

Factors influencing the distribution and activity of *Dermacentor reticulatus* (F.) ticks in an anthropopressure-unaffected area in central-eastern Poland

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Abstract

Introduction and objectives. Despite the increasing prevalence and significant role of *D. reticulatus* in pathogen transmission, the factors influencing its distribution and activity are still poorly known. The paper presents for the first time a study of the *D. reticulatus* ecology in a protected area, in which the dependence of *D. reticulatus* occurrence and activity on the ecological habitat type is analysed.

Materials and methods. Tick collection and environmental monitoring were conducted from March – November 2012 and 2014 in five ecologically-diverse habitats in the Polesie National Park of central-eastern Poland.

Results. The study shows that the most beneficial habitats for *D. reticulatus* are provided by meadow ecosystems dominated by plants from the class *Molinio-Arrhenatheretea*: *Poa pratensis*, *P. trivialis*, *Festuca rubra*, *F. pratensis*, and *Alopecurus pratensis*, as well as *Carex* sp. sedges (64.78% of the tick collections). Less favourable conditions are offered by the fresh habitat covered by deciduous forest with dominance of *Betula* sp., *A. pratensis*, and *Carex* sp. sedges (15.27%), in the wet habitat dominated by *Alnus glutinosa* and *Urtica dioica* (9.00%), *Betula humilis*, *Salix* sp., and *Carex* sp. (8.44%), and in the subcontinental hornbeam forest *Tilio-Carpinetum* with poor undergrowth (2.50%). *D. reticulatus* avoids wet habitats with bog birch *Betuletum pubescentis* communities and moist pine forest *Molinio-Pineteum*, as well as habitats dominated by heather *Calluna vulgaris*.

Conclusions. The distribution and dynamics of the activity of adult *D. reticulatus* depends on the biological and geoclimatic conditions prevailing in tick habitats. The structure of the flora colonising a habitat may be an indicator of the presence of this tick species.

Key words

Dermacentor reticulatus, activity, abundance, habitat preferences, protected area

INTRODUCTION

The geographical range of *Dermacentor reticulatus* covers a vast area in Eurasia. Over the last 10 years, expansion of this tick species into new habitats has been reported [1]. The abundance of *D. reticulatus* population has also been increasing [2]. The extension of the occurrence range and increased quantity of *D. reticulatus* raises the risk of animal and human health infections with tick-borne pathogens transmitted by the tick species, e.g. protozoa from the genus *Babesia* [3, 4], tick-borne encephalitis viruses [5, 6] rickettsiae [7], and other bacteria [8]. Therefore, identification of factors that influence *D. reticulatus* distribution and activity is fundamental for epidemiological investigations of tick-borne diseases in Europe.

Particularly interesting are observations carried out in protected areas where natural environmental conditions are conserved, since they allow determination of the habitat preferences of *D. reticulatus*. Moreover, comparison of results of field studies conducted in habitats affected by anthropopressure with those obtained in legally protected areas, can indicate the extent to which human activity influences the spread of this tick species, and the growing threat to animal and human health posed by tick attacks.

The study was undertaken in the Polesie National Park in central-eastern Poland, which is situated within the occurrence range of the eastern *D. reticulatus* population, and is simultaneously unaffected by anthropopressure. The research was focused on investigation of the impact of habitat type on the distribution and the-host seeking behaviour of this tick species.

MATERIALS AND METHOD

Study area. The Polesie National Park (PNP) with an area of 97.6 km² is located in lowland areas of central-eastern Poland (51°27'N 23°10'E) (Fig. 1) in a zone with temperate climate

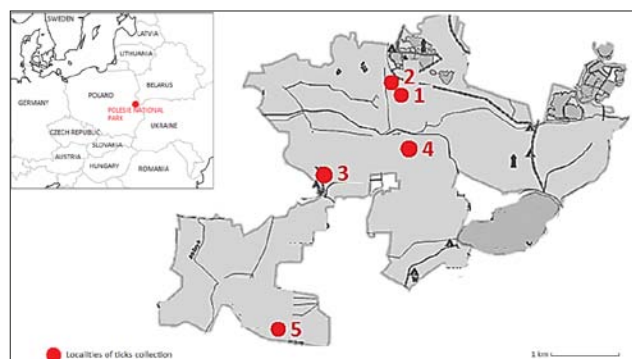


Figure 1. Location of *D. reticulatus* tick habitats in the Polesie National Park

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and continental features. The mean annual temperature in the area is 7.4 °C, with relatively high mean air temperatures in summer and low values in winter; this makes the annual temperature amplitude one of the highest in Poland. The mean annual precipitation figures in this area are 542 mm and represent the lowest values in eastern and central Europe. In autumn, precipitation levels exceed those noted in spring. The mean relative air humidity is 67% [9].

