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Original papers

Infestation of dairy cows by ticks *Dermacentor reticulatus* (Fabricius, 1794) and *Ixodes ricinus* (Linnaeus, 1758) in eastern Poland

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ABSTRACT. *Ixodes ricinus* and *Dermacentor reticulatus* are widely distributed tick species in Europe. The main hosts for immature stadia of these ticks are small rodents, mostly the genera *Apodemus* and *Microtus*, whereas for adult specimens – wild ungulates. These species of ticks are also found on companion and breeding animals including dairy cows. Researches were conducted in the eastern Poland from 2013 to 2014. During consecutive days bodies of cows grazing on the pasture were surveyed and all ticks were collected. Simultaneously ticks were collected from plants with flagging method. Significant preponderance of ornate cow ticks over bean castor ticks infesting cows was observed. The most frequent spot of the tick location on cows' bodies was neck. Statistically a significant impact of air temperature and humidity on the risk of tick's attacks on cattle grazing in open countryside and lack of difference between number of ticks collected from animals and plants had been confirmed. Dairy cows could be considered as one of the preferred hosts for adult specimens of *D. reticulatus*. Grazing of dairy cows in the area of ticks' occurrence causes risk of tick infestation and possible tick-borne pathogen transmission and progression of vector-borne disease. In human case TBE (tick-borne encephalitis) could be transmitted by ingestion of infected milk.

Keywords: tick infestation of dairy cows, ticks, *Dermacentor reticulatus*, *Ixodes ricinus*

Introduction

Ornate cow ticks, *Dermacentor reticulatus* and castor bean ticks, *Ixodes ricinus* (Ixodida: Ixodidae) belong to species with wide range of hosts. Co-occurrence of these tick species on the same host is commonly observed [1]. This is favoured by similar air temperature and humidity requirements and to a lesser extent by habitat preferences of both tick species [2,3]. Co-feeding of the ticks of different species increases their feeding success [4] and favours possibility of pathogen transmission between them [5].

In Europe, immature forms of *D. reticulatus* and *I. ricinus* attack mostly small rodents, primarily murids as *Apodemus agrarius*, *A. flavicollis*, *A. sylvaticus* and voles, i.e. *Microtus arvalis*, *M. oeconomus* and *Myodes glareolus* [6–9]. Nymphs and larvae of *I. ricinus* are regularly found on birds [10] whereas immature forms of *D. reticulatus* – intermittently [11].

Depending on the host availability, adult specimens of *D. reticulatus* and *I. ricinus* prefer middle sized and large ungulates, mostly game animals: European roe deer (*Capreolus capreolus*), red deer (*Cervus elaphus*) and wild boars (*Sus scrofa*) [1,12–14]. In Poland, to this group of hosts for adult specimens of the ornate cow ticks, elk (*Alces alces*) is also included [15].

Ticks *D. reticulatus* and *I. ricinus* often attack companion animals as dogs and cats [13,16,17]. In agricultural areas these ticks are found on breeding animals, mostly on sheep, cows, horses, buffaloes and donkeys [13,18,19].

Especially important for public health are cases of *I. ricinus* ticks (all developmental forms) attacking humans [20] and associated risk of the tick-borne pathogens transmission. This group of

microorganisms contains the most important, from epidemiological perspective, spirochetes of *Borrelia burgdorferi* s.l. and tick-borne encephalitis virus (TBEV) [21]. Human can become an accidental host for adult specimens of *D. reticulatus* [22,23].

Both, immatures and adults of *I. ricinus* and *D. reticulatus*, exhibit high flexibility in selection of their potential hosts. Because of the type of occupied habitat and associated fauna they can prefer diverse species of mammals as main hosts [6,24,25] and play role of the most important vectors of the infectious diseases in Europe.

The aim of this paper was investigation of the risk of tick infestation of dairy cows grazing on pastures in eastern Poland and analysis of potential hazards of humans' and animals' health.

