The density and peaks of seasonal activity of *Dermacentor reticulatus* ticks in eastern Poland

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Abstract

Background: Next to *Ixodes ricinus*, *Dermacentor reticulatus* ticks are the most important reservoir and vector of infectious diseases in Europe. In recent years, progressing expansion of *D. reticulatus* ticks into new territories and an increase in their population size in areas of their previous occurrence have been observed.

Methods: The studies on tick population density were conducted in Lublin Province, eastern Poland. The investigated area was divided into 101 equal squares (plots). Additionally, studies on seasonal activity in the preferred habitat of the ornate cow ticks were conducted from 2014 to 2019 in the Polesie National Park.

Results: Throughout the study on the tick population density, 19559 adult *D. reticulatus* specimens were collected. An average density of the ornate cow ticks in Lublin Province was 96.8 specimens/100 m². A significant impact on the tick population density is exerted by meadow areas ($R_s=0.281$, $p=0.0044$), while weather conditions have no effect ($H=6.08$, $p=0.1931$). The local population of *D. reticulatus* ticks in the eastern and middle regions of Lublin Province exhibits predominance of spring activity peaks, while autumnal seasonal activity peaks prevail in the rest of the region. Multiannual monitoring of the seasonal activity of *D. reticulatus* in the Polesie National Park shows the prevalence of autumnal activity peaks in 2014-2019 and spring activity peaks in 2019.

Conclusions: The area of Lublin Province is characterized by high density of the ornate cow ticks. Local populations of *D. reticulatus* show two peaks of seasonal activity. Dynamics of seasonal activity of *D. reticulatus* is influenced by availability of their hosts. The highest risk of ornate cow tick attacks on animals and humans occurs during spring in the eastern part and in autumn in the western part of Lublin Province.

Background

Dermacentor reticulatus belongs to the most widely spread tick species throughout Europe. According to many authors, two geographically separated populations of ornate cow ticks are distinguished – the “western European” and “eastern European” population. This phenomenon is not found in other widely distributed ticks species in Europe i.e. *Ixodes ricinus* or *D. marginatus* [1, 2].

In Western Europe, the compact area of ornate cow tick distribution includes the southern part of the British Islands, France, Switzerland, Benelux, and Germany [2-5]. Specimens of this species are most commonly collected from plants as well as wild, farm, and domestic animals in Central Europe. The lowlands of the Czech Republic, western Slovakia, Hungary, Slovenia, and Austria should be considered as endemic areas for this species. *D. reticulatus* from the regions mentioned above are included into the western European population of this species [6-10].

The compact range of the eastern European *D. reticulatus* population covers eastern Poland, Ukraine, Belarus, Lithuania, Latvia, Estonia, and the north-west of the European part of Russia. Numerous sites of ornate cow tick occurrence have been reported from the Caucasus and Asia to Omsk and Novosibirsk [4, 11-14].

Until the beginning of the 21st century, it was thought that the compact area of *D. reticulatus* distribution in Poland is limited to the territory located to the east of the Vistula River and in north-eastern part of the country, while western regions were considered free from this species. Simultaneously, it was accepted that the border between the eastern and western population of *D. reticulatus* ran through Poland [12, 15, 16]. Studies on tick ecology and distribution, which have been intensified in recent years by Polish scientists, confirmed the occurrence of the species also in the northern, western and south-western territory of Poland [17-24]. As suggested by some authors, this phenomenon should be referred to as expansion of ornate cow ticks to new areas [21, 25].

Similar changes in the dynamics and distribution of local *D. reticulatus* populations were observed in other European countries, e.g. in Germany where before the 70s of the 20th century its distribution was limited to western Lands, and the described population was not numerous [26]. In the next years, expansion to eastern Lands bordering Poland was observed [7]. Many new *D. reticulatus* occurrence sites were reported in the Czech Republic. In Slovakia, new sites of distribution of ornate cow ticks representing both eastern European and western European populations were reported [27, 28]. In recent years, expansion of *D. reticulatus* ticks was observed in Hungary, where only two sites of occurrence were documented before the 70s of the past century [8]. A similar process of expansion was also observed in Romania [10, 29].

Clustered populations of *D. reticulatus* were confirmed in the south of the continent, i.e. in the Balkan [30-33] and Iberian peninsulas [34]. The spread of ornate cow ticks to Scandinavia [35] and northern Italy, which were regarded as tick-free previously has been observed during the last years [36].

D. reticulatus populations in Europe are separated by a geographical border that has been disappearing lately due to the expansion of the tick species. Since genetic studies on specimens of both populations have shown no differences between their genomes, there is still an unanswered question about gaps in the *D. reticulatus* distribution in Europe [37].

