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SHORT COMMUNICATION

Infestation of urban populations of the Northern white-breasted hedgehog, *Erinaceus roumanicus*, by *Ixodes* spp. ticks in Poland

S. DZIEMIAN¹, J. MICHALIK², B. PIŁACIŃSKA¹, S. BIALIK², B. SIKORA² and R. ZWOLAK¹

¹Department of Systematic Zoology, Faculty of Biology, Adam Mickiewicz University, Poznań, Poland and ²Department of Animal Morphology, Faculty of Biology, Adam Mickiewicz University, Poznań, Poland

Abstract. Infestation by the nest-dwelling *Ixodes hexagonus* Leach and the exophilic *Ixodes ricinus* (Linnaeus) (Ixodida: Ixodidae) on the Northern white-breasted hedgehog, *Erinaceus roumanicus* (Erinaceomorpha: Erinaceidae), was investigated during a 4-year study in residential areas of the city of Poznań, west-central Poland. Of 341 hedgehogs, 303 (88.9%) hosted 10061 *Ixodes* spp. ticks encompassing all parasitic life stages (larvae, nymphs, females). *Ixodes hexagonus* accounted for 73% and *I. ricinus* for 27% of the collected ticks. Male hedgehogs carried significantly higher tick burdens than females. Analyses of seasonal prevalence and abundance of *I. hexagonus* revealed relatively stable levels of infestation of all parasitic stages, with a modest summer peak in tick abundance noted only on male hosts. By contrast, *I. ricinus* females and nymphs peaked in spring and declined steadily thereafter in summer and autumn, whereas the less abundant larvae peaked in summer. This is the first longterm study to evaluate the seasonal dynamics of both tick species on populations of wild hedgehogs inhabiting urban residential areas.

Key words. Erinaceus, Ixodes hexagonus, Ixodes ricinus, hedgehogs, ticks.

Infestations of urban wildlife with ectoparasites are of particular medical and veterinary interest because they influence the likelihood that humans, livestock and pet animals will be exposed to zoonotic agents transmitted by ticks or fleas. Hedgehogs, which are highly adaptable denizens of human-impacted landscapes, may establish and maintain local tick populations. Given the anthropophilic behaviour of these mammals, they can increase the risk for tick exposure directly to humans or to companion animals, which frequently bring ticks into human domiciles (Uspensky, 2014).

The Northern white-breasted hedgehog, *Erinaceus roumanicus*, inhabits Central and Eastern Europe. It has been reported in Poland, Slovakia, the Czech Republic, Austria, Hungary, the Balkan Peninsula, Ukraine and the central and southern regions of European Russia and the northern Caucasus. This hedgehog species is known as an important host of two ixodid tick species: the nidicolous hedgehog tick, *Ixodes hexagonus*, and the sheep tick, Ixodes ricinus, the most prevalent tick in Europe. These tick species may reach very high loads on hedgehogs as a result of the ground-foraging behaviour and ineffective grooming of these spiny mammals (Földvári et al., 2011; Pfäffle et al., 2011). Accordingly, the Northern white-breasted hedgehog may be an important reservoir host of zoonotic agents. It has been found to maintain tick-borne encephalitis virus (TBEV) during hibernation (Kožuch et al., 1967), and it has also been implicated as a potential reservoir of Lyme disease spirochetes (Skuballa et al., 2012). Furthermore, Anaplasma phagocytophilum has been recently detected in I. ricinus ticks removed from Northern white-breasted hedgehogs in Romania (Dumitrache et al., 2013). Nevertheless, despite the widespread distribution of E. roumanicus, data on its role as a host for ectoparasites in urban areas are remarkably scarce, particularly in comparison with information available for the much better-known European hedgehog, Erinaceus europaeus. Exceptions include

Correspondence: Sylwia Dziemian, Department of Systematic Zoology, Faculty of Biology, Adam Mickiewicz University, Umultowska 89, 61-614 Poznan, Poland. Tel.: + 48 61 829 5800; Fax: + 48 61 829 5636; E-mail: sylwia.dziemian@gmail.com a preliminary report (Dziemian *et al.*, 2010) and a recent Hungarian study conducted in a city park in Budapest by Földvári *et al.* (2011). Here, we report the results of a 4-year study of tick parasite loads of *E. roumanicus* in residential areas of the city of Poznań, in west-central Poland.

Poznań is the fifth largest city in Poland; it has about 550 000 inhabitants and an area of 262 km². Hedgehogs were captured alive in four different residential areas with apartment blocks or detached houses, and collected from drainage ditches bordering the route of the Poznań Fast Tramline (PST). Collectively, the study sites covered about 700 ha. Animals were spotted nocturnally with flashlights and captured by hand from April to September in the years 2009–2012. Each session started at sunset and lasted 3–4 h. Sessions were conducted once or twice per month. Drainage ditches were surveyed for hedgehogs during the morning, two or three times per week during each collection month.

