

Research Article

Distribution and identification of tick species on different breeds of livestock in Pakistan

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Citation

Habib Ullah, Muhammad Jamil, Ammar Anwar, Muhammad Imran, Faiqah Ramzan, Muhammad Zarar Khan, Shakirullah, Rashid Manzoor, Mubarak Ali and Norina Jabeen. Distribution and identification of tick species on different breeds of livestock in Pakistan. Pure and Applied Biology. Vol. 12, Issue 2, pp939-947.

<http://dx.doi.org/10.19045/bspab.2023.120095>

Received: 15/12/2022

Revised: 21/02/2023

Accepted: 23/02/2023

Online First: 15/03/2023

Abstract

The agricultural industry is an important part of Pakistan's economy, where the majority of rural population rely on livestock for income. Ticks and tick-borne diseases, on the other hand, are severely impeding livestock production. The identification and prevalence of tick species is very important in the country. Fifteen tick species belong to five genera identified in the current study were *Amblyomma gervaisi*, *A. latum*, *Dermacentor variabilis*, *D. marginatus*, *Hyalomma anatolicum*, *Hy. impeltatum*, *Hy. scupense*, *Hy. dtritum*, *Haemaphysalis montgomeryi*, *Hae. aciculifer*, *Hae. sulcata*, *Hae. punctata*, *Rhipicephalus sanguineus*, *R. annulatus*, and *R. turanicus*. *Hyalomma anatolicum* was most prevalent tick species on the animals in the region at prevalence rate of 16.01%. *Hyalomma* were more abundant (30.55%) followed by *Haemaphysalis* (30.31%), *Rhipicephalus* (22.39%), *Dermacentor* (10.41%), and *Amblyomma* (8%). The overall rates of tick infestation were 52.11, 64.57, 36.46, and 42.25%, respectively on buffalo, cattle, sheep, and goats. Udder and tail were the susceptible sites for tick attachment and neeli ravi, sahiwal, damani, and beetal of buffalo, cattle, sheep, and goat, respectively were highly loaded with tick species. The location, gender, age, breed of animals, and grazing systems are important factors in predicting tick infestations. Results will help to develop informed control measures of ticks.

Keywords: Identification; Livestock; Prevalence; Risk factor; Ticks

Introduction

Pakistan is a developing country and has agricultural land enriched with nutrients; located in South Asia, it is the sixth-largest populous country in the world. 64-75% of its

population is directly or indirectly associated with agriculture [1]. Therefore, agriculture serves as the spine of the economy of Pakistan, and livestock is deemed an integral part of it. It has been reported that 70% of the

population living in rural areas keeps livestock for their existence, and domestic animals are the primary source of income for poor people [2]. As the second most important sector, it generates 21.2% of the gross domestic product (GDP) and employs 45% of the total labor force [3]. In developing and under-developing countries, livestock is plagued by a parasitic infestation. Many poor farming communities in tropical and subtropical regions where more than 70% of animals are raised suffer immensely from vector and vector-borne diseases caused by ectoparasites and endoparasites [4, 5].

Tick is the major obligate, blood-sucking ectoparasite of terrestrial and aquatic life, especially birds and mammals. Ticks are a serious and risky pest of cattle, buffaloes, goats, and sheep all over the world, including Pakistan, except for frozen regions of the globe. Ticks can cause various clinical manifestations, such as body paralysis, anemia, and tissue injury during high infestation on hosts [6, 7]. Ticks directly or indirectly cause economic losses by spreading various bacterial, protozoal, and viral diseases to domestic and wild vertebrates [4, 5].

Nine genera, *i.e.* *Hyalomma*, *Amblyomma*, *Dermacentor*, *Haemaphysalis*, *Rhipicephalus*, *Boophilus*, *Argas*, *Ornithodoros* and *Ixodes* have been found in the country (Pakistan) [8]. Among reported genera, the *Hyalomma* is widely distributed and spreads infectious diseases, especially Crimean-Congo hemorrhagic fever (CCHF) and babesiosis to humans and animals [5]. According to Ramzan *et al.* [5], 53 tick species infesting mammals and humans reported in the scientific literature from different districts of Pakistan [8]. It is imperative to conduct a regular survey for inventory revisions due to increasing reports of geographical expansion of many tick species. In this study, we aimed to identify the tick species currently infesting domestic

animals across various agroecological zones of Pakistan with prevalence percentages.

