TRADITION AND MODERNITY IN VETERINARY MEDICINE, 2023, vol. 8, No 1(14): 71–80

SPECIES COMPOSITION AND GEOGRAPHIC DISTRIBUTION OF HARD TICKS INFESTING GOATS AND SHEEP IN LEBANON

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ABSTRACT

Hard ticks are widely distributed parasites that pose a major threat to both livestock and public health. Minimal studies were done regarding the prevalence of ticks on goats and sheep in Lebanon. The aim of this study was to identify hard tick's species composition and their geographic distribution on small ruminants. The study took place in all Lebanese governorates. The ticks' genus, species, sex, and life stage were obtained via morphological identification using a stereomicroscope. A total of four genera and seven species were identified, being *Rhipicephalus turanicus*, *Rhipicephalus sanguineus*, *Ixodes ricinus*, *Haemaphysalis parva*, *Rhipicephalus bursa*, *Rhipicephalus annulatus* and *Hyalomma excavatum*. *R. turanicus* and *R. sanguineus* were the most abundant tick species, while *R. bursa*, *R. annulatus* and *H. excavatum* were minimally collected. *I. ricinus* and *Hae. parva* were identified for the first time on goats and sheep in Lebanon. Extensive research coupled with molecular analysis of the collected ticks is needed for further studies.

Key words: hard ticks, goats, sheep, Lebanon, species composition.

Introduction

Ticks are common obligate ectoparasites (Nicholson et al., 2019) of a wide range of hosts, most notably domestic animals. This makes them excellent and common vectors of blood borne diseases, including zoonoses.

Their survival is dependent on two factors (Randolph, 2008): animal hosts and their environment. A modification in one factor can impact that survival. Due to climate change, increase in agricultural land areas, urbanization, and increase in host numbers and displacement (Salman & Estrada-Peña, 2013), ticks have now spread to every corner of the world. They can be found at various altitudes, throughout the different seasons, and even in places that previously had unfavourable conditions for them to thrive in.

This growing distribution increases the propagation of tick-borne diseases, especially in the Mediterranean region, an area already suitable for a wide range of tick species. Moreover, different tick species serve as vectors for different diseases. Therefore, the Mediterranean region is a hotspot for tick-borne pathogens. Not only these pathogens and diseases lead to detrimental economical losses when large herds of ruminants are affected, but also, they pose an alarming public health concern.

Lebanon (ESDU, 2020), a Mediterranean country, has many regional climatic variations and a wide range of altitudes at which various tick species can survive. The general climate (ESDU, 2020) is Mediterranean and subtropical, with mild, humid winters and hot, dry summers. More than half of the country's landmass is targeted for agricultural practices. This makes Lebanon heavily reliant on small ruminant livestock, which in turn are hosts for several tick species. All these factors make Lebanon a suitable nidus for tick diversity and survival.

Nonetheless, there are minimal reports and studies on ticks and tick-borne diseases in the country. Only one recent study was conducted by (Dabaja et al., 2017) on cattle and small ruminants during June and September, in which seven different species were reported.

The main objective of this study is identifying tick species composition and their geographic distribution in Lebanon. The study specifically targets goat and sheep final hosts, which are important resources for the economy of the country.

Materials and methods

Lebanon: Geography, Climate and Biodiversity

Lebanon (ESDU, 2020) is a Mediterranean country located at approximately 34˚N, 35˚E. The land is divided into 9 geographic regions called governorates: Beirut, Keserwan-Jbeil, Mount Lebanon, North, South, Nabatieh, Beqaa, Baalbek-Hermel and Akkar.

Lebanon has a narrow coast bordered by the two mountain ranges: the Lebanon Mountains and Anti-Lebanon Mountains, which are separated by the Beqaa Valley.

The climate (Encyclopaedia Britannica Inc., 2021; ESDU, 2020) is Mediterranean and subtropical, with mild, humid winters and hot, dry summers. There are, however, large regional differences. The average temperatures range from the low 30s in July to mid-10s on the coast and low 10s in the Beqaa in January. As the altitudes increase, the temperatures decrease, and the rainfalls increase.

