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Short communication

Wild birds as host of *Borrelia burgdorferi* sensu lato in northwestern Argentina

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ABSTRACT

Borrelia burgdorferi sensu lato (s. l.) spirochetes are associated with a wide range of vectors and hosts. Birds are important hosts in the ecology of some hard ticks (Ixodidae) in northwestern Argentina, where *B. burgdorferi* s.l. have been detected in *Ixodes pararicinus*. We evaluated *Borrelia* infection in ticks collected from wild birds by molecular analysis through the presence of *Borrelia* DNA (by nested-PCR targeting the *fla* gene). A total of 381 ticks (357 larvae and 24 nymphs) belonging to four species (*I. pararicinus*, *Haemaphysalis juxtakochi*, *Haemaphysalis leporispalustris* and *Amblyomma* sp.) were collected. Partial sequences of the *fla* gene of *Borrelia* (100% identical to *Borrelia* sp. haplotype I from Argentina) were detected in 9 of 70 tick pools (6 pools of larvae and 1 pool of nymphs of *I. pararicinus*, and in 2 pools of *H. juxtakochi* larvae) collected on *Turdus rufiventris*, *Syndactila rufosuperciliata* and *Troglodytes aedon*. The results of this study suggest that resident birds have reservoir capacity for *Borrelia* sp. haplotype I.

1. Introduction

The *Borrelia burgdorferi* sensu lato (s. l.) complex constitutes a group of tick-associated bacteria composed by at least 20 genospecies with worldwide distribution (Margos et al., 2011, 2014; Ivanova et al., 2014). *Borrelia burgdorferi* sensu stricto (s. s.), *Borrelia afzelii* and *Borrelia garinii* have been recognized as important human pathogens in the Northern Hemisphere (Neartic and Palearctic Zoogegraphical Regions) (Stanek et al., 2012). Recent research detected in southern South America congeneric organisms phylogenetically related to *B. americana* and *Borrelia* sp. genospecies 1 from United States infecting ticks of the genus *Ixodes* (Barbieri et al., 2013; Nava et al., 2014), *Borrelia chilensis* in *Ixodes stilesi* (Ivanova et al., 2014) and *Borrelia* genospecies closely related to *B. chilensis* in *Ixodes sigelos* and *Ixodes neuquenensis* (Sebastian et al., 2016). Particularly, DNA of *Borrelia* has been detected in *Ixodes pararicinus* ticks collected in the northwestern region of Argentina (Nava et al., 2014; Saracho Bottero et al., 2017; 2018).

Ixodes pararicinus is distributed in Argentina, Colombia and Peru (Keirans et al., 1985; Guglielmone et al., 1992; Mattar and Lopéz-Valencia, 1998; Díaz et al., 2007) In Argentina, *I. pararicinus* inhabits the Yungas ecoregion and has been collected in the provinces of Jujuy, Salta, and Tucuman (Guglielmone and Nava, 2005). In these regions, while larvae and nymphs of *I. pararicinus* feed mainly on rodents and birds (Beldoménico et al., 2003; Flores et al., 2014), adults are found frequently associated with *Mazama gouazoubira*, *Tayassu tajacu* (Mammalia: Artiodactyla), cattle and horses (Guglielmone and Nava, 2005).

In nature, *B. burgdorferi* s. l. is maintained by complex enzootic cycles involving a large variety of mammalian, avian and reptilian hosts and hard ticks of the genus *Ixodes* as vectors (Margos et al., 2011). However, different groups of vertebrates differ in their capacity as host for *Borrelia* genospecies. *Borrelia burgdorferi* s.s. is considered a generalist species, while *B. garinii*, *B. valaisiana* and *B. afzelii* are considered specialist. *Borrelia garinii* and *B. valaisiana* are mainly maintained by avian hosts whereas *B. afzelii* is a rodent specialist (Kurtenbach et al., 2006). Wild birds are among the most mobile tick hosts, and play an important role carrying ticks and their associated pathogens (Elfving et al., 2010; Flores et al., 2016). Some passerine birds act as reservoir hosts for *B. burgdorferi* s.l. (Scott et al., 2001; Norte et al., 2013). In northwestern Argentina, *B. burgdorferi* s.l. have been detected in *I. pararicinus* (Nava et al., 2014; Saracho Bottero et al., 2017; 2018); however, there is no information about the role of autochthonous birds as hosts for *Borrelia* spp. The present study evaluated *Borrelia* infection

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Table 1
Analyzed *Borrelia*-infected ticks collected from avian hosts in El Rey National Park, northwestern Argentina, in 2012.

