



The first outbreak of brucellosis in the region of Šabac

Prva epidemija bruceloze u šabačkom okrugu

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Abstract

Background/Aim. In Serbia brucellosis is a primary disease of the animals in the southern parts of the country. The aim of this study was to describe the first outbreak of human and animal brucellosis in the region of Šabac, Serbia. **Methods.** An epidemiological investigation was conducted to identify a source of outbreak and the ways of transmission of brucellosis infection in human population. A descriptive and analytical epidemiological methods (cohort study) were used. Additional data included monthly reports of the infectious diseases from the Institutes of Public Health and data from the Veterinary Specialistic Institute in Šabac. The serological tests for human brucellosis cases were performed in the Laboratory of the Military Medical Academy; laboratory confirmation of animal brucellosis cases was obtained from the reference laboratory of the Faculty of Veterinary Medicine, Belgrade. **Results.** Twelve cases of brucellosis were recorded from February 9 to September 1, 2004. Total attack rate was 8.1% (7.5% of males, 14.2% of females). Relative risk (RR) of milk consumption was 8.9 (95% confidence interval: 1.63-13.38), and RR for direct contact with animals was 14 (95% confidence interval: 3.5-55.6). The prevalence of seropositive animals in 33 villages of the Mačva region accounted for 0.8%. Regarding animal species, sheep were predominant – 264 (95.7%). Out of a total number of seropositive animals, ELISA results were positive in 228 (88.7%) of them. **Conclusion.** As contact epidemics generally last longer, it is probable that the implemented measures of outbreak control did reduce the length of their duration.

Key words:

brucellosis; serbia; disease outbreaks; epidemiologic methods; serologic test; humans; animals.

Apstrakt

Uvod/Cilj. Bruceloza je prvenstveno oboljenje životinja u južnom delu Srbije. Cilj ovog rada bio je da prikaže prvu epidemiju humane i animalne bruceloze na području Šapca. **Metode.** Sprovedeno je epidemiološko istraživanje u cilju otkrivanja izvora i puteva prenošenja bruceloze u humanoju populaciji. Korišćen je deskriptivni i analitički (kohortna studija) epidemiološki metod. Analizirane su mesečne prijave zaraznih bolesti Zavodu za javno zdravlje Šabac i Veterinarskom specijalističkom institutu u Šapcu. Serološki test za humanu brucelozu rađen je na Vojnomedicinskoj akademiji, a potvrda animalne bruceloze na Veterinarskom fakultetu u Beogradu. **Rezultati.** Dvanaest slučajeva humane bruceloze otkriveno je u periodu od 9. februara do 1. septembra 2004. god. Stopa javljanja iznosila je 8,1% (7,5% kod muškaraca i 14,2% kod žena). Relativni rizik od konzumacije mleka iznosio je 8,9 (interval poverenja: 1,63–13,38), a za kontakt sa životinjama 14 (interval poverenja: 3,5–55,6). Prevalencija seropozitivnih životinja u 33 sela mačvanskog regiona iznosila je 0,8%. Najviše je bilo obolelih ovaca – 264 (95,7%). Od ukupnog broja seropozitivnih životinja, ELISA test bio je pozitivan kod njih 228 (88,7%). **Zaključak.** Iako kontaktne epidemije obično traju dugo, primenjene mere suzbijanja verovatno su doprinele kraćem trajanju ove epidemije.

Ključne reči:

bruceloza; srbija; epidemije; epidemiološki metodi; serološki testovi; ljudi; životinje.