The aim of our research was to conduct simultaneous large-scale studies of the occurrence and density of *D. reticulatus* population in eastern Poland. The investigations were also focused on the seasonal activity rhythms of this species in Lublin Province with their determinants, periods of a high risk of infestation of hosts, and risks of tick-borne pathogen transmission.

Methods

Density and seasonal activity peak rhythms of the *D. reticulatus* population

Determination of the experimental fields

The studies on the density of *D. reticulatus* ticks in eastern Poland were conducted in Lublin Province (Fig. 1). A grid of 101 squares with 15.8-km long sides (plots) was applied to a physical map of the investigated area (Fig. 2). An average area of each square was 250 km², which represents 1% of Lublin Province. Plots 1, 2, 19, 27, 35, 44, 53, 63, 81, 89, 96, 97, 100, and 101 were the exceptions, as they had smaller areas due to the course of administrative boundaries of the Province. At the same time, they constituted more than 60% of the area marked by the described grid.

The next stage included the use of satellite pictures available in Google Maps [38] to determine potential sites of species collection. Meadows undergoing ecological succession located on the edge of forests or in its vicinity and with access to watercourses were regarded as preferred tick habitats (Fig. 3).

Tick collection

The potential areas of tick collection were surveyed previously. Field studies were conducted during seasonal peaks (spring and autumn) of tick activity in Lublin Province. Determination of the activity peaks was based on the literature data [39-41] and on simultaneous monitoring of the *D. reticulatus* activity in different parts of Lublin Province. Ticks were collected during the spring peak between 18.03.2019 and 31.03.2019 and from 12.10.2019 to 24.10.2019 in autumn (square number 93 was excluded due to ongoing construction works).

Ticks were collected with the standard flagging method. Plants were swept with 1-m² flannel fabric on a 100-m² experimental field. During tick collection, weather conditions, i.e. air temperature and relative humidity, were measured with the use of Data Logger R6030.

During the laboratory study, the collected specimens were identified to the species level, developmental stage, and sex with the use of a stereoscopic microscope Zeiss STEMI DV4 and identification guide compiled by Estrada-Peña et al. [42].

Analysis of the land structure of the study area

With use of tools for measuring the distances and surface area in Google Maps [38] and satellite pictures, the percentage structure of the area in each square was calculated. The surface of the cultivated land, forests, and meadows was analyzed. The term "other" means buildings, water resource types, and areas that could not be classified clearly.

Analysis of the weather conditions

The historical data on the weather conditions from 2015-2019 in different regions of Lublin Province were retrieved from the Internet service tutiempo.net [43]. Data of the Terespol weather station were accepted as representative for the north-eastern region of Lublin Province, while the stations in Włodawa, Zamość, and Kozienice provided relevant data on the eastern, southern, and western regions, respectively (Fig. 4).

Multi-annual monitoring of the *D. reticulatus* seasonal activity

The studies on the seasonal activity of *D. reticulatus* ticks were conducted from 2014 to 2019 in the Polesie National Park located in the middle-eastern part of Lublin Province (square number 41). They were a continuation of studies started in 2012 [41].

The methods of collection and identification of specimens to the species level and developmental forms were the same as described above. Ticks were collected in equal periods from March to November. Throughout the study period, ticks were collected in the same habitat. The study was accepted by the Minister of Environment by virtue of permission DOP-WPN.286.312.2019.MD and DLP-III-4102/16/1278/15/MD.

Statistical analysis

Statistical analysis of differences between the number of active *D. reticulatus* females and males within the experimental fields was performed with the use of the Wilcoxon signed-rank test. A multiple regression model was used to assess the impact of the land structure on the tick population density. The significance of the impact of temperatures and air humidity on the tick population density was evaluated with the use of Spearman's Rank correlation coefficient. The weather parameters in the different regions of the province were analyzed with the Kruskal–Wallis test.

The Kruskal–Wallis test was used for statistical analysis of the number of active *D. reticulatus* females and males in the springs and autumns throughout the study period. A multiple regression model was used to assess the significance of differences in the seasonal activity of ticks in 2014-2019. The impact of air temperature and relative humidity on active ticks at the time of collection was checked by Spearman's Rank correlation coefficient.

In all statistical tests, the significance level was $p < 0.05$. The statistical analysis was conducted with Statistica 10PL software.

Study area

Lublin Province has an area of 25,122 km², which accounts for 8.02% of the territory of Poland. The border between three megaregions differing in the geomorphological structure, water features, and climate, i.e. East European Plain, Extra-Alpine Central Europe, and Carpathian Region, runs across the province area [44].

The eastern and north-eastern parts of Lublin Province are located in western Polesie and Volhynian Polesie (Fig. 4). These regions are located in the basin of the Bug – one of the longest rivers in Poland. The subsoil consists mainly of marls and chalk, hence the presence of many lakes and bogs. The regions are characterized by high woodiness and very low average air temperature values. At the same time, this area is exposed to the lowest anthropopressure in Poland [44].

