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Influence of enzootic bovine leukosis virus upon the incidence of subclinical mastitis in cows at a different stage of infection

Nikolay Sandev[1]*, Mariana Koleva[1] , Rumen Binev[1], and Darinka Ilieva[2]

[1]*Trakia University, Faculty of Veterinary Medicine, Stara Zagora, Bulgaria*
[2]*National Diagnostic and Research Veterinary Medical Institute, Sofia, Bulgaria*

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ABSTRACT

Studies were performed on 26 cows, divided into 3 groups on the basis of serological (by immunodiffusion test) and haematological analyses for bovine leukosis: Group I - seropositive without changes in haemogram; group II - seropositive with changes in haemogram and group III (control) - BLV negative. A statistically significant difference was observed in the incidence of subclinical mastitis in cows from group II ($P<0.05$) compared to healthy animals. The most commonly isolated microorganism was *S. agalactiae* (46.80%), followed by *Staphylococcus* spp. (36.06%). The results of our studies, as well as data from other investigators evidenced that the incidence of subclinical mastitis was higher in cows with changes in the white blood cell picture due to the immunosuppressive effect of the enzootic bovine leukosis virus.

Key words: bovine enzootic leukosis, cows, subclinical mastitis

Introduction

Enzootic bovine leukosis (EBL) is a chronic viral infection. The disease develops in three stages. The majority of infected animals (about 60%) remain in the first stage - that of virus carriership. About 30% of animals are in the second (haematological) stage of the infection, characterized by leukocytosis

*Contact address:
Assoc. Prof. Dr. Nikolay Sandev, Department of Veterinary Microbiology, Infectious and Parasitic Diseases, Faculty of Veterinary Medicine, Trakia University, 6000 Stara Zagora, Bulgaria, Phone: +359 42 28 01 29 86; Fax: +359 670 624

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and lymphocytosis. During the third (neoplastic) stage, in about 10% of bovine viscera, tumours are formed (GENOV and TSUTSUMANSKI, 1984). The enzootic bovine leukosis virus suppresses systemic resistance. According to MILOJEVIĆ et al. (1991), RUSOV et al. (1994), PETROV (1997), KOSTADINOVA (2002) the morbidity rate of metritis, mastitis, arthritis and pericarditis is consequently increased.

Others (HUBER et al., 1981; REINCHARDT et al., 1988) do not report significant differences in the incidence of subclinical mastitis in cows infected with BLV or in healthy cows.

The contradictory literature data motivated our study on the incidence of subclinical mastitis in cows in a different stage of enzootic leukosis compared to healthy animals.

Materials and methods

The studies were performed on 26 cows of the Holstein-Fresian and American Brown Cattle breeds for 4 months. All cows were in the same premises, uniformly fed and housed.

Serological examination for the presence of leukosis was done by immunodiffusion reaction (IDR) according to the method of Ouchterlony, modification of MILLER and VAN DER MAATEN (1976) and adapted in Bulgaria by MATEVA and TEKERLEKOV (1978) with glycoprotein antigen gp51 (Sanofi, France).

Total white blood cell (WBC) counts were determined by automated analyser System 150 (Germany). Lymphocyte percentage was detected using routine methods for differential white blood cell counting.

The animals were divided into three groups: group I: serologically positive without haematological changes (total WBC counts 10.3×10^9/L, lymphocyte percentage - 69.7%, 9 cows); group II - serologically positive with haematological changes (total WBC counts 25.9×10^9/L, lymphocyte percentage - 81%, 9 cows) and group III - controls (WBC counts 7.83×10^9/L, lymphocytes 69.6%, 8 cows).

The status of the udder and forms of inflammation were differentiated according to the International Dairy Federation classification. Milk samples were investigated for somatic cells counts in 1 mL milk by a quick mastitis test and directly by the Prescott-Breed method. The samples containing less than 500 000 somatic cells

per 1 mL milk were considered as negative. Microbiological examination was performed by the generally accepted schedule for isolation and differentiation of mastitis microbial agents and the HOLT et al., (1994). Citological and microbiological determination was carried out during the entire period of the study.

Data were statistically processed by the percentage comparisons method.

