

Prevalence of antibodies against leptospire in the wild boar (*Sus scrofa* L., 1758)

F. TREML¹, J. PIKULA², Z. HOLEŠOVSKÁ²

¹Department of Infectious Diseases and Epizootiology, ²Department of Veterinary Ecology and Environmental Protection, University of Veterinary and Pharmaceutical Sciences, Brno, Czech Republic

ABSTRACT: A total of 307 blood sera of the wild boar from different localities of the Břeclav district (Czech Republic) were examined in 1999 to 2002. Antibodies against leptospire were found in 16.9% of blood sera examined. The incidence of antibodies in the wild boar varied both in individual years (26.1, 5.3, 25.0 and 12.0%, respectively) and age categories (7.8, 17.3 and 35.7%, respectively) with older individuals being more often positive. There were found no differences in relation to sex (18.2 and 15.0% of positives in males and females, respectively). Positive reactions at low titres (i.e., 100 and 200) predominated in 71.3%. Higher titres (3 200) were only exceptionally found in older individuals (3.8%). All positive reactions concerned only the *L. grippityphosa* serotype (100%). The other 11 serotypes tested were negative. On the basis of the results it can be stated that the wild boar is susceptible to infection by leptospire and the occurrence of specific antibodies correlates to a large extent with the presence of leptospire in the environment. In our opinion, the wild boar could be used for purposes of monitoring natural foci of leptospirosis.

Keywords: serology; *L. grippityphosa*; game mammals; natural foci; Czech Republic

Leptospirosis is a serious zoonosis of natural foci occurrence. Free living small mammals and rodents, in particular, are the main source and reservoir of pathogenic leptospire in natural foci (Anjanin, 1971; Estavoyer *et al.*, 2001; Levett, 2001; Vinetz, 2001; Valkonen *et al.*, 2002). Considering domestic animals, pigs play an important role in leptospirosis because they frequently harbour the *L. pomona* and *L. tarassovi* serotypes. Apart from these serotypes, other leptospire are found in the pig as well (Malachov and Alechin, 1976; Mraz *et al.*, 1981; Egan and Yearsley, 1987; Power, 1991; Kaszanyitzky *et al.*, 1997; Rocha, 1998). Malachov and Alechin (1976) found 13 serological groups and 17 serotypes of leptospire in the pig. Leptospire of the *L. pomona* and *L. tarassovi* serotypes, however, predominate in the aetiology of leptospirosis in the pig. Domestic pigs as a potential source of leptospire in the Czech Republic have been thoroughly investigated (Sebek *et al.*, 1983; Treml *et al.*, 1984; Treml and Nesnalova, 1995). The above authors found antibodies against

leptospire of various serotypes. Interesting is the fact that they found no antibodies against *L. pomona* and *L. tarassovi* as well as a decreasing percentage of seropositive animals probably due to the improvement of zoo-hygienic conditions. The wild boar has currently a wide distribution in the Czech Republic. It is a question whether the wild boar, like the domestic pig, may serve as a reservoir of leptospire of the *L. pomona* and *L. tarassovi* serotypes and participate in the spread of leptospirosis in natural foci. Examinations of sera from game animals (the wild boar, in particular) for antibodies against leptospire have been performed by a number of authors from the Czech Republic and abroad. In the Czech Republic it was, e.g., Sebek (1960) and Treml and Nesnalova (1993). Hübner and Horsch (1977), Weber and Christoph (1981), Witt *et al.* (1986), Dedek *et al.* (1989) and Lutz (1997) examined game animals for leptospirosis in Germany. Similar studies were performed by Krawczyk (2000) in Poland, Borcic *et al.* (1989) and Kovacic

Supported by the Grant Agency of the Czech Republic (Grant No. 524/00/1122).

et al. (2001) in Croatia and Tagliaube and Farina (1995) in Italy. The above papers document the fact that after contact of game animals with leptospire there form specific antibodies which can easily be confirmed by examination of blood sera.

This paper on leptospirosis is engaged in the examination of blood sera from the wild boar individuals shot during four years in south Moravia where we also confirmed the presence of antibodies against leptospire in small mammals (Tremil *et al.*, 2002).