Hedgehogs were transported to the laboratory and held overnight in individual plastic boxes $(37 \times 47 \times 26 \text{ cm})$. They were fed commercial cat food. Each animal was weighed and sexed. Hedgehogs were examined for the presence of ticks from 15 min to 2 h depending upon the particular animal's parasite load. After this examination, each individual was marked with colour-coded and numbered plastic tubes that were glued to its spines and returned to its site of capture; individuals collected on the PST route were released at least 100 m away from the ditches. The procedures for the catching and handling of hedgehogs were approved by the Polish Ministry of Environment (permission no. DOPozgiz-4200/IV-36/2359/10/km). Attached ticks were removed with tweezers and stored in 75% alcohol. Additionally, each box used by a captured animal was checked for detached ticks. Ticks were identified according to Siuda (1993).

For each tick species, we estimated parasite abundance (mean number of parasites per host) and prevalence. Additionally, we estimated the proportion of hedgehogs that were co-infested with I. hexagonus and I. ricinus. We used generalized linear mixed models (GLMMs) in R (lme4, 2012; R Development Core Team, R Foundation for Statistical Computing, Vienna, Austria), with either Poisson family error terms and a log link function (analysis of parasite abundance and analysis of co-infestation) or binomial family and logit link (parasite prevalence). Predictor variables included fixed effects of season [grouped as spring (April, May), summer (June-August) and autumn (September-November)], sex (male vs. female), and, in the case of abundance and prevalence analyses, tick stage (female, nymph or larva). The initial models included all possible interactions among the fixed effects. The final structure of the models was determined through stepwise elimination of non-significant interactions using likelihood ratio tests.

Random effects included fully or partly correlated effects of tick stage over individual hedgehog, year and study site, and, in abundance and prevalence analyses, a unique identifier for each observation [i.e. for each tick stage within each hedgehog (this method is equivalent to using overdispersed Poisson and binomial models)]. The random effects were evaluated with Akaike's information criterion (AIC) scores. We retained only those random effects that improved the fit of the models.

In total, 341 *E. roumanicus* hedgehogs (170 males and 171 females), including 63 recaptures, were examined for ticks.

A total of 10061 *Ixodes* spp. ticks were collected from 303 (88.9%) infested animals. *Ixodes hexagonus* accounted for 73% and *I. ricinus* for 27% of the collected ticks. No other tick species was found during the study.

Larvae and nymphs prevailed over females and constituted 45% and 41% of total tick numbers, respectively. Hedgehogs were often parasitized concurrently by at least two of the three different tick developmental stages. This occurred in 68% of hedgehogs infested with *I. hexagonus* and 60% of hosts infested with *I. ricinus*. As none of 276 adult males fed actively on the hosts and were found to be *in copula*, we restricted our infestation analysis solely to the parasitic life stages.

Ixodes hexagonus nymphs tended to be more abundant than females and larvae (stage effect in Table 1, Fig. 1A). Infestation patterns of *I. hexagonus* showed relatively weak seasonality. All stages reached their highest abundance in summer, but this peak was apparent only in male hosts (marginally significant season effect and highly significant sex × season interaction in Table 1, Fig. 1A). Male hosts carried more ticks than female hosts in spring and summer, but the opposite trend was observed in autumn (sex and sex × season effects in Table 1, Fig. 1A). Patterns of abundance and prevalence of *I. hexagonus* were similar (Fig. 1C), but seasonal changes in overall prevalence were not statistically significant (Table 1).

With reference to *I. ricinus*, nymphs and females were more abundant than larvae (stage effect in Table 1, Fig. 1B) in contrast with *I. hexagonus*, abundances of *I. ricinus* demonstrated pronounced seasonality, with female and nymphal loads declining from spring to autumn, and abundances of larvae peaking in summer (season and season × stage effects in Table 1, Fig. 1B). In addition, male hosts carried more *I. ricinus* ticks than females (sex effect in Table 1), particularly in spring (season × sex effect in Table 1, Fig. 1B). The prevalence of *I. ricinus* mirrored its pattern of abundance (Table 1, Fig. 1D).

Co-infestations with both tick species were influenced by season ($\chi^2 = 20.06$, d.f. = 2, P < 0.0001), but not by host sex ($\chi^2 = 1.15$, d.f. = 1, P = 0.2833). The estimated proportion of co-infested hedgehogs was highest in spring [0.71, 95% confidence interval (CI) 0.57–0.82], intermediate in summer (0.58, 95% CI 0.44–0.70), and lowest in autumn (0.31, 95% CI 0.17–0.49).