Material and Methods

Study area and study design

A cross-sectional study was conducted in three districts of Pakistan, *i.e.*, Muzaffargarh, Layyah, and Dera Ismail Khan (D.I. Khan), from April 2020 to April 2022. Each district represents different agroecological zones of the country. Muzaffargarh and Layyah lie in hot summer and mild winter while Dera Ismail Khan. The mean annual temperature and the region's average rainfall are 25°C and 127 mm, respectively, with 35.5-67.6% relative humidity (RH). The summer is long and hot, while winter is short and cool in these areas. A multistage approach was used to collect ticks and identify animals for sampling, in which two sub-counties were purposefully chosen from each district based on environmental diversity. Following that, a random selection of one parish from each sub-county was made using the sampling frame provided by the local administrators. Based on geographical spread, 5 villages were identified from each parish. In addition, 5 households with livestock were chosen from each village based on the farmer's convenience and willingness to participate in the study. For tick collection, three animals from the herd were selected from each household based on the farmer's preference and/or those with visible ticks. Ticks were collected from 150 cattle in each district.

Tick collection and identification

The study areas were primarily located in hilly areas, so most of the animal grazing in these areas was infected by various ticks. During the study period, 2361 tick specimens were collected from different breeds of 900 buffaloes, 700 cattle, 523 sheep, and 565 goats. Different animal body parts (tail, brisket, back, testes, udder, ears, dewlap, and hooves) were examined for tick collection. Ticks were collected from each age of the host (kids, adults) and sex (male, female) in

study areas twice a month. Forceps were used to detach the ticks from the body of animals. Data related to the host's location, age, gender, and breed were recorded on a separate page and on the collected vials.

Preservation and identification

Specimens were collected in glass vials containing 70% ethyl alcohol, and collected specimens were brought to the parasitology lab for identification purposes. Ticks identified to species level under a stereomicroscope using morphological keys [9, 10].

Results

A total of 2361 tick specimens were collected from 2688 animals consisting of 900 buffaloes, 700 cows, 523 goats, and 565 sheep in the study areas. Fifteen tick species belonging to five genera were identified. Identified species were *Amblyomma gervaisi*, *A. latum*, *Dermacentor variabilis*, *D. marginatus*, *Hyalomma anatolicum*, and *Hy. impeltatum*, *Hy. scupense*, *Hy. dritum*, *Haemaphysalis montgomeryi*, *Hae. aciculifer*, *Hae. sulcata*, *Hae. punctata*, *Rhipicephalus sanguineus*, *R. annulatus*, and *R. turanicus* (Table 1). All

species identified in the current study belonged to the Ixodidae family. Not a single species of Argasidae and Nuttalliellidae were collected and identified in the present study. Among identified species, *Hyalomma anatolicum* was the dominant species with a 16.01% prevalence. *A. latum* was recorded least infected tick species with a 2.54% prevalence compared to other species, as shown in (Fig. 1).

Area-wise data showed that Dera Ismail Khan (42.52%) was a highly infested district, followed by Layyah (32.65%) and Muzaffargarh (24.81%). Out of 2361 ticks, 1004 were collected from Dera Ismail Khan, which proved the most suitable place for tick distribution (Table 2). Host-wise data showed that cattle carried the maximum ticks, followed by buffaloes, sheep, and goats. Overall tick prevalence was 64.57%, 52.11%, 42.25%, and 36.46% in cattle, buffaloes, sheep, and goats, respectively (Table 3). *A. gervaisi*, *Hae. montgomeryi*, *Hae. aciculifer*, and *Hy. detritum* was not collected from sheep and goats, while *Hy. scupense*, *D. variabilis*, and *R. turanicus* not collected from cattle and goats.

Table 1. Tick species collected from different hosts

Tick species	Buffaloes	Cattle	Goat	Sheep
<i>Amblyomma gervaisi</i>	Yes	Yes	No	No
<i>Amblyomma latum</i>	Yes	Yes	No	No
<i>Dermacentor variabilis</i>	Yes	No	Yes	Yes
<i>Dermacentor marginatus</i>	Yes	Yes	Yes	Yes
<i>Hyalomma anatolicum</i>	Yes	Yes	Yes	Yes
<i>Hyalomma impeltatum</i>	No	Yes	No	No
<i>Hyalomma scupense</i>	Yes	No	Yes	Yes
<i>Hyalomma detritum</i>	Yes	Yes	No	No
<i>Haemaphysalis montgomeryi</i>	Yes	Yes	No	No
<i>Haemaphysalis aciculifer</i>	Yes	Yes	No	No
<i>Haemaphysalis sulcata</i>	Yes	Yes	Yes	Yes
<i>Haemaphysalis punctata</i>	Yes	Yes	Yes	Yes
<i>Rhipicephalus sanguineus</i>	Yes	Yes	Yes	Yes
<i>Rhipicephalus annulatus</i>	No	Yes	No	No
<i>Rhipicephalus turanicus</i>	Yes	Yes	No	Yes