Materials and Methods

The field study was conducted in eastern Poland (51.386°N, 22.981°E) in 2013 and 2014, during spring and autumnal peaks of activity of the *D. reticulatus* and *I. ricinus* ticks [26,27]. Parasites were collected from four, 4–5-year-old dairy cows (Holstein-Friesian type) weighing approximately 400 kg, marked as A, B, C and D, that were grazing on the 1 ha pasture. For 2 years preceding tick collection the pasture (Fig. 1) lied follow and none of the agrarian processes had been conducted there.

Animals were grazing from 7 a.m. to 7 p.m. and animals' bodies were surveyed during consecutive days at 5 p.m. Body part where ticks had been collected from was noted (ears, head, neck, front limbs, back limbs, back and abdomen). Ticks were collected from plant by flagging method simultaneously. To this end, a square 10 m×10 m was marked. Its surface was relevant to one cow daily graze. Before each collection of tick, air temperature and humidity were measured up to 0.1°C and 0.1%, respectively with use of Data Logger R6030.

Figure 1. Pasture, place of the graze of cattle, autumn 2013

Collected ticks were placed in containers with 70% ethanol. During laboratory study the species, developmental form and sex of ticks were identified with use of stereomicroscope Zeiss STEMI DV4 and guide of Estrada-Peña et al. [28].

Results were analysed with use of Statistica ver. 13 software. Statistical analysis included data of activity of ticks collected from plants, level of tick infestation of the particular cow (Kruskal-Wallis test, $p<0.05$), spot of location of ticks on the host body (Chi-squared test, $p<0.05$), impact of weather conditions on the ticks activity and ability of their attack on potential host (Spearman's rank correlation coefficient).

Conducted research did not required permission from the Local Ethics commission.

Results

During term of study significant preponderance of *D. reticulatus* ticks infesting cows over *I. ricinus* ticks was observed (129:1 respectively). Total number of 909 of specimens of ornate cow ticks were collected from animals (Table 1). The mean intensity of *D. reticulatus* females on cattle per day was higher during autumn (14.0±3.1) than spring (4.85±1.8). Similar, more males were collected from animals in autumn (11.5±2.8) than in spring (4.6±1.5). Significant difference of the *D. reticulatus* infesting particular cow was not confirmed ($H=0.349$, $p=0.951$). Preferred spot of their location on the hosts' bodies was neck (486 specimens, 267 females, 219 males), what was 53.46% of all collected ticks (Fig. 2). Similar level of infestation was observed on the front and back limbs (Table 2). Cattle's body part where the lowest number of ticks was collected are back, abdomen and ears (Table 3, Fig. 2). Statistical analysis confirmed significant different between body locations where the ticks of *D. reticulatus* were attached to hosts' skin $\chi^2=45{,}00$; $p<0{,}001$.

Level of the infestation of particular body part by ticks changed because of the year season. During autumn most of the *D. reticulatus* ticks (males and females) were found on the neck. During spring, specimens were collected from front and back limbs and neck that were infested similarly by males (Fig. 3).

The risk of the *D. reticulatus* ticks attack on

Table 1. The occurrence of *D. reticulatus* and *I. ricinus* ticks on dairy cows and their activity

