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## IXODIDAE TICKS OF SMALL RUMINANTS IN THE REGION OF PARVOMAI, SOUTHERN BULGARIA

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#### **Abstract**

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The species composition and the distribution of ticks from the family *Ixodidae*, invading domestic goats (*Capra aegagrus hircus*) and sheep (*Ovis aries*) near Parvomay (region of Plovdiv) were studied. The indicators of an invasion (extension invasion and intensity of invasion) and the seasonal changes in the invasion of goats and sheep by tick species were found out. The daily activity of *Ixodes ricinus* in a habitat near the Gradina village was observed. 637 specimens of goats and 810 specimens of sheep from 12 farms in different villages in the Parvomay Municipality were examined, the study was conducted in all four seasons. It is found out that goats and sheep in the regions investigated is invaded by 7 species of ixodidae ticks – *Rhipicephalus bursa*, *Rhipicephalus sanguineus*, *Ixodes ricinus*, *Dermacentor marginatus*, *Haemaphysalis sulcata*, *Haemaphysalis punctata* and *Hyaloma plumbeum*. The predominant invasive species in both animals is *Rhipicephalus bursa*. The maximum of invasion in both host species was identified – in the spring by *Ixodes ricinus* and *Dermacentor marginatus*; in the summer – the species *Rhipicephalus bursa* and *Rhipicephalus sanguineus*; in the spring and autumn seasons – by *Haemaphysalis sulcata* and *Haemaphysalis punctata*. Single specimens from the species *Hyaloma plumbeum* were found in March and November. Changes in the daily activity of ticks of the species *Ixodes ricinus* were reported – related to the fluctuations of temperature and relative humidity and the solar radiation.

Key words: goats, Ixodidae, sheep

#### Introduction

Ixodid ticks are widely spread in Bulgaria and in many parts of the world, in particular, the greatest species diversity is characteristic of the subtropical and tropical regions. They have important epidemiological and epizootiological significance as vector carriers and natural reservoirs of many major infectious and parasitic diseases in humans and animals. A very important biological feature of the ixodid ticks is that they take part of a transovarial and a three phase transmission of many groups of pathogens. Therefore, they maintain the natural foci of infection of pathogens both in conditions of a constant circulation of the exciter between the ticks and the vertebrates, and in cases of a long-term absence of a susceptible host (Balashov, 1967). This deter-

mines their importance for the health and economy as they are the subject of many studies (Sarbova, 1964; Pavlov et al., 1972a; Hoogstraal and Valdez, 1980; Papadopoulos, 1996; Milutinovic et al., 1997; Granström, 1997; Bouattour et al., 1999; Estrada-Pena and Santos-Silva, 2005; Dantas-Torres, 2010; Dumitrache et al., 2012).

In the 1950s in the region of the town of Parvomai, a natural focus of tick-borne encephalitis occurred (the village of Iskra). It drew attention to the distribution, species composition and biological cycle of ixodid ticks in the area of the infectious centre. Sarbova (1956), while researching tick-borne fauna in Iskra, established that the species *Ixodes ricinus* and *Hyaloma plunbeum* were the main carriers of the disease. Heiny and Rositskii (1962) studied the biocenoses in the area and found that the plant formations of oak, hornbeam and

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alder favour the development of Ixodes ricinus, which is an important factor in the formation of a potential natural focus of tick-borne encephalitis. In an epidemiological study of the disease in the village of Iskra, Rositskii et al. (1962) established that different species of rodents, insectivorous and lizards are hosts of nymphs and larvae of the ixodid ticks. Pavlov et al. (1972b) examined the species composition and seasonal dynamics of ticks of the family Ixodidae in a natural focus of viral encephalomyelitis in sheep and goats near the village of Dobrostan (located near the village of Iskra). According to the authors, the disease vectors are the species Haemaphysalis punctata and Dermacentor marginatus. After this period, for more than 40 years, in the region of the town of Parvomay only one study has been performed on the species composition and distribution of ixodid ticks on domestic animals (cattle and goats) by Pushtinkova (1997). The found species were Dermacentor marginatus, Ixodes ricinus in goats and Hyaloma plumbeum, Haemaphysalis punctata and Boophylus calcaratus in cattle and Rhipicephalus sanguineus was common for both hosts. Insufficient data from the recent years and the fact that the climate changes could lead to drastic changes in the distribution of ixodid ticks (Estrada-Peña et Venzal, 2006), make the studies on species composition and distribution of these ticks urgent.