Northern white-breasted hedgehogs may play an important role in dispersing and increasing abundances of pathogen-transmitting ticks in urban areas. Therefore, understanding the factors that drive patterns of tick infestation in these important hosts can help in assessing the risk for the transmission of tick-borne diseases in cities. We found that I. ricinus and I. hexagonus demonstrate very different infestation patterns on E. roumanicus inhabiting urban areas. These differences were most likely to reflect the distinct habitat preferences of these two tick species. The nidicolous I. hexagonus is a nest-dwelling species and therefore its off-host environment is limited exclusively to the resting or breeding sites of its host (Siuda, 1993). Hedgehogs build two types of nest: longterm nests used for prolonged periods of time (i.e. hibernacula in winter and reproductive nests in summer), and short-term resting or sleeping nests (Reeve, 1994). As both types of nest are inhabited by this tick, hedgehogs are exposed to repeated contact with all tick

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Table 1. Factors influencing the abundance and prevalence of <i>Ixodes hexagonus</i> and <i>Ixodes ricinus</i> ticks on Northern white-breasted hedgehogs
(Erinaceus roumanicus) studied in an urban environment in the city of Poznań, Poland.

Variable*	Abundance			Prevalence		
	χ^2 -value	d.f.	<i>P</i> -value	χ^2 -value	d.f.	P-value
Ixodes hexagonus						
Season	6.98	2	0.0305	3.75	2	0.1532
Stage	30.98	2	< 0.0001	22.33	2	< 0.0001
Sex	8.32	1	0.0039	7.87	1	0.0050
Season × sex	12.00	2	0.0025	12.02	2	0.0024
Ixodes ricinus						
Season	67.89	2	< 0.0001	33.63	2	< 0.0001
Stage	75.47	2	< 0.0001	48.50	2	< 0.0001
Sex	18.97	1	< 0.0001	9.63	1	0.0019
Season × stage	62.51	4	< 0.0001	6.03	2	0.0490
Season × sex	6.72	2	0.0348	60.25	4	< 0.0001

*Season is a categorical variable with three levels (spring, summer, autumn); stage denotes tick stage (larva, nymph, female); sex denotes the sex of the hedgehog. See text for further explanation.

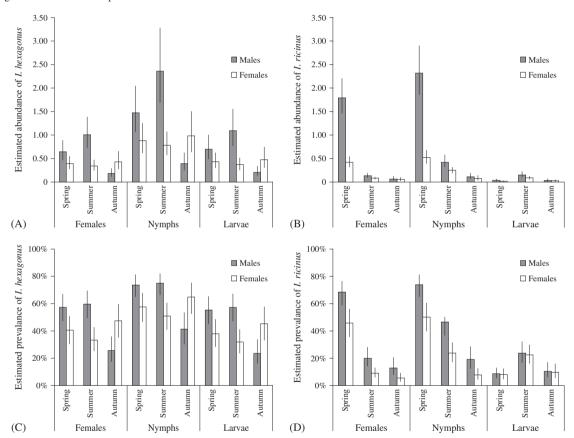


Fig. 1. Seasonal patterns of infestation of Northern white-breasted hedgehogs with three developmental tick stages: (A) abundance of *Ixodes hexagonus*, (B) abundance of *Ixodes ricinus*, (C) prevalence of *I. hexagonus*, and (D) prevalence of *I. ricinus*. Estimates are presented with standard errors.

life stages (Pfäffle *et al.*, 2011). Because burrows and nests represent enclosed microhabitats, they provide stable temperature and humidity, and thus *I. hexagonus* is less dependent on microclimatic conditions and therefore is active throughout the year. This tick has a narrower host range than *I. ricinus* and parasitizes only 15 mammalian species, especially red foxes and Mustelidae

(Siuda, 1993). In urban environments, this tick infests cats and dogs, and therefore hedgehogs can amplify *I. hexagonus* populations and enhance tick exposure to companion animals.

By striking contrast, the exophilic tick *I. ricinus* has extremely generalist feeding habits and an extraordinary suite of 300

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host species (Pfäffle et al., 2011). It requires habitats with >80% relative humidity for survival as its off-host stages quest in open areas exposed to highly variable environmental conditions. Mixed and deciduous stands carpeted with litter are optimal ecosystems for its maintenance (Siuda, 1993). In open urban-suburban areas with highly dispersed vegetation cover, the distribution of *I. ricinus* is patchy; off-host tick density is usually low and the tick is restricted spatially to private gardens, public parks and various forested green spaces (Michalik et al., 2003). Our study sites were located in residential areas with large apartment blocks, in which green spaces comprised only short lawns, small private gardens of semi-detached houses or small parks. Hence, in our opinion these habitat conditions were much more suitable for I. hexagonus than I. ricinus and accounted for the fact that the former tick species represented the majority of ticks collected from E. roumanicus in this study.