Forest communities with the largest cover in the PNP are represented by continental mixed coniferous forest *Quercus roboris-Pinetum* (30%), mixed alder fen forest *Sphagno squarrosi-Pinetum* (26%), sub-continental hornbeam forest *Tilio-Carpinetum* (25%), bog birch forest *Betuletum pubescentis* (23%), and black alder forest *Ribo nigri-Alnetum* (15%). Non-forest ecosystems are represented by aquatic-bog habitats that are unique in Europe, i.e. carbonate peat bogs with *Molinia* meadow communities, and transitional mire vegetation. A large area is covered by meadows dominated by *Poa pratensis-Festuca rubra* communities. The area is inhabited by 48 species of mammals, with rodents as the most numerous group [10].

Study localities. The investigations were carried out for two years in ecologically different habitats: in 2012, in five localities (localities 1–5), and in 2014, in only four localities due to the persisting high level of ground waters in one locality (Locality 5), which prevented tick collection. An inventory of vascular plants was made in each locality. The species were identified using the key developed by Szafer et al. [11], and dominant plant communities recognised [12].

Figure 2A. Locality 1.

The transect along which ticks were collected, extended across meadows with heath enclaves, *Betula* sp. groups, and single *Pinus sylvestris* specimens. It was dominated by meadow vegetation from the class *Molinio-Arrhenatheretea*, as well as the community *Poa pratensis-Festuca rubra* with substantial cover of *Poa pratensis*, *P. trivialis*, *Festuca rubra*, *F. pratensis*, *Alopecurus pratensis*, *Holcus lanatus*, and *Phleum pratense* growing on slightly acidic, often calcareous, peat substrates. This is fresh vegetation which can also colonise drier areas. This habitat also comprised patches of vegetation from the community *Sieglingio-Agrostietum*, characterised by a high share of *Calluna vulgaris* heather, as well as *grostis capillaris*. This habitat type exhibits a moderate moisture level. The banks of the lakes were colonised by vegetation from the sedge rush association *Caricetum paniculata* and the fen sedge association *Cladietum marisci*, characteristic for fresh and moist habitats.

Figure 2B. Locality 2.

The transect was situated along a mid-forest path and a tract between two water reservoirs. The tree stand in this locality was dominated by birch *Betula* sp., *Alnus glutinosa*, *Salix* sp. and species characteristic for the sub-continental hornbeam forest *Tilio-Carpinetum*, as well as shrubs, primarily deciduous tree saplings. The most frequent grass species included *A. pratensis* and *Carex* sp. sedges. A part of the path was a mid-forest clearing with a considerable share of plants characteristic for the class *Molinio-Arrhenatheretea* and the community *Poa pratensis-Festuca rubra*. Areas near the water reservoirs were overgrown by low grass species. The locality had the character of a fresh and moist habitat.

Figure 2C. Locality 3.

The transect extended across a forest and along the bank of Lake Moszne. The first section mentioned was dominated by deciduous forests – sub-continental hornbeam forest *Tilio-Carpinetum* (low and typical hornbeam forest) with single coniferous specimens. The understorey and groundcover were poor. The area located closer to the lake was dominated by the moist pine forest *Molinio-Pineteum*. The area was also covered by a hummock-hollow alder forest. The lake banks were dominated by the bog birch forest *Betuletum pubescentis*, as well as birch *Betula humilis* scrubs and *Salix* sp. shrubs.

Figure 2D. Locality 4.