That is why the aim of this study was to establish the species composition, seasonal changes and the development of ticks of the family Ixodidae, parasitising sheep and goats near the town of Parvomay.

#### **Materials and Methods**

#### Field study area

The area of the city of Parvomay is located in the Plovdiv district in the eastern part of valley of Pazardzhik and Plovdiv within the Thracian valley. The geographical coordinates of the administrative centre are: 25° 14' E longitude and 42° 07' N latitude (fig. 1). The southern part of the studied region falls within the northern slopes of the Rhodopes. The landscape is diverse - flat to hilly and mountainous. The altitude of the area ranges from 110m to 800m. The town and its surrounding villages are situated on both banks of the river Maritsa. The climate is humid continental (Galabov, 1982) and is slightly influenced by the Mediterranean. The territory municipality falls within the only intersection area of the isolines of the lowest January and highest July temperatures in Europe, and this causes the significant temperature amplitudes - on an annual basis they exceed 70°C. The seasonal distribution of precipitation shows a spring and summer maximum in May and June and a minimum in August and September. The average annual rainfall amount is 520-550mm for the lowland areas and 750mm for the higher parts of the area (the average amount for the area is 600mm). Depending on the location and hydrological conditions in the district, different types of vegetation are formed. The majority of the territory is occupied by crop plantations and meadows. Along the rivers Maritsa and Mechka, prevail willow and poplar riverside forests and dense vegetation. In the hilly areas, the natural oak vegetation prevails (Gruev and Kuzmanov, 1994).

#### Tick collection

The parasitic invasion on 637 goats and 810 sheep was studied. Animals are owned by 12 farms in six villages of the Parvomay municipality – the towns of Parvomay, Gradina, Byala Reka, Pravoslaven, Dragoynovo and Bukovo. The hosts were investigated twice a month, in all four seasons. The scrutiny of the animals for invasion was performed in the following order: head (between the horns and ears), neck, back, under the tail, groin, udder.

After the collection, the tick samples were separately stored in 70% ethanol and labelled with the date and the field name until the species determination. Tick species were identified under a binocular magnifier (Karl Zeiss) according to the identification keys (Pomerantsev, 1950, Estrada-Pena et al., 2004).

The quantity determination of the invasion of hosts by ixodid ticks was represented by the following parameters:

- The density of the invasion among hosts (extension invasion). It is the percentage of occurred parasitic invasion of the investigated ovine and caprine animals in the area as a whole and of the settlements in it. It is determined by the formula: number of invaded hosts / number of examined hosts x100;
- The frequency of occurrence of arachnids (intensity of invasion). It represents the number of ticks of one species found on one invaded animal (sheep or goat).

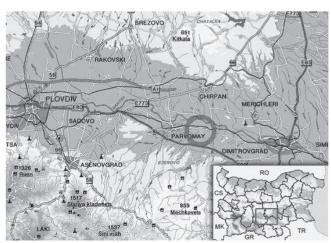


Fig. 1. Location of the field study area

The size and daily activity of *Ixodes ricinus* were determined using the white flag method (Pavloskiy, 1959). In a habitat near the village of Gradina were collected and tested 1 337 individuals (600 males, 562 females and 175 nymphs).

The study was conducted in 1999-2000 and in 2009-2011.

#### Results

# Species composition and invasion indices of ixodid ticks invading on sheep and goats in the region of the town of Parvomay

7 ixodid tick species were found – Rhipicephalus bursa, Rhipicephalus sanguineus, Ixodes ricinus, Dermacentor marginatus, Haemaphysalis sulcata, Haemaphysalis punctata and Hyaloma plumbeum (Table 1). The predominant species, Rhipicephalus bursa, parasitizes on both goats and sheep. It was established in all investigated settlements and the corresponding indices of the extension of invasion were 17.4% for goats and respectively 22.1% for sheep; the intensity of the invasion indices was 1-7 for the goats and 1-8 for the sheep.