Date	Average diurnal temp [°C]	Weather parameters during ticks collection		*D. reticulatus* collected from plants			*I. ricinus* collected from plants			*D.reticulatus* ticks collected from cows				*I. ricinus* ticks collected from cows			
		T [°C]	H [%]	F	M	Total	F	M	Total	Individual	F	M	Total	Individual	F	M	Total
12.10.2013	10.7	14.1	95.0	29	35	64	1	3	4	A	4	3	7	A	0	0	0
										B	7	2	9	B	0	0	0
										C	11	9	20	C	0	0	0
										D	2	10	12	D	0	0	0
13.10.2013	12.1	16.0	88.0	27	41	68	0	0	0	A	15	8	23	A	0	0	0
										B	7	12	19	B	0	0	0
										C	13	5	18	C	0	0	0
										D	6	4	10	D	0	0	0
14.10.2013	11.1	13.5	77.9	18	25	43	0	3	3	A	19	12	31	A	0	0	0
										B	25	9	34	B	0	0	0
										C	12	7	19	C	0	0	0
										D	7	5	12	D	0	0	0
15.10.2013	10.3	10.9	81.3	31	27	48	0	0	0	A	6	11	17	A	0	0	0
										B	23	11	34	B	0	0	0
										C	32	14	46	C	0	0	0
										D	20	15	35	D	0	0	0
16.10.2013	9.5	10.0	80.9	30	32	64	2	4	6	A	16	21	37	A	0	0	0
										B	19	15	34	B	0	0	0
										C	13	9	22	C	0	0	0
										D	11	16	27	D	0	0	0
17.10.2013	5.1	9.9	90.1	28	20	48	0	0	0	A	9	15	24	A	0	0	0
										B	11	11	22	B	0	0	0
										C	23	14	37	C	0	0	0
										D	19	15	34	D	0	0	0
18.10.2013	8.5	12.0	87.3	36	30	66	1	0	0	A	12	21	33	A	0	0	0
										B	16	13	39	B	0	0	0
										C	10	17	27	C	0	0	0
										D	25	17	42	D	0	0	0

Table 1. The occurrence of *D. reticulatus* and *I. ricinus* ticks on dairy cows and their activity

Date	Average diurnal temp [°C]	Weather parameters during ticks collection		*D. reticulatus* collected from plants			*I. ricinus* collected from plants			*D. reticulatus* ticks collected from cows				*I. ricinus* ticks collected from cows			
		T [°C]	H [%]	F	M	Total	F	M	Total	Individual	F	M	Total	Individual	F	M	Total
24.04.2014	–	–	–	–	–	–	–	–	–	A	3	0	3	A	1	0	1
										B	7	2	9	B	0	0	0
										C	0	0	0	C	0	0	0
										D	3	4	7	D	0	0	0
25.04.2014	14.1	23.1	77.3	11	9	20	4	0	4	A	8	1	9	A	0	0	0
										B	1	1	2	B	0	0	0
										C	4	4	8	C	0	0	0
										D	5	7	12	D	0	0	0
26.04.2014	15.3	22.1	69.0	20	17	37	3	7	10	A	6	8	14	A	3	0	3
										B	2	8	10	B	0	0	0
										C	6	3	9	C	0	0	0
										D	0	2	2	D	0	0	0
27.04.2014	13.5	19.8	70.0	14	6	20	0	0	0	A	4	0	4	A	0	0	0
										B	16	19	35	B	0	0	0
										C	4	1	5	C	2	0	2
										D	9	17	26	D	0	0	0
28.04.2014	14.2	21.7	82.3	21	14	35	1	0	0	A	7	9	16	A	0	0	0
										B	2	0	2	B	0	0	0
										C	0	0	0	C	0	0	0
										D	10	6	16	D	1	0	1

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

Table 2. Statistical analysis of dependence between number of questing ticks on pasture and ticks collected from animals

Dermacentor reticulatus								
	Cows			Pasture			Statistical analysis	
	Me	Md	SD	Me	Md	SD	Z	p
F	40.83	23.76	37.00	23.18	7.49	21.00	-1.479	0.139
M	34.42	20.23	31.00	23.27	11.08	25.00	-1.200	0.230
T	76.08	43.64	70.00	46.45	17.41	48.00	-1.449	0.147
Ixodes ricinus								
	Cows			Pasture			Statistical analysis	
	Me	Md	SD	Me	Md	SD	Z	p
F	0.58	1.00	0.00	1.09	1.38	1.00	-1.026	0.305
M	0.00	0.00	0.00	1.55	2.38	0.00	-2.236	0.025
T	0.58	1.00	0.00	2.64	3.20	1.00	-1.833	0.067

F - females, M - males, T - total, Me - mean, Md - median, SD - standard deviation

breeding animals in open countryside is significantly affected by air temperature and humidity (rho=0.377; p=0.023).