The locality was situated in a moist habitat. On one side, it bordered a watercourse and on another side it was adjacent to wet areas covered by forests. *Ribeso nigri-Alnetum* was the dominant plant community. This type of vegetation community was characterised by dominance of *Alnus glutinosa* and the so-called hummock-hollow structure with hollows filled with water. The elevated part of the area was colonised by birch *B. humilis*, willow *Salix* sp., elongated sedge *Carex elongata*, crested wood-fern *Dryopteris cristata*, black currant *Ribes nigrum*, and peat moss *Sphagnum* sp. Additionally, wild shrubs of raspberry *Rubus* sp., dog-rose *Rosa canina*, and common nettle *Urtica dioica* were found in this locality.

Figure 2E. Locality 5.

There were two distinct ecological habitat types in this locality. In one of the sections of the transect, there was a moist habitat dominated by the community *Ribeso nigri-Alnetum* with a substantial share of *A. glutinosa* and single specimens of *Betula humilis* and willow *Salix* sp. A great number of *Carex* sedge species and *U. dioica* were also noted. A fresh habitat constituted the other part of the transect, i.e. a midforest clearing delineated by a watercourse on one side and a forest on the other, covered by grasses, and a *Sphagnetum magellanicum* peat bog with single specimens of *Betula* sp. and *P. sylvestris*.

The transects from which ticks were collected for the presented study coincided with the didactic-ecological path, i.e. a popular recreational site for the residents of the Lublin Province and tourists from all over Europe. The locality is frequently visited by Polish schoolchildren for field biology classes.

Tick collection. The flagging method was used for the collection of unfed *D. reticulatus* adults. After sweeping a several-meter long fragment of vegetation with a flannel cloth, the flag was viewed, attached specimens collected and placed in transport containers. Throughout the observation period, the plants were always swept by the same person in all localities. Each time, ticks were collected for one hour. In the laboratory, the specimens in each sample were counted and the tick species and gender identified.

The field study was conducted at ca. 14-day intervals throughout the vegetation period for this part of Poland, i.e. from mid-March – mid-November. During the longer daylight hours, between May and mid-September, the field study was carried out twice a day. In turn, during the shorter daylight hours in autumn, the ticks were collected once a day. The choice of the time of tick collection followed





Figure 2. Ecologically different habitats of *Dermacentor reticulatus* located in the Polesie National Park: A- locality 1; B – locality 2; C – locality 3; D – locality 4; E – locality 5.

previous studies [13]. Ticks were usually collected only on clear days without precipitation and with moderate or no wind.

During each collection round, air temperature and humidity were measured using electronic devices. The temperature and humidity of the air were measured at three levels, i.e. at ground level (0 cm), at the mean height of plants growing at the collection site (30 cm), and at a height of 70 cm above the ground. At the time of analysis of the results, the measurement data were averaged. Information about the length of daylight on the tick collection days was obtained from a database [14].

The study was conducted with Permission No. 7/2012, 6/2013, 3/2014 of the Directorate of the PNP.

Statistical analysis. Differences between two groups were analysed with the Mann-Whitney U test. In the case of more than two groups, the comparison was based on the chi-square

Kruskal-Wallis test. A significance level of $p=0.05$ was assumed.

RESULTS

Distribution and abundance of *D. reticulatus* in various habitats. During the two-year field study conducted in the PNP, a total of 4,276 *D. reticulatus* specimens were collected: 2,436 females and 1,840 males (Tab. 1). The greatest number of specimens (64.79%) was collected in the open ecosystem covered by meadow plants from the class *Molinio-Arrhenatheretea*, community *Poa pratensis-Festuca rubra*, bordering the mixed forest and watercourses (Locality 1). A four-fold lower number of ticks (15.27%) than that in Locality 1 was noted for the mid-forest paths in forests dominated by deciduous trees in Locality 2. Adult *D. reticulatus* stages were also abundant in Locality 5, where



Table 1. Number of adult *D. reticulatus* stages collected in the different localities in the Polesie National Park in the period of tick seasonal activity in 2012 and 2014