Differences in the invasion of sheep and goats in relation to the different altitudes of their breeding places were found. In lowland and foothill locations (Gradina, Parvomay, Byala Reka and Pravoslaven), the density and the frequency of occurrence of Rh. bursa and Rh. sanguineus were much higher compared to places of more than 600 meters above sea level (Dragovnovo and Bukovo). Dermacentor marginatus and Hyaloma plumbeum were found in sheep and goats inhabiting areas with an altitude above 600 meters (Dragoynovo and Bukovo). Dermacentor marginatus individuals were found only on single individuals of sheep and goats which were led to graze in damp, overgrown with thick bushes places in the lowlands (Byala Reka and Pravoslaven). Haemaphysalis sulcata and Haemaphysalis punctata were not found in lowland settlements (the town of Parvomay and the village of Gradina) but only in the foothill places (*Haemaphysalis sulcata* - in the village of Byala reka and the village of Pravoslaven and Haemaphysalis punctata – in the village of Pravoslaven) and the highest places in the research area (the villages of Dragoynovo and Bukovo).

*The seasonal changes* of the invasion of both hosts have the following features:

Rhipicephalus bursa and Rhipicephalus sanguineus occur in the spring and in the summer (from March to August). Single individuals of these species were found in the autumn (September and October) and in the winter (December in sheep). The maximum of the invasion by both arachnids is reached in June.

Table 1
Ixodid ticks extension invasion (EI%) and intensity of invasion (II) on goats and sheep in the region of the town of Parvomay

Ixodid ticks	Rh. bursa		Rh. sanguineus		Ix. ricinus		Derm. marginatus		Haem. sulcata		Haem. punctata		H. plumbeum	
	EI %	II	EI %	II	EI %	II	EI %	II	EI %	II	EI %	II	EI %%	II
Gradina														
goats	12.5	1-5	21.66	1-5	59.2	3-5	0	0	0	0	0	0	0	0
sheep	28.75	1-6	11.25	1-4	37.5	1-2	0	0	0	0	0	0	0	0
Parvomay														
goats	27.27	1-5	22.72	1-5	14.6	1-2	0	0	0	0	0	0	0	0
sheep	39.6	1-7	14.19	1-5	0	0	0	0	0	0	0	0	0	0
Byala reka														
goats	31.11	1-7	25.55	1-6	15.6	1-3	10	1-3	5.55	1-2	0	0	0	0
sheep	50	1-8	23	1-5			6	1-3	9	1-2	0	0	0	0
Pravoslaven														
goats	19.95	1-5	20.95	1-5	10.5	1-4	18.09	1-5	4.78	1-2	10.48	1-2	0	0
sheep	25.71	1-6	14.81	1-4	5.93	1-3	16.3	1-4	8.88	1-3	9.62	1-2	0	0
Dragoynovo														
goats	8.41	1-4	9.35	1-4	8.41	1-2	42.99	1-8	1-3	1-3	9.35	1-3	2.8	1-2
sheep	12.14	1-4	7.86	1-3	6.43	1-2	30	1-6	10.28	1-3	5.71	1-2	4.28	1-2
Bukovo														
goats	9.52	1-4	11.42	1-4	6.66	1-2	36.19	1-7	5.71	1-2	7.62	1-2	3.81	1-2
sheep	11.42	1-4	8.33	1-4	5.83	1-2	28.33	1-7	7.5	1-2	8.33	1-2	2.5	1-2

Two distinct peaks of invasion – one in the spring with a maximum in March and April and one in the autumn, were reported for *Ixodes ricinus* (with a maximum in September), *Haemaphysalis sulcata* (with a maximum in October) and *Haemaphysalis punctata*.

*Dermacentor marginatus* can be found in the spring (March, April and May). Single individuals were found on sheep in the autumn (September) and on goats (September and October) and in the winter on goats (February).

In March and in October, only single individuals of *Hyaloma plumbeum* were found in hosts (sheep) bred in areas with an altitude above 600 meters (the villages of Dragoynovo and Bukovo).

### Changes in the daily activity of *Ixodes ricinus* in the habitat near the village of Gradina

As part of the present study, it was found out that ticks have the highest activity between 8 a.m. and 10 a.m., when the air temperature is moderate and the humidity is high. After these hours, along with an increase in the temperature and a decrease of the relative humidity, the activity of ticks decreases. In the interval from 1p.m. to 4p.m., only single individuals were found. After 4p.m., the tick activity is restored but after 6p.m., the arachnids head back to their shelters regardless of the temperature and the relative humidity.