In autumn, an average of 57.2 adult *D. reticulatus* ticks were collected from vegetation and 1.8 of *I. ricinus*, while in spring 28.0 and 3.5 respectively (Table 1). There is a lack of the significant difference between number of *D. reticulatus* and *I. ricinus* ticks collected from plants and cattle (Table 3).

Discussion

Dairy cows are attacked by multiple tick species. In Europe the most frequently found ticks on those animals are *I. ricinus*, *D. reticulatus*, *Rhipicephalus bursa*, *R. turanicus*, *Hyalomma m. marginatum*, *H. lusitanicum*, *Boophilus annulatus*. Tick infestation

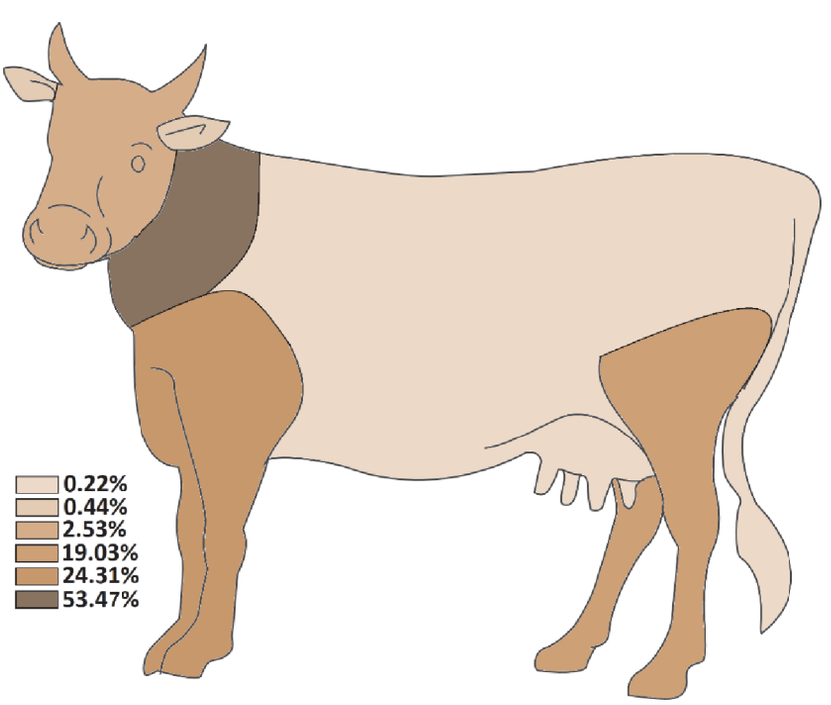

Figure 2. The percentage participation of location sites of *D. reticulatus* ticks on dairy cattle (Holstein-Friesian type)

Table 3. Number of *D. reticulatus* ticks collected from animals and location spots of ticks on dairy cows' body parts (Holstein-Friesian 1 type) during the study

Body part	Females	Males	Total
Ears	3	1	4
Head	10	13	23
Neck	267	219	486
Front limbs	103	118	221
Back limbs	112	61	173
Back and abdomen	0	2	2
Total	495	414	909

of dairy cows contributes to economic loss, primarily to decrease in milk and meat production and in extreme cases can lead to the death of breeding animal [23,29–36].

The results of our study exhibit substantial advantage of *D. reticulatus* over *I. ricinus* ticks in specimens collected in pasture and also in infestation level in cattle in study area. That level was higher in autumn than in spring (Table 1). These dependences are caused by seasonal activity of found ticks species, that in eastern Poland in natural habitats, present threefold higher activity in autumn [26,37,38].

