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RELATIONSHIP BETWEEN SOMATIC CELL COUNT AND INTRAMAMMARY INFECTION IN EGYPTIAN ZARAIBI GOATS

U. M. El-Saied[1], R. E. Al-Ahwal[1], M. E. Ahmed[1], Ferial Hassan[1] and B. M. Edrise[2]

1- Animal Production Research Institute, Ministry of Agriculture and Land Reclamation, Dokki, Giza, Egypt, 2- Faculaty of Veterinary Medicine, Cairo University, Giza, Egypt.

SUMMARY

A total of 206 milk samples from half udders of 103 hand milked Egyptian Zaraibi goats raised at an experimental farm belonging to Animal Production Research Institute (El-Serw, Damietta Governorate, Egypt) were collected at midlactation to study the relationship between Somatic Cell Count (SCC) and Intramammary Infection (IMI) status. Bacteriological tests were carried out to investigate udder health status and type of IMI followed by determination of SCC for all samples. Eight bacterial types were isolated in this study. Samples were classified into five groups according to the bacterial infection as follows: infected with single major pathogen (Staphylococcus aureus, Coliform spp., Streptococus uberis, Streptococcus agalactiae or Streptococcus dysgalactiae), infected with single minor pathogen (Corinebacteria, Bacilli or other Staphylococci spp.,), infected with double major pathogens, infected with double minor pathogens and non-infected. Twenty-six samples were excluded from the statistical analysis because they were triple contaminated by minor pathogens. Therefore, statistical analysis of the data was performed for a total of 175 milk samples from half udders of 90 does.

The most frequent bacterial groups were Staphylococci spp. and Bacilli. Log SCC was higher for the infected group with single major pathogen (5.70) or single minor pathogen (5.73) than the non-infected one (5.48). SCC was affected significantly ($P < 0.05$) by udder status and the interaction between udder status and parity number and non-significantly by parity number. Prevalence of subclinical IMI was 27.4% of half udders. The obtained results primarily illustrated that the threshold of 1'600,000 cells/ml showed the best indication for intramammary health status of the Egyptian Nubian (Zaraibi) Goat breed.

Keywords: *Somatic Cell Count, Intramammary Infection, Zaraibi Goats.*

INTRODUCTION

In Egypt goat milk is not usually direct consumed fresh except in the northwestern desert and in Sinai where small ruminants, goats in specific, play an important role in the animal agricultural development of these regions. Almost all

goat milk produced by the commercial sector is used by the dairy industry for cheese making mainly for export and partially to cover the little demand of the domestic market. It is known that the cheese-making process is largely affected by milk quality, especially casein percentage.

Dairy industry generally necessitates adoption of strict hygienic measures in order to secure healthy milk. Mastitis is one of the major reasons for culling dairy animals. It causes great economic losses to the dairy industry as it affects milk productivity and quality (Contreras *et al.*, 1993; Sanchez *et al.*, 1997). Moreover, it needs extra labor and increases veterinary cost and animal replacement.

Somatic cell counting has been studied for possible application in identifying individuals or herds with mastitis and is now being included in many monitoring programs for dairy goat herds. It is considered as an efficient/cost-effective indirect indicator of udder health and milk quality in the dairy industry.

The application of SCC monitoring for early diagnosis of subclinical mastitis needs to be institutionalized for dairy goats in Egypt as one of the tools for food safety. The literature presents a wide range (400,000 to 1.750 million cells/ml of milk) for the discrimination of SCC threshold between healthy and infected udders for dairy goats (Dulin *et al.*, 1983; Poutrel and Lerondelle, 1983; Maisi, 1990; Rigau *et al.*, 1991; Lerondelle *et al.*, 1992; Wilson *et al.*, 1992, 1993; Dorke *et al.*, 1993; De Cremoux *et al.*, 1994; Zeng and Escobar, 1995; 1996; Contreras *et al.*, 1996; Haenlein and Hinckley, 1996; Hinckley, 1990; Paape and Capuco, 1997; Sanchez *et al.*, 1999; White and Hinckley, 1999; Scuton *et al.*, 2000). However, most of the thresholds fall in the range between 400,000 to 800,000 cells/ml of milk. The present work aimed at studying the relationship between SCC and IMI of the Egyptian Zaraibi Goat (also called Egyptian Nubian) in a pilot attempt to provide valid interpretation for criteria in detecting subclinical mastitis.