The western part of the study area belongs to the South Podlasie Plain. This region is typically agricultural with high ratio of wastelands. The climate is more temperate, especially in the western regions. The central area of Lublin Province is part of Lublin Upland. Fawn and podzol soils form the subsoil, while brown soils are the substrate in the river valleys. Landform is pleated. This part of the province is intensively used for agriculture and has been almost deforested [44].

The southern outskirts of the study area are part of Roztocze. This is the highest area of the region, with climate close to continental and marls, opoka rocks, and limestone in its subsoil. Rivers in this area have a mountain character [44].

Results

The density and peaks of *D. reticulatus* population activity

In total, 19559 adult *D. reticulatus* specimens were collected throughout the study period, including 11598 females and 7961 males (Table 1). An average density of the ornate cow tick population in Lublin Province was 96.8 specimens/100m² with statistically significant dominance of females ($Z=8.19$, $p<0.001$). The ticks population density was higher in autumn than in spring (Table 2). The greatest population density was observed in the northern part of the study area (plots 1-18). It varied from 86 specimens to 311 specimens/100 m² throughout the study period. The lowest density of the *D. reticulatus* population was noted in the central part of the province. However, the lowest number of ticks were collected in square 67, where the population density was 22 specimens/100 m² (Table 1, Fig. 5).

The predominance of active *D. reticulatus* ticks in autumn was observed in the northern, southern, and western part of Lublin Province, while in the middle and eastern parts of region, the highest number of ticks were collected in spring (Table 1, Fig. 2).

In the regression model of the density of the ornate cow ticks, none of the land structure variables (arable lands, forests, meadows) has statistical significance ($F_{(3,97)}=1.05$, $p=0.3728$). However, Spearman's Rank correlation coefficient indicates that the density of active *D. reticulatus* females and males rises with the increase in the meadow area ($R_s=0.281$, $p=0.0044$).

The weather conditions observed in the particular parts of the region in 2019 were not significantly different (average air temperature $H=0.18$, $p=0.9962$; relative air humidity $H=2.00$, $p=0.7354$; total precipitation $H=0.82$, $p=0.9361$; number of days with snow cover $H=0.06$, $p=0.9995$; number of days with fog $H=6.08$, $p=0.1931$). These parameters had no statistically significant impact on differences in the tick density between the particular study areas (average air temperature $p=0.2848$; relative air humidity $p=0.1881$; total precipitation $p=0.3910$; number of days with snow cover $p=0.3910$).

The statistical analysis of the weather parameters in 2015-2019 did not show statistically significant differences between the parts of Lublin Province ($H=0.91$, $p=0.9901$).

Multiannual monitoring of the seasonal activity of *D. reticulatus* ticks

In the Polesie National Park (51°22'N, 30°30'E), a tendency towards an increase in the number of active *D. reticulatus* ticks was observed in 2014-2018. The substantial dominance of the average number of active ticks in autumn over spring was revealed. Simultaneously, more females were collected. In 2019, more active ticks were collected in spring than in autumn (331 to 93 specimens, respectively) (Table 3).

In each year from 2014 to 2018, a statistically significant difference between the number of active *D. reticulatus* ticks in spring and autumn was confirmed ($H=6.98$, $p=0.047$). However, there was no statistically significant difference in the number of active ticks between the study years ($H=3.94$, $p=2678$). Moreover, a statistically significant difference in the number of active ticks in 2014-2018 was revealed ($H=4.87$, $p<0.014$). The seasonal activity of *D. reticulatus* ticks is significantly influenced by the air temperature prevailing during tick collection ($H=12.887$, $p=0.002$).

Discussion

The dynamics and size of the *D. reticulatus* population, as in the case of other hard ticks, depend on many biotic and abiotic factors. The most important include the presence of potential hosts, ecological type of occupied habitats, climatic factors, photoperiod, excreted semiochemical substances and hormones controlling the rhythms of seasonal activity, diapause periods, and biology of tick reproduction and development [13, 39, 45, 46].

In eastern Poland, the greatest impact on the seasonal and diurnal rhythms of *D. reticulatus* activity is exerted by air temperature prevailing during tick collection [40, 41]. Together with air humidity, it can affect the questing behavior [47] and locomotor abilities of this species [48]. This dependence is confirmed by the results of our long-term studies on seasonal tick activity in the Polesie National Park ($H = 12.887$, $p = 0.002$).