Background

Brucellosis is one of the most common zoonotic infections of domestic and wild animals caused by members of the genus *Brucella*¹. The humans become infected by di-

rect contact with infected animals or inhalation of infected aerosols, or through the consumption of raw animal products such as unpasteurized milk or cheese. In humans, ovine and caprine brucellosis caused by *B. melitensis* is the most important clinically apparent disease. Reported inci-

dence and prevalence of the disease vary widely from country to country. According to annual reports from the Centers for Disease Control and Prevention (CDC), the eradication campaign resulted in elimination of cattle brucellosis and substantial decline in the incidence of human disease in the USA. During 1993–2002, 46 states reported at least one case, mainly attributed to *B. melitensis*². Mexico is one of the most important reservoirs of human brucellosis in Latin America³. The disease remains a major problem in the Mediterranean region, Western Asia and parts of Africa⁴. North European countries declared as “officially bovine and/or ovine and canine brucellosis free”⁵. Incidence of disease was reported high in the Balkan Peninsula, Greece, Macedonia, Albania and Bosnia-Herzegovina^{4,6}. In Serbia, brucellosis used to be a primary disease of animals in the territories of Kosovo and Metohija and southern parts of the country.

The aim of this study was to describe the first outbreak of human and animal brucellosis in the region of Šabac, Mačva, Serbia.

Methods

An epidemiological investigation was conducted to identify a source of outbreak and the ways of transmission of the brucellosis infection in human population. In addition, the sources of data included monthly reports on infectious diseases from the Institute of Public Health, Šabac⁷ as well as the official reported data about infectious diseases for Serbia⁸. A descriptive and analytical epidemiological method (cohort study) were used. Relative risk (RR) and 95% confidence interval (CI) were calculated. Moreover, data were obtained from the Veterinary Specialistic Institute in Sabac, e.g., decisions on verified household infection and implemented measures were used⁹.

Brucellosis was diagnosed on the basis of clinical symptoms and signs compatible with brucellosis, supported by laboratory results, as it was recommended by the World Health Organisation¹⁰. A regional health center sent blood samples to the laboratory of the Military Medical Academy in Belgrade. In Serbia, PCR test, as a gold standard for diagnosis of brucellosis, was not in use during the period of the study. Consequently, a rapid agglutination reaction – BAB test (in Serbian: *brza serumska aglutinacija* – spead serra agglutination) for brucella antibodies with antigen solution prepared from brucella species of interest in region of Serbia was used. Detection of antibodies to brucellas was done in some patients by Wright serological reaction. This method was performed in the Institute of Public Health of Serbia in Belgrade.

The veterinarian investigation was conducted simultaneously with epidemiological investigation of human population. As serological test was positive in a animal sample, a laboratory confirmation of animal brucellosis cases was obtained from the reference laboratory of the Faculty of Veterinary Medicine, Belgrade, using the enzyme-linked immunosorbent assay (ELISA).

Results

Twelve cases of human brucellosis were recorded during the outbreak, which developed in the Šabac district from February 9 to September 1, 2004. Figure 1 shows the distribution of patients presented with human brucellosis. The epidemic curve suggested common and persistent source of contamination. The largest number of patients was recorded in March. Ten patients were males and two patients females, and their age ranged from 20 to 59 years. A severe clinical picture was manifested in eight patients (fever, muscular pain, headache, spleno- and hepatomegaly), so they were hospitalized. A prompt response to antibiotics was seen in all the cases but one. No case had any former history of brucellosis.

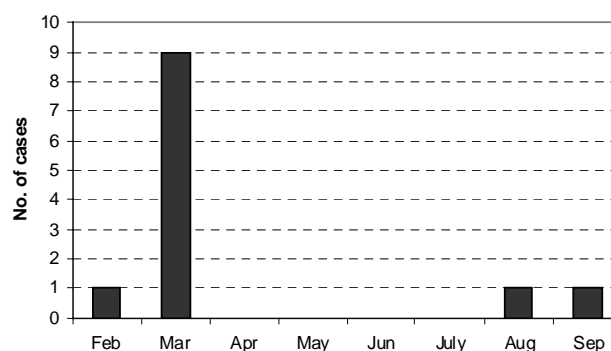


Fig. 1 – The distribution of patients with brucellosis in Šabac district (February 9 to September 1, 2004)

All the cases were positive by BAB test with titers ranging from 1/80 to 1/1280. These findings with positive antibodies tests in some patients were diagnostic criteria for *Brucella* infections. Isolation of *Brucella spp* organisms were not done routinely in human material, but it was done in animals.