Bird family	Bird species	No. birds with infected Ticks /No. infested birds (%)	Ticks			
			ID[a]	Species	Stage[b]	No. of ticks analyzed in one pool
Furnaridae	*Syndactyla rufosuperciliata*	1/3 (33)	Rey28	*Ixodes pararicinus*	L	4
Troglodytidae	*Troglodytes aedon*	1/4 (25)	Rey18	*Ixodes pararicinus*	L	7
Turdidae	*Turdus rufiventris*	5/10 (50)	Rey17	*Ixodes pararicinus*	L	2
				Ixodes pararicinus	N	1
				Haemaphysalis juxtakochi	L	4
			Rey27	*Ixodes pararicinus*	L	7
				Ixodes pararicinus	N	3
			Rey31	*Ixodes pararicinus*	L	10
				Haemaphysalis juxtakochi	L	14
			Rey33	*Ixodes pararicinus*	L	13
				Ixodes pararicinus	N	1
				Haemaphysalis juxtakochi	L	39
				Haemaphysalis juxtakochi	N	3
			Rey36	*Ixodes pararicinus*	L	4

[a] **ID**: Bird identification.
[b] **L**: larvae; **N**: nymphs.

in ticks collected from wild birds in northwestern Argentina.

2. Materials and methods

2.1. Study site and bird captures

The study was conducted in the Yungas ecoregion (El Rey National Park: 24°43′S, 64°38′W), northwestern Argentina (Burkart et al., 1999) during May of 2012. Birds were captured using mist nets that remained active during morning and twilight hours as previously reported (Flores et al., 2014). Each individual bird was identified following Narosky and Yzurieta (2010), classified according to Clements et al. (2016) criteria and examined for ticks using fine-tipped tweezers. After being processed, the birds were released, and the obtained ticks were stored frozen until specific determination in the laboratory. Identification was carried out following Kohls (1960), Venzal et al. (2005) and Nava et al. (2017), and by comparison with laboratory-reared material deposited at the tick collection of the Instituto Nacional de Tecnología Agropecuaria, Estación Experimental Agropecuaria Rafaela, Argentina. Although there are not formal descriptions to differentiate immature stages of *I. pararicinus* and *I. aragaoi*, we have assumed that our findings correspond to I. pararicinus since the ticks processed in this study were collected in an area which corresponds to the distributional area of *I. pararicinus* in Argentina (see Nava et al., 2017). The results of the ticks infesting these birds have been published elsewhere (Flores et al., 2014). To determine the probable participation of the bird species as reservoir host, the infection in engorging or engorged larvae collected was determined (Gern et al., 1998).

2.2. Molecular detection and characterization of Borrelia species

For each individual bird, ticks were separated according to the developmental stage and homogenised with a pestle in pools of 1–39 larvae and 1–4 nymphs. DNA extraction from each pool was performed using the AxyPrep Body Fluid Viral DNA/RNA Miniprep Kit (Axygen Biosciences, USA) following the manufacturer's instructions. The presence of *Borrelia* DNA was tested by nested-PCR targeting the flagellin gene (*fla*), which is present in all *Borrelia* species, using primers Fla LL (5′-ACA TAT TCA GAT GCA GAC AGA GGT- 3′) and FLA RL (5′-GCA ATC ATA GCC ATT GCA GAT TGT-3′) for the first reaction, and primers Fla LS (5′-AAC AGC TGA AGA GCT TGG AAT G-3′) and FLA RS (5′-CTT TGA TCA CTT ATC ATT CTA ATA GC-3′) for the nested reaction. The first reaction targets a 665-bp fragment, and the nested reaction targets a 354-bp fragment. PCR conditions were adopted as described elsewhere (Barbour et al., 1996). For all PCR reactions, water was included as negative control, and *Borrelia anserina* DNA (Ataliba et al., 2007) was included as positive control. PCR products were purified using ExoSAP-IT® (USB Corporation, Cleveland, OH), and submitted to DNA sequencing in an ABI automatic sequencer (Applied Biosystems/Thermo Fisher Scientific, model ABI 3500 Genetic Analyser, Foster City, CA). DNA sequences were assembled, aligned and compared to each other using Geneious R9 software (Kearse et al., 2012), and also compared with sequences deposited in GenBank through BLAST analyses (www.ncbi.nlm.nih.gov/blast/).