MATERIAL AND METHODS

In 1999 to 2002 a total of 307 blood sera from the wild boar individuals shot in various hunting grounds of the Břeclav district (Czech Republic) were obtained and examined. Blood was collected by heart puncture or from the thoracic cavity during veterinary inspection of venison. Following sampling blood was kept in a refrigerator and delivered on the next day to the laboratory of the Department of Infectious Diseases and Epizootiology (Veterinary and Pharmaceutical University, Brno) for further processing. Here it was centrifuged to obtain serum and stored frozen until testing. Serological examination was based on the reaction of agglutination lysis using standard methods (Sebek, 1979). Following serotypes and strains of leptospire were used for the testing: 1. *L. grippotyphosa* – P 125, 2. *L. ictero haemorrhagiae* – Fryšava, 3. *L. sejroe* – M 84, 4. *L. canicola* – C 7, 5. *L. pomona* – Šimon, 6. *L. bratislava* – Jež Bratislava, 7. *L. arborea* – M 7, 8. *L. sorex jalna* – Sorex Jalná, 9. *L. bataviae* – Moldava, 10. *L. tarassovi* – S 42, 11. *L. bulgarica* – Nikolaevo, 12. *L. pyrogenes* – Salinem. Sera reacting in the standard dilution of 1 : 100 were examined with the respective serotype up to the titre.

On inspection of venison veterinarians were not in any case suspicious of clinical leptospirosis.

RESULTS

Examination of 307 blood sera from the wild boar individuals for antibodies against leptospire resulted in finding 52 positive cases, i.e., 16.9%. The highest percentage of positive reactions was found in 1999 and 2001 (26.1 and 25.0%, respectively), while in 2000 and 2002 it was only 5.3 and 12.0%, respectively (Table 1). Considering the age

Table 1. Numbers of examined and positive wild boar individuals in years 1999 to 2002

Year	Numbers of examined wild boars	Positive	%
1999	65	17	26.1
2000	75	4	5.3
2001	84	21	25.0
2002	83	10	12.0
Total	307	52	16.9

structure of the examined collection of wild boar individuals, there were 65 individuals less than 1 year old, 214 individuals in the age of 1 to 2 years and 28 individuals over 2 years (i.e., adult ones). Antibodies against leptospire were found in all three age groups examined. The highest percentage of positive reactions was found in the group of adult animals with 10 positive sera, i.e., 35.7% of the examined. Among the wild boar individuals from 1 to 2 years of age there were 37 positive sera representing 17.3% of the examined. Only 5 positive cases (i.e., 7.8% of the examined) were found in the youngest age group of individuals less than one year old (Table 2). In all, 187 males and 120 females

Table 2. Numbers of examined and positive wild boar individuals in individual age groups

Age	Numbers of examined wild boars	Positive	%
Less than 1 year	65	5	7.8
1–2 years	214	37	17.3
Over 2 years	28	10	35.7
Total	307	52	16.9

were examined. Males and females were positive for antibodies against leptospire in 34 and 18 cases, respectively, representing 18.2% and 15.0% of the examined (Table 3). All positive cases concerned only the *L. grippotyphosa* serotype. Results of test-

Table 3. Numbers of examined and positive males and females of the wild boar

	Numbers of examined wild boars	Positive	%
Males	187	34	18.2
Females	120	18	15.0
Total	307	52	16.9

Table 4. Serotypes of leptospire used for testing and positive titres found by serology in the wild boar

Serotypes of leptospire	Positive/%	Titres					
		100	200	400	800	1 600	3 200
<i>L. grippityphosa</i>	52/100	27/51.9	10/19.4	4/7.6	4/7.6	5/9.7	2/3.8
<i>L. icterohaemorrhagiae</i>	0	0	0	0	0	0	0
<i>L. sejroe</i>	0	0	0	0	0	0	0
<i>L. sejroe</i>	0	0	0	0	0	0	0
<i>L. canicola</i>	0	0	0	0	0	0	0
<i>L. jež bratislava</i>	0	0	0	0	0	0	0
<i>L. pomona</i>	0	0	0	0	0	0	0
<i>L. sorex jalna</i>	0	0	0	0	0	0	0
<i>L. bulgarica</i>	0	0	0	0	0	0	0
<i>L. bulgarica</i>	0	0	0	0	0	0	0
<i>L. arborea</i>	0	0	0	0	0	0	0
<i>L. tarassovi</i>	0	0	0	0	0	0	0
<i>L. bataviae</i>	0	0	0	0	0	0	0
<i>L. pyrogenes</i>	0	0	0	0	0	0	0
Total	52	27	10	4	4	5	2