During the period of an active search for a host, the arachnids would climb on grass plants or bushes at a height from 25cm – 50 cm to 1m from the soil surface, ready to attack. They would settle on top of a stem in a vertical position and on the bottom or top side on the leaves. When a host would approach, the ticks would occupy the characteristic position of "active waiting" – where the front pair of legs is moved up and forward and the second, third and fourth pairs are attached to the stem or leaves. In the absence of a host, ticks would maintain the condition of "passive position" by which the front legs were moved back.

#### **Discussion**

In the present study, *Rhipicephalus bursa* had the highest frequency among the collected tick species from both goats and sheep. This can be explained by the fact that it is a thermophilic species (Hoogstraal and Valdes, 1980) and the range of its distribution is mainly restricted to the Mediterranean region (Estrada-Pena and Santos-Silva, 2005). The climate conditions in the research area are influenced by the Mediterranean through the valley of the river Maritsa, which clearly has a beneficial influence on the development of this arachnid. The most important factor limiting the distribution of *Rh. bursa* is the decrease in humidity, and respectively

the precipitation (Yeruham et al., 1998). The average annual rainfall in the region is well above the critical minimum (300 mm) which would limit the development of this species. The decrease in the invasion of sheep and goats during the dry summer months generally coincides with the usual minimum of rainfall in the area in August and September. *Rhipicephalus bursa* is the most common tick in a neighbouring geographical area (Macedonia region in Greece) in domestic animals, including sheep and goats (Papadopolus et al., 1996).

**Rhipicephalus sanguineus** is the most widespread tick in the world (Dantas-Torres, 2011). In the present study, the widespread invasion of goats and sheep by this arachnid was also found, although its main host is the dog. The established increase of the activity of the arachnid in the spring and summer months and its very weak activity in the fall and winter resembles the biological activity of the arachnid under the influence of the Mediterranean climate. According to Dantas-Torres et al. (2010), the influence of the Mediterranean on the biological cycle of *Rh. sanguineus* is expressed by an intense nutrition and molting of the larvae and nymphs in the summer and egg production in the fall.

*Ixodes ricinus* is epidemiologically the most important arthropod in Northern Europe (Dumitrache et al., 2012). This tick is a vector of one of the most important thick-borne diseases except for tick-borne encepahalitis - Lyme disease, tularemia, babasiosis (Goddard, 2003) but also other pathogenic microorganisms. The invasion of *I. ricinus* by B. burgdorferi, various species Rickettsia, Babesia and Ehrlichia is found in various degrees in various European countries (Dumitrache et al., 2012).

It is a well known species, widely distributed in Central Europe and the British Isles (Gray, 1991). But it has an unequal distribution as a consequence of the high humidity requirement for survival (Estrada-Pena et Santos-Silva, 2005).

*Dermacentor marginatus* (another species established by this study) has been reported from countries in Europe and northern Africa (Giliot, 1985). This species is able to colonize several series of vegetation (mainly Quercus spp.) and can be found from the sea level areas to 800-1000m of altitude (Estrada-Peña et Santos-Silva, 2005). This explains its discovery by us mainly in areas with an altitude above 600 meters and natural oak vegetation.

The invasion of sheep and goats with other tick species (*Haemaphysalis sulcata*, *Haemaphysalis punctata* and *Hyaloma plumbeum*) is limited and depends on the altitude, humidity and temperature in the settlements in the region. It should be noted that *Hyaloma plumbeum* has high epidemiological significance because it is the main vector of tickborne encephalitis in the established focus of infection in the region, along with *Ixodes ricinus* (Sarbova, 1960).

To us, the differences in the invasion of goats and sheep by different types of ixodid ticks are due to the following maior reasons:

The type of the host. Rhipicephalus bursa manifests lower density and frequency in goats due to the lower susceptibility of this host to a tick invasion (Sarbova, 1964). On the contrary, Rhipicephalus sanguineus has higher values of the intensity of the invasion and extension invasion on goats than on sheep.

The way of animal breeding. The values of the intensity of the invasion and extension invasion on goats by *Ixodes ricinus* in the village of Gradina are very high compared to sheep, which is related to the fact that goats are led to graze in the area which is also a habitat of *Ixodes ricinus*.

#### **Conclusion**

The results, obtained in this study, generally coincide with the results of Pushtinkova (1997) and Sarbova (1956) who have carried out similar studies in the same area. There were, however, some differences, such as the predominant parasitic species and the number of the identified ixodid tick species. We consider that these differences are due to the scope of the study (which is different in the three studies within the Parvomay municipality) and to the difference in the studied hosts (goats, sheep and cattle).

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