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Short note

Comparison of the body mass of *Dermacentor reticulatus* ticks from two ecologically varied habitats located in a close vicinity

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ABSTRACT. *Dermacentor reticulatus* as hematophagous arthropod has a great veterinary and epidemiological significance in Europe as a vector and reservoir of numerous pathogens including bacteria, rickettsia, viruses and protozoa. Because of biological and ecological traits, i.e. ability to survive prolonged periods of starvation, ecological plasticity, adaptability and resistance to fluctuating and adverse environmental conditions *D. reticulatus* ticks are an important parts of enzootic cycles, thus have been studied intensively since last years. Genetic studies of ticks collected in Poland suggest existence of the overlapping zone between two European populations of this species. The aim of this study was to compare the body mass (as reduced body mass) of *D. reticulatus* females and males collected in two plots, representing different ecological characteristics, located in close vicinity, placed in Lublin province (eastern Poland). The results indicate that the reduced body mass values of *D. reticulatus* adults collected in diverse microhabitats did significantly vary. The cause of this phenomenon is ambiguous, so it need to be elucidated in further studies.

Keywords: ticks, *Dermacentor reticulatus*, tick body parameter, tick population

Introduction

Dermacentor reticulatus ticks are, next to *Ixodes ricinus,* arthropods of the greatest medical importance in Europe. It is caused mainly by the ability of these species to maintain pathogens in enzootic cycles. Additionally, especially in case of *D. reticulatus*, it is favored by the wide range of hosts and an ability to adapt to fluctuating conditions in their habitats, that allow this species to survive in periods of adverse values of temperature and relative humidity or temporary lack of host [1,2]. *D. reticulatus* often occurs in habitats such as meadows, fallow lands as well as in urban and suburban and ecotone zones. An increase in the size of *D. reticulatus* population and an expansion into new territories in Poland and Europe have been observed since last decades [3–9]. This is probably the result of changes in agricultural structure what leads to increase in area of unused meadows and fallow lands. Other reasons of *D. reticulatus* expansion are fragmentation and deforestation of forests, ongoing urbanization as well as climate changes and increase in the size of potential hosts population, i.e. deer species [3,4]. Nowadays *D. reticulatus* occurs in the most of territory of Poland and published data indicates that the most numerous populations of this tick species are found in the eastern part of the country [7,10,11]. Results of genetic studies on expansion of *D. reticulatus* in Poland suggest that this territory is the overlapping zone between distribution of its two European populations – Western and Eastern [12–14]. Moreover, genetic studies have shown a moderate differentiation between ticks derived from different sites in Poland [14]. Observable signs of genetic variability among ticks can be visible as varied values of morphometric traits (weight and body size) as described by de la Fuente et al. [15] in case of *D. andersoni.*

The aim of this study was to compare the body mass of *D. reticulatus* adults (in term of the reduced

