SEROLOGICAL STUDY ON *BRUCELLA SPP.* AND SMALL RUMINANT LENTIVIRUS IN DAIRY GOATS IN VOJVODINA

I. Pusic1, D. Lupulovic1, J. Prodanov-Radulovic1, M. Urosevic2, Z. Grgic1

1Veterinary Scientific Institute “Novi Sad”, Rumenci put 20, Novi Sad, Republic of Serbia
Corresponding author: ivan@niv.ns.ac.rs
2High school for agriculture “Futog”, CariceMilice 2, Futog
Original scientific paper

Abstract: The objective of this study was to determine seroprevalence of small ruminant lentivirus (SRLV), *Brucella abortus* and *Brucella melitensis* infections among dairy goats in South Bačka and Srem region in Vojvodina Province. The other aim was to determine the risk factors associated with SRLV infection in investigated herds. A total of 971 serum samples were collected from 8 commercial dairy goat herds distributed in these two districts. All collected samples were examined for antibodies against *Brucella abortus* and *Brucella melitensis* by an competitive enzyme linked immunosorbant assay (cELISA), and all yielded negative result. Sera from 216 goats older than six months were tested for antibodies against *Brucella abortus* and *Brucella melitensis* by an competitive enzyme linked immunosorbant assay (cELISA), and all yielded negative result. A total of 73 goats in six herds tested seropositive for SRLV with overall seroprevalence of 33% indicating that the infected herds are evenly spread throughout these two regions. The prevalence of positive animals within the herds ranged from 0 to 100% and the prevalence rate of seropositive goats increased with age. On the herd level import of goats, herd size and rearing with sheep were the most significant risk factors. The result of present study indicates that SRLV infection exists in the goat herds in Vojvodina Province. It also provides an overview about most important risk factors connected with the disease in the examined herds. Specific antibodies against *Brucella abortus* and *Brucella melitensis* infection were not detected.

Keywords: dairy goats, lentivirus, brucella, serology, prevalence

Introduction

Estimated goat population in Serbia is about 320,000, with a large number of non-commercial husbandries predominantly located in mountainous regions (Zujovic et al., 2011). Interest for intensive dairy goat farming in Vojvodina Province has significantly increased in recent years. Despite growing population of
In intensive farming systems, the milk production in most operations remains unsatisfactory. Among other factors, infectious diseases such as brucellosis or caprine arthritis-encephalitis (CAE), may contribute to impaired milk production, and represent a significant trade barrier. Small ruminant lentiviruses (SRLV), including ovine maedi-visna virus (MVV) and caprine arthritis-encephalitis virus (CAEV), are considered genetically as a single pathogen (Shah et al., 2004). Infections caused by SRLV are widespread in many countries and are responsible for significant economic losses (Peterhans et al., 2004). Disease usually takes years to develop and is progressive, causing chronic and inflammatory lesions in the brain, lungs, joints and mammary glands of sheep and goats (Dawson, 1989).

Brucellosis is one of the most important bacterial diseases that can affect goat production and cause significant economic losses. During the last decade in Vojvodina Province there were several outbreaks of caprine brucellosis (Lalic et al., 2004), which is last time reported in 2005. The purpose of this study was to document the presence and to determine seroprevalence of CAEV and to elucidate risk factors connected to infection. The other aim was to determine brucella status of the investigated herds.

**Material and Methods**

During the 2008 samples were taken from 8 commercial goat farms situated in South Backa and Srem region of Vojvodina Province. The goat flocks were chosen according to production type and geographical area in order to represent both regions. The goats are predominantly Alpine, Saanen or Toggenburg breeds, and are all kept under intensive conditions for milk production. A total of 971 blood samples were collected in vacutainer tubes from the jugular vein of each goat. Blood samples were chilled and transported to the laboratory where they were centrifuged at 1000g for 10 minutes. Serum was stored in 1.5 ml Eppendorf tubes and stored at -20ºC for batch testing. In order to detect anti Brucella sp. agglutinins, all the sera were tested with Rose Bengal serum agglutination test (VZ «Zemun») and competitive ELISA (Brucella-AbC-ELISA, Swanowa, Sweden). Sera from 216 goats older than six months (which represents about 20% of the total number of goats in each herd) were evaluated for antibodies against SRLV using commercially available ELISA (CAEV/MVV antibody test kit Chekitt, IDEXX, Netherlands).