The size of the tick population in given area largely depends on the presence of their potential hosts. During 2 years preceding our research the pasture was left as a wasteland where none of the agrarian processes and graze of animals were done. Additionally, in the study area beginning of the ecological succession had been observed – presence of volunteers of deciduous trees – mostly birch (*Betula pendula*). These conditions and vicinity of forest created possibility of migration of large and middle sized ungulates between the forest islands surrounding the study area. Group of these animals includes mostly elks (*A. alces*), European roe deer (*C. capreolus*) and wild boars (*S. scrofa*). No exploitation of meadow created possibility of evolvement of peculiar ecological niche and development of optimal conditions for rodents of the genera *Apodemus*, *Myodes* and *Microtus* that are competent hosts for immature forms of *D. reticulatus* [39,40]. Finally, those processes lead to the recolonization of agricultural areas by animals and are essential environmental factors influencing increase in size of the ticks populations [41]. Similar dependences, resulting in sevenfold increase in density of ornate cow tick population in the area of

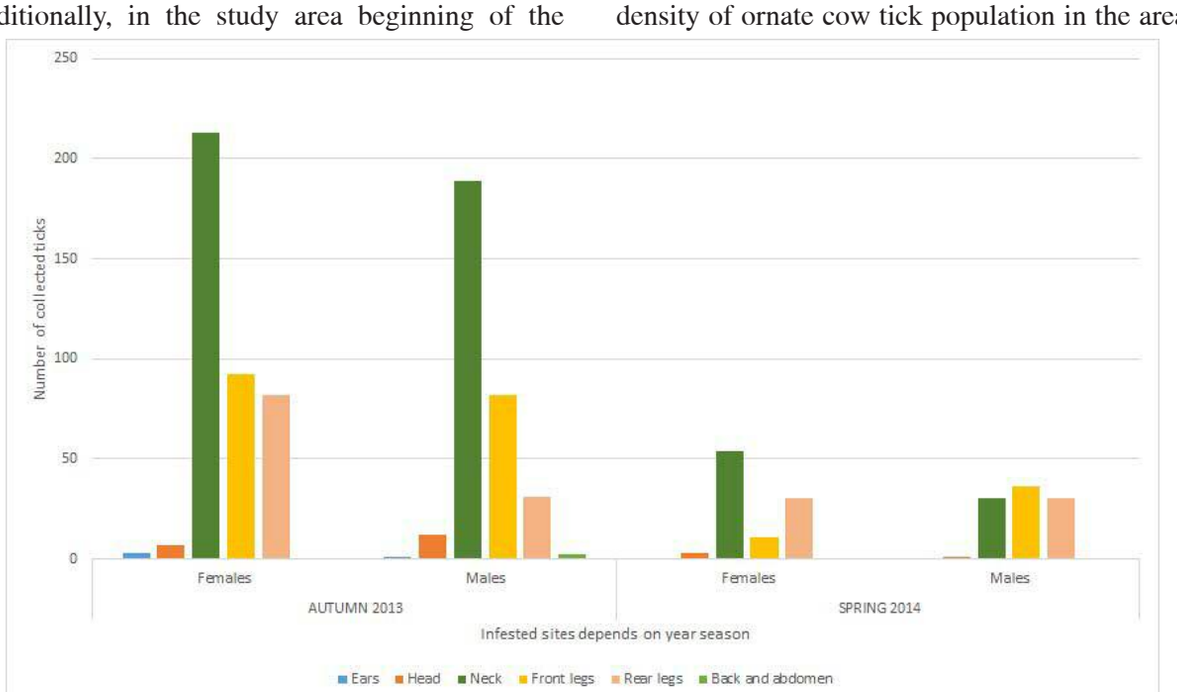

Figure 3. The occurrence of *D. reticulatus* on dairy cattle (Holstein-Friesian type) during seasons of the year

100 m^2 during 4 years, on wastelands were observed by Mierzejewska et al. [42]. Re-graze of cattle on pastures where ticks *D. reticulatus* occur can lead to the increase in size of their population. Dairy cows can be considered as one of the competent host species for adult specimens of ornate cow ticks [42]. High level of dairy cows infestation by *D. reticulatus* hovers 26 specimens per one cow and concurrent average number of the ticks collected from pasture is 57. Thus, the lack of the statistically significant difference between ticks collected from plants and animals. Those factors indicate high aggressiveness during questing a host (Table 1). All-day-long and sustained graze of cows infested by ticks without regular surveys of cattle's bodies can lead to detachment of engorged females in pasture, laying eggs and then originate of the new tick generation. It can raise the level of intensity and extensity of the infestation of cattle by ticks in the next seasons.