In the present study, both tick species showed differences in seasonal patterns of infestation on *E. roumanicus*. To our knowledge, this is the first longterm study to evaluate the seasonal dynamics of *I. hexagonus* and *I. ricinus* infestations in wild hedgehog populations inhabiting residential urban areas. In a previous 3-year investigation, both ticks were evaluated under semi-natural conditions in a captive *E. europaeus* population (Pfäffle *et al.*, 2011). As less is known of the seasonal activity of *I. hexagonus* under natural conditions in comparison with *I. ricinus*, our findings on the population dynamics of this tick on *E. roumanicus* expand our knowledge in relation to its occurrence in urban settings.

The overall seasonal activity pattern of *I. ricinus* revealed only one distinct peak in spring, after which tick numbers gradually decreased to reach their lowest values in autumn. This trend was especially apparent in nymphs and females. Only larvae revealed relatively comparable abundances in the spring and summer months. The lack of the second smaller autumn peak, which is commonly recorded in temperate climates for I. ricinus (Michalik et al., 2003), might be explained by unfavourable microclimatic conditions at our study sites. A comparable descending pattern in the prevalence of *I. ricinus*, with peak emergence in spring and a decrease in summer and autumn, was recorded in urban hedgehog populations in Switzerland (Egli, 2004). Only in the German study by Pfäffle et al. (2011), nymphs and adult I. ricinus showed a typical bimodal seasonal distribution (larvae showed a summer peak, which coincides with our results).

Analyses of the prevalence and abundance of the dominant *I. hexagonus* revealed weaker seasonal differences, with a summer peak occurring on male but not female hosts. Similarly, relatively little change in the overall population density of *I. hexagonus* was reported by Pfäffle *et al.* (2011). The trend towards smaller seasonal variations in the prevalence of this tick on hedgehogs, in comparison with *I. ricinus*, clearly confirms differences in the reproductive cycles of the two tick species. The fluctuating but relatively high occurrence of all stages of *I. hexagonus* on the host throughout the year seems to be facilitated by the stable microclimate of nests and shelters. Alternatively, because the host ranges of *I. ricinus* and *I. hexagonus* are very different, the dissimilarities in the seasonal dynamics of these ticks may be related to fluctuations in the abundances of non-hedgehog hosts.

In each season except for autumn, male hedgehogs carried higher numbers of I. hexagonus ticks than females. This difference may be attributed to the larger home ranges of E. roumanicus males and the fact that they may travel longer distances during the mating season (mostly in May and June), which can increase their tick-encounter rates in open habitats (Boyer et al., 2010). An alternative explanation is that male hedgehogs end hibernation earlier than females and therefore have a greater likelihood of making contact with host-seeking ticks in urban environments. Finally, this pattern could be caused by the association between testosterone and the immune system; sexually mature male vertebrates are often more susceptible to infection and carry higher parasite burdens (Zuk & McKean, 1996). If this explanation is correct, a post-mating season decline in testosterone levels [documented in the closely related E. europaeus (Fowler, 1988)] and associated changes in behaviour or in the hedgehog's immune system may contribute to the drop in the tick loads of male hedgehogs in autumn.

We found that the frequency of co-infestation with both tick species was strongly seasonal: in spring, the proportion of co-infested hedgehogs was over twice as high as in autumn. This pattern may influence the circulation of disease because the co-infestations could result in co-infections by pathogens vectored by *I. hexagonus* and *I. ricinus* and in the transmission of disease agents between the two species of ticks.

We conclude that wild populations of Northern white-breasted hedgehogs inhabiting residential areas in the city of Poznań are effective hosts for all parasitic stages of I. hexagonus and I. ricinus ticks. As E. roumanicus has been recognized recently as a probable reservoir for Lyme disease spirochetes, and A. phagocytophilum appears to infect this mammal species with a high prevalence (Földvári et al., 2014), not only do urban populations of this species influence the densities of both competent tick vectors, but they may establish and maintain local foci of zoonotic bacteria. Given that our hedgehogs were frequently co-infested by both sympatric tick species and different life stages, co-feeding transmission (Randolph et al., 1996) between infected nymphs or females and uninfected larvae infesting this host may enhance the transmission cycles of tick-borne pathogens in urban habitats. Exploitation of both endophilic and exophilic tick species may increase the circulation of shared pathogens (e.g. A. phagocytophilum) in particular biotopes. Moreover, although I. hexagonus rarely bites humans, it is feasible that this tick could serve as an efficient vector capable of transmitting infectious agents to companion animals [see Smith et al. (2011) for rates of infestation of pets]. Further studies monitoring populations of E. roumanicus hedgehogs in anthropogenic ecosystems are essential to elucidate their contributions to the enzootiology of tick-borne agents inducing zoonotic infections in humans and domestic animals.

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