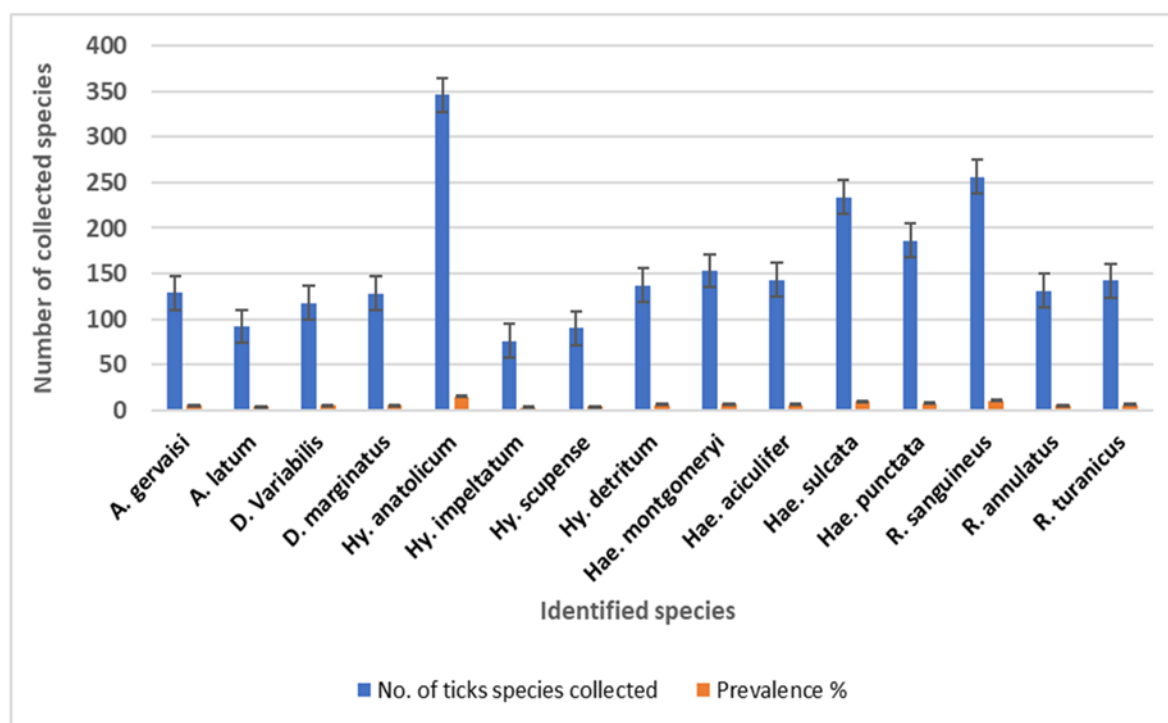


Figure 1. Prevalence of most dominant tick species

Table 2. Details of caught tick species of Pakistan from study areas

Tick species	Study areas/districts			Total	Prevalence %
	Muzaffargarh	Layyah	Dera Ismail Khan		
<i>A. gervaisi</i>	23	43	63	129	5.46
<i>A. latum</i>	12	0	48	60	2.54
<i>D. Variabilis</i>	15	37	66	118	4.99
<i>D. marginatus</i>	21	42	65	128	5.42
<i>Hy. anaticum</i>	99	147	132	378	16.01
<i>Hy. impeltatum</i>	0	26	33	76	3.21
<i>Hy. scupense</i>	27	38	42	107	4.53
<i>Hy. detritum</i>	34	47	56	137	5.8
<i>Hae. montgomeryi</i>	41	53	59	153	6.48
<i>Hae. aciculifer</i>	56	25	62	143	6.05
<i>Hae. sulcata</i>	64	77	93	234	9.91
<i>Hae. punctata</i>	43	69	74	186	7.87
<i>R. sanguineus</i>	71	86	99	256	10.84
<i>R. annulatus</i>	38	45	48	131	5.54
<i>R. turanicus</i>	42	36	64	142	6.01
Total	586	771	1004	2361	100
Percentage (%)	24.81	32.65	42.52	100	100

