

Study of Somatic Cell Count in Milk of Pregnant and Non-Pregnant Buffaloes

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Abstract

Sixty nine samples of milk from apparently healthy buffaloes in early, middle and late lactation stages were collected and 21 samples of normal milk from apparently healthy pregnant buffaloes in early and late stages were collected. All samples were from South of Capital Baghdad areas and were stained with Newman-Lampert stain and the somatic cells count was recorded for each sample. The results revealed that there is a range between 370000 and 1540000 cells/ ml in all lactation stages, while in early and late of pregnancy stages, the range is between 1700000 and 5800000 cells/ ml. There are no significant differences among the somatic cell counts in early, middle and late non-pregnant lactation stages. In pregnancy stages there was a highly significant difference ($P \leq 0.01$) between the early and the late stages, and all non-pregnant lactation stages. In the lactation stage SCC was low due may be to high production of milk, while in late stage was high due may be to decrease in the milk production and the effect of hormones during pregnancy. In conclusion, somatic cell count (SCC) in buffaloes' milk may play an important role in the defense mechanism of the body against mastitis agents.

دراسة أعداد الخلايا الجسمية في حليب الجاموس الحوامل وغير الحوامل

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الخلاصة

جمعت تسعة وستون عينة من حليب الجاموس الصحي ظاهرياً في مرحلة الرضاعة المبكرة والمتوسطة والمتأخرة و 21 عينة من الحليب الطبيعي من جواميس حوامل صحية ظاهرياً في وقت مبكر وفي المراحل المتأخرة من الحمل. صبغت جميع العينات بوصمة لاميرت- نيومان وتسجيل متوسط إعداد الخلايا الجسمية في كل عينة. وجدت الأعداد 370000 و 1540000 خلية/ مل في جميع مراحل الرضاعة بينما كانت الأعداد في مراحل الحمل المبكرة والمتأخرة من الجواميس الحوامل والأعداد في جميع مراحل الرضاعة في الجواميس غير الحوامل. تتخفف في مرحلة الرضاعة الأولية الخلايا الجسمية بسبب ارتفاع إنتاج الحليب بينما في أواخر مرحلة الرضاعة خلال فترة الحمل تنخفض الخلايا الجسمية للتغيرات الفسلجية ووجود هرمونات الحمل في الجسم. ونستنتج ان الخلايا الجسمية (SCC) في حليب الجاموس ستلعب دوراً هاماً في آلية الدفاع عن الجسم ضد جراثيم التهاب الضرع.

Introduction

Mastitis is a disease of dairy animals caused by different types of microorganisms. This disease is an inflammation of mammary gland tissues in dairy animals usually infected one or more quarters of the udder (1) Mastitis is still the most costly infection of dairy cattle for

the milk industry and causes a significant reason of financial loss in the economy worldwide (2). The studied factors were determined as risk factors affecting mastitis as breed (3) season, age (4) management and environment (5). There are many tools for evaluating health of the udder and the milk quality on dairy farms. First, the inflammatory process in the mammary gland will be observed at quarter, cow, and herd and population level (6). Second, somatic cells (SC) play a vital role in the dairy animal's body defense against pathogenic agents. The somatic cells refer to leukocytes, epithelial cells and other immunity components (7). The term "Somatic Cells" has come to replace the word leukocytes as a more meaningful expression of those body cells that are usually associated with inflammation of the udder (8). Somatic cells resulted from normal death of the epithelial cells (apoptosis) (9). The aim of this study is to use the SCC of buffalo's milk for determination of mastitis during lactation and early and late pregnancy periods.

Materials and Method

Milk samples: In a clean environment, thoroughly wiping of the teats with 70% ethyl alcohol with paying extra attention to teat orifice was applied. After discarding the first few milk squirts, a total of 90 milk samples from apparently healthy buffaloes from February to May 2012, in the areas around the South Capital Baghdad. In this study 69 out of 90 milk samples were collected during stages of lactation period and 21 milk samples were collected from early and late stages of pregnant animals. Then the samples were transferred within ice box to the laboratory. After the samples received, 0.01 ml of the milk was smeared on one cm² of the slide, and stained with Newman-Lambert stain to count the somatic cells (10). Under light microscope at 100× oil immersion lens, 30 fields were counted then the mean of these readings were recorded to each sample and multiply by 400. ANOVA statistical analysis was applied to analyse the results, according to the method recorded by (11).

