

Tick distribution along animal tracks: implication for preventative medicine

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Abstract

Introduction. Tick abundance and the prevalence of the pathogens they carry have been increasing worldwide in the last decades, and is projected to increase even further. Despite the fact that problem is global, there still remain many gaps in the diagnosis and treatment of tick-borne diseases. The best protection from tick-borne pathogens, therefore, is prevention and avoidance of bites. Ticks mobility is limited so that their spatial distribution is strongly correlated with the presence of, especially with large mammals. In this study, the hypothesis was tested that tick abundance is higher on animal tracks in the forests than in adjacent habitats. This is an important issue because there are still several human habits and practices that can decrease the zoonoses risk. For example, during recreation in forest, people should always walk on the paths (including narrow animal's tracks) instead of wading through bushes.

Materials and method. Flagging of animal trails and near control transects were performed simultaneously. Next, collected ticks were counted, sexed and aged.

Results. The abundance of ticks was almost 5-fold (*Ixodes ricinus*) and 3-fold (*Dermacentor* spp.) higher on animal trails than on adjacent control transects.

Conclusions. The results obtained support the hypothesis that ticks are more abundant on pathways than in adjacent habitats. Most likely, the pattern emerges because large mammals, like deer, which are the most important ticks hosts, use forest paths to move across the landscape and frequently move along the same routes. This research sends an important public message that these forest trails are hotspots of disease risk and should be avoided.

Key words

One Health, tick-borne diseases, spatial distribution, preventive medicine, *Ixodes*, human behaviour

INTRODUCTION

Ticks as pathogen vectors are major ectoparasites of humans [1] and have a crucial influence on the epidemiology of zoonoses, such as Lyme disease [2, 3] and ticks-borne encephalitis [4], babesiosis [5] as well as numerous others less frequent diseases [6, 7]. Moreover, both tick abundance and the prevalence of pathogens the ticks carry has increased worldwide in the last decades, and is projected to increase even further [8–11]. Therefore, numerous studies have explored how their abundance and associated disease risk changes in time and over large spatial scales [12–17]. However, the distribution of ticks at the microhabitat scale, in conjunction with co-use of these habitats by humans, will define the disease risk observed at the landscape scale.

The mobility of ticks is limited as nymphs are able to move only ca. 1 m, while adults can move ca. 3.5 m [18, 19].

Therefore, their spatial distribution is strongly correlated with the presence of hosts, especially large mammals [20, 21]. This non-random distribution is used in disease risk management to reduce ticks and humans co-occurrence, e.g. by creating 'ticks free' areas around houses and schools [22]. That concept is intensively studied as a vital component of the One Health conception [23, 24], i.e. sustainable management practices to protect humans from zoonoses. Nonetheless, there are still several human habits concerning practices to decrease zoonoses risk. For example, during recreation in forests, people should always keep to the paths (including narrow animal's tracks) instead of wading through the bushes. However, since the presence of ticks is strongly shaped by their host activity, not all forest pathways may be safe, and some, in fact, may be hotspots of tick abundance at the microhabitat scale. Thus, this study tests the hypothesis that tick abundance is higher on animal tracks in the forests than in adjacent habitats.

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MATERIALS AND METHOD

Sampling was performed during the active season of ticks, March – December, in 2014–2016, at 2 geographical distant areas. The first location was situated in the High Tatras in the northern part of Slovakia (N 49.161823594, E 20.269952694), with an average altitude of 900 m a.s.l. The second location was situated in southern part of Slovakia, in Slovak Karst (N 48.589756604, E 20.697413282), with average altitude of 200 m a.s.l. In both locations, 39 sites (in total) were selected for tick sampling which had evidence of the presence of an animal trail at least 100 meters long. Each trail was paired with a control transect that was parallel to the animal trail, and spaced approximately 5 m away. It was ensured that the control trails did not cross other animal trails, residential signs of animals, piles of droppings or noticeable resting areas of animals, and were located in the same habitats as animal trails. Tick populations were sampled by flagging questing ticks. Each flag consisted of a 1 m² piece of white cotton flannel attached to a 1.2 m wooden dowel [25]. The flag pulled along the canopy cover, grassy vegetation, shrubs and leaf litter. Every 2 m the flag was examined for the presence of ticks, and collected if present. The flagging of animal trails and control transects were carried out simultaneously, but no samples were collected during rain or on wet vegetation. Fields workers were regularly swapped between treatments to avoid potential bias. All ticks were released back at the place of capture. Sampled ticks were identified to species level (*Ixodes ricinus*) or genus level (*Dermacentor* spp.), gender and developmental stage were determined.