ing other eleven serotypes were negative. Titres of antibodies in positive cases varied from 100 to 3 200 (Table 4). Most reactions were at the dilution of 100 and 200 (71.3%), while only 2 examined sera (i.e., 3.8%) reacted in the highest titre of 3 200. Higher titres were found only in older wild boar individuals.

DISCUSSION

The wild boar is currently at relatively high population levels in the Czech Republic. It likes to inhabit deep deciduous and mixed forests, marshland covered with reed as well as field habitats. Serology for antibodies against leptospire in the wild boar from such areas of the Břeclav district confirmed positive cases in 16.9%. The results witness the fact that the wild boar individuals come in their environment into contact with various serotypes of leptospire which cause production of specific antibodies. Similar results differing only in the incidence of antibodies against leptospire were obtained by a number of authors studying these problems, for example (Sebek, 1960; Hübner and Horsch, 1977; Borcic *et al.*, 1989; Saliki *et al.*, 1998;

Krawczyk, 2000; Deutz *et al.*, 2002; Vicente *et al.*, 2002). In this study there predominates the *L. grippityphosa* serotype (100% of positive reactions) which is in accordance with other studies by, for example (Sebek, 1960; Witt *et al.*, 1986; Tremel and Nesnalova, 1993). A number of other authors, however, found positive reactions against more serotypes of leptospire. It was, for example, Kovacic *et al.* (2001) who in Croatia confirmed the presence of antibodies against up to 8 serotypes of leptospire with the *L. tarassovi* and *L. pomona* serotypes prevailing. New *et al.* (1994) found in the wild boar individuals from a national park antibodies against five serotypes of leptospire. Mason *et al.* (1998) tested blood sera of wild boars with leptospire of 14 serotypes and detected antibodies against 9 of them. In their examinations Saliki *et al.* (1998) found prevailing reactions against leptospire of the *L. bratislava* and *L. pomona* serotypes. From the above data it is clear that in the wild boar there are antibodies against leptospire which currently circulate in the territory. Considering the fact that the wild boar belongs to omnivores feeding on plants, invertebrates and various small vertebrates as well as cadavers, it can easily get into contact with leptospire and become infected in natural foci.

Antibodies were found in all age categories of the wild boar both in males and females. On the basis of these results we can suppose a general susceptibility of the wild boar to infection by leptospires with the incidence being strongly dependent on the frequency of contact with reservoir animals or the contaminated environment. Similar results are presented by Mason *et al.* (1998) examining wild boars from New South Wales.

Interesting is the fact that the titres found in the wild boar do not reach high levels. Most often we found titres of 100 and 200 (71.3%) and only exceptionally of 3 200 (3.8%). Similar results were those by, e.g., Schoneberg *et al.* (1999) in Germany and Krawczyk (2000) in Poland. With respect to the prevailing low titres we can state that the wild boar is susceptible to infection by various serotypes of leptospires, nevertheless, it is not the main reservoir animal. It is most probably only a potential reservoir infected in direct or indirect contact with main reservoir animals of particular serotypes of leptospires. Under conditions of the Czech Republic it is known that natural foci of the *L. grippotyphosa* serotype prevail; this fact being also documented in our study. This serotype is responsible for up to 90% of positive reactions in animals (Sebek and Rosicky, 1974). The main reservoir of leptospires of this serotype is the common vole (*Microtus arvalis*) in which the seropositivity amounts up to 42.5% (Sebek, 1985). Because this small rodent is widely distributed throughout biocoenoses in our country, natural foci of leptospirosis of the grippotyphosa serotype can be expected anywhere in the Czech Republic and the wild boar can get infected as confirmed in our study. In the area under study we have already confirmed the presence of leptospiral foci of the grippotyphosa type in small mammals (Treml *et al.*, 2002).