Lebanon has a rich fauna and flora; there are around 4,600 plant species and 4,400 animal species. Typical Mediterranean vegetation that can be found (Encyclopaedia Britannica Inc., 2021) includes but is not limited to oaks, pines, firs, and junipers. Lebanon has mostly small wild animals such as hedgehogs, squirrels, martens, dormice, and hares. It is also a stop for many migratory birds.

Around 66 percent of the total area (ESDU, 2020) consists of agricultural land, of which half of it is in the Beqaa, irrigated by the Litani River. The other half is dispersed among the coast and northern mountains that have reddish topsoil with high clay content, accounting for the fertility of the land. These lands herd numerous sheep and goats.

Experimental Plan

Ticks were collected from different breeds of both goats (Baladi, Saanen, Baladi Saanen mix, Shami and Alpine) and sheep (Turkish, Awassi and Bella) from farms in all governorates except Beirut, where there are no farms and no agricultural practices (Table 2). The collection spanned a total of 5 months. It started in December 2020, continuing in March, April and June 2021 and ending in July 2021. It took place at various altitudes, beginning at the low 226 meters and ending at the high 1500 meters.

Tick Collection

Ticks were removed at the attachment site using fine tip forceps. Most ticks were collected from the animals' ears where they were easily seen and quickly removed. Eppendorf tubes half filled with 70% ethanol were used to preserve the collected ticks. With a lead pencil, each tube was identified on the field before being placed in a small Ziploc bag stapled to an identification paper. The paper provided the required following data to be filled out: altitude, location, and date of the collection, collector's name, breed and species of tick host, number of ticks collected and part of the body from where they were collected.

Identification Method

Ticks were observed using a Motic® stereo microscope and a digital electronic stereo microscope. Tick's species, life-cycle stage and sex were identified using a range of keys (Estrada-Peña et al., 2004; Farkas et al., 2013; Nicholson et al., 2019; Roberts & Janovy, 2009; Walker et al., 2014).

Results

The total number of animals included in this study was 176, of which 107 were goats belonging to five different breeds (Baladi, Alpine, Saanen, Shami and Baladi Saanen mix) and 69 were sheep belonging to three different breeds (Awassi, Bella and Turkish). All specimens were outdoor, herding animals of various ages (Figure 3).

The collection took place in the 8 governorates of Lebanon, which are also grouped, for the purpose of this study, into 4 regions: Akkar and North Lebanon as the North Lebanon region, Keserwan-Jbeil and Mount Lebanon as the Mount Lebanon region, Beqaa and Baalbek-Hermel as the Beqaa region and South Lebanon and Nabatieh governorates as the South Lebanon region. The daily mean temperature and relative humidity of each region during the time of the study (WordData.info, 2022) are summarized in the table below (Table 3).

Table 2*:* Distribution of goats and sheep in the governorates of Lebanon with their respective altitude.

Governorate	Region	Nb of Farms	Altitude (m)
Akkar	Kobayat	2	700
North	Bechmezzine	2	275
	Hadath el Jebbeh	2	1500
	Tannourine	1	1500
Keserwan-Jbeil	Ain Kfaa	2	380
Mount Lebanon	Aabadiyeh	1	780
	Hlaliyeh	1	880
	Kahlounieh	1	800
	Ain Mouaffaq	1	750
	Barouk	1	1000
Baalbek-Hermel	Ras Baalbek	2	1000
	Qaa	2	650
	Baalbek	1	1100
Beqaa	Rayak	1	900
	Zahleh	1	1000
	Ablah	1	1000
	Terbol	2	900
South	Ghassanieh	1	300
	Qnaitra	2	300
Nabatieh	Khiam	1	700
	Marjaayoun	1	850
	Mazraat Doumiyat	1	250
	Blat	1	680
	Rab el Talathine	1	620

Table 3*:* Comparison between the different climatic features of the different regions visited during 5 different months (Weather and Climate 2021).