**Results and Discussion**

Seventy-three goats out of 216 tested, were found seropositive to CAE virus infection. On the herd level 6 out of 8 tested herds presented at least one.
seroreactive animal, revealing that infection is spread uniformly through two regions (Table 1.).

Table 1. Goat herds examined with ELISA for CAEV antibodies in two Vojvodina regions

<table>
<thead>
<tr>
<th>Region</th>
<th>Number of herds</th>
<th>Positive</th>
<th>Percent (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>South Backa</td>
<td>4</td>
<td>3</td>
<td>75</td>
</tr>
<tr>
<td>Srem</td>
<td>4</td>
<td>3</td>
<td>75</td>
</tr>
<tr>
<td>Total</td>
<td>8</td>
<td>6</td>
<td>75</td>
</tr>
</tbody>
</table>

Although seroconversion does not necessarily mean that the animal was clinically affected, CAE seem to be widespread amongst goat flocks from South Backa and Srem region, and probably represents an important factor that contributes to the sub optimal productivity. Additionally, in herd 5 where the prevalence of CAE seropositive animals was amongst the highest (Table 2.), the clinical findings of encephalitis in kids 2-4 months old were strongly suggestive of CAE and caused high mortality rates.

Table 2. Prevalence of antibodies against CAEV in tested goats from eight herds

<table>
<thead>
<tr>
<th>Flock Number</th>
<th>Number of animals tested</th>
<th>Positive</th>
<th>Percent (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>59</td>
<td>3</td>
<td>5</td>
</tr>
<tr>
<td>2</td>
<td>12</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>3</td>
<td>40</td>
<td>32</td>
<td>80</td>
</tr>
<tr>
<td>4</td>
<td>10</td>
<td>10</td>
<td>100</td>
</tr>
<tr>
<td>5</td>
<td>27</td>
<td>24</td>
<td>88</td>
</tr>
<tr>
<td>6</td>
<td>8</td>
<td>3</td>
<td>37</td>
</tr>
<tr>
<td>7</td>
<td>24</td>
<td>1</td>
<td>4</td>
</tr>
<tr>
<td>8</td>
<td>36</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Total</td>
<td>216</td>
<td>73</td>
<td>33</td>
</tr>
</tbody>
</table>

Virus isolation was not attempted. However, the detection of CAEV antibodies in goats is considered to be diagnostic for the infection (Adams et al., 1983, Cutlip et al., 1992), and decreased birth weights, delayed weight gains and increased mortality before weaning have been demonstrated in offspring (Peterhans et al., 2004). The high overall seroprevalence within the most of herds under the study implied that infection might have been present for several years. Seroconversion may be delayed, ranging from a few weeks up to 2 years post infection, and is well documented (Adams et al., 1983, Blacklaws et al. 2004). Herd seroprevalence in this study (75%) was higher than that reported in Jordan (23%), Great Britain (10%), Wales (57%), but lower than that estimated in studies conducted in Norway (86%), Australia (82%) (Al-Quadah et al., 2006, Dawson and Wilesmith 1985, Nord K et al., 1998, Greenwood PL et al. 1995). This finding
might be due to a small number of herds included in the study, but a higher seroprevalence was recorded where intensive husbandry is generally practiced (Cutlip et al., 1992). Information about prevalence of SRLVs in the goat population are less updated, but no EU country can be considered to be SRLV infection free (Peterhans et al., 2004). In this study, goats older than three years were more likely to be seropositive than younger goats. The observed association between the age and seroreactivity is consistent with findings in previous surveys (Nord K et al., 1998, Al-Quadah et al., 2006). The animals that escaped infection in neonate period, are at permanent risk of contracting infection later in life through horizontal spread of disease. The main route of transmission between flocks appear to be ingestion of infected colostrum or direct contatact with infected animal (Blacklaws et al. 2004). Epizootiological investigation in this study revealed that single most important risk factor for CAE infection at herd level was import of goats. In 5 of 6 herds where seropositive goats were detected, replacement goats and bucks were imported from three different European countries. The exchange of live animals across the national boundaries is considered to be an important cause of horizontal transmission of SRLV (Blacklaws et al., 2004). In one herd (herd 8), where no seropositive animals were found there was no history of buying replacements from abroad. In herd No1 where three seropositive animals were detected, the most probable source of infection was rearing goats in a close proximity with sheep flock that was previously diagnosed with clinical MVV and pulmonary adenomatosisis (Pusic et al.,2008). Evidence of cross-species transmission is documented earlier (Castro et al., 1999, Shah et al., 2004) and can be experimentaly induced (Peterhans et al., 2004). Recent sequence comparisions of natural isolates of SRLV from sheep and goats suggested that horizontal cross-species infection can occur and that it is common (Roland et al., 2002). In herd 7 only one buck that was purchased few months earlier from infected flock tested positive. Live animal trading is considered a major risk factor in spread of SRLV infection between herds. Presence of seropositive bucks in a herd poses a risk for spreading the pathogen through direct contact between infected buck and uninfected does, although sexual route of transmission seems not to be well documented yet (Blacklaws et al., 2004). Our data suggest that addition of new animals, contact with sheep flocks, herd size and stocking density are associated with high seropositivity to CAEV in intensive goat rearing. Out of 971 serum samples tested for anti- Brucella sp. aglutinins, neither one yielded a positive reaction. Occurence of brucellosis in goat population in Vojvodina during last decade was documented (Lalic et al., 2004), but since 2005 when 145 infected goats were culled there is no further evidence of seropositive animals in South Backa and Srem region, as our study also suggest.
Conclusion

