

## Prevalence of enzootic bovine leukosis in the Republic of Bulgaria in 1997-2004

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**SANDEV, N., D. ILIEVA, I. SIZOV, N. RUSENOVA, E. ILIEV: Prevalence of enzootic bovine leukosis in the Republic of Bulgaria in 1997-2004. Vet. arhiv 76, 263-268, 2006.**

### ABSTRACT

The serological investigations using the agar gel immunodiffusion (AGID) test performed in the Republic of Bulgaria in the period 1997-2004 showed a high prevalence of enzootic bovine leukosis (EBL), from 8.47% in 1997 to 22.26% in 2004. The highest percentage of seroreagents was observed in the regions of Silistra (48.61%), Dobrich (47.57%) and Burgas (47.32%), with the lowest, in the regions of Pazardjik (0.28%), Kyustendil (1.89%) and Smolyan (1.95%). Leukosis infection was found to be more prevalent in Eastern than in Western Bulgaria. The total EBL epidemic foci in 2004 numbered more than 600. Depending on the severity of the epidemiological process, the eradication of leukosis in the country could be achieved in 5 to 10 years with sufficient financial funding and strict realization of the National programme for prophylaxis and control of EBL.

**Key words:** enzootic bovine leukosis, agar gel immunodiffusion test, prevalence, Bulgaria

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### Introduction

Enzootic bovine leukosis (EBL) is a chronic viral infection characterized by neoplastic growth of erythropoietic organs and perivascular proliferates in the liver, kidneys and lungs. Some 30-40 years ago the disease was encountered in almost all European countries and caused considerable economic losses to cattle breeding. Thus, the creation of national programmes for EBL prophylaxis and control was necessary. As a result of adequate

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financial support and the strict performance of eradication programmes, after 10 to 15 years the disease disappeared completely in most EC countries (Belgium, The Netherlands, Denmark, Germany, France, Great Britain, Austria, Finland (FORSCHNER et al., 1988; KNAPEN et al., 1993; NUOTIO et al., 2003) and in other countries only sporadic cases were encountered (Italy, Portugal, Lithuania, Latvia, Estonia (POLET, 2004).

In Bulgaria, EBL was distributed during the 1970s mainly via import of highly productive animals infected with the bovine leukaemia virus from some West European countries: the former German Democratic Republic, as well as Israel, USA, Canada, etc. (GENOV et al., 1986). Despite the instructions for EBL control approved by the Ministry of Agriculture and Forestry in 1986, only partial success in EBL control was achieved in some settlements in the regions of Burgas, Sofia and Stara Zagora due to a number of objective and subjective causes (GENOV, 1986; SANDEV, 1988). The serological studies carried out by us later in the regions of Stara Zagora and Russe and in south-eastern Bulgaria (SANDEV et al., 2001a; SANDEV et al., 2001b) confirmed the broad distribution of the infection under the new conditions of cattle breeding. The percentage of positive seroreagents in some regions varied on average between 15.62% and 33.07%.

The aim of the present study was to perform a national representative report about the prevalence of EBL in the Republic of Bulgaria for the period 1997-2004.

### **Materials and methods**

The annual reports of serological analyses for EBL using the agar gel immunodiffusion test (AGID), provided by the National Veterinary Service, accompanied by the reports of Regional Veterinary Services in Stara Zagora, Silistra and Smolyan for 2004, were used. The AGID test was performed with glycoprotein antigen (gp 51) using the method of Ouchterlony, modification of MILLER and VAN DER MAATEN (1976). The results were statistically processed by the method of percentage comparisons.