D. reticulatus ticks have a wide range of ecological tolerance to environmental factors. Adult specimens can be active during winter months [49, 50] even at negative ambient temperature values, while in laboratory conditions they are able to survive up to 150 days at -10 °C [51]. As demonstrated by Hubalek et al. [52], the limit temperature value allowing ticks for host seeking up to -0.1 °C.

The area of Lublin Province has the highest density (96.8 specimens/100 m²) of tick populations at the peak of their seasonal activity of all areas studied in Poland (Table 1). The number of active adult *D. reticulatus* specimens recorded in Lublin Province is 13.2 times higher than in Mazovia (central Poland; an average of 7.4 specimens/100 m² in 2012–2014), 15.6 times higher than in Lubuskie Province (western Poland), and 32.3 time higher than in the West Pomeranian Province (north-western Poland) [21].

The density of the *D. reticulatus* tick population in Lublin Province is high but unevenly distributed (Fig. 5). On average, most ticks were collected in the experimental fields of squares located in the northern and north-eastern part of the region (South Podlasie Lowland, Polesie Lubelskie) (Fig. 4, Table 1). These areas have the largest meadows areas in the entire province. An increase in the meadow area positively correlates and has a statistically significant impact on the number of collected ticks ($R_s = 0.281$, $p = 0.0044$). It should be noticed that a significant percentage of this area is occupied by wastelands and forest islands (Table 1). The mosaic character of the landscape offers beneficial conditions for *D. reticulatus* populations. Forests provide availability of host species for adult specimens, while meadows and wastelands are preferred habitats for rodents – hosts of juvenile forms of ticks.

The greatest density of *D. reticulatus* ticks throughout the whole study period was observed in the experimental field located in plot 69 placed in Lublin Upland (Fig. 4). During single collection in the spring activity peak, 516 specimens of ornate cow ticks were harvested (297 females and 219 males) per 100 m² area (Table 1, Fig. 5, Fig. 6). Attention should be drawn to the lowest density of tick populations in the neighboring sites (located in the same macroregion) ranging from 10 to 100 specimens/100 m² (Fig. 5). Therefore, the high density of ornate cow ticks is a result of the ecological character of the habitat. The site where the greatest number of ticks was collected is surrounded by an uncut meadow, a watercourse, and a forest. The forest cover is < 8% in the scale of the whole macroregion and 22% for the province (Table 1). With its fertile soils, the area is intensively used for agriculture.

As indicated by Mierzejewska et al. [52] the spread of *D. reticulatus* is associated with the loss of forest area. We agree with this opinion but with the provision that deforestation cannot lead to transformation of woodlands into an agricultural area because this would limit the range of ticks distribution and decrease the population size. Large woodland areas have an important impact on tick population density in Lublin Province. The number of ticks collected in squares 73 and 74 (woodland > 61%, meadow area 1.93%) located within the biggest forest of Lublin Province ("Janów Forest") was significantly lower than an average of the region, i.e. 29 and 23 specimens/100 m² in spring and 46 and 30 specimens/100 m² in autumn, respectively (Table 1). The results of our studies show that *D. reticulatus* populations can endure and steadily grow only in areas with appropriate proportions between forests and meadows/wastelands. However, small isolated populations of *D. reticulatus* ticks can exist in habitats where these proportions are disturbed.

D. reticulatus ticks mostly exhibit two outstanding peaks of seasonal activity: in spring and autumn. In the French Alps, the European part of Russia, and north-eastern Poland, a higher number of ticks are active during spring [13, 53, 54]. The data published so far indicate that the ornate cow tick population in eastern Poland is characterized by even three-fold higher number of ticks collected in autumn [39, 40, 41, 47]. During our study, we observed predominance of the spring peak in the central and eastern parts of the region (Fig. 2) and the autumn peak in the other parts of the province. The seasonal activity of *D. reticulatus* during the year and the population density are not influenced by weather conditions, which were not significantly different in the whole study area over the last 5 years ($H = 6.08$, $p = 1931$). Similar dependences were described in studies on colonization of new areas by ornate cow ticks in western Poland [52]. The results of our study demonstrate that changes in the pattern of seasonal activity may occur in the same population that is stable in terms of its seasonal activity rhythms and growth (ongoing long-term investigations). The *D. reticulatus* population in the Polesie National Park (square 41) monitored for the last 7 years [41] exhibited a significantly higher number of active ticks in autumn in 2012–2018 and dominance of the spring peak of activity over autumn in 2019 with a ratio of 331:93 (Table 3). Because the experimental field is located within a protected area, the anthropogenic impact is limited to a minimum. We believe that the size and dynamics of the *D. reticulatus* seasonal activity are influenced mostly by the availability of preferred hosts occurring in investigated area, i.e. roe deer *Capreolus capreolus*, deer *Cervus elaphus*, elks *Alces alces*, and wild boars *Sus scrofa*. Our unpublished observations demonstrate the highest tick infestation in wild boars. These animals occur in the whole province and their population size has been growing during the last years, even up to 178% (from 6,400 to 17,800 specimens within 5 years). For comparison, the elk population in the same time comprised 2,300 and deer 7,600 individuals [55].