Some authors from abroad found antibodies against leptospires of the *L. pomona* and *L. tarassovi* serotypes in the wild boar (Mason *et al.*, 1998; Saliki *et al.*, 1998; Shoneberg *et al.*, 1999; Vicente *et al.*, 2002). It is common for pigs to be reservoirs of leptospires of the above two serotypes. The role of the wild boar, however, is questionable with respect to these serotypes. In our collection of samples from the wild boar we found no antibodies against the above-mentioned two serotypes as well as in domestic pigs kept in farms in the Czech Republic (Treml *et al.*, 1984). Our results confirm that the wild boar is susceptible to infection by leptospires circulating in natural foci. In our opinion, it would

be possible to use the examination of blood sera of the wild boar to monitor leptospiral foci in the environment.

Acknowledgements

We are indebted for the support to MVDr. D. Bill – director of the District Veterinary Service Břeclav, and for collection of blood specimens to a number of hunters and veterinarians MVDr. M. Hnilička and MVDr. Vl. Janík – staff members of the District Veterinary Service Břeclav.

REFERENCES

- Anjanin V.V. (1971): Leptospirosis in humans and animals (in Russian). Medicina, Moskva. 351 pp.
- Borcic B., Raos B., Sebek Z., Krancelic D., Eldan J.A., Filipovic V. (1989): Leptospiral antibodies in large game animals in northern Croatia (in Croatian). Vet. Arhiv, 59, 117–123.
- Dedek J., Loepelmann H., Kokles R. (1989): Ergebnisse flächendeckender serologischer Untersuchungen beim Schwarzwild (*Sus scrofa*) in einem Bezirk der DDR. Erkrankunden der Zootiere. In: Verhandlungsbericht des 31. Internationalen Symposiums über Erkrankunden der Zoo – und Wildtiere, Dortmund 1989, 309–314.
- Deutz A., Fuchs K., Schuller W., Müller M., Kerbl U., Klement C. (2002): Studies on the seroprevalence of antibodies against *Leptospira interrogans* in hunters and wild boar from south-eastern Austria (in German). Z. Jagdwiss., 48, 60–65.
- Egan J., Yearsley D. (1987): Serological survey of leptospiral antibodies in cattle, sheep and pigs in the Republic of Ireland. Irish Vet. J, 41, 213–214.
- Estavoyer J.M., Tran T.A., Hoen B. (2001): Leptospiroses. Rev. Prat., 51, 2086–2090.
- Hübner J., Horsch P. (1977): Untersuchungen zum Leptospirosegeschehen unter heimischen Wildtieren. Mh. Veter.-Med. (Jena), 25, 634–639.
- Kaszanyitzky J.E., Bajmocy E., Bascadi A., Matiz K. (1997): Observation on the abortion caused by leptospires in domestic farm animals (in Hungarian). Mag. Allatorv. Lap., 119, 415–419.
- Kovacic H., Karlovic M., Frkovic A. (2001): Investigation on the prevalence of antibodies against *Leptospira interrogans* in game in the territory of Gorski Kotar (in Croatian). Veterinarska stanica, 32, 69–77.
- Krawczyk M. (2000): Serological studies on leptospirosis in wild boar (in Polish). Med. Weter., 56, 440–443.