### **Results**

Data on serological investigations are shown in Fig. 1. They show considerable incidence of EBL in the country, varying between 8.49% in 1997 and 14.02% in 2000. From 2001 on, a slight decrease was observed (11.84% in 2001, 10.94% in 2002 and 7.6 % in 2003) but thereafter the prevalence of EBL rose sharply to 22.26% in 2004. By regions, the highest percentage of seropositive reagents was found in the regions of Burgas (33.19%), Russe (23.29%), Stara Zagora (22.62%), Sliven (21.03%) and Razgrad (20.17%). The lowest incidence was found in the regions of Smolyan (0.26%), Blagoevgrad (0.63%), Kyustendil (1.18%), Pernik (1.63%) and Vratsa (4.39%).

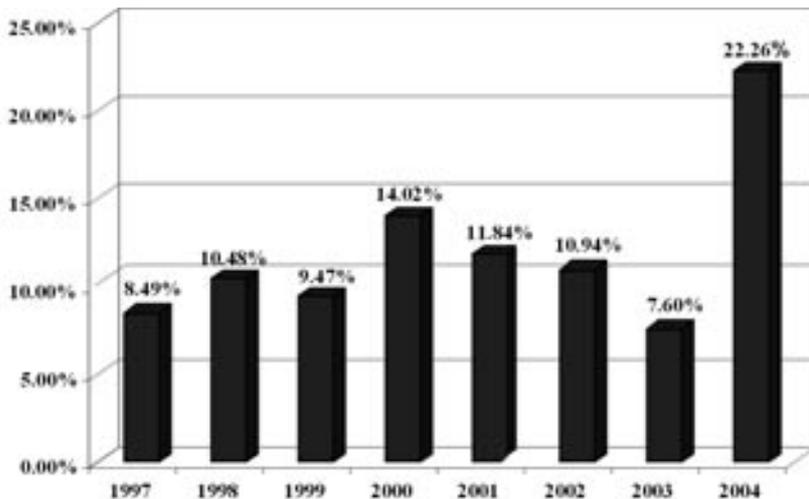


Fig. 1. Incidence of bovine enzootic leukosis in the Republic of Bulgaria during 1997-2004



Fig. 2. Incidence of bovine enzootic leukosis in the Republic of Bulgaria in 2004

Fig. 2. presents data on serological analyses made in 2004 by regions. From 254,124 tested animals, the number of seropositive reagents was 56 529 (22.26%). The highest percentage of seroreagents was observed in the regions of Silistra (48.61%), Dobrich

(47.57%), Burgas (47.32%) Razgrad (46.61%) and Russe (43.24%), whereas the lowest prevalence of EBL was found in the regions of Pazardjik (0.28%), Kyustendil (1.89%), Smolyan (1.95%) and Sofia District (2.93%). From a geographical point of view leukosis infection was more common in Eastern than in Western Bulgaria. The most numerous foci of enzootic leukosis were found in the region of Kardjali (177), followed by the region of Stara Zagora (136). The total number of infection foci for the country was over 600.

## **Discussion**

Analysis of results showed that enzootic bovine leukosis was widely distributed in the Republic of Bulgaria, especially in 2004. Considering that serological analyses had been performed in about half of the national cattle population, the number of seropositive animals appears to be much higher. In our opinion this severe epidemic status is due to disregard of the Instruction for prophylaxis and control of EBL, accepted in 1986 and further revised in 1993 and 2002. Over a 20-year period only partial serological investigations have been performed. Seropositive animals are registered but no other action is performed for either limitation or elimination of the leukosis infection. Nevertheless, according to our observations, after the reformation of cattle breeding in Bulgaria and the creation of farms with 20, 50 and up to 100 cattle, the percentage of seropositive animals is very low, or else those farms are completely leukosis-free. And yet the majority of animals are owned by farmers with limited financial resources and it was among these that the percentage of seropositive reagents was the highest.