In 2017–2018, a decrease in the size of wild boar populations was observed in Lublin Province due to the confirmed outbreaks of ASF (African swine fever). Before April 2018, 1942 specimens were found dead in eastern Poland because of this viral disease [56]. Furthermore, during this period, the wild boar population was reduced preventively. This led to a significant decrease in the number of animals that are one of a main preferred hosts of *D. reticulatus* [17, 57]. This situation is reflected in the structure of the size and dynamics of seasonal activity of eastern Poland populations of *D. reticulatus* and explains the spatial distribution of the activity peaks of this tick species in Lublin Province. Considering the survivability of adult specimens in a habitat over 2 years [13], the predominance of tick activity peaks in spring in areas where the autumn peak was presented previously [41] (Table 3) can be caused by an insufficient amount of hosts for adult ticks. Nevertheless, an impact of other factors on this phenomenon should not be excluded and further observations and research are required.

The high population density of *D. reticulatus* ticks in Lublin Province is associated with a greater risk of tick attack on animals and sporadically on humans than in other country regions and increases the possibility of transmission of tick-borne pathogens. Diverse pathogens have been confirmed in ornate cow ticks, i.e. *Anaplasma phagocytophilum*, *Rickettsia raoultii*, *Borrelia burgdorferi* s. l., *Babesia* spp., and TBE virus [58]. The increase in the tick population density in this area can result in a future increase in the incidence of tick-borne diseases [59]. Numerous cases of canine babesiosis and domestic animal borreliosis have been currently reported in Lublin Province [60, 61, 62].

Conclusions

D. reticulatus ticks occur in the whole area of the Lublin Province. This region is characterized by a high level of activity of ornate cow ticks, which however is unevenly distributed. Local *D. reticulatus* populations exhibit differences in the peaks of seasonal activity determined mostly by availability of potential hosts.

The highest risk of *D. reticulatus* attacks on animals and humans occurs in spring in the eastern and central parts of the region but in autumn in the north and western parts of the province.

Declarations

Ethics approval and consent to participate

The study was accepted by the Minister of Environment by virtue of permission DOP-WPN.286.312.2019.MD and DLP-III-4102/16/1278/15/MD.

Consent for publication

Photos used in Fig. 3 and Fig. 6 were taken by Zbigniew Zająć, corresponding author of this manuscript

Availability of data and materials

The climate data analyzed during the current study are available in <https://en.tutiempo.net/climate/poland.html>

Competing interests

The authors declare that they have no competing interests

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Authors' contributions

ZZ: study design, ticks collection, data analysis, writing manuscript; AW: ticks collection, data analysis, writing manuscript, JK: ticks collection, data analysis, writing manuscript

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Tables

Table 1. Structure of the landscape in the study area and *D. reticulatus* ticks density per 100 m²