MATERIAL AND METHODS

Animals and management:

Egyptian Zaraibi is the most pronounced dairy goat amongst the local breeds (Baladi, Barki and Black desert goats) in Egypt. It is considered to be of high genetic potential as a dairy and prolific breed (Aboul-Naga *et al.*, 1993). Milk production averages 250 kg in a lactation of 210 days. The present work was carried out at El-Serw Animal Production Research Station, Damietta Governorate, located in the north east of the Nile River Delta and belonging to Animal Production Research Institute (APRI), Ministry of Agriculture and Land Reclamation, Egypt. Animals are housed in semi-roofed yards. Feeding allowances are calculated according to NRC (1981). Roughage (Berseem, *Trifolium alexandrinum* hay and bean straw) to concentrate ratio was 40:60. The ration was offered twice daily at 8 a.m. and 3 p.m. and clean water was available all times.

Data and sampling:

A total of 206 milk samples were collected at midlactation from half udders of 103 Egyptian Nubian goats (Zaraibi). Kidding season started in November 2001 and sampling was carried out in February 2002 (100-120 d postpartum). Animals were hand-milked twice daily. Dry-off treatment is practiced for all animals in the herd, however, postdipping activity is not practiced.

The teats of sampled animals were cleaned using 70% ethanol. The initial milk stripped from each half was discarded and then milk samples were taken from each half udder (approximately 15 ml) in sterile prelabeled tubes and kept in an icebox at 4°C until bacteriological analysis. Delivery to the laboratory took place within 3 h after sampling. Udders that showed signs of clinical mastitis and milk samples that showed obvious abnormal physical aspects were not included in the study.

Bacteriological analysis and SCC determination:

Bacteriological tests were carried out to investigate the status and type of IMI by the Standard Plate Count (SPC) according to Houghtby *et al.*, (1992). Detection and enumeration of specific organisms or groups of organisms were performed by serial dilution of milk samples that were spread-plated onto MacConkey agar (Difco, Detroit, MI) for presumptive Gram-negative (*Coliform spp.* count); modified Edwards medium (Oxoid, Hampshirre, England) for presumptive *Streptococcus* spp., (*agalactiae, dysglactiae, ubris*). Staph. 110 media was used for presumptive *Staphylococcus* spp., Baird paker agar (Oxford, Hampshire, England) for presumptive *Staphylococcus aureus* and blood agar with Tellurite for presumptive *Corinebacteria.* Blood agar was used for presumptive *Bacillus* spp.

All plates were incubated at 37 °C ± 2 °C and examined after 24 and 48 hours. These selective and differential media were chosen for the isolation and identification of mastitis inducing pathogens according to Collins and Lyne (1979). The results were expressed as colony forming unit (CFU)/ml of milk.

Staphylococcus aureus, Coliform spp., Streptococus uberis, Streptococcus agalactiae and *Streptococcus dysgalactiae* were considered as major pathogens where, *Corinebacteria,* Bacilli and other Staphylococci spp., were considered as minor pathogens. Twenty-six milk samples were excluded from the statistical analysis due to the likelihood of contamination with triple minor pathogens. Neither triple infections by major pathogens nor quadruple infections by minor pathogens were determined. Subclinical IMI was defined as 300 CFU/ml.

SCC was determined for each milk sample with a Fossomatic machine (Foss Electric, Hillerϕd, Denmark) 24h postcollection following the rules of the International Dairy Federation (1984) and the machine was calibrated for goat milk. Original scale SCC values were transformed to logarithmic form (Ali and shook, 1980) to meet the characteristics of hypothesis testing. The bacteriological investigation and SCC determination were performed in the Dairy Services Unit, which belongs to the Animal Production Research Institute, Sakha, Kafr El-Shaiek governorate, Egypt.

Statistical Model:

The repeated measures of the studied trait (log SCC) was analyzed using the MIXED procedure of the SAS (SAS, 1996) with the following model:

$Y_{ijklm} = \mu + US_i + P_j + US*P_{ij} + A(US\ P)_{ijk} + UH_l + e_{ijklm}$

Where:

Y_{ijklm} = the studied dependent variable (log SCC),

μ = the overall mean,

US_i = the effect of udder status represented in 5 levels as: infected with single major pathogen, infected with double major pathogens, infected with single minor pathogen, infected with double minor pathogens or non-infected,
P_k = the effect of parity (five levels representing the first five parities),
$US*P_{ij}$ = the interaction between udder status and parity,
$A(US\ P)_{ijk}$ = the random effect of the animal k nested within udder status i & parity j,
UH_l = the fixed effect of udder half (2 levels) and
e_{ijklm} = the residual effect.

RESULTS AND DISCUSSION

Average log SCC for the total samples was 5.58 (637 x 10^3 cells/ml). No previous values were found in the literature for the same breed but this average is comparable with that (753 x 10^3 cells/ml) reported by Park (1991) for the Nubian breed. The average log SCC for group infected with single major pathogen (5.70) and for the group infected with single minor pathogen (5.73) were obviously higher than that for the non-infected group (5.48). Of the total half udders, 72.6% were non-infected, where, 20.0% and 7.4% of them were infected by major and minor pathogens, respectively.