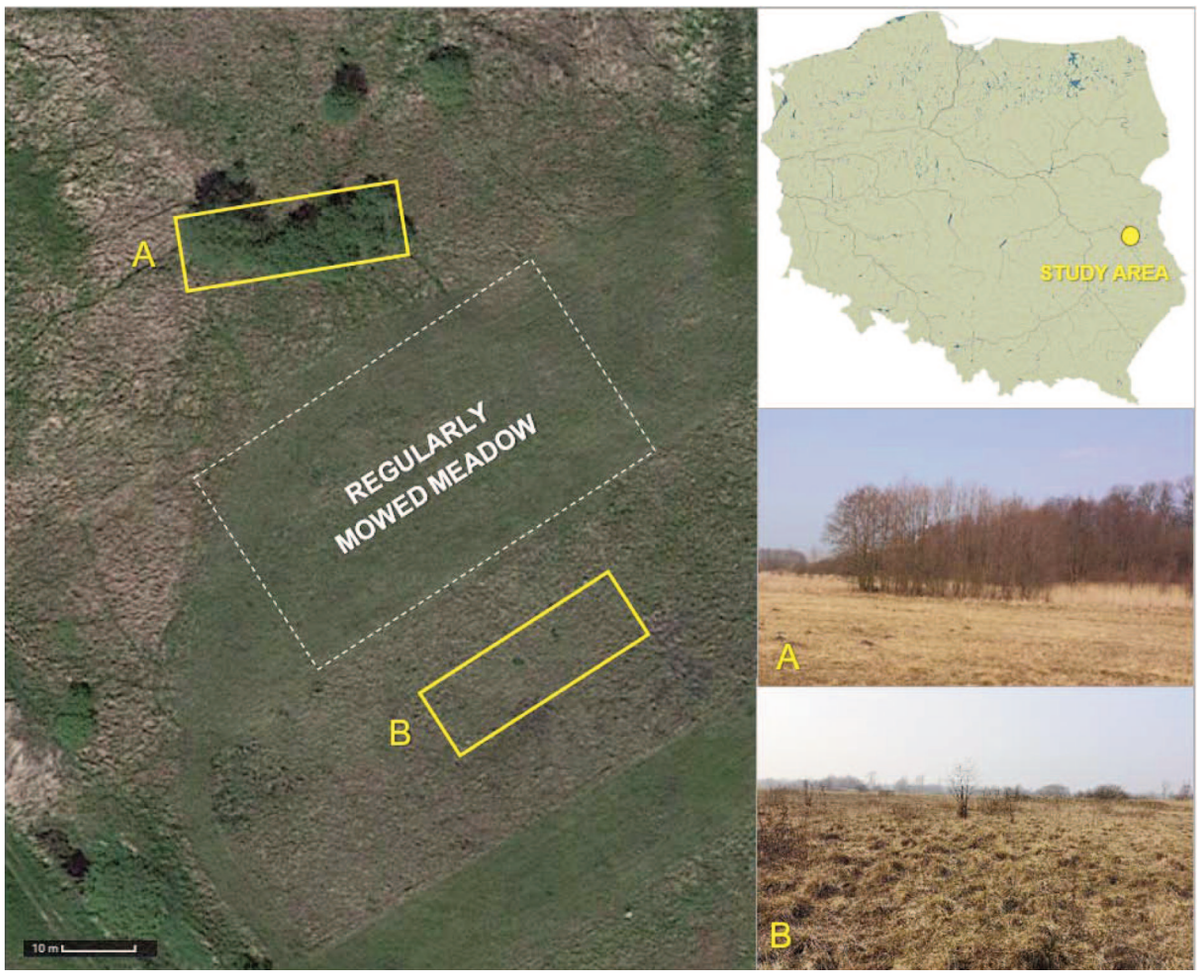

Figure 1. Location of the study area (plot A and B). Based on Google Maps [16] and Wikipedia [17] with own modifications.

body mass) collected in the ecologically divergent habitats localized in a close vicinity, placed in Lublin province.

Materials and Methods

Ticks collection and study area

Ticks were collected during the autumn activity peak (in November 2020), to assure representative sample of specimens for analysis. Ticks were collected in two plots of the approximately area of 300 m^2 each: A and B, localized in Zawieprzyce (51°35'N 22°76'E; Lublin province) (Fig. 1) [16,17]. Plots are located in a close vicinity but represent ecologically diverse habitats. Plot A consists mostly of community of shrubs and trees of *Salix* spp., *Alnus* spp. and *Betula* spp., while plot B represents communities of plants belonging to class Molinio-Arrhenatheretea with progressive ecological succession.

Ticks were collected with a flagging method [18] and placed in the plastic containers. Collected ticks were immediately transferred to a laboratory and placed in a low-temperature freezer at –80°C (ULTF, Arctico, Esbjerg, Denmark).

Laboratory processing of ticks

Frozen ticks were identified to species and sex with an identification guide compiled by Nowak-Chmura [18] and with the use of Zeiss STEMI DV4 stereoscopic microscope (Carl Zeiss Light Microscopy, Göttingen, Germany). Among the frozen ticks, independently for A and B plot, 70 females and 70 males were chosen by random, that were undergoing further processing.