3. Results

Overall, 85 birds were captured, of which 42 (49%) were infested by ticks. Parasitized birds belonged to 14 of 27 species, 9 of 12 families and three orders. A complete list of these tick-host associations has been published elsewhere (Flores et al., 2014). A total of 381 ticks (357 larvae and 24 nymphs) belonging to the following four species were collected: *I. pararicinus* (236 larvae and 11 nymphs), *Haemaphysalis juxtakochi* (89 larvae and 9 nymphs), *Haemaphysalis leporispalustris* (12 larvae and 4 nymphs), *Haemaphysalis* sp. (10 larvae), and *Amblyomma* sp. (10 larvae). *Ixodes pararicinus* was the most abundant species, collected on 37/42 (88%) birds.

Among 70 evaluated tick pools (42 of *I. pararicinus*, 15 of *H. juxtakochi*, 5 of *H. leporispalustris*, 4 of *Haemaphysalis* sp. and 4 of *Amblyomma* sp.), DNA of *Borrelia* was detected in 6 pools of larvae and 1 pool of nymphs of *I. pararicinus*, and in 2 pools of *H. juxtakochi* larvae. *Borrelia*-positive tick pools were collected on 5 *Turdus rufiventris*, 1 *Syndactila rufosuperciliata* and 1 *Troglodytes aedon* (Table 1). In 4 out of 5 *Turdus rufuventris*, 1 out of 1 *Syndactila rufosuperciliata*, and 1 out of 1 *Troglodytes aedon*, *Borrelia*-positive larvae were found, while no nymphs or positive nymphs were detected on the same bird individuals (excluding/ reducing the probability of co-feeding transmission) suggesting that infection was acquired from the birds.

Partial sequences of the *fla* gene from tick pools were 100% identical to each other and to the sequence of *Borrelia* sp. haplotype I from Argentina (GenBank accession number: KJ994335). The *fla* gene partial sequences generated in the present study have been deposited in GenBank under the accession numbers KY984010-KY984018.

4. Discussion

In the present work we detected the presence of *B. burgdorferi* s. l. DNA in two of the four species of tick collected on free ranging birds. Engorged larvae and nymphs of *I. pararicinus* and *H. juxtakochi* were found to contain *Borrelia* DNA. These ticks were found feeding on *T. rufiventris*, *S. rufosuperciliata* and *T. aedon*. The sequences obtained from a fragment of the *Borrelia fla* gene were 100% identical to sequences

previously detected in *I. pararicinus* adult ticks collected on vegetation in the same region of the present study (*Borrelia* sp. haplotype I) (Nava et al., 2014).

The present detection of *Borrelia* DNA in *I. pararicinus* larvae is complementary to the studies of Nava et al. (2014) and Saracho Bottero et al. (2017), who detected infection in unfed adults collected on vegetation and in semi-engorged nymphs collected on birds, respectively. In addition, we report for the first time that individuals of *H. juxtakochi* contained *Borrelia* DNA; however, this detection alone does not prove any vector role. Sun and Xu (2003) observed that *B. garinii* did not survive to the digestion process in *Haemaphysalis concinna,* precluding transstadial survival of the bacterium in this tick species. Therefore, at this moment, our findings for *H. juxtakochi* do not imply that this species has any epidemiological role.

The three bird species harboring *Borrelia*-positive ticks, *T. rufiventris*, *S. rufosuperciliata* and *T. aedon*, are resident species. Similarly, Saracho Bottero et al. (2017) obtained positive samples for *Borrelia* from *I. pararicinus* collected on *T. rufiventris*, *T. nigriceps* and *S. rufosuperciliata*. In the present work, no *T. nigriceps* was captured, since this species is a latitudinal migrant and its density decreases during the winter season (Blake and Rouges, 1997). The importance of birds, including those belonging to the genus *Turdus* and *T. aedon*, in the ecology of the genus *Borrelia* is recognized in the Northern Hemisphere (Anderson et al., 1990; Taragel'ová et al., 2008; Hamer et al., 2012; Scott et al., 2012; Norte et al., 2013; Rudenko et al., 2014); however, it is poorly known in South America. The detected *Borrelia* sp. haplotype I was described as closely related to *B. americana* by Nava et al. (2014). Based on new evidences, *B. americana* is a bird-associated agent closely related to *B. garinii* and *B. valaisiana* (Rudenko et al., 2014). Gern (2008) postulated that the evolution of *Borrelia* is driven by a specific association among the parasite and the host. Based on this hypothesis we can speculate that this *Borrelia* is maintained by birds as reservoir hosts. However, we cannot discard rodents as potential host since they are found frequently infested by *I. pararicinus* (Beldoménico et al., 2003).