The information obtained on seroprevalence in this study reveals that SRLV infections among goat herds in South Backa and Srem region are evident. It also indicates that in future more clinical outbreaks are to be expected, and goat breeders should be informed about the role of SRLV infection on animal welfare and potential economic losses. Carefully tailored eradication strategies are required to prevent dissemination of disease, and SRLV-free certified flocks should be the ultimate aim. We found no evidence of brucellosis infection in investigated goat farms, and present surveillance programs should be continued.

Acknowledgment

This paper is a result of the research within the project TR31084 “Wild animal health monitoring and introduction of new biotechnology procedures in detection of infectious and zoonotic agents – risk analysis for human health, domestic and wild animal health and for environmental contamination”, financed by the Ministry of Education, Science and Technological Development, Republic of Serbia.

Serološko ispitivanje prisustva infekcije mlečnih koza lentivirusima i brucella vrstama u Vojvodini

I. Pušić, D. Lupulović, J. Prodanov-Radulović, M. Urosević, Ž. Grgić

Rezime

Cilj istraživanja bio je da se ustanovi seroprevalencija infekcije lentivirusima i bakterijama iz roda Brucella (Brucella abortus i Brucella melitensis) u zapatima mlečnih koza na teritoriji Južnobačkog i Šremskog okruga u Vojvodini. Takođe, cilj je bio i da se ustanove najvažniji faktori rizika povezani sa pojavom infekcije lentivirusima malih preživara (SRLV) u ispitivanim gazdinstvima. Ukupno je pregledan 971 uzorak krvnog seruma koza sa 8 komercijalnih farmi koje su bile smeštene u ova dva regiona. Svi uzorci ispitani su kompetitivnom ELISA metodom na prisustvo specifičnih antitela protiv Brucella abortus i Brucella melitensis i svi su dali negativan rezultat. Krvni serumi 216 koza starih od 6 meseci ispitani su na prisustvo specifičnih antitela protiv lentivirusa malih preživara ELISA testom. Od ukupno ispitanog broja uzoraka seropozitivna su bila 73 grla iz 6 zapata, sa prosečnom seroprevalencijom od 33%, a inficirani zapati bili su ravnomerno raspoređeni u oba regiona. Prevalencija seropozitivnih
jedinki unutar zapata kretala se od 0 do 100%, pri čemu je rasla sa starošću životinja. Kao najvažniji faktori rizika za pojavu infekcije SRLV u zapatu identifikovani su: uvoz koza iz inostranstva, veličina stada i odgajanje u kohabitaciji sa ovca. Rezultati istraživanja ukazuju da infekcija lentivirusima na farmama koza u odabranim regionima Vojvodine, perzistira i predstavlja značajan problem. Isto tako, ukazuju na najvažnije fakto rizika koji su povezani sa prisustvom i raširenosti SRLV infekcije u pojedinačnim zapatima. Prisustvo infekcije vrstama *B. abortus* i *B. melitensis* na ispitivanim farmama koza nije ustanovljeno.

References