Bacteriological isolation results revealed that eight types of microorganisms were found in the milk of Zaraibi goats under farm operational conditions in El-Serw Animal Research Station. The most dominant bacterial types were *Staphylococci* spp., (18.9%), *Streptococcus dysgalactiae* (2.9%) and *Corinebacteria* (1.7%). Bacilli as a less hazardous microorganism (minor) were found in 11.4% of the samples.

Similar bacterial isolates were reported for goat milk worldwide. For Greek goats, Kalogridou-Vassiliadou (1991) reported that 59% of mastitis-related pathogens present in normal milk were of staphylococci type (*S. aureus* 17%, *S. epidermidis* 14%, *S. capitis* 13%, *S. hyicus* 11%), 30% bacilli, 4% *Coliform spp.*, 3% *Micrococci*, 2% streptococci, 1% *Corinebacteria* and 1% *Pseudomonas*. In France, however, Lerondelle *et al.* (1992) detected *S. aureus* (2%) and coagulase negative *staphylococci* (23%). In Spain, for Murciano-Granadian goats, the dominant pathogens were: coagulase negative staphylococci (70%), coagulase positive staphylococci (1%), *Corinebacteria* 12%, *Mycoplasma* 9% and 8% for *Enterobacteria, Pasteurella,* streptococci and yeast (Contreras *et al.*, 1996).

In this study, a total of 48 samples had more than 300 CFU/ml of the pathogens isolated. According to this criterion, the prevalence of subclinical IMI was 27.4% of half udders.

Table 1 presents the results of analysis of variance of the factors affecting log SCC of half udders (udder status, parity number, the interaction between udder status and parity number and udder half).

As shown in Table 1 both udder status and its interaction with parity number had a significant effect ($P < 0.05$) on log SCC, however, the rest of the effects had a non-significant effect on the studied trait.

Table 1. Analysis of Variance for log SCC from mixed model

Source of variance	df	F	*P*
Udder status (US) [1]	4	2.89	0.0261[3]
Parity (P)	4	0.57	0.6883[NS]
Udder half	1	1.63	0.2043[NS]
US*P	11	2.29	0.0150[3]
Doe[2] (Random)	---	---	---

[1] Udder status levels are: infected with single major pathogen, infected with double major pathogens, infected with single minor pathogen, infected with double minor pathogens or non-infected.
[2] Levels of random effect (doe) = 90.
[3] $P < 0.05$.
[NS] = not significant.

Table 2 shows least squares means (±SE) of log SCC by udder status. Log SCC differs significantly ($P < 0.05$) for the non-infected group against the rest of the infected groups. Therefore, elevated SCC in milk is associated with the subclinical IMI of the udder.

Table 2. Least squares means (±SE) of log SCC by udder status

Udder status	Log SCC Least squares means	SE	Number of records
Infected with single major pathogen [1]	5.70	0.15	10
Infected with double major pathogen	---	---	3
Infected with single minor pathogen [2]	5.73	0.12	25
Infected with double minor pathogen	---	---	10
Non infected	5.48	0.05	127

[1] *Staphylococcus aureus, Coliform spp., Streptococus uberis, Streptococcus agalactiae* or *Streptococcus dysgalactiae*
[2] *Corinebacteria,* Bacilli or other Staphylococci spp.,

Udder status and parity number are within the most important factors affecting udder health and levels of SCC in milk. Sanchez *et al.* (1999) studying the effect of parity as a risk factor for caprine subclinical IMI reported a positive statistical association between IMI and parity number in half udders of older than fifth parity does. In this study, does were between the first and the fifth parity.

Results in this study show that, across the levels of udder status, log SCC clearly increased in the fifth parity (from 5.71 to 5.94) in comparison with the first one (from 4.97 to 5.65). The relatively low number of records available in each class did not give a clear idea of the ovulation of log SCC within all levels of interaction between udder status and parity number. In general, the fifth parity had the highest log SCC. This result is in good agreement with those reported for dairy goats (El-Edrissi *et al.*, 1994; Sanchez *et al.* (1999); dairy sheep (Poutrel, 1983; Watkins *et al.*, 1991); and dairy cattle (Oliver, 1988).

Table 3 shows percentage distribution of correct and false classification (as based upon total SCC) and sensitivity with specificity on the different SCC thresholds for goats. Based on the results of this table for correctly classified samples, false positives, false negatives, sensitivity and specificity, the threshold of 1 600,000 cells/ml showed the best indication for IMI / health status. Such result is somewhat lower but comparable with that (1 750,000 cells/ml) reported by Sanchez *et al.* (1999).