Tests were carried out for the differences in abundance of ticks using zero-inflated negative binomial (ZINB), logit link, mixture models implemented via 'pscl' package [26, 27] in the R software [28]. Negative binomial was used, rather than Poisson error distribution, because preliminary analysis showed that the former strongly outperformed the latter in the goodness of fit (comparison of full models with different error distribution; $\Delta\text{AICc} = 445.47$). Mixture models analyze the data in 2 steps, with the first one being a binomial process that estimates the probability of measuring a zero, while in the second process, counts are modeled by negative binomial model [29]. Thus, to limit the number of models fitted, the logistic part of the model was first specified, while keeping the count part constant (intercept only). A complete model was built with tick count as the response variable and following the independent factors: treatment (trail vs. off-trail), species (*I. ricinus* and *Dermacentor* spp.), developmental stage (adults vs. nymphs), and interaction term between treatment and species. The study site was included to account for nested data structure, all of which was retained in all candidate models. Next, the Akaike information criterion corrected for the small sample size (AICc) was used to specify the optimal model structure [30]. As the next step, the model that received the highest AICc support was used, and the count part of the model was specified by including different combinations of the same set of independent effects, as before, and evaluated their fit with the AICc. The model with the lowest AICc within the set was considered the best [30]. In the analysis, genders were pooled together and their summed count was treated as 'adults', because preliminary data exploration showed no differences in the spatial distribution of males and females (see Supplementary Material, Fig. 1S). The 'MuMIn' package was used for the AICc model selection process [31].

RESULTS AND DISCUSSION

During the study, 351 *Ixodes ricinus* and 570 *Dermacentor* spp. ticks were collected (Tab. 1).

The best model included the effects of treatment, tick species, developmental stage, and the interaction between treatment and species in both the logistic and binomial count part (Tab. 2a). The interaction term in the count part of the best model suggests that *I. ricinus* responded more strongly to the trail presence (Tab. 3, Fig. 1). However, the second-best model did not include the interaction term and received similar AICc support ($\Delta\text{AICc} = 1.13$) [32] (Tab. 2A).

Generally, the overall abundance of ticks was almost 5-fold (*I. ricinus*) and 3-fold (*Dermacentor* spp.) higher on animal trails than on adjacent control transects (Tab. 3, Fig. 1).

Table 1. Total number of ticks collected on animal trails and transects along animal trails

	<i>Ixodes ricinus</i>				<i>Dermacentor</i> spp.			
	males	fe-males	nymphs	to-gether	males	fe-males	nymphs	to-gether
animal trail	49	51	208	308	191	244	0	435
transect	9	12	22	43	80	55	0	135

Table 2. Model selection table. A) specification of the count part of the model, b) specification of the logistic part of the model. To limit the number of models fitted we first specified the logistic part of the model, while keeping the count part constant (intercept only). Next, we defined the count part of the model. Models are ranked according to the AICc value; logLik – log-likelihood; AICc – Akaike's information criterion adjusted for small sample size; $\Delta\text{AICc} = \text{AICc} - \text{minAICc}$; wi – model weight. × denotes interaction term

Rank	Fixed effects	Df	logLik	AICc	ΔAICc	wi
<i>logistic part of the model</i>						
1	Site + Treat + Species + Stage + Treat × Species	15	-442.6	917.0	0	0.99
2	Site + Treat + Species + Stage	14	-449.3	928.2	11.16	0.01
3	Site + Treat + Species	13	-484.0	995.3	78.32	0
4	Site + Treat	12	-489.3	1003.7	86.72	0
5	Site	11	-493.8	1010.6	93.53	0
<i>count part of the model</i>						
1	Site + Treat + Species + Stage + Treat × Species	27	-399.1	857.7	0	0.64
2	Site + Treat + Species + Stage	26	-400.9	858.8	1.13	0.36
3	Site + Treat + Species	25	-413.1	880.8	23.10	0
4	Site + Treat	24	-427.7	907.6	49.95	0
5	Site	23	-435.1	920.2	62.56	0

Table 3. Output of the count part of the model that received highest AICc support (see Table 1). Coefficients for study sites and for the logistic part of the model are omitted

Variable	Regression coefficient	SE
Intercept	3.78	0.52
Trail	0.98	0.33
Species (<i>I. ricinus</i>)	-2.17	0.41
Stage (Adult)	-2.02	0.43
Trail × Species (<i>I. ricinus</i>)	0.98	0.52

Note: 'Trail' represents the difference in abundance between trails and adjacent transects for *Dermacentor*. 'Species' represents the difference in abundance between *Dermacentor* spp. *I. ricinus* ticks. 'Stage' represents the difference between adult and nymph abundance. The interaction term represents the difference in the effect of trail on *I. ricinus* abundance in comparison to *Dermacentor*. The coefficients represent changes in abundance expressed on a logarithmic scale.