Locality	Range of temperature (T) and humidity (H)		Number of collected ticks			
	T [°C]	H [%]	Females (F)	Males (M)	Total	Ratio F:M
1	9.1–34.6	33.4–80.2	1,519	1,251		1.21
2	11.0–31.1	32.4–82.6	390	263	653	1.48
3	11.8–32.6	41.5–88.3	75	32	107	2.34
4	9.7–33.2	32.1–82.7	225	160	385	1.40
5	14.0–28.2	50.9–82.9	227	134	361	1.69
Study area	9.1–34.6	32.1–88.3	2,436	1,840	4,276	1.33

8.44% of all specimens collected during a two-fold shorter time (one-year investigations in 2012). A bigger number of adult ticks was found in the fresh habitat dominated by plants from the class *Molinio-Arrhenatheretea*. In the other localities (3 and 4), the tick abundance was substantially lower and reached 2.5% and 9.0%, respectively, of all specimens collected during the study (Tab. 1).

The statistical test confirmed the differences in tick abundance in the different habitats during the two study years. No statistically significant difference was demonstrated between the number of active ticks in 2012 and 2014, neither in the entire study area ($Z=-0.268$; $p=0.789$) nor in each analysed locality (Locality 1: $Z=0.190$; $p=-0.849$; Locality 2: $Z=-0.199$; $p=0.842$; Locality 3: $Z=-0.024$; $p=0.842$; Locality 4: $Z=-1.627$; $p=0.104$).

The proportions of females and males differed between the localities. The greatest differences in the number of specimens of both genders were revealed in Locality 3, where the number of females was 2.34-fold higher than that of male specimens. In the other localities, the dominance of females over males was nearly two-fold lower. As shown by the study, similar temperature and humidity conditions prevailed during the collection of *D. reticulatus* in all the tick habitats (Tab. 1).

Seasonal activity of *D. reticulatus*. A bimodal rhythm of the seasonal activity of adult *D. reticulatus* ticks was demonstrated in both study years in all the investigated localities. The spring activity began in March and exhibited a peak in May when, on average, 20.90 females and 15.02 males were collected per hour in the two-year study period. Males were found to be more active in March and the first half of April, whereas females dominated over males from late May to late June (Tab. 2).

The second activity peak in the analysed area began in mid-August (Tab. 2). From the beginning of September, the activity of the adult stages increased nearly 12-fold in comparison to August. The peak of autumn activity was noted at the turn of October – November. Active females dominated over males from early October to the end of autumn *D. reticulatus* activity.

A similar rhythm of the seasonal activity of adult *D. reticulatus* was noted in both study years (Tab. 2). The statistical test confirmed statistically significant differences in the activity of adult *D. reticulatus* in spring and autumn in the entire study area, in both 2012 and 2014 ($p=0.007$; $Z=-2.676$).

Within the investigated habitats, statistically significant differences between the spring and autumn activities were only noted in Locality 1 in 2012 ($Z=-2.148$; $p=0.032$) and in Locality 2 in 2014 ($Z=-2.344$; $p=0.019$).

In the Polesie National Park, ticks were active at a temperature ranging from 9.1 – 34.6°C (mean temperature: 22.9±5.57°C) and air humidity – 31.10–88.30% (mean humidity: 59.92%±11.28) (Tab. 1). Statistical analysis performed for the entire study area showed that air temperature had a significant effect on the activity of adult *D. reticulatus* ticks ($\chi^2=28.386$; $p<0.001$). However, no correlations were found between the number of collected ticks and air humidity in the entire protected area ($\chi^2=7.134$; $p=0.068$) and in the phytosociologically different localities (Locality 1: $\chi^2=4.690$; $p=0.196$; Locality 2: $\chi^2=1.632$; $p=0.652$; Locality 3: $\chi^2=6.377$; $p=0.095$; Locality 4: $\chi^2=2.864$; $p=0.413$; Locality 5: $\chi^2=0.513$; $p=0.774$).

Table 2. Seasonal activity of adult *D. reticulatus* stages at the different localities in the Polesie National Park in 2012 and 2014.