	December 2020: North Lebanon	March 2021: Mount Lebanon	March 2021: South Lebanon	April 2021: Mount Lebanon	June 2021: North Lebanon	June 2021: Beqaa	July 2021: North Lebanon	July 2021: South Lebanon
Daily mean temperature (degrees Celcius)	17	16	15	19	26	23	29	28
Relative humidity (%)	59	63	64	63	63	51	65	60

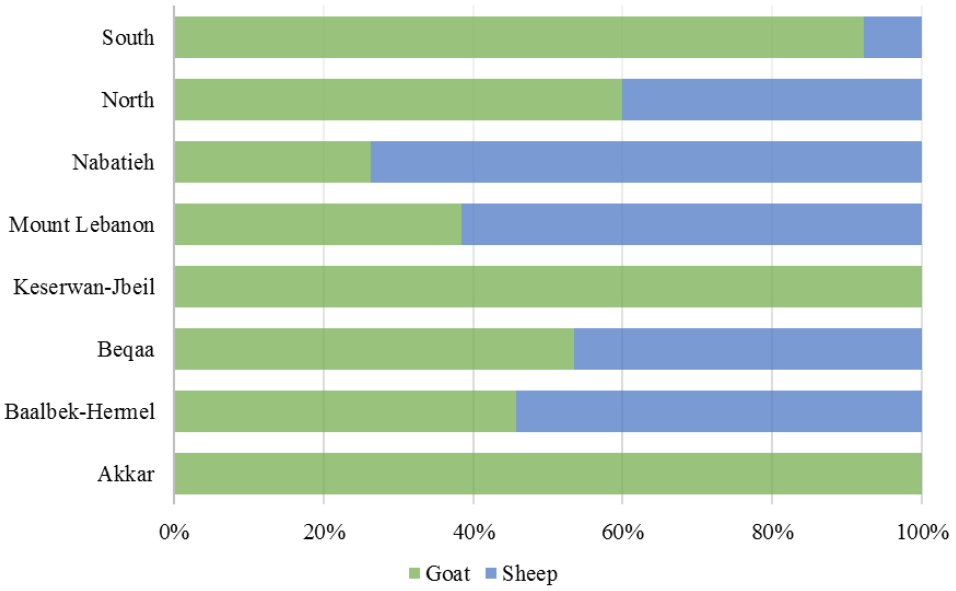

Figure 3*:* Graph Showing Distribution of Small Ruminants by Lebanese Governorates.

A total of 286 ticks were removed from goats, and 124 from sheep, totalling 410. Seven species belonging to the order Ixodidae were identified: *Rhipicephalus turanicus*, *Rhipicephalus sanguineus*, *Ixodes ricinus*, *Haemaphysalis parva*, *Rhipicephalus bursa*, *Rhipicephalus annulatus* and *Hyalomma excavatum* (Table 4).

Table 4*:* Ticks species composition according to host

Tick species	Animal Host		Total Number	Percentage (%)
	Goats	Sheep		
Hae. parva	13	10	23	5.61
H. excavatum	2	1	3	0.73
I. ricinus	43	0	43	10.49
R. annulatus	6	0	6	1.46
R. bursa	5	2	7	1.71
R. sanguineus	69	33	102	24.88
R. turanicus	148	78	226	55.12
Total percentage (%)	69.76	30.24	100	

Two tick species dominated at most regions, namely *R. turanicus*, and to a lesser extent *R. sanguineus. R. turanicus* was the most abundant at 55.12% (226/410) of the total collection of ticks. It was mostly collected in Keserwan-Jbeil (Table 5), in the Eumediterranean zone (500–1000 m altitude) (Table 6), and during spring (Table 7). The second most identified tick was *R. sanguineus*, with a prevalence of 24.88% (102/410). It was most abundant in the North, in the Eumediterranean zone, and during the summer season. *I. ricinus* made up 10.49% (43/410) of total tick collection. It was only collected in Akkar, in the Eumediterranean zone, during winter. *Hae. Parva* had a prevalence of 5.61% (23/410). It was most abundant in Akkar, in the Eumediterranean zone, during winter. *R. bursa* accounted for 1.71% (7/410) of total ticks. It was most abundant in Baalbek-Hermel in the Eumediterranean zone during summer. *R. annulatus*'s prevalence was 1.46% (6/410) and was only found in Nabatieh, in the Eumediterranean zone, during spring. *H. excavatum* was the least abundant at 0.73% (3/410) of total collection. It was collected in Baalbek-Hermel, in the Eumediterranean zone, during summer.