Table 3. Number of animals and infestation percentage observed during the study areas

Species	Buffaloes		Cattle		Goat		Sheep	
	Positive	%	Positive	%	Positive	%	Positive	%
<i>A. gervaisi</i>	8	0.88	2	0.28	0	0.00	0	0.00
<i>A. latum</i>	2	0.22	4	0.57	0	0.00	0	0.00
<i>D. Variabilis</i>	11	1.22	0	0.00	13	2.3	6	1.14
<i>D. marginatus</i>	15	1.66	21	3.00	6	1.06	14	2.67
<i>Hy. anatolicum</i>	79	8.77	81	11.57	49	8.67	47	8.98
<i>Hy. impeltatum</i>	0	0.00	14	2.00	0	0.00	0	0.00
<i>Hy. scupense</i>	23	2.55	0	0.00	25	4.42	38	7.26
<i>Hy. detritum</i>	43	4.77	31	4.42	0	0.00	0	0.00
<i>Hae. montgomeryi</i>	32	3.55	35	5.00	0	0.00	0	0.00
<i>Hae. aciculifer</i>	36	4.00	29	4.14	0	0.00	0	0.00
<i>Hae. sulcate</i>	53	5.88	57	8.14	36	6.37	29	5.54
<i>Hae. punctata</i>	59	6.55	61	8.71	34	6.01	31	5.92
<i>R. sanguineus</i>	67	7.44	75	10.71	43	7.61	33	6.3
<i>R. annulatus</i>	0	0.00	11	1.57	0	0.00	0	0.00
<i>R. turanicus</i>	41	4.55	31	4.42	0	0.00	23	4.39
Total	469	52.11%	452	64.57%	206	36.46%	221	42.25%

The majority of animals were infested with more than one tick species. The most preferable sites for tick attachment were the udder, followed by the tail, ears, external genitalia, testes, dewlap, and face (Table 4).

The current study showed that the cattle were highly susceptible to tick infestation. Udder is the most sensitive part of animals, so infected most as compared to other body parts.

Table 4. Predilection sites of animal for tick attach

Tick species	Predilection sites of host
<i>A. gervaisi</i>	Dewlap, Neck, udder, external genitalia, tail
<i>A. latum</i>	Ears, around eyes, tail, udder
<i>D. variabilis</i>	Testes, tail, udder
<i>D. marginatus</i>	Udder, testes, tail
<i>Hy. anatolicum</i>	Dewlap, face, flank, Neck, udder, external genitalia, testes, tail, ears
<i>Hy. impeltatum</i>	Flank, udder, ears
<i>Hy. scupense</i>	Tail, ears, udder
<i>Hy. detritum</i>	Udder, flank, tail
<i>Hae. montgomeryi</i>	Udder, flank, tail
<i>Hae. aciculifer</i>	Flank, udder, ears, testes
<i>Hae. sulcata</i>	Dewlap, Neck, udder, external genitalia, testes, tail, ears
<i>Hae. punctata</i>	Dewlap, Neck, udder, external genitalia, flank, testes, tail, ears
<i>R. sanguineus</i>	Dewlap, Neck, udder, external genitalia, testes, tail, ears
<i>R. annulatus</i>	Tail, eyes, ears, udder
<i>R. turanicus</i>	Face, dewlap, udder, tail

It was observed that Neeli Ravi was the most infested breed with tick species as compared to Kundhi, and no significant difference was noted between the breed and gender (Table 5).

Gender, breed, and age-wise tick prevalence on cattle are presented in (Table 6). Crossbred animals carried a smaller number of ticks than local and exotic breeds.

Breed-wise tick prevalence in goats showed that the beetel breed was recorded as more infested (13.09%) with ticks followed by dera din panah (11.15%), teddy (9.02%), and gulabi (3.36%). No significant difference was recorded in the gender and breed of goats. Young goats (1-5 years) carried less number of ticks as compared to adult of 10-12 years' age (Table 7).

