nov-Jan). Data were analyzed by one-way Analyses of Variance (ANOVA). The differences among the means of the analyses data were compared at a significance level of $p<0.05$.

RESULTS AND DISCUSSION

The data presented in Table 1 reveals the SCC was significantly higher in wet season compared to dry season in cow milk. According to our findings most researchers reported on higher SCC in milk in winter (Dobranic *et al*., 2008; Rajcevic *et al*., 2003). Contrary to the reports by Hanuo and Gabriel (1991) and Agabriel *et al*. (1995) who found that somatic cell counts are generally lowest during the Winter and highest during the summer which coincides with an increased incidence of clinical mastitis during the summer months (Smith and Hogan, 1995; Smith *et al*., 1985; Todhunter *et al*., 1991, 1995). Our results suggested that the cold stress could have increased the susceptibility to infection as well as increased the numbers of pathogens to which cows were exposed. Additional data support the association of rates of clinical mastitis with bacterial counts in bedding (Smith and Hogan, 1995; Todhunter *et al*., 1995). These findings support the concept that the cold stress per se is not the cause of increased SCC but the increased SCC is a result of greater exposure of teat ends to pathogens, resulting in more new infections and clinical cases during the wet months. Precipitation also poses a series of problems and ingestion of pathogens resulting in environmental mastitis (Bramley, 1992; Hogan and Smith, 2003).

Table 1: Total somatic cell count in camel's milk and cow's milk (mean±SE)

Parameters	Wet season	Dry season	p-values
Camel milk			
SCC (10^3 cell/mL)	171.44±83.86	47.1±18.29	0.13
Log_{10} SCC	2.23±1.92	1.67±1.21	0.97
Cow milk			
SCC (10^3 cell/mL)	260.30±76.67	38.77±11.80	0.00
Log_{10} SCC	2.41±0.88	1.58±0.52	0.03

In this study, SCC in camel milk did not change with season. These results are consistent with Coleman and Moss (1989) who found that the season does not affect the SCC.

We could not correlate this seasonal change in SCC to any previous data in camels. However, Nagy *et al*. (2013) showed for log SCC, the mean was lowest in summer and highest in autumn. However, such a seasonal pattern has been also demonstrated in both dairy cows and sheep. Berry *et al*. (2006) reported a highly seasonal change of SCC in Irish dairy herds with low values from spring to summer and high values during the autumn to winter period. Others also described seasonal change in bulk milk SCC but the variation of the geometric mean was lower ($<100\times10^3$ cell/mL) and the peak occurred from July to September (Olde Riekerink *et al*., 2007; Elmoslemany *et al*., 2009). In dairy sheep month and month within herd were significant variation factors and maximum SCC values were observed from July to September (Gonzalo *et al*., 2005). We concluded that the season has significant influence on the frequency of IMI cases was larger in the rainy season than in the dry

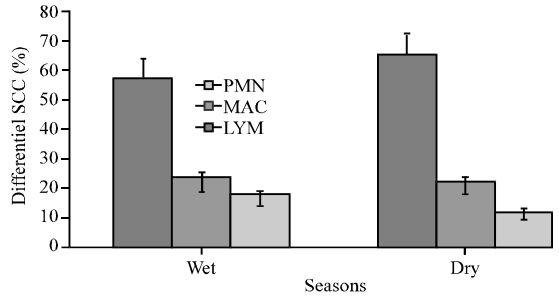

Fig. 1: Variation of Macrophage (MAC), Lymphocyte (LYM) and Polynuclear Neutrophil (PMN) with season of camel's milk

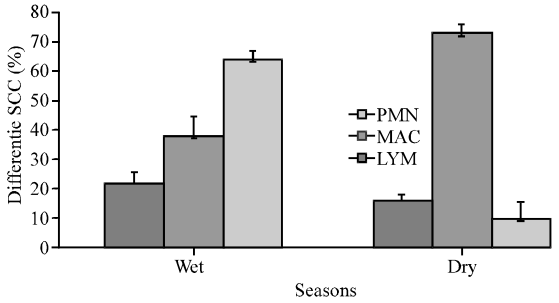

Fig. 2: Variation of Macrophage (MAC), Lymphocyte (LYM) and Polynuclear Neutrophil (PMN) with season of cow milk

season for cows (Nobrega and Langoli, 2011). There seems to be differences between dromedary camels and cows in some aspects of immune response.

As shown in Fig 1, the proportion MAC, LYM and PMN in wet season averaged ~57, ~24 and ~18% respectively in camel milk. These results are consistent with the role of MAC as the first immunological responder against invading pathogens (Hamed *et al*., 2010; Sordillo and Streicher, 2002). During dry season, the relative proportion of MAC increased significantly, while LYM and PMN level decreased (Fig. 1).

Thus in healthy quarters, MAC was the predominant cell type in consistent with (Lee *et al*., 1980; Riollet *et al*., 2001). Particularly in camels where such changes were associated with significant increase in MAC, it may represent physiological differences in the innate immunity response to mammary gland infection.

In cows milk, the dominant cell type was PMN (64%), followed by LYM (38%) and MAC (22%) throughout wet season. Cow's milk on dry season, had higher proportion of LYM and low level of PMN (Fig. 2). Our results suggest an increased incidence of clinical mastitis during the rainy months. Thus, in uninfected cows, the predominant cell type is MAC; however, during chronic mastitis, the predominant cell type switches to PMNs (Lee *et al*., 1976; Riollet *et al*., 2001).

Poly Morpho Nuclear cell percentage (PMN %) tended ($p<0.1$) to be greater in cows during the wet season when compared to those during the hot season, there was an effect on the PMN percentage on positively diagnosed animals between the hot season and the rainy season. In addition, the proportion of MAC in camel's milk particularly at wet and dry was notably greater ($p<0.05$) than in cow's milk (Fig. 1 and 2). As MAC belongs to the acquired immune system it is additional indication that the immune system in camels may be different from the one in cow's one and may relate the famous camel's resistance to stress and disease.

The lowest incidence of clinical mastitis in winter and in dry season in dromedary has been attributed to anatomical structure of mammary gland which renders mammary gland more resistant to cold and heat stress.

CONCLUSION

On the basis of this investigation, it is concluded that the seasonal variations in this part of subcontinent influences the total and differential somatic cell count. It was concluded that wet season significantly increases mastitis incidence in cows. The cows were more venerable to mastitis while camels can withstand rigors of adverse dry and wet season. However, the higher proportion of LYM in camel's than cow's milk at different season suggest that the innate and acquired immune system of camels is potentially different from that of cows and may relate to their famous capacity to resist stress and infections.

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