Table 5*:* Distribution of tick species per Lebanese governorates

Tick species	Akkar	Baalbek-Hermel	Beqaa	Keserwan-Jbeil	Mount Lebanon	Nabatieh	North	South
Hae. parva	13	0	0	0	0	0	10	0
H. excavatum	0	2	0	0	1	0	0	0
I. ricinus	43	0	0	0	0	0	0	0
R. annulatus	0	0	0	0	0	6	0	0
R. bursa	0	5	2	0	0	0	0	0
R. sanguineus	0	20	6	13	6	24	33	0
R. turanicus	0	21	25	85	58	9	11	17
Total	56	48	33	98	65	39	54	17
%	13.66	11.71	8.05	23.9	15.85	9.51	13.17	4.15

Table 6: Distribution of Ticks According to Altitude

Tick species	Thermomediterranean zone (0–500 m altitude)	Eumediterranean zone (500-1000 m altitude)	Supramediterranean zone (1000–1500 m altitude)
Hae. parva	10	13	0
H. excavatum	0	3	0
I. ricinus	0	43	0
R. annulatus	0	6	0
R. bursa	0	6	1
R. sanguineus	19	49	34
R. turanicus	107	109	10
Total	136	229	45
%	33.17	55.85	10.98

Table 7: Obtained tick species from December 2020 to July 2021

Tick species	Winter			Spring			Summer		
	Dec	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug
Haemaphysalis parva	23	0	0	0	0	0	0	0	0
Hyalomma excavatum	0	0	0	0	1	0	2	0	0
Ixodes ricinus	43	0	0	0	0	0	0	0	0
Rhipicephalus annulatus	0	0	0	6	0	0	0	0	0
Rhipicephalus bursa	0	0	0	0	0	0	7	0	0
Rhipicephalus sanguineus	0	0	0	10	15	0	26	51	0
Rhipicephalus turanicus	1	0	0	0	143	0	73	9	0

Discussion and conclusion

In this study, the most abundant tick was *Rhipicephalus turanicus*, which is an exophilic three-host species (Estrada-Peña et al., 2004) that thrives in the warm climate of the Mediterranean region where it primarily infests sheep. It was mostly collected during spring in the month of April (64.55% of total collect), from the Mount Lebanon region, where the daily mean temperature was 19 degrees Celsius and relative humidity was 63%. The remaining ticks were all collected during summer (most notably in the month of June). *R. turanicus* was found almost equally in both the Thermo- (47.34%) and Eumediterranean zones (48.23%). This finding is in agreement with a study done by Dabaja et al. (Dabaja et al., 2017) in Lebanon during June and September where *R. turanicus* was only collected during the former, at altitudes ranging between 270 and 960 meters. The results are also in agreement with studies made in neighbouring countries Cyprus (Campbell et al., 1974) and Palestine (Yeruham et al., 1996), where *R. turanicus* was also collected during the Mediterranean spring and summer seasons, which have all the optimal factors for it to thrive. No ticks were collected during the winter season when the temperatures are too low for it to survive (Guglielmone et al., 2016). *R. turanicus* is the most prevalent tick of both ruminant hosts in the current study. It has also been previously found on both hosts in Lebanon (Dabaja et al., 2017), neighbouring Arab countries (Perveen et al., 2021) and Cyprus, and its numbers have been rapidly increasing for the past decade. Its abundance in the region raises concern for its role as a vector of diseases, especially for zoonoses, which implicate public health. *R. turanicus* was proven to transmit spotted fever group rickettsioses (SFGR) in a study in Lebanon (Fernández de Mera et al., 2018), Palestine (Ereqat et al., 2016) and Cyprus (Chochlakis et al., 2011). SFGR can cause severe rashes, fever, and headache in humans.