Results and discussion

In the first group of cows (Table 1), 16 milk quarters (44.44%) reacted positively to the quick mastitis test; the somatic cells counts in 1 mL milk were above the reference range. Six *S. agalactiae* strains, two of each *S. dysgalactiae* and *S. aureus* and three *S. epidermidis* strains were isolated. In the second group of animals, 77.78% of studied milk quarters were with subclinical mastitis. The most prevalent among microbial isolates was *S. agalactiae* (n = 13), followed by *S. epidermidis* (n = 5), *S. aureus* (n = 3), *S. dysgalactiae* (n = 3) and *Corynebacterium* spp. (n = 2). The incidence of subclinical mastitis in control cows was 25%. A statistically significant difference ($P<0.05$) was observed only between the second and the third (control) groups, whereas those between groups I-II and I-III were not significant.

According to some authors the primary aetiological role in bovine subclinical mastitis was that of *Staphylococcus* spp. (TSOLOV, 1989; FILIPOV, 1993). They showed that the involvement of *S. agalactiae* was 1.37% and 11.35%, respectively. Despite that, the most frequently isolated organism in our study was *S. agalactiae* - 46.80%, while the total contribution of *S. aureus* and *S. epidermidis* was 36.06%.

Our results confirmed the studies of MILOJEVIĆ et al. (1991), RUSOV (1993), RUSOV et al. (1994) who reported a higher percentage of subclinical mastitis in cows with haematological changes. However, SCOTT et al. (1991) have reported a lower percentage of serologically positive cows with changes in white blood cells which, in their opinion, was due to the enhanced immune response against the leukosis virus, providing an indirect protection of cows against mastitis.

On the basis of the data resulting from our study it is possible to summarize that bovine enzootic leukosis virus had an immunosuppressive effect that could result in an increased incidence of subclinical mastitis in infected cows with changes in leukocyte haematology.

Table 1. Incidence of subclinical mastitis in cows with enzootic leukosis

Group Number of animals (n)	Quick mastitis test milk quarters (n)	Microbial isolates milk quarters (n)	Status of udder
Group I BLV seropositive without changes in the haemogramme (n=9)	+ n = 16 44.44%	*S. dysgalactiae*(2) *S. agalactiae* (6) *S. aureus* (2) *S. epidermidis* (3)	Subclinical mastitis
Milk quarters (n=36)	- n = 20	-	Healthy
Group II BLV seropositive with changes in the haemogramme (n = 9)	+ n = 28 77.78%	*S. dysgalactiae* (3) *S. agalactiae* (13) *S. aureus* (3) *S. epidermidis* (5) *Corynebacterium* spp. (2)	Subclinical mastitis
Milk quarters (n = 36)	- n = 8	-	Healthy
Group III Healthy cows (n = 8)	+ n = 8 25%	*S. uberis* (2) *S. agalactiae* (3) *S. epidermidis* (2) *S. aureus* (1)	Subclinical mastitis
Milk quarters (n = 32)	- n = 24	-	Healthy

Legend: (+) positive result - reacted by (++) or (+++) to the Quick mastitis test; (-) negative result

Conclusions

1. The incidence of subclinical mastitis in leukosis-seropositive cows with changes in the haemogram was higher ($P<0.05$) compared to healthy animals.

2. The most commonly isolated microorganism was *S. agalactiae* (46.80%), followed by *S. epidermidis* and *S. aureus* (both 36.06%).

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SAŽETAK

Istraživanje je provedeno na 26 krava podijeljenih u tri skupine na osnovi seroloških (imunodifuzijski test) i hematoloških pretraga na leukozu goveda. U prvoj skupini bile su serološki pozitivne krave bez promjena u hemogramu. U drugoj skupini bile su serološki pozitivne krave s promijenjenim hemogramom, a u trećoj kontrolnoj skupini bile su krave serološki negativne za virus enzootske leukoze goveda. Statistički značajna razlika u pojavi supkliničkog mastitisa u usporedbi sa zdravim kravama dokazana je u krava druge skupine ($P<0,05$). Najčešće izdvojena bakterija bila je *S. agalactiae* (46,80%) te *Staphylococcus* spp. (36,06%). Rezultati istraživanja potvrđuju da je pojava mastitisa bila češća u krava s poremećajima u bijeloj krvnoj slici, što se pripisuje učinku virusa enzootske leukoze goveda.

Ključne riječi: enzootska leukoza goveda, krave, supklinički mastitis