The first (index) case in the outbreak was performed in a sheep and goat farm in the village Miokus near Šabac. The patient mentioned that, during his work in the farm, he used to drink freshly milked milk. Upon joining the veterinarian in the outbreak inspection, it was found that sheep from the aforementioned farm in Miokus had been sold in three more villages before the first case of brucellosis was diagnosed. Laboratory analyses of blood cultures collected from sheep, goats and cows of these farms revealed infected 93 head of cattle. On the basis of these results, epidemiologic cohort study was initiated in the families with the established infection of animals. A total number of the subjects exposed to epidemic, *ie* family members where animal infection had been verified were 148. Considering that 12 persons were affected during the epidemic, a total attack rate was 8.1% (7.5% of males, 14.2% of females). The highest attack rate was in the age group from 20 to 29 years (33.3%). All the cases and family members were interviewed to discover the mode of transmission. With the exception of the index case in the outbreak, other patients could not confirm a consumption of non-boiled milk, so RR of milk consumption was 8.9

(95% confidence interval: 1.63–13.38). Direct contact with animals was reported in 9 patients, RR = 14 (95% confidence interval: 3.5–55.6). The distribution of cases according to place of residence showed the geographical grouping in three villages (Figure 2).

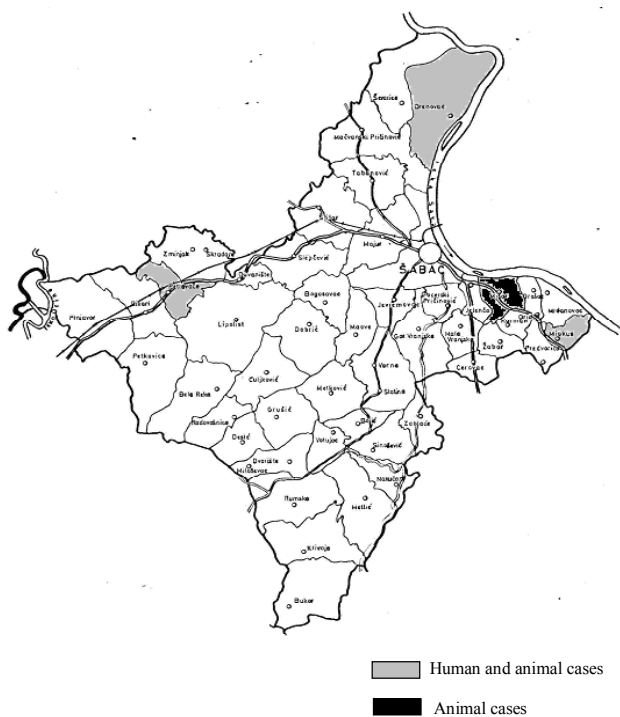


Fig. 2 – Geographical case distribution by residential ward, Šabac district

As soon as the first case was diagnosed, the Veterinary Service started with laboratory analyses of sheep, goat and cow blood samples collected from the farms in three aforementioned villages. Thereafter, investigation was expanded to 33 villages of Mačva region, where 29.294 goats and sheep were analyzed. They detected 257 infected animals (out of which 246 sheep and the rest were goats) in 152 home-steads. The prevalence of seropositive animals accounted for 0.8%. Regarding animal species, sheep were predominant – 264 (95.7%), out of which 225 (91.4%) were destroyed. Out of a total number of seropositive animals, ELISA results were positive in 228 (88.7%) of them.

Discussion

Since the end of 1980s when human brucellosis spreaded from the region of Kosovo and Metohija and southern parts of the central Serbia, the number of the affected people in the central Serbia ranged from 1 to 4 until 1994, while no similar cases were recorded in Vojvodina. For the first time in 2001, brucellosis in humans was reported in several parts of Vojvodina (20 cases), where outbreaks were evident in the subsequent period⁸. There was no outbreak of this disease in the central Serbia till 2003, when an epidemic was described on the territory of Belgrade with 16 cases. According to the information of veterinary services, brucellosis

had been confirmed that year in 1 218 animals in central Serbia¹¹.