In conclusion, the presence of *Borrelia* DNA in larvae of *I. pararicinus* and *H. juxtakochi* off four *T. rufiventris*, one *S. rufosuperciliata* and one *T. aedon* and the unlikely occurrence of co-feeding suggest that these birds are reservoir hosts, although transovarial transmission of *Borrelia* sp. in these ticks cannot be totally discarded (Rollend et al., 2013). Future research is needed to confirm the biological elements that integrate the transmission cycle of *B. burgdorferi* s. l. in northwestern Argentina.

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References

Anderson, J.F., Magnarelli, L.A., Stafford III, K.C., 1990. Bird-feeding ticks transstadially transmit *Borrelia burgdorferi* that infect Syrian hamsters. J. Wildl. Dis. 26, 1–10.

Ataliba, A.C., Resende, J.S., Yoshinari, N., Labruna, M.B., 2007. Isolation and molecular characterization of a Brazilian strain of *Borrelia anserina*, the agent of fowl spirochaetosis. Res. Vet. Sci. 83, 145–149.

Barbieri, A.M., Venzal, J.M., Marcili, A., Almeida, A.P., González, E.M., Labruna, M.B., 2013. *Borrelia burgdorferi* sensu lato infecting ticks of the *Ixodes ricinus* complex in Uruguay: first report for the Southern Hemisphere. Vector Borne Zoonotic Dis. 13, 147–153.

Barbour, A.G., Maupin, G.O., Teltow, G.J., Carter, C.J., Piesman, J., 1996. Identification of an uncultivable *Borrelia* species in the hard tick *Amblyomma americanum*: possible agent of a Lyme disease-likeillness. J. Infect. Dis. 173, 403–409.

Beldoménico, P.M., Baldi, C.J., Antoniazzi, L.R., Orduna, G.M., Mastropaolo, M., Macedo, A.C., Ruiz, M.F., Orcellet, V.M., Peralta, J.L., Venzal, J.M., Mangold, A.J., Guglielmone, A.A., 2003. Ixodid ticks (Acari: Ixodidae) present at Parque Nacional El Rey. Argentina. Neotrop. Entomol. 32, 273–277.

Blake, J.G., Rouges, M., 1997. Variation in capture rates of understory birds in El Rey National Park, northwestern Argentina. Ornitol. Neotrop. 8, 185–193.

Burkart, R.N., Bárbaro, N., Sánchez, R., Gómez, D., 1999. Eco-regiones de la Argentina. Administración de Parques Nacionales, Programa de Desarrollo Institucional Ambiental, Buenos Aires, Argentina.

Clements, J.F., Schulenberg, T.S., Iliff, M.J., Roberson, D., Fredericks, T.A., Sullivan, B.L., Wood, C.L., 2016. The eBird/Clements Checklist of Birds of the World. v2016. Downloaded from. http://www.birds.cornell.edu/clementschecklist/download/.

Díaz, M.M., Nava, S., Venzal, J.M., Sanchez, N., Guglielmone, A.A., 2007. Tick collections from the Peruvian Amazon, with new host records for species of *Ixodes* Latreille, 1795 (Acari: Ixodidae) and *Ornithodoros* Koch, 1844 (Acari: Argasidae). Syst. Appl. Acarol. 12, 127–133.

Elfving, K., Olsen, B., Bergström, S., Waldenström, J., Lundkvist, A., Sjöstedt, A., Mejlon, H., Nilsson, K., 2010. Dissemination of spotted fever rickettsia agents in Europe by migrating birds. PLoS ONE 5, e8572.

Flores, F.S., Nava, S., Batallán, G., Tauro, L.B., Contigiani, M.S., Diaz, L.A., Guglielmone, A.A., 2014. Ticks (Acari: Ixodidae) on wild birds in north-central Argentina. Ticks Tick-borne Dis. 5, 715–721.

