

Short Communication

Detection of *Amblyomma mixtum* (Acari: Ixodidae) in Germany on a Human Traveler Returning From Cuba

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Abstract

The importation of exotic ticks to Germany on infested humans is rarely reported. Here we describe the detection of an *Amblyomma mixtum* nymph harboring *Rickettsia amblyommatis* by a tourist returning from a holiday trip to Cuba. Tick infestation may be neglected by tourists. Therefore, careful anamnesis and diagnosis should be carried out when tourists return from tropical and subtropical countries suffering from nonspecific symptoms, such as fever and malaise.

Key words: *Amblyomma mixtum*, *Rickettsia* spp., Cuba, Germany

In the New World, the *Amblyomma cajennense* complex comprises *A. cajennense* sensu stricto (Fabricius, 1787); *Amblyomma interandinum* Beati, Nava and Cáceres, 2014; *Amblyomma mixtum* Koch, 1844; *Amblyomma patinoi* Labruna, Nava and Beati, 2014; *Amblyomma sculptum* Berlese, 1888; and *Amblyomma tonelliae* Nava, Beati and Labruna, 2014 (Beati et al. 2013, Nava et al. 2014). These species are among the most important human-biting ticks in South America (Nava et al. 2017, Guglielmo and Robbins 2018). The *A. cajennense* complex includes potential vectors of *Rickettsia rickettsii* Ricketts, 1907, the agent of Rocky Mountain spotted fever (Krawczak et al. 2014, Labruna et al. 2014, Faccini-Martinez et al. 2015, Tarragona et al. 2016). The members of this complex are very similar morphologically; thus, a combination of morphological, molecular, and distributional data may be necessary to ensure correct species determinations (Nava et al. 2014). Here, we report the detection of one member of the *A. cajennense* complex—*A. mixtum*—in Germany while attached to a human traveler and its infection with *Rickettsia*.

Material and Methods

On 5 March 2019, an adult male resident of Kassel (State of Hesse, Germany) returned from a holiday trip to Cuba, having visited Playa Jibacoa and Havana, where he is presumed to have been parasitized by a nymphal tick. The day after his return, he recognized the tick on his shoulder, removed it, and sent it to the Museum of Natural History at Kassel, where it was forwarded to the Bundeswehr

Institute of Microbiology for identification and screening for rickettsiae in early May. The tick was identified on the basis of morphological descriptions of nymphs of the *A. cajennense* complex provided in Keirans and Durden (1998), Martins et al. (2016), and Nava et al. (2017). (The nymph described as *A. cajennense* in Keirans and Durden (1998) corresponds to *A. mixtum*, according to the known geographical distribution of this species [see Beati et al. 2013, Nava et al. 2014].) Additionally, the 16S rDNA sequence was amplified according to Mangold et al. (1998). DNA analysis using pan-*Rickettsia* real-time PCR to amplify part of the *gltA* gene (Wölfel et al. 2008), followed by a 23S-5S intergenic spacer region PCR (Chitimia-Dobler et al. 2018), served to identify the *Rickettsia* species, and *ompA* PCR (Fournier et al. 1998) and *ompB* PCR (Roux and Raoult 2000) provided further molecular characterization. Obtained sequences were edited using BioEdit Sequence Alignment Editor (Hall 1999) prior and then compared with those available in GenBank using BLAST (<http://www.ncbi.nlm.nih.gov>).

Results and Discussion

The tick was morphologically determined to be an *A. mixtum* nymph (Fig. 1a and b), and the 16S rDNA sequence subsequently obtained from this specimen (GenBank MN106364) showed high similarity (98–100% identity) to sequences available in GenBank from *A. mixtum* ticks collected at different localities (MG930061; MG930058; KM519935). *Amblyomma mixtum* is currently known to have a range extending from Texas to Colombia and Ecuador



Fig. 1. *Amblyomma mixtum* nymph: (a) dorsal—scutum and capitulum; (b) ventral—coxae.

(Estrada-Peña et al. 2014, Nava et al. 2014, Rivera-Páez et al. 2016). This tick has been shown to harbor *Rickettsia* spp. (Noda et al. 2016a).

The *A. mixtum* nymph tested positive for *Rickettsia* sp. by panRICK PCR. Furthermore, 23S-5S intergenic spacer region (GenBank MN447535), *ompA* (GenBank MN447533) and *ompB* (GenBank MN447534), sequences of the *Rickettsia* detected in the nymph were highly similar (98.9 to 100%) to sequences of *Rickettsia amblyommatis* available in GenBank (23S-5S intergenic spacer region: CP015012, KJ796425; *ompA*: CP012420; *ompB*: CP015012). *Rickettsia amblyommatis* (also referred to as *Rickettsia amblyommii* and ‘*Candidatus Rickettsia amblyommii*’) is the most commonly detected *Rickettsia* in Central America and has been reported in 10 species of ticks and one flea species in six of the seven countries in that region (Bermúdez and Troyo 2018, Noda et al. 2016a). Commonly more than 25% of *A. cajennense* populations are infected with *R. amblyommatis*. (Bermúdez et al. 2009). The medical importance and pathogenicity for humans of *R. amblyommatis* have not yet been fully elucidated. Moreover, some cases of Rocky Mountain spotted fever (RMSF) diagnosed in the United States may be due to *R. amblyommatis* rather than *R. rickettsii* (Apperson et al. 2008). It is also possible that previous infection with *R. amblyommatis* (*R. amblyommii*) may prevent severe disease during subsequent infection by the more highly pathogenic *R. rickettsii* (Blanton et al. 2014, Rivas et al. 2015).

To the best of our knowledge, this is the first record of *A. mixtum* having been detected in Germany; however, the unintentional importation of an exotic tick was only detected because of the individual’s background in science, causing him to be aware that such events occasionally occur and promptly delivered the specimen to the Kassel museum. Some tick species are regularly introduced from other countries or continents, on avian hosts (Petney et al. 2012, Chitima-Dobler et al. 2019) or domestic animals traveling with tourists (Petney et al. 2012). *Amblyomma mixtum* is the only member of the *A. cajennense* complex known from Cuba (Estrada-Peña et al. 2014, Nava et al. 2014). Physicians should be aware of the potential for exotic diseases in patients who report fever and other nonspecific symptoms, especially if they have recently returned from tropical or subtropical countries.

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Conflict of Interest

The authors declare no competing personal or financial interests. M.B. had consultancy agreements with Pfizer and GlaxoSmithKline.

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