In the period from 1994 to 2004, trichinellosis was the most frequent zoonotic disease in Šabac district. Q-fever epidemic with nine patients was noted in 1996. Other zoonoses were recorded in single cases. In this period, there was no case of human brucellosis.

The described outbreak is the first epidemic of human brucellosis in Mačva region which occurred simultaneously with epizootic of domestic animals in this region. In comparison with the largest outbreak in Serbia in 2001 that has been a food-borne¹², that outbreak was not so large by a number of cases. However, alimentary transmission cannot be fully ruled out in this epidemic, although only one patient reported that he had drunk fresh milk. It is possible that other cases used to eat cheese made of non-boiled milk from infected animals, at least until the outbreak was officially announced. Spain reported the epidemic with 11 sick people¹³ who had consumed cheese made of unpasteurized milk. The epidemic ended as soon as selling of this cheese was banned. Similar outbreak, but with much bigger number of cases, eg 81, and high relative risk (RR = 311.9), has been recorded in the same country. At that time, mode of transmission was home-made cottage cheese, and the pertinent livestock turned out to be infected with *Brucella melitensis*¹⁴. Nevertheless, the most common transmission way is *via* abraded skin contact with infected animals. It is the leading way of transmission in Asian countries, before all in Korea where, in distinction from Mediterranean countries, goats, sheep and camels are raised to produce meat and not milk¹⁵. High RR for contact as way of transmission suggests that it was the principle mode of transmission in our outbreaks. However, an epidemic curve does not look like other curves of contact outbreak. During described epidemic, all subjects contracted disease within 6 weeks. Two patients (one got ill in August, another on September 1) came from households with no elimination of infected animals upon official outbreak announcement. There are two explanations for their contracting the disease: either the incubation time was longer or the infection was not caused by contact with sheep during their lambing but subsequently during everyday handling with animals (eg with urine-contact).

In favor of contact as the way of transmission is the fact that the outbreak occurred in February–March. The most voluminous excretion of brucellosis from the animals is during their lambing (January–February), and the incubation period of human brucellosis is estimated as five days to three months¹. After 1995, China evidenced a sudden increase of a number of human brucellosis cases and the outbreaks. The largest number of patients was reported in the period February–June¹⁶. The results of our epidemic studies are compatible with reference ones. As contact epidemics generally last longer, it is probable that the implemented measures of the outbreak control did affect the length of their duration. There were no sick people in the third village, although three household had bought the sheep from the same farm. All sheep had no clinical picture of disease, lambing did not take place in these households, so there was no direct contact of people with the

excretions. By veterinary service regulations, the sheep were slaughtered already at the beginning of March.

Detection of a larger number of infected animals in the villages of Mačva suggests that the mode of transmission was most probably from animals having been bought in other part of Serbia or neighboring countries. The prevalence of the infected animals in this region (0.8%) was significantly lower than the prevalence of seropositive animals in Kosovo, where in 26 out of 29 districts 6.3% of seropositive sheep, 7.2% of goats and 0.6% of bovine cattle were recorded¹⁷. There is an increase in the number of reported outbreaks in the ruminants of Bosnia and Herzegovina¹⁸. An outbreak of human brucellosis in southern Croatia in 2004, the region with no brucellosis history, was

accounted for illegal transport of goats and sheep from Bosnia and Herzegovina¹⁹. The possible illegal trafficking of animals and their products across the borders of surrounding countries could be responsible for the persistence of foci of brucellosis in the area²⁰.

Conclusion

This outbreak shows selling of animals without any adequate control measures higher risk of spread of disease in animals. Contact with animals without any appropriate preventive measures may lead to infection of people, and consequently, to epidemic.

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