## Theses of the Doctoral (PhD) Dissertation

# INVESTIGATIONS OF THE ROLE OF BIRDS IN THE EPIDEMIOLOGY OF TICK-BORNE AND VECTOR-BORNE PATHOGENS

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#### Background and objectives of the doctoral thesis

The role of birds in the transportation of arthropods with vector potential has long been known. This aspect of avian ecology—as hosts and dispersal agents of parasites—is expected to gain increasing importance due to ecological changes, particularly those associated with ongoing climate change. Among the arthropods transported by birds, hard ticks (Acari: Ixodidae) are of particular concern. Notably, species of the genus *Hyalomma*, such as *Hyalomma rufipes* and *Hyalomma marginatum*, are principal vectors of the Crimean-Congo haemorrhagic fever virus. In addition, birds contribute significantly to the spread of local tick species, including *Ixodes ricinus*, *Ixodes frontalis*, and *Haemaphysalis concinna*.

Louse flies (Diptera: Hippoboscidae) are common hematophagous ectoparasites of birds. However, their potential role in pathogen transmission remains largely unexplored and poorly understood.

The aim of this doctoral thesis is to map the spatial and temporal distribution of hard ticks and louse flies arriving in Hungary via migratory birds, and to identify non-native species among them, primarily those originating from Africa. Furthermore, we aimed to detect piroplasms found in *Haemaphysalis concinna*, a tick species frequently carried by birds, and to assess the role of louse flies (Hippoboscidae: Ornithomyinae) in the transmission of certain bacteria and protozoan parasites of veterinary importance.

### 2. Summary

The role of birds in the dispersal of ticks, other ectoparasitic vectors, and vector-borne pathogens has been the subject to extensive research. In Hungary, several studies have been carried out over the past decades to explore these dynamics. This thesis aims to provide a better understanding of the role birds play in the epidemiology of pathogens transmitted by ticks and other blood-feeding parasites. To gain a comprehensive understanding of this process, it is equally important to explore both the interactions between birds and blood-feeding vectors, and the potential vector roles of these arthropods. This thesis is based on five peer-reviewed articles published in scientific journals. To provide a more comprehensive overview, the thesis also incorporates findings from an unpublished study focusing on selected pathogens identified in avian louse flies.

A comprehensive review was conducted to assess the composition and size of the tick fauna infesting European birds. Over 200 published studies were analysed for this purpose. Based on our findings, 37 tick species have been identified so far on European birds, belonging to 16 different orders. Five widely distributed tick species—*Ixodes arboricola, Ixodes frontalis, Ixodes ricinus, Haemaphysalis concinna*, and *Hyalomma marginatum*—were further analysed for their host associations, revealing clear preferences for certain bird species based on ground-feeding behaviour and habitat use.

Following this, independent research took place examining the temporal relationship between birds and ticks. Ticks, collected from birds captured for ringing purposes at the Ócsa bird observatory between March 2014 and November 2022 were analysed. A total of 5,833 ticks from 10 species were collected from 2,395 infested birds. The dominant tick species were *I. ricinus* and *H. concinna*. *I. ricinus* was more common on ground-feeding, resident, and short-distance migratory birds in forest habitats, while *H. concinna* was found more frequently on long-distance migrants in reed habitats. Seasonal patterns showed that *I. ricinus* nymphs peaked in spring, larvae in autumn, while both larval and nymph stages of *H. concinna* peaked in summer. This study represents the first long-term bird-tick investigation in Central Europe, highlighting how bird ecology and tick life cycles jointly influence the spread of ticks. Notably, the Savi's Warbler (*Locustella luscinioides*) was identified as a key host for *H. concinna* in the region.

Parallel to the temporal distribution, the spatial distribution of ticks feeding on birds in Hungary was also studied. Ticks were collected from 38 species of passerine birds at seven locations in Hungary, resulting in 956 ixodid ticks. The most common species were *I. ricinus*, *H. concinna* 

and *I. frontalis*. Interestingly, 12 *Hyalomma* ticks (11 engorged nymphs, and one non-engorged larva) were identified as *H. rufipes* based on three mitochondrial markers. This species was only found in the western and southeastern regions of Hungary. This study represents the first European observation of a reproducing *H. rufipes* population and offers a new ornithological explanation for its century-long presence in the Transdanubian region of the Carpathian Basin.

