International Journal for Parasitology 42 (2012) 739–745

Contents lists available at SciVerse ScienceDirect

International Journal for Parasitology

journal homepage: www.elsevier.com/locate/ijpara

Spatio-temporal trends and risk factors for *Trichinella* species infection in wild boar (*Sus scrofa*) populations of central Spain: A long-term study

M. Boadella [a,*], J.A. Barasona [a], E. Pozio [b], V. Montoro [a], J. Vicente [a], C. Gortazar [a], P. Acevedo [c,d]

[a] *Instituto de Investigación en Recursos Cinegéticos (CSIC-UCLM-JCCM), Ronda de Toledo s/n, 13071 Ciudad Real, Spain*
[b] *Department of Infectious Parasitic and Immunomediated Diseases, Istituto Superiore di Sanità, viale Regina Elena 299, 00161 Rome, Italy*
[c] *Biogeography, Diversity, and Conservation Research Team, Department of Animal Biology, Faculty of Sciences, University of Malaga, Campus de Teatinos s/n, 29071 Malaga, Spain*
[d] *CIBIO/UP – Centro de Investigação em Biodiversidade e Recursos Genéticos da Universidade do Porto, Campus Agrario de Vairão, 4485-661 Vairão, Portugal*

ARTICLE INFO

Article history:
Received 16 April 2012
Received in revised form 2 May 2012
Accepted 6 May 2012
Available online 4 June 2012

Keywords:
Biogeography of disease
Disease monitoring
Favourability function
Risk factor modelling
Time trend
Trichinella
Spain

ABSTRACT

In south-central Spain, the harvest of Eurasian wild boar (*Sus scrofa*) has increased significantly during recent decades in association with more intensive management actions to increase hunting yields and with consequent effects on the health status of the wild boar populations. We investigated the spatio-temporal trends and the risk factors related to the prevalence of *Trichinella* spp. in wild boar in order to obtain the annual probability of occurrence for these parasites in the Ciudad Real province of south-central Spain. Based on muscle samples collected during the hunting seasons from 1998/1999 to 2009/2010, the mean prevalence for *Trichinella* spp. in 95,070 wild boar was 0.2% (95% confidence interval 0.17–0.23). A subsample of 1,432 wild boar was also tested by ELISA. No correlation was observed between the prevalence of infection detected by serology and by the artificial digestion of muscle. The presence of *Trichinella* infections in wild boar showed a decreasing trend during the study period and was negatively related with fenced wild boar populations. The predicted 'favourability' for *Trichinella* infections disappeared almost completely after the 2006/2007 hunting season. Risk maps based on biogeographical tools showed, however, that most hunting estates presented favourable risk factors for these parasites during at least one of the hunting seasons studied.

1. Introduction

Nematodes of the genus *Trichinella*, the causal agents of trichinellosis in humans, are among the most widespread of zoonotic pathogens (Gibbs, 1997; Murrell et al., 2000; Pozio, 2007). Globally, the most important source of the human infection with these parasites is the domestic pig (Murrell and Pozio, 2011), and in industrialised areas of the European Union, efforts focused on removing *Trichinella* from the pig food chain have been highly successful (EFSA, 2011). Nonetheless, trichinellosis in Europe is still a problem (Murrell and Pozio, 2011) due to the sylvatic cycle in which the red fox (*Vulpes vulpes*) and the Eurasian wild boar (*Sus scrofa*) play important roles as wildlife reservoirs of *Trichinella britovi* and *Trichinella spiralis*, respectively (Pozio et al., 2009) making it almost impossible to eradicate this zoonosis (Rafter et al., 2005).

In the European Union, all animals susceptible to *Trichinella* infection are tested for these pathogens by digestion of muscle if the meat is intended for human consumption (European Community, 2005). Despite the sanitary regulations, pork and pork derived products from wild boar represent the second most important source of trichinellosis for humans (Murrell and Pozio, 2011). Outbreaks of trichinellosis associated with the consumption of pork from wild boar by hunters, their households and friends, are repeatedly documented (Arévalo Velasco et al., 2009; García-Sánchez et al., 2009).