Month and year	Locality/Average number of ticks collected by 1 person during 1 hour									
	1		2		3		4		5	
	F	M	F	M	F	M	F	M	F	M
April 2012	nd	nd	nd	nd	7.00	1.00	nd	nd	42.00	11.75
April 2014	26.00	27.30	8.00	8.30	nd	nd	6.33	8.00	nd	nd
May 2012	59.75	25.50	41.00	12.50	8.00	1.00	16.25	7.00	22.00	7.00
May 2014	40.50	59.50	10.50	10.10	3.66	2.33	2.50	2.75	nd	nd
June 2012	5.80	3.00	3.80	4.70	1.50	0.25	6.25	1.25	1.50	1.00
June 2014	6.00	3.00	2.00	3.00	0.00	3.00	0.00	0.00	nd	Nd
July 2012	nd	nd	0.00	0.00	4.00	0.00	0.00	0.00	0.00	0.00
July 2014	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	nd	nd
August 2012	0.00	0.00	0.00	2.00	0.00	0.00	0.00	0.00	0.00	0.00
August 2014	nd	nd	4.00	6.00	0.00	0.00	nd	nd	nd	nd
September 2012	67.75	34.00	16.50	10.50	2.00	0.25	20.50	15.75	21.75	18.50
September 2014	24.67	33.67	12.50	8.20	6.00	3.00	0.50	2.50	nd	nd
October 2012	121.00	82.00	11.50	9.00	5.00	7.00	13.00	8.00	22.00	11.00
October 2014	131.00	75.00	12.50	20.00	0.00	2.00	3.00	2.00	nd	nd
November 2012	96.50	83.50	nd	nd	nd	nd	nd	nd	nd	nd
November 2014	100.00	80.00	nd	nd	nd	nd	0.00	4.00	nd	nd

F – females; M – males; nd – no data



Comparison of the study results obtained in the different habitats indicates a statistically significant correlation between temperature and the number of active ticks only in Locality 1 ($\chi^2=12.887$; $p=0.002$), but not in others (locality 2 $\chi^2=4.841$; $p=0.089$; locality 3 $\chi^2=0.415$; $p=0.813$; locality 4: $\chi^2=5.122$; $p=0.077$; locality: 5 $\chi^2=0.356$; $p=0.837$).

The greatest number of specimens was collected in a period with less than 11 hours of daylight (i.e. October and November); in turn, the lowest number of ticks was found on days with more than 15 hours of daylight (i.e. June, July and August). The statistical test confirmed the correlation between the length of daylight and activity of *D. reticulatus* ($\chi^2=45.027$; $p<0.001$).

DISCUSSION

Results of field studies on the distribution, abundance, and activity of *D. reticulatus* in environments characterised by varied degrees of anthropopressure in various geographical regions, have so far been analysed primarily with reference to selected climatic indicators noted during the study periods, i.e. mainly temperature and humidity [2, 13, 15, 16, 17, 18], some habitat traits [17, 19, 20, 21], or in terms of the host species composition [22, 23].

As indicated in this study, in a protected area, distribution, abundance, and activity of *D. reticulatus* depend on the type of habitat, which is defined by characteristic plant groups determined by the climate and soil conditions, and presence of specific animal species. Meadow ecosystems with fresh vegetation bordering forested areas and watercourses offer the most favourable conditions for *D. reticulatus* development in anthropopressure-unaffected areas. Such habitats ensure the frequent presence of many animals that can be hosts for immature tick stages [19, 24, 25, 26, 27] and adult ticks [28, 29, 30].

In the area of the PNP, the accumulation of adult *D. reticulatus* ticks was observed on meadow vegetation from the class *Molinio-Arrhenatheretea*, which serves as food for such animals as the elk (*A. alces*), deer (*C. elaphus*), wild boar (*S. scrofa*), and, to a lesser extent, roe deer (*C. capreolus*) [31, 32, 33]. An inventory made in two hunting districts and comprising the presented study area, revealed the presence of numerous animals (Tab. 3) regarded as the

Table 3. Occurrence of hosts of adult *D. reticulatus* stages in the study area, habitat and feeding preferences

Species	No. of individuals	Habitat preferences	Feeding preferences
Elk <i>Alces alces</i>	150 ¹ 1,543 ²	Open meadows, banks of water bodies and rivers	Herbaceous plants, aquatic plants, grasses, sedges
Deer <i>Cervus elaphus</i>	100 ¹ 4,040 ²	Meadows, deciduous and mixed forests	Shoots, leaves, and bark of trees and shrubs, grasses
Wild boar <i>Sus scrofa</i>	200 ¹ 8,798 ²	Deciduous and mixed forests with beech and oak, riparian forests, bogs, and meadows	Omnivorous species; mainly acorns, plant rhizomes, grasses, tubers
Roe deer <i>Capreolus capreolus</i>	600 ¹ 21,465 ²	Deciduous and mixed forests, scrubs, meadows	Shoots, leaves, and bark of trees and shrubs, grasses

¹No. of individuals in the Polesie National Park (area of 97.6 km²) [19].