Among the different age groups of cattle, buffaloes, goats, and sheep, the highest tick

infestation was observed in old animals (10-12 years), while the lowest was in young animals (1-5 years). Among infested breeds of sheep, Damani sheep were more highly infested with tick burden than other examined breeds of sheep (Table 8). It was also observed that females of all infested animals were more susceptible to tick infestation than males. Tick infestation between sex groups (male and female) of sheep and goat animals was insignificant ($p=0.056$ and $p=0.069$). Tick infestation was significantly varied between the different age groups of all infested animals ($p = 0.000$).

In the grazing system, significantly ($p < 0.001$) higher tick infestation was observed in animals from semi-grazing (18.19%) and free grazing (25.06%) areas than tick infestation in animals that were kept in zero grazing areas (6.88%) given in (Table 9).

Table 5. Gender, breed and age wise tick prevalence on buffaloes

Factor	Groups	Prevalence %		Statistical Analysis
		Positive	Negative	
Gender	Male	201 (22.33)	132 (14.66)	$\chi^2=2.548$, df=1, p=0.076
	Female	268 (29.77)	299 (33.22)	
Breed	Neeli ravi	293 (32.55)	160 (17.77)	$\chi^2=4.978$, df=2, p=0.081
	Kundhi	176 (19.55)	271 (30.11)	
Age in year	1-5	56 (6.22)	186 (20.66)	$\chi^2=19.87$, df=2, P=0.000
	5-8	78 (8.66)	121 (13.44)	
	8-10	130 (14.44)	45 (5.00)	
	10-12	205 (22.77)	79 (8.77)	

Table 6. Gender, breed and age wise tick prevalence on cattle/cows

Factor	Groups	Prevalence %		Statistical Analysis
		Positive	Negative	
Gender	Male	221 (31.57)	118 (16.85)	$\chi^2=2.532$, df=1, p=0.075
	Female	231 (33.00)	130 (18.57)	
Breed	Sahiwal	156 (22.28)	79 (11.28)	$\chi^2=5.198$, df=2, p=0.062
	Cholistani	123 (17.57)	66 (9.42)	
	Crossbred	71 (10.14)	56 (8.00)	
	Exotic	102 (14.57)	47 (6.71)	
Age in year	1-5	51 (7.28)	87 (12.42)	$\chi^2=17.96$, df=2, P=0.000
	5-8	91 (13.00)	74 (10.57)	
	8-10	129 (18.42)	47 (6.71)	
	10-12	181 (25.85)	39 (5.57)	

Table 7. Gender, breed and age wise tick prevalence on goats

Factor	Groups	Prevalence %		Statistical Analysis
		Positive	Negative	
Gender	Male	100 (17.69)	218 (38.58)	$\chi^2=2.211$, df=1, p=0.069
	Female	106 (18.76)	141 (24.95)	
Breed	Dera Din Panah	63 (11.15)	84 (14.86)	$\chi^2=5.201$, df=2, p=0.059
	Beetal	74 (13.09)	96 (16.99)	
	Teddy	51 (9.02)	72 (12.74)	
	Gulabi	19 (3.36)	107 (18.93)	
Age in year	1-5	21 (3.71)	77 (13.62)	$\chi^2=18.84$, df=2, P=0.000
	5-8	53 (9.38)	82 (14.51)	
	8-10	56 (9.91)	97 (17.16)	
	10-12	76 (13.45)	103 (18.23)	

Table 8. Gender, breed and age wise tick prevalence on sheep

Factor	Groups	Prevalence %		Statistical Analysis
		Positive	Negative	
Gender	Male	96 (17.69)	165 (38.58)	$\chi^2=2.108$, df=1, p=0.056
	Female	125 (18.76)	137 (24.95)	
Breed	Kajli	42 (11.15)	79 (14.86)	$\chi^2=5.311$, df=2, p=0.070
	Lohi	55 (13.09)	103 (16.99)	
	Buchi	61 (9.02)	54 (12.74)	
	Damani	63 (3.36)	66 (18.93)	
Age in year	1-5	15 (3.71)	106 (13.62)	$\chi^2=19.43$, df=2, p=0.001
	5-8	54 (9.38)	72 (14.51)	
	8-10	64 (9.91)	57 (17.16)	
	10-12	88 (13.45)	67 (18.23)	