Given that the wild boar is an important source of *Trichinella* for humans, understanding the spatio-temporal distribution of this host–parasite relationship and the associated risk factors is important for disease control and policy (Boadella et al., 2011). Some studies have described temporal trends of *Trichinella* infections in European wildlife (Ramisz et al., 2001, 2011; Kurdova-Mintcheva et al., 2009), and an increasing trend in the prevalence of *Trichinella* infections has recently been recorded in wild boar populations from Slovakia (Hurníková and Dubinský, 2009), north-eastern Germany (Mecklenburg–Western Pomerania) (Pannwitz et al., 2010) and Poland (Ramisz et al., 2011). In Poland, a recent increase in fox densities has been linked with an increase in the prevalence of *Trichinella* among wild boar (Ramisz et al., 2011), whereas in Mecklenburg–Western Pomerania the increase has been associated with a spread of raccoon dogs (*Nyctereutes procyonoides*) in the region (Pannwitz et al., 2010). Spatial trends in wild boar infection with *Trichinella* have been reported in Slovakia, where infected animals have been found in areas where these parasites had not

* Corresponding author. Tel.: +34 926295450; fax: +34 926295451.
E-mail address: mariana.boadella@gmail.com (M. Boadella).

0020-7519/$36.00
http://dx.doi.org/10.1016/j.ijpara.2012.05.003

been previously recorded in wild boar populations (Hurníková and Dubinský, 2009).

In south-central Spain (SCS), wild boar are often raised under intensive management conditions on commercial hunting estates that tend to maintain overabundant wild boar populations in order to maximise their profits. This high density of wild boar is maintained by fencing, artificial watering in summer and supplementary feeding throughout the year, creating an ecosystem dominated by large ungulates (Gortazar et al., 2006; Acevedo et al., 2007). In this region, the prevalence of *Trichinella* infection in wild boar (0.3–0.8%) (Pérez-Martín et al., 2000; García-Sánchez et al., 2009) was higher than the mean prevalence (0.2%) in Spain for the period 2007–2009 (EFSA, 2011). In wild boar from SCS, the prevalence of *T. spiralis* (74–100%) is higher than that of *T. britovi* (0–21%) (Pérez-Martín et al., 2000; García-Sánchez et al., 2009). However, the presence of 5% mixed *Trichinella* infections is noteworthy (Pérez-Martín et al., 2000; Rodríguez et al., 2008).

The aim of this work was to investigate the spatio-temporal trends and the risk factors explaining the detection of wild boar infected with *Trichinella* under different hunting management strategies in the province of Ciudad Real, SCS. We hypothesised that the prevalence of *Trichinella* infection in wild boar in Ciudad Real would be affected by recent trends towards the more intense management of fenced hunting estates.

2. Materials and methods

2.1. Study area and samples

The study area was Ciudad Real, a 19,813 km^2 province of SCS (Fig. 1). This province is characterised by a Mediterranean climate. The habitat is typically Mediterranean and characterised by evergreen oak (*Quercus ilex*) forests and scrublands (dominated by *Cystus* spp., *Pistacia* spp., *Rosmarinus* spp., *Erica* spp., and *Phyllirea* spp.) with scattered pastures and small areas with crops. Most of this territory is devoted to hunting, and the wild boar is widely distributed in the region (e.g., Rosell and Herrero, 2007) where it cohabits with the domestic pig (Palo et al., 2007; Ruiz-Fons et al., 2008). Wild boar population densities vary greatly in the province (up to 90 animals/km^2) due to hunting management actions (Acevedo et al., 2007).

We used two types of data and samples. The results of the artificial digestion of muscle-tissue samples from 95,070 wild boar hunted in 711 game estates during the period 1998–2010 was obtained from the Official Veterinary Services (see Section 2.3) of the province of Ciudad Real. Furthermore, sera from 1,432 wild boar were collected in 25 of the above-mentioned hunting estates between the 1999/2000 and 2010/2011 hunting seasons. Blood was drawn from the heart or the thoracic cavity during field sampling, and the serum was collected and frozen at −20 °C until required for analysis. For more details on sampling see Boadella et al. (2012a).