²No. of individuals in the Biala Podlaska (area: 5,348 km²) and Chełm (area: 3,866 km²) Hunting Districts comprising the Polesie National Park [53].

major *D. reticulatus* hosts in Europe. These animals do not exhibit territorial behaviour; therefore, they can be assumed to migrate in search of food to the habitats situated in the analysed protected area.

Meadow ecosystems adjacent to forests offer favourable temperature and humidity conditions required for maintenance of water balance, which is indispensable for the survival and development of the immature and adult stages of *D. reticulatus* [17, 18]. Temperature and humidity conditions prevailing in meadow ecosystems satisfy *D. reticulatus* preferences defined in laboratory studies.

In the protected areas in eastern Poland, no *D. reticulatus* ticks were found in the habitats of the moist pine forest *Molinio-Pineteum*, bog birch forest *Betuletum pubescentis*, and in transect sections covered by heather (*C. vulgaris*).

The current study shows not only a considerable abundance of *D. reticulatus* populations, but also the greatest numbers of adult specimens during the autumn activity peak in the meadow ecosystem bordering the forest complexes. For this type of habitat, a statistically significant correlation was found between the *D. reticulatus* activity and temperature, which was not reported from the other habitats.

The absence of a statistically significant correlation between the number of active ticks and temperature in the localities situated on the mid-forest paths (Locality 2 and 4), in the deciduous forest (Locality 3), and in the clearing partly covered by alder forest (Locality 5), may be caused by the more stable abiotic conditions offered by these habitats in the different seasons of the year, compared with open areas. Both during the morning and afternoon collection sessions, greater temperature fluctuations were observed in the meadow ecosystems than in the other localities.

The abundance of *D. reticulatus* ticks in the protected area in central-eastern Poland is substantially higher than that in anthropopressure-affected regions in other parts of the Lublin macroregion [26, unpublished data]. The differences in the methodologies employed in this study, and in investigations carried out by other researchers in different parts of Europe, make it impossible to compare their results and determine the impact of anthropogenic factors on *D. reticulatus* populations. In other tick species originating from contaminated regions, differences were noted in their abundance and seasonal activity rhythm in areas with a varied degree of anthropopressure [34, 35, 36], as well as developmental distortions leading to morphological anomalies in many specimens [37, 38].

The dominance of active females over males reported from the shady areas of the deciduous moist sub-continental hornbeam forests (locality 3), compared with their numbers in the open areas of the moderately moist meadows (Locality 1) noted during the presented study, can be explained by the differences in the morphology (degree of chitinisation of the body surface) and physiology between the two genders, and their different temperature and humidity requirements. Since they are exposed to higher risk of water loss, females reduce their activity in adverse conditions. In an earlier study by the authors conducted in various types of habitats in eastern Poland, it was found that females and males exhibited the highest activity at a temperature of 20°C and 65%, and 18°C and 60% humidity, respectively (Buczek et al. unpublished data).

The presented and previous results [2, 13, 17, 18] of studies conducted in eastern Poland, and those reported by other



authors [15, 16, 20, 39], confirm the bimodal rhythm of the activity of adult *D. reticulatus* ticks, irrespective of the habitat type and climatic conditions. In turn, they indicate dependence of the dynamics of the activity of adult stages of this species on climatic and weather [18] conditions. The current study was carried out in the same period and in different habitats of the protected area and indicates the impact of the habitat type on the activity of adult *D. reticulatus* ticks in different seasons of the year.

The wide range of temperature and humidity tolerance, as well as the habitat requirements of *D. reticulatus* determined in this study, may explain the expansion of the species into new areas [40], including recreational suburban areas [41, 42].

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