- Levett P.N. (2001): Leptospirosis. Clin. Microbiol. Rev., 14, 296–326.
- Lutz W. (1997): Serological evidence of antibodies against Toxoplasma and Leptospira in wild boar. (in German). Z. Jagdwiss., 43, 283–287.
- Malachov Ju.A., Alechin R.M. (1976): Leptospirosis in pigs (in Russian). Kolos – Moskva: 146 pp.
- Mraz A., Halasa M., Novomestský L. (1981): Latent form of leptospirosis of the pig (in Slovak). Zborník štatnej veterinárnej spravy MPVz SSR. Priroda, Bratislava. 175–184.
- Mason R.J., Fleming P.J.S., Smythe L.D., Dohnt M.F., Norris M.A., Symonds M.L. (1998): Leptospira interrogans antibodies in feral pigs from New South Wales. J. Wildl. Dis., 34, 738–743.
- New J.C., Delozier K., Barton C.E., Morris P.J., Potgieter L.N.D. (1994): A serological survey of selected viral and bacterial diseases of European wild hogs, Great – Smoky Mountains National Park, USA. J. Wildl. Dis., 30, 103–106.
- Power S.B. (1991): Diagnosing leptospira in pigs. Vet. Rec., 128, 43–45.
- Rocha T. (1998): A review of leptospirosis in farm animals in Portugal. Rev. Sci. Techn. OIE, 17, 699–712.
- Saliki J.T., Rodgers S.J., Eskew G. (1998): Serosurvey of selected viral and bacterial diseases in wild swine from Oklahoma. J. Wildl. Dis., 34, 834–838.
- Schoneberg A., Lutz W., Kampe U. (1999): Investigation of serum samples of wild boar (Sus scrofa L., 1758) for Leptospirosis. (in German) Z. Jagdwiss., 45, 262–265.
- Sebek Z. (1960): Leptospire – untersuchungen beim Jagdwild. Angew. Parasitol., 1, 52–56.
- Sebek Z. (1979): Standard methods of laboratory diagnostics of leptospires (in Czech). Acta Hyg. Epidemiol. Microbiol., 18 pp.
- Sebek Z. (1985): Problems of natural focality, epizootiology and epidemiology of leptospires in the Czech Republic (in Czech). Veterinářství, 35, 542–544.
- Sebek Z., Rosický B. (1974): On the occurrence, characteristics and structure of the foci of leptospirosis in Czechoslovakia (in Czech). Čs. Epidemiol. Mikrobiol. Imunol., 23, 10–21.
- Sebek Z., Tremel F., Valova M. (1983): Experimental infection with the virulent, central-European, murine Leptospira pomona strain in the pig (in German). Fol. Parasitol., 30, 269–275.
- Tagliaube S., Farina R. (1995): Sero-epidemiological study on prevalence of leptospirosis in domestic and wild animals (in Italian). Selezione Vet., 36, 941–952.
- Tremel F., Nesnalova E. (1993): The occurrence of antibodies to Leptospira in the blood serum of game (in Czech). Vet. Med. – Czech, 38, 123–127.
- Tremel F., Nesnalova E. (1995): Leptospirosis in slaughter pigs – serological and bacteriological examinations (in Czech). Veterinářství, 45, 216–217.
- Tremel F., Sebek Z., Hejlíček K. (1984): Territorial dissemination of antibodies against leptospirae at individual farms throughout a district, detected on the basis of serological testing of slaughter cattle and pigs (in Czech). Vet. Med. (Praha), 29, 531–537.
- Tremel F., Pejšoch M., Holešová Z. (2002): Small mammals – natural reservoir of pathogenic leptospires. Vet. Med. – Czech., 47, 309–314.
- Valkonen M., Klemets P., Nuorti P., Siikamäki H., Valtonen V. (2002): Leptospirosis -an increasingly common zoonosis (in Finnish). Duodecim, 118, 379–383.
- Vicente J., Leon-Vizcaino L., Gortazar C., Cubero Mj., Gonzales M., Martin-Atance P. (2002): Antibodies to selected viral and bacterial pathogens in European wild boars from southcentral Spain. J. Wildl. Dis., 38, 649–652.
- Vinetz J.M. (2001): Leptospirosis. Curr. Opin. Infect. Dis., 14, 527–538.
- Weber A., Christoph H. (1981): Seroepidemiologische Untersuchungen zum Vorkommen von Leptospirosen bei Schalenwild in der Bundesrepublik Deutschland. Z. Jagdwiss., 27, 283–287.
- Witt W., Friedel U., Winter E. (1986): Zum Vorkommen von Leptospiren - Antikörpern bei Wildschweinen. Mh. Veter. – Med., 41, 153–155.

Received: 02–12–13

Accepted after corrections: 03–04–07

Corresponding Author

Prof. MVDr. František Tremel, CSc., University of Veterinary and Pharmaceutical Sciences, Palackého 1–3, 612 42 Brno, Czech Republic
Tel. +420 541 562 307, e-mail: tremelf@vfu.cz