Table 9. Effect of grazing on tick infestation

Category	Variables	Number infested	Tick infestation (%)	95% CI	Chi-square
Grazing	Free grazing	674	25.067	21.02-32.41	$\chi^2 = 13.32$
	Semi-grazing	489	18.19	16.10-24.26	df=2
	No grazing	185	6.88	3.76-9.11	p=0.0001

Discussion

Small and large ruminants are essential contributors to world food production, especially in Pakistan. Farming and livestock production is the primary source of capital income for poor communities. Animals are reared in the whole country (Pakistan), but sheep are primarily found in hilly areas. The production of domestic animals is decreasing daily due to various

constraints, especially pathogens spread by ectoparasites. The main ectoparasite of livestock ticks spreads many diseases to humans and animals [11-13]. The country's population of ticks is increasing due to Pakistan's favorable environmental and climatic conditions for tick reproduction and migration. Ecological change, particularly climate warming, governs not only the variation in the density of ticks but also the

population predominance of their hosts, changes in active periods, and variations in geographical distribution.

Ixodidae, *Nuttalliellidae*, and *Argasidae* are the prominent tick families having approximately 896 species. *Ixodidae*, *Argasidae*, and *Nuttalliellidae* contain 702, 193, and 1 species, respectively. The family *Nuttalliellidae* consists of only one species, *Nuttalliella namaqua* [14]. To date, 53 tick species belonging to the first two families have been identified throughout Pakistan [8]. Nine tick genera have been reported from the country but need to identify ticks from fewer studies areas like hilly or current study areas. To full fill the need of the study, the present cross-sectional study was performed to identify tick species, their distribution, and seasonal fluctuation.

The current study provides evidence about the distribution, identification, and prevalence of tick species on domesticated animals in different districts of Pakistan. Maximum tick infestation (64.57%) was recorded on cattle in the study area, possibly due to hosts, vegetation, and most suitable climatic conditions. These are the risk factors for tick infestation on domestic and wild animals, as previously reported by many early researchers [15, 16]. Zeb *et al.* [12] reported 77.9% tick infestation in cattle. Ramzan *et al.* [5, 22] reported buffaloes as the most infested animal, followed by cattle, goats, and sheep. They had reported similar reasons (climate change, host, and vegetation availability) [8]. These factors are the main reasons for tick growth and development in the study area, which leads to higher prevalence rates. Lack of knowledge among farmers about the peak point of tick infestation or season on animals is another reason for high tick prevalence in the current study areas [17]. The primary factor is the poor extension services and lack of education in the present study areas.

Hyalomma anatolicum is widely distributed worldwide, especially in Pakistan [18]. It was recorded most prevalent tick species in the current study was *H. anatolicum*, while Zeb *et al.* [12] reported *R. microplus* followed by *Hy. anatolicu*. Our present study findings are almost in line with the results of previous researchers [19-21]. For example, a study was conducted by Ramzan *et al.* [5], Ullah *et al.* (2022), and Jamil

et al. (2022) in various districts of Pakistan and reported *Hy. anatolicum*, a dominant species [9, 10, 22]. Many researchers reported *R. microplus* is the second most dominant tick species after *Hy. anatolicum* [2, 23]. The different ecological conditions in other provinces play a vital role in the variation of tick species. For example, arid and semi-arid zone areas provide favorable climatic conditions for tick reproduction and distribution [24]. Our current findings also support the higher prevalence of *H. anatolicum* in semi-arid agroecological zones, which aligns with Rehman *et al.* [2] results. The tail and udder of all animals were highly infested sites for tick attachment. These are sensitive body parts of hosts. Local animals were loaded with tick species than crossbred animals. Crossbred animals may be resistant to ectoparasites. Stall-feeding animals were less infected with tick species as compared to grazing animals. Our findings are similar to those of previous researchers [2].

Conclusion

Our results concluded that *H. anatolicum* was the predominant tick species in domesticated animals in Pakistan. Gender, host breed, age, and grazing systems were the significant risk factors for higher tick infestation in the study areas. Females were more highly infested hosts than males. Tail and udder were recorded as the most suitable site for tick attachment.

Authors' contributions

Conceived and designed the experiments: H Ullah & M Jamil, Performed the experiments: M Jamil, MZ Khan, R Manzoor, M Ali & N Jabeen, Analyzed the data: A Anwar & M Imran, Contributed materials/ analysis/ tools: F Ramzan & Shakirullah, Wrote the paper: H Ullah, M Jamil, M Ali & N Jabeen.

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