Flores, F.S., Costa, F.B., Nava, S., Diaz, L.A., Labruna, M.B., 2016. Rickettsial infection in ticks infesting wild birds from two eco-regions of Argentina. Rev. Bras. Parasitol. Vet. 25, 378–382.

Gern, L., 2008. *Borrelia burgdorferi* sensu lato, the agent of Lyme borreliosis: life in the wilds. Parasite 15, 244–247.

Gern, L., Estrada-Peña, A., Frandsen, F., Gray, J.S., Jaenson, T.G.T., Jongejan, F., Kahl, O., Korenberg, E., Mehl, R., Nuttall, P.A., 1998. European reservoir hosts of *Borrelia burgdorferi* sensu lato. Zentralbl. Bakteriol. 287, 196–204.

Guglielmone, A.A., Nava, S., 2005. Las garrapatas de la familia Argasidae y de los géneros *Dermacentor*, *Haemaphysalis*, *Ixodes* y *Rhipicephalus* (Ixodidae) de la Argentina: distribución y hospedadores. Rev. Inv. Agropec. 34, 123–141.

Guglielmone, A.A., Mangold, A.J., Aufranc, C.R., 1992. *Haemaphysalis juxtakochi*, *Ixodes pararicinus* (Ixodidae) and *Otobius megnini* (Argasidae) in relation to the phytogeography of Argentina. Ann. Parasitol. Hum. Comp. 67, 91–93.

Hamer, S.A., Goldberg, T.L., Kitron, U.D., Brawn, J.D., Anderson, T.K., Loss, S.R., Walker, E.D., Hamer, G.L., 2012. Wild birds and urban ecology of ticks and tick-borne pathogens, Chicago, Ilinois, USA, 2005–2010. Emerg. Infect. Dis. 18, 1589–1595.

Ivanova, L.B., Tomova, A., González-Acuña, D., Murúa, R., Moreno, C.X., Hernández, C., Cabello, J., Cabello, C., Daniels, T.J., Godfrey, H.P., Cabello, F.C., 2014. *Borrelia chilensis*, a new member of the *Borrelia burgdorferi* sensu lato complex that extends the range of this genospecies in the Southern Hemisphere. Environ. Microbiol. 16, 1069–1080.

Kearse, M., Moir, R., Wilson, A., Stones-Havas, S., Cheung, M., Sturrock, S., Buxton, S., Cooper, A., Markowitz, S., Duran, C., Thierer, T., Ashton, B., Meintjes, P., Drummond, A., 2012. Geneious Basic: An integrated and extendable desktop software platform for the organization and analysis of sequence data. Bioinformatics 28, 1647–1649.

Keirans, J.E., Clifford, C.M., Guglielmone, A.A., Mangold, A.J., 1985. *Ixodes (Ixodes) pararicinus*, n. sp. (Acari: Ixodoidea: Ixodidae), a South American cattle tick long confused with *Ixodes ricinus*. J. Med. Entomol. 22, 401–407.

Kohls, G.M., 1960. Records and new synonymy of New World Haemaphysalis tick, with descriptions of the nymph and larva of H. juxtakochi Cooley. J. Parasitol 46, 355–361.

Kurtenbach, K., Hanincová, K., Tsao, J.I., Margos, G., Fish, D., Ogden, N.H., 2006. Fundamental processes in the evolutionary ecology of Lyme borreliosis. Nat. Rev. Microbiol. 4, 660–669.

Margos, G., Vollmer, S.A., Ogden, N.H., Fish, D., 2011. Population genetics, taxonomy, phylogeny and evolution of *Borrelia burgdorferi* sensu lato. Infect. Genet. Evol. 11, 1545–1563.

Margos, G., Piesman, J., Lane, R.S., Ogden, N.H., Sing, A., Straubinger, R.K., Fingerle, V., 2014. *Borrelia kurtenbachii* sp. nov., a widely distributed member of the *Borrelia burgdorferi* sensu lato species complex in North America. Int. J. Syst. Evol. Microbiol. 64, 128–130.

Mattar, S., Lopez Valencia, G., 1998. Searching for Lyme disease in Colombia: a preliminary study on the vector. J. Med. Entomol. 35, 324–326.

Narosky, T., Yzurieta, D., 2010. Aves de Argentina y Uruguay. Vázquez Mazzini, Buenos Aires.

