

Molecular identification of *Babesia* sp. in questing *Ixodes ricinus* ticks in the Warmia-Masuria Province in north-eastern Poland

Hanna Szymańska, Katarzyna Kubiak, Ewa Dzika

University of Warmia and Mazury in Olsztyn, Faculty of Health Sciences, Department of Medical Biology, Żołnierska 14C, 10 - 561 Olsztyn

The *Ixodes ricinus* tick is the most widespread in Europe as an important vector of pathogens of medical and veterinary importance. In Poland, the most common human pathogens transmitted by *I. ricinus*, in addtition to TBEV and *Borrelia spirochetes*, are protozoa of the genus *Babesia* sp. They are etiological factor of babesiosis – dangerous, invasive zoonosis, leading in severe cases to anemia, hemoglobinuria and possible organ failure. In Poland, there were noted symptomatic and asymptomatic cases of *B. microti*-associated babesiosis, imported from the tropical countries and infections with *B. venatorum*, coexisting in patients with other tick-borne diseases. However, according to the National Institute of Public Health, cases of asymptomatic babesiosis in Poland are more frequent than expected, as indicated by the level of IgG antibodies against *B. microti* antigens, which were detected in 4.4–5% of foresters and 2.6% of farmers in the eastern parts of country. In addition, the incidence of *Babesia* sp. in *I. ricinus* ticks in our country, depending on the geographical region is varied, and in the eastern region it can reach up to 5%.

The aim of the study was to determine the prevalence of *Babesia* sp. in questing *I. ricinus* ticks in the Warmia-Masuria Province.

A total of 1070 *I. ricinus* ticks (186 adults and 884 nymphs) collected by flagging method from April to May 2016 in 15 sites located in the Warmia-Masuria Province were examined. Genomic DNA from ticks was isolated with Sherlock AX kit (A&A Biotechnology, Poland) according to the manufacturer's protocol, from individual adults and from pools of 5 nymphs. Molecular identification of *Babesia* sp. in ticks was determined on the basis of nested polymerase chain reaction (nested-PCR) using 18S rRNA gene-specific primers: outer – CRYPTO F/CRYPTO R and inner – Bab GR2/Bab GF2 (Bonnet *et al.,* 2007) and confirmed by sequencing reaction.

Molecular identification of *Babesia* sp. in ticks was carried out in 361 samples (99 males, 87 females and 175 pools of nymphs). DNA of *Babesia* sp. was identified in 4.2% (15/361). It was demonstrated that the dominant pathogen species were *B. microti* (40.0%, 6/15) and *B. venatorum* (40.0%, 6/15). Whereas, 2 out of 15 (13.3%) isolates were similar to the sequences of *B. divergens* and *B. capreoli*, with identities of 99.5% and 94.5%, respectively. The last one *Babesia*-positive tick sample was assigned only to the genus *Babesia* sp. (1/15, 6.7%). No co-infections were found.

Our study indicates the presence of genetic material of *Babesia* sp. in *I. ricinus* ticks in the Warmia-Masuria Province and suggests their participation in the transmission of these pathogens to humans and animals.