2.2. Serological testing

Wild boar sera were tested using a commercial ELISA (ID Screen® *Trichinella* Indirect, IDVet, Montpellier, France), based on the excretory/secretory (E/S) antigens and allowing the detection of anti-*Trichinella* spp. IgG, according to the manufacturer's instructions. Briefly, the cut-off was calculated as the sample to positive (S/P) ratio = 100 × [(OD sample − OD negative control)/(OD positive control − OD negative control)]. Sera with an S/P ratio > 60% were considered positive. Thirty ELISA positive sera were sent to the European Union Reference Laboratory for Parasites (Rome, Italy) to confirm the result using a western blot (Gomez-Morales et al., 2008).

2.3. Spatial and temporal trends

The number, origin and *Trichinella* spp. from wild boar tested by artificial digestion were entered into a database based on hunting estate and hunting season. Data from a hunting estate where 141 *Trichinella*-infected wild boar were recorded out of 1,888 examined were excluded from the analysis in order to avoid the risk that the model overfits the local conditions of this hunting estate with a consequent loss of generality of the model.

The spatio-temporal trends and the risk factors for the detection of *Trichinella*-infected wild boar were assessed using a logistic regression (Hosmer and Lemeshow, 2000). The epidemiological information useful to predict the risk for *Trichinella* infection in wild boar was obtained from three sources: (i) the database of Official Veterinary Services which provided the prevalence of *Trichinella* infections in wild boar per hunting estate and hunting season, and information on the relative abundance of wild boar, (i.e., the number of animals hunted per 100 ha (Acevedo et al.,

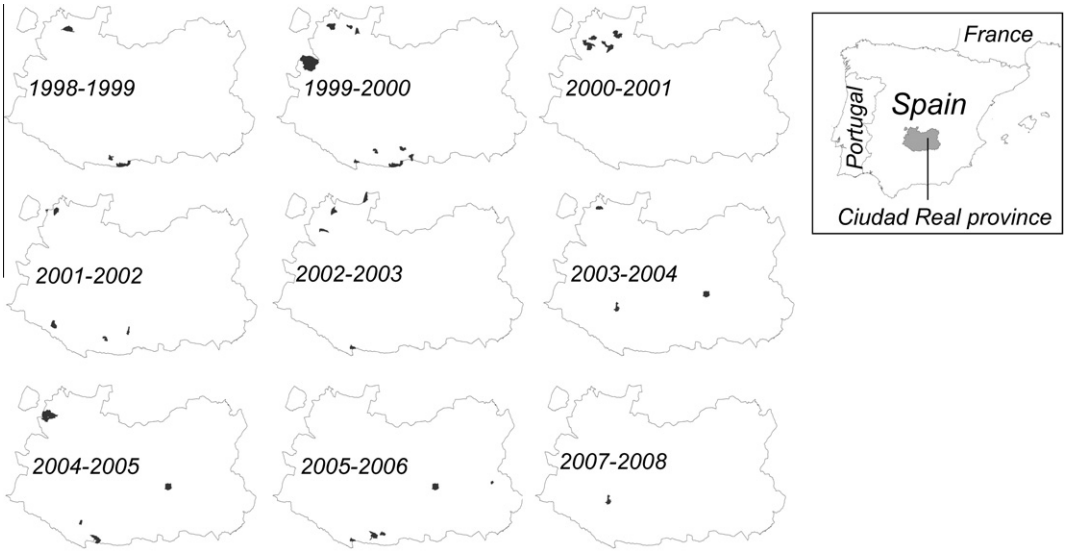

Fig. 1. Wild boar *Trichinella* spp. infections in province of Ciudad Real, Spain. Map of the province of Ciudad Real showing the hunting estates (in black) where *Trichinella* spp. infections were detected in wild boar (*Sus scrofa*) by the Official Veterinary Services, in each hunting season during which at least one infected animal was detected.