The principal cause for disregard of the Instructions for EBL control is a lack of adequate funding. Nor is there any material stimulus for either farmers or veterinarians who are engaged in its realization. Another important aspect in the realization of this programme is the performance of individual serological analyses with a diagnostic test, standardized by the European Union (AGID test or ELISA) of all cattle older than 6 months, in order to know the exact number of seropositive animals and the foci of infection. The performance of those analyses would be greatly facilitated by the individual marks of all cattle and the supply of farmers with veterinary sanitary passports for the healthy condition of animals. On the basis of this information, a specific plan for every settlement and farm would be elaborated, with the participation of specialists from the Regional veterinary services, the Regional Diagnostic and Research Veterinary Medical Institutes as well as regional veterinarians for restriction and eradication of the leukosis infection. The activities contained in the eradication programme should be co-ordinated and controlled by an expert committee of the National Veterinary Service and the Ministry of Agriculture and Forestry. In the beginning, such activities could be carried out in separate regions (i. e. 3 regions) with EBL incidence <10%, between 10-30%, and >30%, using Epizooties funds for the purpose. At a later stage, after the country's membership of the European Union, this model could be realized on a national scale with the financial support of the EU-25

programme. In 2005, this programme gave a total of 775,000 EUR for EBL control in Estonia, Italy, Lithuania, Latvia and Portugal (POLET, 2004).

Our experience (SANDEV, 1988; SANDEV et al., 1998; TSUTSUMANSKI, 1999), as well as the experience of our colleagues from abroad (SMIRNOV, 1991; PETROV, 1997; RUSSINOVICH and DROGUN, 1998; DI GIACOMO, 1992; PELZER and SPRECHER, 1993; SPRECHER et al., 1991; STOKKA et al., 1998; TEKES, 1989) showed that depending on the severity of the epidemiological process, eradication of leukosis from bovine herds could be achieved in 5 or 10 years.

### Conclusions

1. Results of serological tests for EBL carried out in the period 1997-2004 showed that the infection was widespread in the Republic of Bulgaria.

2. Eradication of leukosis in the country could be achieved in 5 to 10 years given sufficient financial funding and strict realization of the National programme for prophylaxis and control of EBL.

### References

- DI GIACOMO, R. F. (1992): The Epidemiology and control of bovine leukemia virus infection. *Vet. Med.* 3, 248-257.
- FORSCHNER, E., I. BÜNGER, H. P. KRAUSE (1988): Überwachungsuntersuchungen in Brucellose-, Leukose- und BHV1-freien Rinderherden. ELISA-gestützte Tankmilchuntersuchung anstelle der Einzeltierprobenuntersuchung mit herkömmlichen Testsystemen. Sicherheit und Kosten. *Dtsch. tierärztl. Wschr.* 95, 214-218.
- GENOV, I. (1986): Enzootic bovine leukosis (Contemporary methods of prophylaxis and control), a review, Sofia. pp. 3-4.
- GENOV, I., B. TSUTSUMANSKI, A. DIMITROV (1986): Enzootic Bovine Leukosis, Zemizdat, Sofia. pp. 7-8.
- KNAPEN, K., P. KERKHOFS, M. MAMMERICKX (1993): Eradication de la leucose bovine enzootique en Belgique: bilan du dépistage de masse réalisé sur l'ensemble du cheptel national en 1989, 1990 et 1991. *Ann. Med. Vet.* 137, 197-201.
- MILLER, J. M., M. J. VAN DER MAATEN (1976): Serological detection of bovine leukemia virus infection. *Vet. Microbiol.* 1, 195-202.
- NUOTIO, L., H. RUSANEN, L. SIHVONEN, E. NEUVONEN (2003): Eradication of enzootic bovine leukosis from Finland. *Prev. Vet. Med.* 59, 43-49.
- PELZER, K. D., D. J. SPRECHER (1993): Controlling BLV infection on dairy operations. *Vet. Med.* 3, 275-281.
- PETROV, N. I. (1997): Healing of farms affected from enzootic bovine leukosis. *Veterinaria* 9, 10-12.
- POLET, Y. (2004): EU-25 Livestock and Products: EU Approves €188 Million to Fight Animal Diseases in 2005. USDA Foreign Agricultural Service, GAIN Report Number: E34084, 3.