Previously selected ticks had been drying (24h, 70°C) to remove water and obtain the value of reduced body mass (RBM). It is the most reliable parameter to determine body mass of analyzed specimens. Subsequently, each tick was individually weighted with the use of an AS.R2 Plus analytical balance (RADWAG, Radom, Poland) with an

Table 1. Reduced body mass (mg) of *Dermacentor reticulatus* adults collected in plot A and B

Processed ticks	Plot A		Plot B	
	F (mg)	M (mg)	F (mg)	M (mg)
1	2.29	2.72	3.29	2.41
2	1.98	1.46	2.50	2.48
3	1.81	2.79	2.82	2.66
4	1.98	2.53	3.28	1.62
5	2.18	2.31	2.58	2.43
6	1.79	2.08	2.22	3.01
7	2.17	1.96	3.58	1.78
8	1.83	2.41	2.49	2.14
9	1.88	2.15	3.15	2.43
10	3.28	0.91	1.91	2.45
11	2.52	1.17	3.00	1.76
12	2.13	2.41	4.06	1.74
13	2.38	2.88	2.26	1.69
14	2.20	2.30	2.56	2.43
15	1.64	2.47	2.34	2.51
16	2.50	2.07	2.28	1.00
17	2.22	2.72	2.30	3.21
18	2.15	2.34	3.47	2.27
19	1.43	2.68	3.24	2.43
20	1.87	1.25	2.77	1.42
21	2.08	1.53	2.28	1.56
22	2.24	2.40	2.04	1.36
23	1.78	2.80	2.44	3.06
24	2.12	2.35	2.75	3.56
25	2.10	1.81	2.96	3.00
26	2.02	1.93	2.00	3.03
27	1.93	1.26	2.04	3.03
28	1.94	1.42	2.80	2.78
29	1.71	0.97	3.25	2.64
30	1.77	1.25	1.74	2.86
31	1.86	2.74	2.51	2.27
32	2.09	1.90	2.88	2.14
33	1.31	2.81	3.12	1.86
34	1.48	1.98	3.15	2.31
35	2.54	1.14	2.66	2.18
36	1.45	1.71	2.78	2.66
37	2.10	2.67	2.52	2.14
38	2.59	1.71	1.99	2.13
39	0.92	1.35	3.03	1.78
40	1.71	1.84	2.12	2.52
41	2.41	1.60	2.70	1.81
42	2.18	1.69	3.01	2.06
43	2.79	2.55	3.20	2.20
44	2.02	1.08	3.06	1.15
45	1.87	1.27	2.77	2.23
46	2.49	0.89	2.74	1.97
47	2.42	2.81	3.52	1.90
48	1.96	1.67	2.42	1.96
49	1.85	1.38	3.61	2.17
50	1.38	1.92	2.43	2.27
51	2.23	2.40	1.63	1.28
52	1.47	1.09	2.56	1.87
53	1.53	1.15	2.07	1.15
54	1.50	1.28	2.42	1.67
55	2.08	2.43	2.49	1.24
56	1.44	1.83	2.78	2.71
57	1.38	1.73	2.59	1.66
58	1.00	1.49	2.53	0.88
59	1.03	1.77	1.57	1.91
60	1.08	1.82	1.45	1.43
61	1.52	2.34	2.67	2.63
62	2.26	1.72	2.27	0.89
63	2.25	2.84	2.12	1.92
64	1.81	1.46	2.10	1.46
65	1.49	1.56	2.36	1.36
66	1.30	2.31	1.90	1.42
67	1.55	1.49	2.23	2.21
68	1.97	1.44	1.36	1.92
69	2.02	1.04	1.35	1.33
70	1.91	0.94	2.15	2.71
Mean±SD	1.92±0.44	1.89±0.59	2.56±0.56	2.09±0.60
Min.	0.92	0.89	1.35	0.88
Max.	3.28	2.88	4.06	3.56
Median	1.95	1.83	2.53	2.14

Explanations: F – females; M – males; SD – standard deviation; Min. – minimal; Max. – maximal

accuracy of 0.01 mg.

Statistical analysis

The Mann-Whitney U Test was used to determine if there was any statistically significant difference in the RBM of *D. reticulatus* adults collected in A and B plot, and in the RBM value of females between plot A and B, and in the RBM of males collected in plot A and B. The Single Sample t-Test was used to analyze if collected females and males varied significantly among their groups because of the RBM value.

The value of $p<0.05$ was considered statistically significant. Statistical analysis was performed using the STATISTICA 10 PL statistical package (StatSoft, TIBCO Software Inc, Palo Alto, CA, USA).

Results

During this study 263 females and 220 males of *D. reticulatus* were collected, of which 108 females and 100 males were collected in plot A, and 155 and 120 in plot B, respectively.