Square number	Landscape structure				Ticks density per 100 m ²													Total study period		
	Farmlands [%]	Forests [%]	Meadows [%]	Other [%]	Spring					Autumn					Average ticks density					
					Weather parameters		Ticks density			Weather parameters		Ticks density								
					T [°C]	H [%]	F	M	F+M	T [°C]	H [%]	F	M	F+M	F	M	F+M			
1	67.21	13.5	5.21	14.08	12.1	88.8	84	54	138	22.5	60.0	98	61	159	91	57.5	148.5			
2	68.00	20.65	5.98	5.37	10.8	87.0	105	75	180	21.8	70.6	150	122	272	127.5	98.5	226			
3	61.00	15.59	5.23	18.18	13.4	90.6	29	14	43	23.2	70.1	100	69	169	64.5	41.5	106			
4	55.11	29.96	7.99	6.94	13.5	80.5	74	51	125	23.1	70.6	112	82	194	93	66.5	159.5			
5	79.56	5.66	2.05	12.73	14.1	86.0	153	93	246	19.6	81.0	177	106	283	165	99.5	264.5			
6	65.12	18.20	6.78	9.90	15.1	75.4	45	25	70	20.5	67.5	130	96	226	87.5	60.5	148			
7	80.12	10.10	4.11	5.67	14.8	81.2	105	66	171	22.5	66.0	164	102	266	134.5	84	218.5			
8	64.99	18.70	6.78	9.53	14.7	70.0	201	51	252	23.4	72.5	248	122	370	224.5	86.5	311			
9	40.00	45.03	6.50	8.47	17.5	77.0	96	48	144	21.0	77.8	190	134	324	143	91	234			
10	71.20	11.96	8.98	7.86	16.1	86.0	44	30	74	19.8	69.0	116	81	197	80	55.5	135.5			
11	55.10	30.11	8.21	6.58	14.5	70.5	93	93	186	20.0	54.2	170	110	280	131.5	101.5	233			
12	73.30	16.41	6.10	4.19	12.9	77.9	26	19	45	18.5	70.5	145	98	243	85.5	58.5	144			
13	85.23	4.29	2.70	7.78	15.0	84.2	31	36	67	17.5	81.2	56	36	92	43.5	36	79.5			
14	77.40	13.04	1.80	7.76	12.1	81.8	75	39	114	16.1	84.2	88	24	112	81.5	31.5	113			
15	83.90	3.61	6.67	5.82	13.7	74.0	123	87	210	15.1	75.9	196	108	304	159.5	97.5	257			
16	65.21	18.47	7.45	8.87	11.9	86.1	84	42	126	13.9	77.7	99	55	154	91.5	48.5	140			
17	60.21	25.00	7.00	7.79	15.6	82.0	102	81	183	18.8	78.3	190	100	290	146	90.5	236.5			
18	55.12	30.56	6.12	8.20	16.1	76.0	42	35	77	20.5	71.0	64	31	95	53	33	86			
19	65.12	24.87	3.15	6.86	18.5	67.0	68	56	124	16.9	79.9	133	70	203	100.5	63	163.5			
20	65.18	17.30	7.87	9.65	22.0	66.0	166	122	288	21.0	65.7	180	135	315	173	128.5	301.5			
21	80.99	6.68	2.10	10.23	19.5	69.9	177	69	246	23.0	69.0	75	50	125	126	59.5	185.5			
22	70.36	21.67	3.98	3.99	15.4	78.9	129	99	228	22.0	54.9	60	64	124	94.5	81.5	176			
23	71.33	18.90	5.00	4.77	16.7	69.2	100	65	165	19.8	55.0	62	41	103	81	53	134			
24	60.12	25.53	7.00	7.35	14.8	67.0	141	75	216	17.5	87.0	36	18	54	88.5	46.5	135			
25	45.00	39.56	7.11	8.33	10.5	90.5	15	13	28	16.4	80.0	20	50	70	17.5	31.5	49			
26	70.01	12.24	3.98	13.77	11.0	82.0	45	39	84	17.0	81.0	26	13	39	35.5	26	61.5			
27	50.98	32.11	3.00	13.91	19.3	82.8	22	11	33	13.6	88.0	66	22	88	44	16.5	60.5			
28	70.11	17.53	6.18	6.18	18.5	71.6	49	22	71	17.4	80.2	66	50	116	57.5	36	93.5			
29	70.96	11.72	5.23	12.09	17.1	75.9	75	35	110	23.1	65.0	59	66	125	67	50.5	117.5			
30	55.00	25.84	8.01	13.15	16.5	65.0	45	25	70	22.0	53.2	58	44	102	51.5	34.5	86			
31	40.19	43.67	5.00	11.14	10.7	83.0	61	50	111	24.8	55.0	84	68	152	72.5	59	131.5			
32	71.11	18.31	5.45	5.13	10.9	81.0	40	34	74	25.6	45.7	37	10	47	38.5	17	55.5			
33	35.23	56.19	4.00	4.58	13.5	73.5	25	18	43	24.0	58.0	62	35	97	43.5	26.5	70			
34	14.18	72.00	9.12	4.70	13.0	79.0	18	11	29	20.9	74.8	15	5	20	16.5	8	24.5			
35	69.27	19.45	5.00	6.28	14.0	65.8	26	17	43	14.5	80.0	30	32	62	28	24.5	52.5			
36	78.16	8.00	7.12	6.72	13.7	58.9	69	96	165	15.6	69.0	60	58	118	64.5	77	141.5			
37	80.50	2.03	5.90	11.57	11.0	70.0	58	60	118	20.5	73.3	34	26	60	46	43	89			
38	80.10	9.06	4.17	6.67	9.5	79.0	144	81	225	22.0	60.0	14	12	26	79	46.5	125.5			
39	68.00	16.29	5.22	4.49	12.5	78.0	181	98	279	23.0	62.4	84	66	150	132.5	82	214.5			
40	60.05	25.24	7.01	7.70	13.1	85.0	120	111	231	21.0	55.8	58	30	88	89	70.5	159.5			
41	65.19	14.06	8.08	12.67	13.0	68.9	194	137	331	22.1	67.0	57	36	93	125.5	86.5	212			
42	50.00	40.00	5.23	4.77	14.9	60.1	80	60	140	20.5	73.8	15	6	21	47.5	33	80.5			
43	30.12	60.00	4.00	5.88	12.0	68.3	44	22	66	23.5	73.0	16	22	38	30	22	52			
44	61.87	23.32	2.55	12.26	14.7	67.0	63	66	129	16.9	83.8	85	80	165	74	73	147			
45	80.99	9.26	2.99	6.76	13.0	78.0	35	18	53	18.0	80.6	4	12	16	19.5	15	34.5			
46	90.01	1.35	1.81	6.83	11.0	77.0	10	8	18	23.5	75.5	32	20	52	21	14	35			
47	50.09	6.01	5.00	38.90	9.1	85.0	46	36	82	25.7	49.0	90	61	151	68	48.5	116.5			
48	58.98	29.05	4.90	7.07	8.5	88.0	28	18	46	23.0	57.0	20	26	46	24	22	46			
49	70.76	13.03	6.98	9.23	12.0	80.0	82	60	142	21.9	69.8	80	71	151	81	65.5	146.5			
50	76.32	13.50	4.12	6.06	12.9	83.5	44	40	84	22.0	74.0	16	11	27	30	25.5	55.5			
51	64.95	18.73	4.76	11.56	12.6	81.1	30	18	48	17.0	87.0	10	9	19	20	13.5	33.5			
52	65.85	18.31	5.98	9.86	13.0	78.8	21	12	33	12.9	88.0	12	7	19	16.5	9.5	26			
53	75.66	13.53	1.90	8.91	10.8	90.0	26	9	35	15.6	76.0	74	77	151	50	43	93			
54	76.98	13.04	1.40	8.58	12.0	81.5	18	24	42	19.9	73.1	32	20	52	25	22	47			
55	80.01	6.03	1.00	12.96	15.9	75.0	12	16	28	17.8	72.7	29	12	41	20.5	14	34.5			
56	82.03	6.20	3.09	8.68	17.5	74.1	36	20	56	17.0	60.6	10	2	12	23	11	34			
57	79.06	8.30	5.99	6.65	14.0	81.0	56	48	104	13.0	90.0	14	9	23	35	28.5	63.5			
58	71.11	13.06	6.89	8.94	18.3	70.0	40	22	62	11.9	93.2	28	12	40	34	17	51			
59	70.20	15.74	6.34	7.72	16.4	65.3	18	16	34	15.7	85.2	16	22	38	17	19	36			
60	40.56	39.62	8.00	11.82	17.5	53.8	35	23	58	20.1	74.8	7	11	18	21	17	38			
61	74.58	10.24	6.12	9.06	15.0	67.0	78	75	153	20.0	67.0	25	29	54	51.5	52	103.5			
62	50.75	35.65	4.00	9.60	14.8	74.9	22	10	32	23.1	63.0	25	17	42	23.5	13.5	37			
63	80.00	8.16	2.98	8.86	10.5	80.3	45	12	57	19.5	76.9	84	76	160	64.5	44	108.5			

