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PCR-based screening of pathogenic genotypes of Borrelia burgdorferi sensu lato complex in Ixodes ticks in different regions of Ukraine

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Introduction. Lyme borreliosis (LB) is a common tick-borne disease caused by spirochetes grouped in the *Borrelia burgdorferi sensu lato* complex (*B. burgdorferi s.l.*), which includes about 21 genotypes. The most common genotypes of *B. burgdorferi s.l.* causing LB are *B. burgdorferi sensu stricto*, *B. garinii* and *B. afzelii*, but the genotypic composition may vary depending on the geographical location. In the United States, the annual incidence of LB is 10 or more cases per 100,000 population. In Europe, the average rate is 22 cases per 100,000 population, although the data varies across European countries. In Ukraine, in 2018-2022, the incidence of LB was 9.03 cases per 100,000 population. Passive surveillance suggests that the incidence of LB in Ukraine is significantly underestimated. Increasing incidence, insufficient reporting and lack of data on *Borelia* genotypes in Ixodes ticks in Ukraine make it difficult to control the disease.
Previous studies have shown that the mean combined prevalence of *B. burgdorferi s.l.* in Ixodes ticks from 5 cities in Ukraine was 26%. Here, we set out to determine the prevalence of *B. burgdorferi* genotypes in Ixodes ticks in different regions of Ukraine.
Methods. Ticks were collected by drag-flag method and identified microscopically. Ticks were homoginized in 200 ?L of 0.9% sodium chloride solution, spiked with 25 ?L of proteinase K incubated for 1.5 hours at 56ºC. PCR reactions were performed in a reaction mixture containing One Taq Quick-Load 2X Master Mix with standard buffer (New England Biolabs, USA), primer sets developed by Marconi et al., 1993, deionized water, and tick DNA. Amplification products were detected by electrophoresis in a 2% agarose gel containing ethidium bromide. Primary pools of isolated tick DNA were analyzed: 7 pools of 10 *I. ricinus* ticks from Kyiv region and 7 pools of 10 *D. reticulatus* ticks from Cherkasy region.
Results. Initial screening showed that 7 primary pools of *I. ricinus* ticks from Kyiv region were PCR-positive for the *B. burgdorferi s.l.* complex and *B. afzelii* genotype, and 1 of 7 was positive for the *B. burgdorferi sensu stricto* genotype. In 3 of 7 primary pools of *D. reticulatus* ticks from Cherkasy region, DNA of the *B. burgdorferi s.l.* complex and *B. afzelii* DNA were detected in 2 pools and *B. burgdorferi sensu stricto* - in 1 pool.
Conclusions. The results of the screening indicate that the *B. burgdorferi s.l.* complex was present in *I. ricinus* ticks from Kyiv region and in *D. reticulatus* ticks from Cherkasy region. Pathogenic genotypes of *B. afzelii* and *B. burgdorferi sensu stricto* were also identified. PCR screening of individual ticks for LB genotypes is ongoing and will be presented together with the initial data on pooled ticks.
Detection of *B. burgdorferi s.l.* in ticks and knowledge of the distribution of specific genotypes will help in the diagnosis and treatment of LB. Overall, this study emphasizes the importance of tick surveillance and genotypic screening for LB control.