Rhipicephalus sanguineus, whose primary host is the dog, is a three host species (Estrada-Peña et al., 2004) that is widespread in the Mediterranean region where ruminants in close contact with canines can become infested with it. It has been previously identified in Lebanon (Dabaja et al., 2017), Jordan and Syria (Qablan et al., 2012) on livestock. In this study, all visited livestock were guarded by herding dogs, which most probably served as a reservoir for these ticks. This could explain why it was the second most prevalent tick species on both hosts. It was mostly collected during summer (75.49%), most notably in July. It was most abundant in the Northern region where the daily mean temperature was 29 degrees Celsius and relative humidity was 65%. It was also found in lower numbers during the spring in the Mount Lebanon region, where the humidity was 63%. The temperatures however were averaging between 16 and 19 Degrees Celsius. This endophilic tick is known to adapt to living indoors, which helps it evade the lower temperatures. *R. sanguineus* was collected at all altitudes, most abundantly at the range of 500 to 1000 meters (48%) where it is still humid in the summer, so optimal for tick survival (Estrada-Peña et al., 2004). This is in accordance with the earlier study made in Lebanon (Dabaja et al., 2017), where *R. sanguineus* was only collected in the summer, at an altitude ranging from 270 to 963 meters. Other studies conducted in Cyprus (Campbell et al., 1974) and Palestine (Yeruham et al., 1996) shared similar results. Similarly to *R. turanicus*, *R. sanguineus* also transmits SFGR. It also transmits *Babesia* protozoa to humans (Brites-Neto et al., 2015) causing human babesiosis.

During identification, *R. turanicus* and *R. sanguineus* were sometimes hard to differentiate from each other. These two species share similar morphological characteristics and have minimal differences, except for the different spiracle plate shapes which were still oftentimes hard to differentiate. These two ticks still represent the most abundant species in this study (81.97% of all ticks).

Ixodes ricinus is a three-host tick known as the sheep tick or wood tick in Europe (Estrada-Peña et al., 2004). It is typically found in the cool humid European woodlands. This tick is not endemic to the Mediterranean region (Spickler, 2009) which is warmer and drier than preferable. To the best of our knowledge, this is the first report of *I. ricinus* on small ruminants in Lebanon. It was only collected during December in Kobayat, Akkar at an altitude of 700 meters. The temperature and relative humidity of the region were 17 degrees Celsius and 59% respectively. This is in agreement with a study made in Northern Algeria (Dib et al., 2009), a Mediterranean region that has a climate similar to Akkar, where the species was only collected during December as well.

These ticks were found in a region with heterogeneous deciduous woods, good shrub cover and adequate litter depth. These factors create a suitable microclimate (Estrada-Peña, 2001) for these ticks to thrive in, and even if relative humidity is not high enough to suit the species, it is in fact higher at the ground level (Spickler, 2009). This implies that a thorough search at the base of vegetation coupled with tick dragging might result in a collection of adults *I. ricinus*. All the collected ticks were nymphs, with the exception of 4 larvae. Since at least one female *I. ricinus* needs to lay eggs that hatch into larvae, their presence proves that this species is established in the country, at least in the northern part of it (in the Akkar governorate bordering Syria, where there is a lack of data and studies on ticks in the latter). *I. ricinus* poses a new threat to public health because it is the major vector of Lyme disease. Similarly to *R. sanguineus*, *I. ricinus* also transmits human babesiosis (Brites-Neto et al., 2015). Furthermore, *Ixodes* spp. can transmit rickettsia (Estrada-Peña et al., 2012) like *Anaplasma phagocytophilum*, which causes human granulocytic anaplasmosis, and viruses such as tick-borne encephalitis virus, which causes tick-borne encephalitis in mammals.

