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Short communication

Prevalence of *Coxiella burnetti* infection in wild and farmed ungulates

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Abstract

The aim of this study was to evaluate by serology and PCR analyses the prevalence of *Coxiella burnetti* infection in ungulates in Spain. Sera were collected from red deer (*Cervus elaphus*; $n = 116$), roe deer (*Capreolus capreolus*; $n = 39$), fallow deer (*Dama dama*; $n = 13$) and cattle ($n = 79$). Sera were tested for anti-*C. burnetii* antibody detection by means of an immunofluorescence antibody assay (IFA) and *C. burnetii* DNA was amplified by PCR in samples from ungulates that had antibodies to phase II antigens. Twenty-nine, 15 and 39 percent of the red deer, roe deer and cattle had antibodies against *C. burnetii*, respectively. None of the fallow deer sera tested positive. Seroprevalence was statistically higher in farmed than in wild red deer and higher in northern than in southern populations, whereas an inverse pattern was observed for the roe deer. Most of the seropositive animals had only anti-*C. burnetii* phase II antibodies, thus showing the acute nature of infections in the sampled ungulates. These results show that *C. burnetii* circulates in wild ungulates in Spain and suggest that they can act as pathogen reservoirs for both domestic animals and humans.

Keywords: *Coxiella*; Q fever; Tick; Vector-borne diseases; Wildlife

1. Introduction

Q fever is a worldwide zoonosis produced by the Gram-negative bacteria, *Coxiella burnetii*. Multiple hosts can serve as reservoir of infection, including many wild and domestic mammals, birds and ticks (Willeberg et al., 1980; Marrie et al., 1986; Webster et al., 1995; Dunbar et al., 1998; Maurin and Raoult, 1999; Komiya et al., 2003). However, domestic ruminants represent the most frequent source of *C. burnetii* infection in humans (Maurin and Raoult, 1999).

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Over 40 tick species are naturally infected with *C. burnetii*, including *Rhipicephalus*, *Haemaphysalis*, *Amblyomma*, *Dermacentor*, *Ixodes* and *Otobius* species. However, ticks are not considered essential in the natural cycle of *C. burnetii* in livestock because other sources of infection are more important in animals that live in close contact (Maurin and Raoult, 1999). In contrast, ticks may play a significant role in the transmission of coxiellosis among wild vertebrates (Marrie et al., 1986; Maurin and Raoult, 1999).

Despite efforts to eradicate coxiellosis from cattle, goat and sheep herds, the disease remains a serious risk for human and animal health in Spain (Tellez et al., 1988; Maurin and Raoult, 1999; Maltezou and Raoult, 2002; Bolaños et al., 2003; Marrie, 2004; Cardenosa et al., 2006; Oporto et al., 2006; Sanz et al., 2006). The disease seems to be more prevalent in the Basque and Navarra provinces in northern Spain than in the central and southern regions of the country (Tellez et al., 1988; Maurin and Raoult, 1999). Recent research efforts have been focused on the characterization of *C. burnetii* infection in domestic ruminants and humans (reviewed by Woldehiwet, 2004; Kazar, 2005). However, little is known about the prevalence of *C. burnetii* in wild ungulates (Enright et al., 1971; Ejercito et al., 1993; Marrie et al., 1993) and to our knowledge, *C. burnetii* infection has not been characterized in Spanish wild and farmed cervids. Wild and farmed cervid populations are growing in Spain and in other countries due to their value as game animals, thus increasing the risk for disease transmission to humans and domestic animals (Gortázar et al., 2006; Vicente et al., 2005).

The objective of the study reported herein was to evaluate by serology and PCR analyses the prevalence of *C. burnetii* infection in wild and farmed cervids from two regions in northern and southern Spain.

2. Materials and methods

2.1. Study sites

In southern Spain, we collected sera of Iberian red deer (wild and farmed), European roe deer and cattle in a 3000 ha fenced hunting estate (LO) located in the Natural Park of ''Los Alcornocales'' in the province of Cádiz, Andalucía. We also collected European roe deer and cattle sera from a neighbouring hunting estate (OJ). In northern Spain, Iberian red deer, European roe deer and fallow deer serum samples were collected in Asturias region. The number of samples through ruminant species and habitat are summarized in Table 1.

Table 1
Results of IFA test to phases I and II *C. burnetii* antigens and *htpB* gene nested PCR in wild and domesticated cervids and cattle

Host	Habitat	Location	No. of samples	Seropositive (% ± S.E. at 95% CI)		Nested PCR positive/total tested (%)[a]
				Phase I	Phase II	
Red deer	Wild	Northern Spain	21	0 (0 ± 0)	2 (9.5 ± 12.9)	1/2 (50)
	Wild	Southern Spain	15	0 (0 ± 0)	0 (0 ± 0)	None tested
	Farm LO	Southern Spain	80	0 (0 ± 0)	32 (40 ± 10.8)	4/32 (12)
Subtotal red deer			116	0 (0 ± 0)	34 (29.3 ± 8.2)	5/34 (14.7)
Roe deer	Wild	Northern Spain	21	0 (0 ± 0)	0 (0 ± 0)	None tested
	Wild	Southern Spain	18	0 (0 ± 0)	6 (33 ± 22.4)	0/6 (0)
Subtotal roe deer			39	0 (0 ± 0)	6 (15.4 ± 11.6)	0/6 (0)
Fallow deer	Wild	Northern Spain	13	0 (0 ± 0)	0 (0 ± 0)	None tested
Cattle	Farm LO	Southern Spain	49	3 (6 ± 4.9)	26 (53 ± 10.6)	3/26 (12)[b]
	Farm OJ	Southern Spain	30	0 (0 ± 0)	2 (7 ± 5.1)	0/2 (0)
Subtotal cattle			79	3 (3.8 ± 4.3)	28 (35.4 ± 10.6)	3/28 (10.7)

[a] Only samples from seropositive animals were tested by *C. burnetii htpB* gene nested PCR.
[b] Samples from animals seropositive for phase I *C. burnetii* antibodies were negative for the *htpB* gene nested PCR assay.