72	44.87	42.16	6.00	6.97	15.3	78.3	60	54	114	25.0	62.2	45	22	67	52.5	38	90.5
73	10.00	85.00	1.93	3.07	19.5	75.0	16	8	24	19.0	70.5	26	20	46	21	14	35
74	29.09	61.00	4.09	5.82	18.0	65.8	15	14	29	18.7	76.0	18	12	30	16.5	13	29.5
75	75.00	7.45	1.31	16.24	18.0	51.2	12	11	23	16.5	79.1	19	6	25	15.5	8.5	24
76	80.34	10.86	3.05	5.75	16.5	55.5	42	16	58	20.2	78.5	36	10	46	39	13	52
77	70.91	15.23	5.09	8.77	14.7	69.0	52	37	89	20.5	68.0	23	2	25	37.5	19.5	57
78	75.03	7.80	6.99	10.18	17.7	64.7	7	6	13	22.8	70.0	41	31	72	24	18.5	42.5
79	79.00	7.54	6.28	7.18	18.5	70.5	17	6	23	25.6	71.4	28	25	53	22.5	15.5	38
80	83.00	5.67	4.00	7.33	20.8	70.0	66	60	126	24.0	72.8	88	46	134	77	53	130
81	5.21	82.12	2.00	10.67	21.0	61.0	21	9	30	20.0	70.0	18	12	30	19.5	10.5	30
82	20.09	70.10	3.65	6.16	22.0	73.2	33	12	45	21.9	65.2	24	10	34	28.5	11	39.5
83	45.67	40.87	4.01	9.45	19.0	65.0	9	13	22	25.5	73.6	26	24	50	17.5	18.5	36
84	38.87	53.00	3.01	5.12	22.3	76.8	54	40	94	23.0	77.0	30	23	53	42	31.5	73.5
85	59.91	18.97	7.98	13.14	16.5	80.0	41	14	55	19.0	78.9	52	78	130	46.5	46	92.5
86	75.67	10.00	6.22	8.11	17.9	62.3	11	7	18	23.0	72.3	61	50	111	36	28.5	64.5
87	67.99	7.00	8.88	16.13	15.4	62.2	19	6	25	21.2	70.4	54	55	109	36.5	30.5	67
88	69.11	6.18	18.90	5.81	14.1	77.8	34	15	49	19.0	80.5	41	19	60	37.5	17	54.5
89	15.43	2.33	58.95	23.29	19.6	59.0	29	14	43	24.0	70.0	44	30	74	36.5	22	58.5
90	21.09	61.01	5.00	12.90	21.0	70.0	48	54	102	23.0	67.0	10	1	11	29	27.5	56.5
91	40.99	48.15	4.87	5.99	19.0	66.6	61	55	116	21.0	79.0	12	30	42	36.5	42.5	79
92	49.23	37.11	3.69	9.97	17.5	70.2	38	30	68	17.8	74.2	58	47	105	48	38.5	86.5
93	67.12	15.27	7.21	10.40	16.0	79.5	20	10	30	-	-	-	-	0	20	10	15
94	65.00	6.88	10.00	18.12	14.2	77.8	32	24	56	19.5	77.0	29	27	56	30.5	25.5	56
95	18.99	1.19	38.00	41.82	13.3	77.1	46	33	79	23.1	70.9	58	40	98	52	36.5	88.5
96	8.09	1.27	70.07	20.57	15.1	80.5	18	20	38	22.0	65.3	36	22	58	27	21	48
97	65.00	18.45	4.00	12.55	16.0	80.1	9	7	16	19.6	62.1	20	14	34	14.5	10.5	25
98	78.99	11.35	2.54	7.12	13.0	73.0	14	10	24	22.0	70.0	16	19	35	15	14.5	29.5
99	18.98	70.00	3.10	7.92	11.9	79.5	14	20	34	18.8	69.3	28	6	34	21	13	34
100	45.05	37.09	8.14	9.72	17.0	77.7	40	29	69	22.8	66.8	88	63	151	64	46	110
101	57.21	30.12	6.00	6.67	16.7	73.6	34	16	50	23.9	75.5	82	50	132	58	33	91