Haemaphysalis parva is a three-host exophilic tick (Guglielmone et al., 2016; Walker et al., 2014) of domestic animals. It is usually found in the Mediterranean region. In this study, it was collected from both goats and sheep, and mostly in Akkar in the woods of Kobayat (56.52%) (700 meters). It was also found in Bechmezzine in the North (43.48%) (275 meters). It made up around 5.6% of total collected ticks. Those results are in agreement with a study done in Palestine (Ereqat et al., 2016) where *Hae. parva* was also collected from both hosts, with the low prevalence of 8.9%. It marked its first appearance on small ruminants in Lebanon in the present study, however it has been previously reported (Raad et al., 2020) on a human host. Similarly to *I. ricinus*, it was only collected during winter in the Northern region, where the temperature and relative humidity were also 17 degrees Celsius and 59% respectively. This is in accordance with another study conducted in Jerusalem (Uspensky, 1998) where *Hae. parva* was only collected between January and February. *Hae. parva* poses a threat to the public health since it can transmit Lyme disease and babesiosis (Orkun et al., 2020). Similarly to *R. turanicus* and *R. sanguineus* (Ereqat et al., 2016), it can also transmit SFGRs.

Although low in numbers and statistically insignificant, *Rhipicephalus bursa*, *Rhipicephalus annulatus* and *Hyalomma excavatum* were also reported. *R. bursa* is a two-host tick of livestock (Estrada-Peña et al., 2004) that is abundant all throughout the Mediterranean, but is less thermophilic than other *Rhipicephalus spp.*, so it is usually present at altitudes higher than 600 meters. *R. bursa* was only collected during the month of June most notably at the altitude of 657 meters (71.42%). This is in accordance with a study conducted in Lebanon (Dabaja et al., 2017) where *R. bursa* was only collected in June at an altitude of 650 meters. *R. annulatus* which is also known as the cattle tick (Estrada-Peña et al., 2004) is a one host tick that can parasitize goats and sheep. It prefers the Mediterranean region's high temperatures and humidity with a peak in activity during the autumn. Only six *annulatus* were collected from one goat host. This result can be explained by the fact that

the autumn season was not included in the collection. Moreover, none of the visited farms herded cattle, the primary host of this tick. *H. excavatum* is a robust two or three-host tick that parasitizes livestock all throughout the Mediterranean region (Estrada-Peña et al., 2004), even adapted to the steppe climates of North Africa. Only three *H. excavatum* ticks were identified on one goat and one sheep. In one study in Cyprus (Chochlakis et al., 2011), 7.6% of total ticks were *H. excavatum* and in another one in Lebanon (Dabaja et al., 2017), 6.25% of total ovine ticks were *R. annulatus*. The limited number of ticks that were collected for this study could account for those differences.

Dermacentor marginatus, *Hyalomma anatolicum* and *Haemaphysalis punctata* were previously reported on livestock by Dabaja et al. (Dabaja et al., 2017) in Lebanon, however none of the three species were identified in this study. Instead, the most prevalent goat tick was *R. turanicus* which suggests a strong discordance with Dabaja's study in which *Hae. punctata* (7.7%) and *D. marginatus* (7.36%) were significantly more abundant than *R. turanicus* (5.16%) on small ruminants. *H. excavatum* and *H. anatolicum* could have been confused with each other in one or both studies since the two are closely related subspecies (Guglielmone et al., 2016; Perveen et al., 2021) that are both present in the region. On the other hand, *Hae. parva* and *Hae. punctata* could not have been confused since they have significantly different spur lengths on their 4th coxae (short for *parva*, long for *punctata*) and the number of festoons is different (9 for *Hae. parva* and 11 for *Hae. punctata*). This suggests that this truly is the first report of *Hae. parva* on livestock in Lebanon.

There are several limitations to this study. It was conducted in only five months, and every governorate was visited once in one season. No ticks were collected in the autumn season either.

More extensive research in each governorate at various altitudes in each season, would result in a much larger, varied sample. This could result in different percentages for each tick. For instance, the sex ratios would be different, and immature stages would be more numerous. Larger numbers of *I. ricinus* and *Hae. parva* are expected to be obtained, maybe in different regions during different seasons.

The current study serves as a starting point for further studies concerning the species that were identified, especially that some of them had not been previously reported on livestock in Lebanon. Since morphological identification can be subjective, molecular analysis of these ticks is recommended, as it can confirm all the doubtful samples. Moreover, it is important to study which pathogens these ticks carry. Control of the abundance of ticks, in addition to the study of the prevalence of associated tick-borne pathogens is mandatory in order to determine the risks of these diseases on public health, and to take actual precautionary steps to ensure the preservation of both human and animal health.

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