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Tick-borne Diseases in Syntopic Populations of Fallow Deer and Axis Deer in Northern Mexico

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ABSTRACT: We harvested 21 fallow deer (*Dama dama*) and 17 axis deer (*Axis axis*) in northern Mexico. Two fallow deer were positive for *Babesia bigemina* and one for *Babesia bovis*. Amplicons had the expected 170 and 291 base pairs and were identical to *B. bigemina* (S45366) and *B. bovis* (M38218), respectively.

Populations of exotic ungulates have been established outside their native range in many parts of the world (Chapman and Chapman 1975; Feldhammer et al. 1988), including Mexico, even though current regulations ban the transport and release of exotic ungulates in Mexico. These introductions have potential detrimental effects on health of livestock and native wildlife and on resource availability for native species. Additionally, exotic species may be more resistant to some diseases, conferring a competitive advantage over native species (Davidson and Crow 1983; Davidson et al. 1985; Flynn et al. 1990).

Babesia bovis and *Babesia bigemina* are transmitted by cattle fever ticks (*Rhipicephalus* spp.) and cause bovine babesiosis. Fever ticks are one-host ticks that preferentially feed on cattle (*Bos primigenius*) but also feed on other ungulates, such as white-tailed deer (*Odocoileus virginianus*; Pound et al. 2010). The role of exotic ungulates in cattle fever tick host-vector-pathogen dynamics is poorly known.

Cattle ranches in northern Mexico increasingly use recreational ventures to generate income. Consequently, native populations of white-tailed deer and exotic

species such as fallow deer (*Dama dama*) and axis deer (*Axis axis*) are increasing and may serve as hosts for ticks that are vectors for *Babesia* spp. Molecular and serologic evidence show *Babesia* spp. in white-tailed deer from northern Mexico and southern Texas (Cantu et al. 2009; Ramos et al. 2010), and Cardenas-Canales et al. (2011) reported *B. bovis* and *B. bigemina* in nilgai (*Boselaphus tragocamelus*) in northern Mexico. Thus, exotic ungulates may harbor *Babesia* spp. Serologic monitoring of domestic livestock and wildlife on ranches will help determine the prevalence, incidence, and possible patterns of spread of *Babesia* and other tickborne pathogens.

Axis and fallow deer were introduced into central and northern Mexico for hunting. Axis deer occur in at least 50 extensive operations, with a total area of 160,100 ha; fallow deer exist in 44 operations on 116,000 ha (Álvarez-Romero and Medellín 2005). No information exists on the epidemiology of *Babesia* in cervids in Mexico. We used serology to determine whether axis and fallow deer had been exposed to *B. bovis* and *B. bigemina* and nested PCR to detect *Babesia* DNA on a ranch in northern Mexico.

This study was conducted on a 400-ha, high-fenced, private ranch in Soto la Marina, Tamaulipas, Mexico, (23°34'108"N, 97°53'W). Ungulate species living on the ranch are axis and fallow deer, sika (*Cervus nippon*), blackbuck (*Antilope cervicapra*), Barbary sheep (*Ammotragus lervia*), native

white-tailed deer, and domestic cattle. Cattle have been allowed to graze pastures periodically since 2007 as a tick control strategy. Exotic wildlife and cattle use the same source of water at a water control structure.

Twenty-one fallow deer and 17 axis deer of different sexes and ages were captured using drop nets. Nets were set and baited with corn 3 d before the capture. Blood samples were collected from the jugular vein in ethylenediaminetetraacetic acid-K3 anticoagulant and centrifuged to obtain serum at the laboratory of the Instituto de Investigaciones Forestales, Agrícola y Pecuarias-Aldama, Tamaulipas, and stored at 4 C.

Blood samples were analyzed by nested PCR following Figueroa et al. (1993) using specific primers for *B. bovis* that identified the gene *Rap-1* and *B. bigemina*. Amplified PCR products were electrophoresed on agarose gels, stained with ethidium bromide, visualized, and photographed under ultraviolet light. Amplicons from two *B. bigemina*-positive and one *B. bovis*-positive blood samples were sequenced.

Indirect immunofluorescence antibody (IFA) test was used to test for evidence of exposure to *B. bovis* and *B. bigemina*. *Babesia bovis* and *B. bigemina*-infected bovine erythrocytes were obtained from in vitro culture as described by Levy and Ristic (1980) and Vega et al. (1985) and were smeared and kept at -70 C until used. The IFA slides were fixed with acetone and incubated with each of the serum samples at a screening dilution of 1:100 for 30 min at 37 C. This dilution was chosen because there is no cross-reactivity between *B. bovis* and *B. bigemina* in cattle. The reaction was detected with protein G conjugated with Alexa 488 (Molecular Probes, Eugene, Oregon, USA) at 1:100 followed by incubation for 30 min at 37 C. As positive controls, bovine sera from cattle experimentally infected with *B. bovis* or *B. bigemina* were used at the same dilution. Sera from uninfected deer were used as negative

controls. Finally, a Giemsa-stained blood smear from each blood sample was examined under oil immersion at 1,000 \times to detect hemoparasites.

We found two positive samples for *B. bigemina* based on amplicon size (170 base pairs [bp]), and one positive sample for *B. bovis* (291 bp) in fallow deer. Amplicons from positive samples had the expected length and were identical to published sequences of both species (*B. bovis* accession M38218; *B. bigemina* accession S45366). No positive samples were found from axis deer.

Four samples were antibody-positive for *B. bovis*, two from axis deer and two from fallow deer, whereas 19 samples were *B. bigemina* antibody-positive, five axis and 13 fallow deer. Only one fallow deer sample was positive for antibodies to both *B. bigemina* and *B. bovis*. No *Babesia* parasites definitively identifiable by piriform shape in joined pairs were observed by microscopic examination of the Giemsa-stained blood films.

Evidence of *Babesia* spp. in fallow deer has not been reported previously. Bovine babesiosis is widespread in Tamaulipas and has been reported in white-tailed deer (Cantu et al. 2009) and nilgai (Cardenas-Canales et al. 2011). The presence of other non-cattle hosts such as fallow deer complicates the epidemiology of bovine babesiosis. Subclinical infection may occur in white-tailed deer, providing a reservoir of infection (Gallatin et al. 2003). According to Holman et al. (2000), subclinical infections with *Babesia odocoilei* were not found in fallow and axis deer; rather, white-tailed deer resident within the ranch appeared to be the reservoir of infection.

The presence of antibodies and DNA of *B. bovis* and *B. bigemina* raises the still unanswered question of whether fallow deer and axis deer are susceptible to subclinical disease.

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