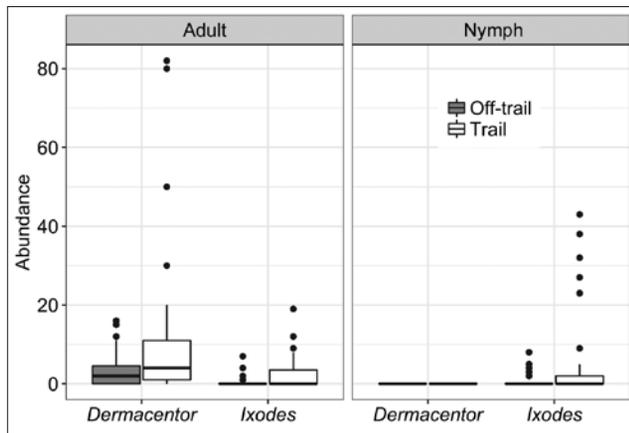


Figure 1. Spatial differences in abundance of studies ticks.

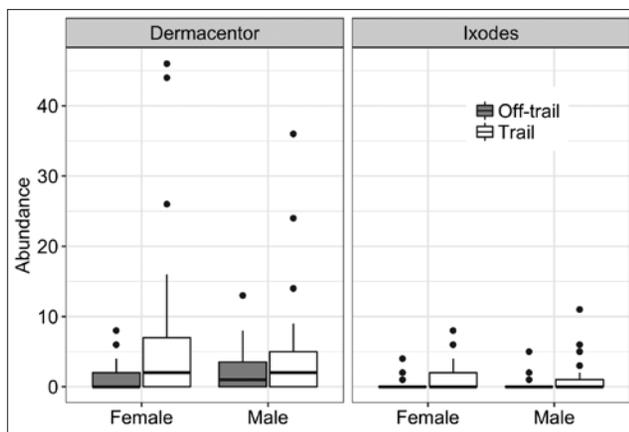


Figure 15. Boxplots differentiating spatial distribution of tick genders

These results support the hypothesis that ticks are more abundant on pathways than in adjacent habitats. Most likely, this pattern emerges because large mammals, like deer, the most important ticks hosts, [33,34], use forest paths to move across the landscape, and frequently move along the same routes [35].

Ticks leave the host after feeding by dropping on the vegetation, and molting (larvae or nymphs) or lay eggs (adult females) [36]. Since their dispersal range is very limited [18], most of them stay near the forest path, creating strong spatial differences in their abundance at the microhabitat scale. Furthermore, this study shows spatial differences in abundance among species and life stages. Lack of the *Dermacentor* spp. nymphs in the study sample is a consequence of the method of collection as nymphs of this species are nidicolous [37], contrary to *I. ricinus* nymphs [38]. One likely factor responsible for the different response of adult tick species to trail presence is different habitat preferences of the focal species. *Dermacentor* spp. prefer dry, open spaces, like xerophilic plant communities and open

meadows, hence their occurrence within forests is generally low [39]. In contrast, *I. ricinus* prefers forest edges [40, 41], which likely contributes to its higher abundance on animal trails than in adjacent habitats.

CONCLUSION

The best protection from tick-borne pathogens is prevention [23, 42–44]. One simple but very effective practice is avoidance of habitats with a heavy occurrence of ticks [45]. Numerous, recent studies have investigated how the abundance of ticks varies among habitats (e.g. forests, meadows, or city parks) [46–48], but the crucial role of the distribution of the parasites inside these habitats remains largely unexplored. The presented study aimed at filling that gap, and we disproved the popular notion that hiking on animal trails can help lower the risk of acquiring a tick. In fact, their research sends an important public message – that these forest trails are hotspots of disease risk and should be avoided. This fact should be treated as important information for implementing preventive medical and veterinary medicine.

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