F - females, M- males, T - temperature, H - humidity

Table 2. Descriptive statistics of *D. reticulatus* ticks density per 100 m²

Study period	Sex	n	Average density	SD.	Median	Min.	Max.
Spring	F	101	56.5	50.6	41.0	4.0	297.0
	M	101	38.4	34.8	25.0	1.0	219.0
	F+M	101	94.9	83.4	68.0	5.0	516.0
Autumn	F	100	58.9	51.3	41.0	2.0	248.0
	M	100	40.9	33.5	30.0	1.0	135.0
	F+M	101	98.9	83.3	67.0	0.0	370.0
Whole study period	F	101	57.5	44.4	43.5	3.0	224.5
	M	101	39.5	28.3	31.5	2.5	128.5
	F+M	101	96.8	71.7	73.5	5.5	311.0

SD - standard deviation, n - number of plots, F - females M- males, Min- minimum, Max - Maximum

Table 3. Average collections of *D. reticulatus* ticks in the Polesie National Park (square 41)

Year	Spring						Autumn					
	n	T [°C]	H [%]	F	M	F+M	n	T [°C]	H [%]	F	M	F+M
2014	6	11.8	77.2	62	41	103	3	23.0	72.2	80	68	148
2015	7	14.9	69.3	78	49	127	4	20.8	76.3	84	77	161
2016	6	12.8	81.0	61	50	111	4	18.1	68.2	108	70	178
2017	6	13.4	77.2	83	79	162	5	17.8	75.4	128	97	225
2018	3	12.7	79.0	124	111	235	2	21.0	68.0	155	120	275
2019	2	13.5	80.2	194	137	331	2	19.8	74.6	57	36	93

n- number of tick collection, T - temperature, H- humidity, F - females, M- males

Figures

Figure 1

Localization of the study area, based on Wikipedia [63, 64], changed

Figure 2

Visualization of grid squares and spatial distribution of seasonal activity peaks of *D. reticulatus* in Lublin Province. Data are presented as absolute values, see table 1.

Figure 3

Method of selection of *D. reticulatus* collection sites, plot 17, based on Google Maps [38], changed

Figure 4

Physico-geographical regions of Lublin Province, location of the weather stations, based on Kondracki [44], changed

Figure 5

Spatial distribution of the average density of *D. reticulatus* populations in Lublin Province

Figure 6

D. reticulatus ticks collected on an experimental field located in square 41. 96 adult specimens were harvested in an area of 10 m²