The RBM of the *D. reticulatus* adults, collected in plot A and B, was significantly different, that was confirmed by Mann-Whitney U Test (z = -5.5179; p < 0.0001) (Tab. 1). Difference between the RBM of *D. reticulatus* females collected in plots A and B was statistically significant (z= -6.6202; p < 0.0001) (Tab. 1). The RBM of the *D. reticulatus* males collected in A and B plots varied significantly (z= 1.7774; p = 0.0375) (Tab. 1).

The RBM values of the *D. reticulatus* females and males collected in plot A ranged from 0.92 to 3.28 mg and from 0.89 to 2.88 mg, while in plot B from 1.35 to 4.06 mg and from 0.88 to 3.56 mg, respectively. An average RBM values of the *D. reticulatus* females and males in plot A reached 1.92 and 1.89 mg, while in plot B reached 2.56 and 2.09 mg, respectively (Fig. 1). The RBM values of females (t = -0.0654; p = 0.9480) and males (t = -0.0264; p = 0.9790) collected in plot A and females (t = 0.0043; p = 0.9966) and males (t = -0.0380; p = 0.9698) collected in plot B did not vary significantly among those groups of ticks (Tab. 1).

Discussion

The results of the studies that had been previously conducted in the same region indicate that activity and abundance of *D. reticulatus* ticks in area where this research was established is very high when compared to an average values for Lublin province and other parts of the Poland [7,11]. Moreover, an estimated size of the population of this species is higher than in habitats localized in suburbs of Lublin [10], what proves that environmental conditions in the study area are optimal for development of *D. reticulatus* ticks, thus the structure and the size of population assure reliable samples and results. The vicinity of plot A and B in a distance of 50 m ensure interval that prevents mixing of these two micropopulations, because the locomotor activity of *D. reticulatus* ticks is about 60 cm [19]. The presence of regularly mowed meadow between the study plots is an efficient barrier that limits an abundance and moving of ticks [20].

Comparison of the sizes of ticks in term of the RBM value limits an impact of relative humidity, that can cause short-term fluctuations in their body mass causing a water content change. Substances remaining after drying are mainly nutrients, as lipids and proteins and substances that are not exhausted during off-host stage, as e.g. chitin [21].

D. reticulatus ticks collected in plot B had significantly higher RBM value (z = -5.5179; p < 0.0001), than specimens collected in plot A. Statistical analysis indicated that this difference was more significant between groups of collected females (z = -6.6202; p < 0.0001), than males (z = -1.7774; p = 0.0375) (Tab. 1). The significantly higher RBM values of females and males collected in plot B (Tab. 1) probably indicates the presence in that microhabitat the factor that causes a beneficial effect on growth and development of ticks. Because of the ecological differences between microhabitats we can suppose that biotic and abiotic environmental conditions can favour diverse range of species as potential hosts for *D. reticulatus*. Available results of studies state that the host species is one of the most important factors influencing efficiency of females feeding, thus can affect development of the next tick generation [22]. The presence of potential hosts (e.g. rodents – their presence was confirmed in this locality [23]) can induce ticks–host–non-tick parasite interaction and affect tick feeding process and development [24]. The *D. reticulatus* adults with higher RBM could have had higher energetic reserves because of shorter activity period or could have been infected by pathogens that affect metabolism of lipids [25], but this preliminary study did not reveals these reasons. The possibility to complete the entire life cycle during one year by *D. reticulatus* does not

exclude its duration for few years with prolonged periods of starvation, what in turn leads to decrease in ticks body weight [2,26]. The structure of vegetation cover, which differs between plot A and B, plays an important role in the occurrence and activity of *D. reticulatus* ticks, regulating relative humidity, mitigating adverse environmental conditions and being a scene for displaying their host seeking behavior [3]. Nevertheless, differences in morphometric parameters as body size and mass could be a result of genetic diversity among ticks in local population [15]. Moreover, localization of the sampling sites in the overlapping zone between Western and Eastern European populations of *D. reticulatus* suggests existence of genetic variability among ticks [13,14].

Differences between the RBM values of *D. reticulatus* ticks collected in two microhabitats located in close vicinity suggest an impact of internal and/or external biotic and/or abiotic factors affecting ticks biology. Nevertheless, further investigations are required to reveal the reasons of this phenomenon.

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