Results

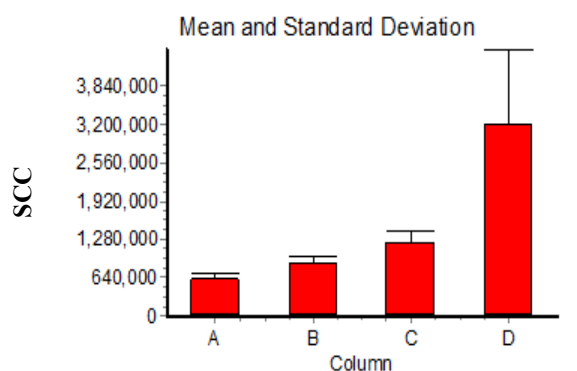
The average of somatic cell counts of milk samples in the early stage of lactation was between 400000- 736000 cells/ ml and the mean was 568000 while it was elevated in the middle stage of lactation to 800000- 1000000 cells/ ml and the mean was 900000. It was recorded in the late stage of lactation to 1200000-1520000 cells/ ml and the mean was 1280000. In case of the late pregnant animals or called drying stage, the average of the somatic cell counts were between 1800000- 5500000 cells/ ml and the mean was 2250000. Non-pregnant buffaloes results divided into three groups: First group was in pro-oestrus phase range from 370000 to 738000 cells/ ml and the range was 400000 to 736000 cells/ ml. Second group was in oestrus phase in which the SCC ranged from 700000 to 1100000 cells/ ml and the range was 800000 to 1000000 cells/ ml. Third group was in dioestrus range from 1000000 to 1540000 cells/ ml and the range was 1040000 to 1520000 cells/ ml (Table 1). Pregnant results divided into two groups: First group was early pregnancy stage range from 1700000 to 2900000 cells/ ml and the mean was 1800000 to 2700000 cells/ ml. Second group was late pregnancy stage range from 3600000 to 5800000 cells/ ml and the mean was 3400000 to 5500000 cells/ ml (Table 2). Group A of somatic cell counts is represented pro-oestrus lactation stage somatic cells have been compared with Group B the oestrus lactation stage somatic cells and Group C dioestrus stage somatic cells. There are no significant differences among these groups. In case of Group D, the early and late stages of pregnancy somatic cells, there are highly significant differences when it compared Group A and D, Group B and D, group C and D by one way ANOVA (Figure 1).

Table (1) The Somatic Cell Counts in Various Phases of Oestrus Cycle

Non- Pregnant buffaloes			
Pro-estrus phase	Estrus phase	Dioestrus phase	
370000-738000	700000 - 1100000	1000000 -1540000	Range
568000	900000	1280000	Mean

Table (2) The Somatic Cell Counts in Early and Late Stages of Pregnancy

Pregnant buffaloes		
Early stage	Late stage	
1700000- 2900000	3600000 - 5800000	Range
2250000	4450000	Mean



Groups of Lactation and Pregnant Stages

Fig. (1) Group A: Pro-oestrus lactation stage, Group B: Oestrus lactation stage, Group C: Di-oestrus stage. Group D: Early and late stages of pregnancy

One-way Analysis of Variance (ANOVA)

The P value is < 0.001, considered extremely significant.

Variation among column means is significantly greater than expected by chance.

Tukey-Kramer Multiple Comparisons Test

If the value of q is greater than 3.901 then the P value is less than 0.05.

Mean Comparison	Difference	q	P value	
Column A vs Column B	-290857	1.224	ns	P>0.05
Column A vs Column C	-637714	2.684	ns	P>0.05
Column A vs Column D	-2592000	10.910	***	P<0.001
Column B vs Column C	-346857	1.460	ns	P>0.05
Column B vs Column D	-2301143	9.685	***	P<0.001
Column C vs Column D	-1954286	8.226	***	P<0.001

Mean 95% Confidence Interval

Discussion

The somatic cell counts in buffaloes reflected the inflammation of the mammary gland. The somatic cell count for “normal” milk is ranged between 400,000 and 1500000 cells/ml (Table 1). Higher counts are considered abnormal and indicate probable infection. Factors such as late lactation, old age and environmental stress may cause slight elevations of SCC but such increases are inconsequential when compared to the elevation which results from infection (12). Higher SCC counts are also associated with decreased production in which the somatic cell counts are shifted to the high level in the late stage of pregnancy due to the physiological and hormonal changes (Table 2). The high results of SCC indicated that these SCC in buffaloes related to inflammatory responses to intra-mammary infection or another trigger to immune system (12). Leukocytes accumulate at the inflamed site to combat

invading bacteria. Somatic cells are mostly cells of the immune system in mastitic quarters (13). In the non-pregnant buffaloes the somatic cell counts results indicate that the animal showed a high resistant to infection due to the consistency of the somatic cell counts. These somatic cells are part of the natural defense mechanism which includes lymphocytes, macrophages, polymorphonuclear cells and some epithelial cells (14). The high number of the SCC in the pregnant animals related to the decrease of the milk production, the physical and physiological changes due to the hormonal production for animal pregnancy requirement. The SCC in early and late pregnancy buffaloes have high SCC more than usual so some investigations were done to make sure these buffaloes did not exposed to bacterial infection which shifted SCC to higher level due to normal factors, in this case these results are disagreed with the results recorded by (15) who showed that the SCC in buffaloes were due to differences in management practices, methods of detection, breeds of the animals, immune response of animals and climatic conditions. Some authors recorded that an extreme weather favors the problem and creates stress to the body, hence immunity decreases, leading to increased sub-clinical incidence associated with contagious and environmental conditions (16). In a previous study performed in our laboratory(17) found mean of somatic cells in cows in early (580.000) cell/ ml and late stages of lactation period (6.870.000) cell/ml as well as in pregnancy period the mean SCC were 4480000 cell/ml in early stage and 960.000 in dioestrus phase. The results have been shown in this study are lower somatic cell counts comparing to somatic cell counts in the same period of lactation of non-pregnant and pregnant cows. In conclusion, SCC in this study reflected the health of the animals during non-pregnant early, middle and late lactation stage, as well as in the early and late stage of the pregnancy. In comparison with the results of SCC in cow the buffaloes have low SCC in all abovementioned stages due to the high defense mechanisms which the buffaloes have in the milk, and these animals are characterized by high resistant to infection with mastitis-causing agents.

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