- RUSSINOVICH, A., A. DROGUN (1998): Parameters of the epizootological process of enzootic bovine leukosis in the Republic of Belarus. *Veterinarnaya nauka - proizvodstvu*, Scientific works, Minsk, 33, 33-36.
- SANDEV, N. (1988): Studies on some epidemiological and immunological features of bovine enzootic leukosis. PhD Thesis, Trakia University, Faculty of Veterinary Medicine, Stara Zagora, Bulgaria.
- SANDEV, N., N. NIKOLOVA, M. NIKOLOVA, B. TODOROV (1998): Application of ELISA and immunodiffusion method (IDM) for detection of bovine leukosis virus in highly affected farms. *Bulg. J. Vet. Med.* 1, 83-88.
- SANDEV, N., I. SIZOV, S. PANDAROV, S. ALEXANDROVA, T. DOJCHEV, V. VASILEV, T. TANCHEV, L. GEORGIEVA (2001a): Prevalence of enzootic bovine leukosis in South-eastern Bulgaria during the period 1998-2000. *Vet. arhiv* 71, 215-221.
- SANDEV, N., V. HVARCHILKOV, I. SIZOV (2001b): Data from the serological investigation about the prevalence of enzootic bovine leukosis in two regions in Bulgaria during 1994-1998. *Animal Sciences (Sofia)*, 3-4, 53-56.
- SMIRNOV, Y. P. (1991): Efficacy of leukosis control depending on the frequency and the intervals of serological analyses. *Veterinaria* 11, 29-30.
- SPRECHER, D. J., K. D. PELZER, P. LESSARD (1991): Possible effect of altered management practices on seroprevalence of bovine leukemia virus in heifers of a dairy herd with history of high prevalence of infection. *JAVMA* 199, 584-588.
- STOKKA, G. L., J. SMITH, J. SHIRLEY, T. FALKER, TR. VAN ANNE (1998): Bovine leukosis. Kansas State University, Agricultural Experiment Station and Cooperative Extension Service, 1-3.
- TEKES, L. (1989): Leucosisfertőzöttség felszámolásának lehetősége fertőzött és negatív állatok közös légtérben tartása mellett. *Magyar Állatorvosok Lapja* 44, 69-72.
- TSUTSUMANSKI, V. (1999): Epizootological evaluation of the prophylaxis and control of enzootic bovine leukosis in modern husbandry. *Vet. News* 3, 15-17.

Received: 18 March 2005

Accepted: 2 May 2006

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**SANDEV, N., D. ILIEVA, I. SIZOV, N. RUSENOVA, E. ILIEV: Proširenost enzootske leukoze goveda u Republici Bugarskoj od 1997. do 2004. *Vet. arhiv* 76, 263-268, 2006.**

**SAŽETAK**

Serološka istraživanja enzootske leukoze goveda (ELG) u Republici Bugarskoj u razdoblju od 1997. do 2004. provedena testom imunodifuzije u gelu, ukazala su na visoku zastupljenost te bolesti i to od 8,47% u 1997. do 22,26% u 2004. god. Najviši postotak serološki pozitivnih životinja ustanovljen je u predjelima Silistra (48,61%), Dobrich (47,57%) i Burgas (47,32%), dok je najniži postotak zabilježen u predjelima Pazardjik (0,28%), Kyustendil (1,89%) i Smolyan (1,95%). Ustanovljeno je da je leukoza više proširena u istočnoj nego u zapadnoj Bugarskoj. Ukupan broj ELG epidemijskih žarišta u 2004. god. iznosio je više od 600. Ovisno o jačini epidemiološkog procesa, leukozu će u zemlji biti moguće iskorijeniti za 5 do 10 godina s dostatnim financijskim sredstvima i strogom realizacijom Nacionalnog programa za profilaksu i kontrolu ELG-a.

**Ključne riječi:** enzootska leukoza goveda, imunodifuzija u gelu, proširenost, Bugarska

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