In the Carpathian Basin, *H. concinna*, one of the most common bird parasites in Hungary, occurs in unusually high numbers compared to other European countries. To gain a deeper understanding of the prevalence of piroplasms in these ticks and their potential role as vectors, we examined ticks collected from vegetation. Our study identified 11 distinct *Babesia* genotypes and *Theileria capreoli* within the *H. concinna* population. The results revealed that the peak monthly prevalence of *Babesia* and *Theileria* spp. in questing *H. concinna* ticks may not align with the peak abundance of the ticks themselves. This discrepancy may be influenced by factors such as changes in metabolism, behavior, and the survival rate of infected ticks. Further research is needed to clarify these observations.

In addition to ticks, our research also examined avian louse flies (Hippoboscidae: Ornithomyinae). We confirmed the presence of nine bird-specific hippoboscid species in Hungary, including *Ornithoctona laticornis*, an African tick species, which we identified in Europe for the first time. In several louse fly species, we discovered multiple *Trypanosoma* genotypes, which had not been previously reported in avian louse flies. Furthermore, we identified a strain of *Anaplasma phagocytophilum* in *Ornithomya avicularia*, a species that, according to current knowledge, likely only infects birds. Additionally, the emerging pathogen *Haematospirillum jordaniae* was detected in three specimens of *Ornithomya fringillina*, marking the first report of this pathogen in hippoboscid flies.

#### 3. New scientific results

- 1. The role of Savi's warbler as the most prominent avian host of *Haemaphysalis concinna* in Hungary was revealed for the first time.
- 2. First detection of a reproducing *Hyalomma rufipes* population in Europe.
- 3. First demonstration of the uneven temporal distribution of piroplasms in an urban *Haemaphysalis concinna* population.
- 4. First detection of the African louse fly *Ornithoctona laticornis* in Europe.
- 5. First time detection of *Anaplasma phagocytophilum* in a member of Ornithomyinae subfamily (*Ornithomya avicularia*)
- 6. First time detection of *Trypanosoma* sp., lineage B14 in Hungary and Malta, as well as in *Crataerina hirundinis*.
- 7. First time detection of *Trypanosoma culicavium*, *Trypanosoma* sp. (from the "theileri" group), and *Trypanosoma* sp. (from the "bennetti" group) from members of the Ornithomyinae subfamily
- 8. First time detection of *Haematospirillum jordaniae* in hippoboscid flies.

#### 4. Publications

- <u>Keve G</u>, Sándor AD, Hornok S, 2022. Hard ticks (Acari: Ixodidae) associated with birds in Europe: Review of literature data. FRONTIERS IN VETERINARY SCIENCE, 9, p.928756.
- 2. **Keve G**, Csörgő T, Kováts D, Hornok S, 2024. Long term evaluation of factors influencing the association of ixodid ticks with birds in Central Europe, Hungary. SCIENTIFIC REPORTS, 14(1), p.4958.
- 3. **Keve G**, Csörgő T, Benke A, Huber A, Mórocz A, Németh Á, Kalocsa B, Tamás EA, Gyurácz J, Kiss, O, Kováts D, Sándor AD, Karcza ZS, 2023. Ornithological and molecular evidence of a reproducing Hyalomma rufipes population under continental climate in Europe. FRONTIERS IN VETERINARY SCIENCE, 10, p.1147186.
- 4. **Keve G**, Reynolds C, Takács N, Hornok S, 2025. Uneven temporal distribution of piroplasms (Piroplasmida: Babesiidae, Theileriidae) in Haemaphysalis concinna in an urban biotope of the Western Palearctic focus region of this tick species. TICKS AND TICK-BORNE DISEASES, 16(2), p.102458.
- Keve G, Csörgő T, Kováts D, Benke A, Bende AT, Ágoston H, Mórocz A, Németh, Á, Tamás EA, Huber A, Gyurácz J, Keve G, Kontschán J, Németh A, Hornok S, 2024. Contributions to our knowledge on avian louse flies (Hippoboscidae: Ornithomyinae) with the first European record of the African species Ornithoctona laticornis. PARASITES & VECTORS, 17(1), p.237.