Weather conditions substantially influence ticks activity. Confirmed statistically significant impact of air temperature and humidity (Table 2) on the level of infestation of the dairy cattle with ticks reflect in other authors' papers considering seasonal activity and locomotor abilities [26,37,38]. Air temperature is the main factor determining activity of ornate cow ticks in eastern Poland, whereas air humidity plays a key role in locomotor activity control. Ticks *D. reticulatus* in opposition to some other species belonging to Amblyommidae family attacking cattle can cover modest distances (60 cm) during questing behaviour to the first attempt of attack [43].

Lower, and occurring in spring only, level of infestation of dairy cattle by *I. ricinus* ticks and lower number of hungry specimens collected in pasture (Table 1) are affected mostly by ecological type of the habitat. Open meadow ecosystem, despite the presence of potential host, does not create optimal environmental conditions for development of the immature and adult forms of the castor bean ticks. Vulnerability to fast and substantial loss of water by *I. ricinus* ticks causes that diel fluctuations of air temperature and humidity observed on the pasture (Table 1) greatly limit size of population of castor bean tick and their host questing ability [44].

Ticks *D. reticulatus* locate the most often on dairy cows' neck, afterwards on front and back limbs (Table 3, Fig. 2). The neck is beneficial spot for the tick attachment because is characterized by higher body temperature and blood supply. Moreover, this location prevent removing ticks by host. High percentage of tick attachment to the neck and limbs is caused by foremost contact of those body parts with vegetation during grazing, especially in high grass (Fig. 1). Similar arrangements of *I. ricinus* on cattle were reported by Hostis et al. [29] and Kiffner et al. [45,46] on wild animals.

Infestation of dairy cattle by *D. reticulatus* and *I. ricinus* ticks creates risk of transmission of tick-borne pathogens, e.g. *Borrelia burgdorferi* s.l. – etiological agent of bovine borreliosis [47]. This disease often affects whole herds, but a clinical picture is not completely clear [48]. The presence of joint oedema, body weight loss and fever suggest diagnosis toward borreliosis, especially if there was a risk of tick attacks on cattle. High level of infestation of dairy cattle with ticks can result in lowered (up to 2–3 l) milk production [13]. The presence of antibodies IgG anti-*Borrelia* in cow milk was detected in Poland [49] and Slovakia [50]. The possibility of human infection with *B. burgdorferi* spirochetes via milk ingestion is not clear. Literature data reports cases of infected cat ingesting cow's milk [51] and guinea pigs fed by infected mother [52].

Other bovine tick-borne diseases are e.g. bovine anaplasmosis (*Anaplasma marginale*) [53], bovine babesiosis (*Babesia bovis*) and tick-borne bovine fever (*Rickettsia* sp.). The herds of cattle grazed in eastern Poland are exposed to tick-borne pathogens. In *D. reticulatus* ticks collected in this area occurrence of *R. raoulti* (43.8%), TBEV (8.5%), *Babesia* sp. (2.1%), *B. burgdorferi* (1.6%), *A. phagocytophilum* (1.1%) were confirmed [54].

Infection with some of pathogens, i.e. TBEV, does not cause progression of disease in cattle but can be transmitted via milk ingestion into humans [55]. Milk-borne TBE outbreaks were reported from Russia, Austria, former Czechoslovakia and Hungary [56–59]. In the samples of milk collected in eastern Poland the presence of antibodies anti-TBE and RNA of TBEV were confirmed [60]. This is strongly advised to consume dairy products undergoing pasteurization that successfully exterminate TBEV [61].

Grazing of cattle in the area of ticks' occurrence causes risk of the infestation of animals with these parasites, the possibility of tick-borne pathogens transmission and can lead to progression of vector-

borne diseases, e.g. bovine borreliosis, anaplasmosis and babesiosis. Because of above-mentioned, systematic survey of animals' bodies is strongly recommended. Concurrently, drinking and eating pasteurized dairy products is advised because of public health and safety.

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