Table 3. Percentage distribution of correct and false classification (as based upon total SCC) and sensitivity with specificity on the different SCC thresholds for Egyptian Nubian (Zaraibi) goats

Thresholds (Cells/ml) x10^3	Correctly classified samples	False positives	False negatives	Sensitivity	Specificity	Rank of thresholds
400	61.6	17.0	62.1	37.9	83.0	13
500	61.7	20.2	61.8	38.2	79.8	12
600	62.9	20.4	61.1	38.9	79.6	11
700	65.7	20.5	58.7	41.3	79.5	10
800	66.9	21.2	57.9	42.1	78.8	6
900	67.4	22.0	57.7	42.3	78.0	8
1000	66.9	23.0	59.2	40.8	77.0	7
1100	67.4	24.6	60.0	40.0	75.6	9
1200	68.6	24.5	58.3	41.7	75.5	5
1300	69.7	24.5	56.2	43.8	75.5	4
1400	70.3	25.0	55.6	44.4	75.0	3
1500	70.9	25.2	54.2	45.8	74.8	2
1600	72.0	25.2	50.0	50.0	74.8	1

CONCLUSION

Somatic cell counting of half udder samples of milk taken from the Egyptian Zaraibi goat breed at midlactation can be used as a tool in developing a recording system to indicate the existence of subclinical mastitis. The threshold of 1'600,000 cells/ml gave the best indication for IMI / health status of the Egyptian Nubian (Zaraibi) goat breed. However, more studies using larger data sets and other goat breeds and locations are still needed to support the feasibility of results of this pilot study.

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دراسة عن علاقة تعداد الخلايا الجسدية بصحة الضرع فى الماعز الزرايبى المصرى

أسـامة محمد السعيد[1]، رشاد إبراهيم الأحول[1]، محمد إبراهيم أحمد[1]، فريال عبد الرسول حسن[1]، بهجت مصطفى إدريس[2]

١- معهـد بحـوث الإنتاج الحيوانى، وزارة الزراعة و استصلاح الأراضى، الدقى، جيزه، مصر، ٢- كلية الطب البيطرى، جامعة القاهرة، جيزه، مصر

جمعت ٢٠٦ عينة لبن من أنصاف ضرع من ١٠٣ عنزه مصريه زرايبى فى منتصف موسم الحليب فى قطيع يحلب يدوياً بمحطة بحوث التربية بالسرو التابعة لمعهد بحوث الإنتاج الحيوانى و ذلك لدراسة العلاقة بيـن تعداد الخلايا الجسدية و الحالة الصحية للضرع. أجريت التحاليل البكتيريولوجية لعينات اللبن واتبعت بتقدير تعداد الخلايا الجسدية فيه.

عزلـت ثمانية أنواع بكتيرية و قسمت العينات إلى خمس مجموعات وفقاً لنوع الإصابة كالأتى: عينات مصـابة بـنوع واحـد مـن البكتـيريا الممرضـة (*Staphylococcus aureus, Coliform spp.*, *Streptococcus uberis, Streptococcus agalactiae* or *Streptococcus dysgalactiae*)، عينات مـصـابة بـنوع واحد من البكتيريا البيئية (*Corinebacteria,* Bacilli and other Staphylococci spp.,)، عينات مصابة بنوعين من البكتيريا الممرضة ،عينات مصابة بنوعين من البكتيريا البيئية و عينات غـير مصـابة. اسـتبعدت نتائج ست و عشرين عينة لبن من التحليل الإحصائي لأنة تبين تلوثها بأكثر من نوعين من البكتريا البيئية.

أظهـرت النتائج أن أكثر الأنواع البكتيرية انتشاراً كان Staphylococci ثم Bacilli . ارتفع متوسط لوغاريـتم تعـداد الخلايا الجسدية للمجموعة المصابة ببكتيريا ممرضة (٥,٧٠) أو بيئية (٥,٧٣) بالمقارنة بالمجموعة غير المصابة (٥,٤٨).

بينـت نتائج التحليل الإحصائي تأثر تعداد الخلايا الجسدية معنوياً بكل من حالة الضرع من الإصابة و تأثـير الـتداخل بيـن العامل السابق و ترتيب موسم الولادة بينما لم يتأثر بترتيب موسم الولادة. بلغت نسبة إصـابة أنصـاف الضـرع بالالـتهاب تحت الحاد (Subclinical Mastitis) ٢٧,٤%. بينت النتائج أن ١٦٠٠٠٠٠ خلية جسدية / مل من اللبن أعطى أفضل النتائج كحد فاصل يشير إلى حالة الضرع من الإصابة للماعز الزرايبى المصرى.