Nava, S., Barbieri, A.M., Maya, L., Colina, R., Mangold, A.J., Labruna, M.B., Venzal, J.M., 2014. *Borrelia* infection in *Ixodes pararicinus* ticks (Acari: Ixodidae) from northwestern Argentina. Acta Trop. 139, 1–4.

Nava, S., Venzal, J.M., González-Acuña, D., Martins, T.F., Guglielmone, A.A., 2017. Ticks of the Southern Cone of America: Diagnosis, Distribution and Hosts with Taxonomy, Ecology and Sanitary Importance. Elsevier, Academic Press, London.

Norte, A.C., Lopes de Carvalho, I., Núncio, M.S., Ramos, J.A., Gern, L., 2013. Blackbirds *Turdus merula* as competent reservoirs for *Borrelia turdi* and *Borrelia valaisiana* in Portugal: evidence from a xenodiagnostic experiment. Environ. Microbiol. Rep. 5, 604–607.

Rollend, L., Fish, D., Childs, J.E., 2013. Transovarial transmission of *Borrelia* spirochetes by *Ixodes scapularis*: a summary of the literature and recent observations. Ticks Tick. Dis. 4, 46–51.

Rudenko, N., Golovchenko, M., Belfiore, N.M., Grubhoffer, L., Oliver Jr., J.H., 2014. Divergence of *Borrelia burgdorferi* sensu lato spirochetes could be driven by the host: diversity of *Borrelia* strains isolated from ticks feeding on a single bird. Parasit. Vectors 7, 4.

Saracho Bottero, M.N., Sebastian, P.S., Carvalho, L.A., Claps, L.G., Mangold, A.J., Venzal, J.M., Nava, S., 2017. Presence of *Borrelia* in different populations of *Ixodes pararicinus* from northwestern Argentina. Ticks Tick. Dis. 8, 488–493.

Saracho-Bottero, M.N., Tarragona, E.L., Sebastian, P.S., Venzal, J.M., Mangold, A.J., Guglielmone, A.A., Nava, S., 2018. Ticks infesting cattle and humans in the Yungas Biogeographic Province of Argentina, with notes on the presence of tick-borne bacteria. Exp. Appl. Acarol. 74, 107–116.

Scott, J.D., Fernando, K., Banerjee, S.N., Durden, L.A., Byrne, S.K., Banerjee, M., Mann, R.B., Morshed, M.G., 2001. Birds disperse ixodid (Acari: Ixodidae) and *Borrelia burgdorferi* infected ticks in Canada. J. Med. Entomol. 38, 493–500.

Scott, J.D., Anderson, J.F., Durden, L.A., 2012. Widespread dispersal of Borrelia burgdorferi-infected ticks collected from songbirds across Canada. J. Parasitol. 98, 49–59.

Sebastian, P.S., Bottero, M.N., Carvalho, L., Mackenstedt, U., Lareschi, M., Venzal, J.M., Nava, S., 2016. *Borrelia burgdorferi* sensu lato in *Ixodes* cf. *neuquenensis* and *Ixodes sigelos* ticks from the Patagonian region of Argentina. Acta Trop. 162, 218–221.

Stanek, G., Wormser, G.P., Gray, J., Strle, F., 2012. Lyme borreliosis. Lancet 379, 461–473.

Sun, Y., Xu, R., 2003. Ability of *Ixodes persulcatus*, *Haemaphysalis concinna* and *Dermacentor silvarum* ticks to acquire and transstadially transmit *Borrelia garinii*. Exp. Appl. Acarol. 31, 151–160.

Taragel'ová, V., Koci, J., Hanincová, K., Kurtenbach, K., Derdáková, M., Ogden, N.H., Literák, I., Kocianová, E., Labuda, M., 2008. Blackbirds and song thrushes constitute a key reservoir of *Borrelia garinii*, the causative agent of borreliosis in Central Europe. Appl. Environ. Microbiol. 74, 1289–1293.

Venzal, J.M., Estrada-Peña, A., Barros-Battesti, D.M., Onofrio, V.C., Beldoménico, P.M., 2005. *Ixodes* (*Ixodes*) *pararicinus* Keirans & Clifford, 1985 (Acari: Ixodidae): description of the immature stages, distribution, hosts and medical/veterinary importance. Syst. Parasitol. 60, 225–234.