



## RESEARCH ARTICLE

### Species Distribution and Seasonal Dynamics of Equine Tick Infestation in Two Subtropical Climate Niches in Punjab, Pakistan

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#### ARTICLE HISTORY (19-173)

Received: May 03, 2019  
Revised: August 21, 2019  
Accepted: August 26, 2019  
Published online: September 03, 2019

#### Key words:

Horses  
Infestation  
Prevalence  
Risk factors  
Tick

#### A B S T R A C T

Ticks are major external parasites of horses that affect animal welfare and transmit many infections. Little is known about the epidemiology of ticks in horses in Pakistan. Keeping in view the significance of horses and its importance, a cross-sectional study was designed to investigate species distribution, seasonal dynamics and epidemiology of ticks infesting horses in Pakistan. By convenience sampling, 500 horses in two districts (Sargodha and Lahore) of Punjab were screened for the presence of ticks from January to December 2017. Tick samples were collected from horses and identified to species level. Data of temporospatial, host and husbandry practices-related risk factors were recorded in a separate questionnaire. Ticks representing six species were collected i.e. *Hyalomma impeltatum* ( $n=52$ ), *H. impressum* ( $n=25$ ), *H. excavatum* ( $n=9$ ), *H. anatomicum* ( $n=3$ ), *H. scutense* ( $n=3$ ) and *H. dromedarii* ( $n=3$ ). The sex ratio of collected ticks showed 63 (66.32%) male and 32 (33.68%) female. In both districts, predominant species in horses was *H. impeltatum*. All infested horses had more than one tick species. The overall proportion of tick infested horses was 7% (35/500), which was high in district Lahore (8.15%) than district Sargodha (5.99%). Summer,  $\leq 3$  body condition score, satisfactory nutritional status, same breed rearing system, presence of dogs and absence of birds at farms, and activity were important risk factors associated with high equine tick infestation. This is the first report regarding the presence of *H. dromedarii* in the horses of Punjab.

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**To Cite This Article:** Ali S, Ijaz M, Ghaffar A, Oneeb M, Masud A, Durrani AZ and Rashid MI, 2020. Species distribution and seasonal dynamics of equine tick infestation in two subtropical climate niches in Punjab, Pakistan. Pak Vet J, 40(1): 25-30. <http://dx.doi.org/10.29261/pakvetj/2019.095>

#### INTRODUCTION

Ectoparasites are major threat that affect animal welfare and are the vector of many bacterial, viral, rickettsial and protozoal, including zoonotic, diseases (Abbas *et al.*, 2014, 2018; Khater *et al.*, 2018; Tirosh-Levy *et al.*, 2018). Ticks can affect equine welfare directly through blood loss, skin damage, irritation and discomfort, allergy, tick paralysis, or indirectly through morbidity or mortality caused by infectious organisms transmitted (Duell *et al.*, 2013). Assessment of climate niches of various tick species is now considered necessary to better figure out the relationship among pathogens, vectors and hosts (Estrada-Peña *et al.*, 2012). A coalition

of abiotic and biotic factors comprising host availability, vegetation and climate are needed for the survival of different developmental stages of ticks and their fruitful reproduction. Certain tick species also display ecological plasticity and can easily adjust to changing climate and new niches (Estrada-Peña, 2008). Climate changes in recent years have changed the distribution of a number of tick species and introduced some tick species and infectious agents into previously known unaffected regions (Tirosh-Levy *et al.*, 2018).

World equine population is about 122.4 million, consisting of 43.4, 15 and 40 million horses, mules and donkeys, respectively. Out of this population, 60% of horses, 97% of mules and 98% of donkeys are distributed

in developing countries (Tedla and Abichu, 2018). The overall population of equine in Pakistan is about six million with 0.4 million horse population (Wasti *et al.*, 2018-19). In Pakistan, equines are raised for riding, racing, transportation and companion animal (Javed *et al.*, 2014). Pakistan is situated in Indus river plain of South Asian region with a climate ranging from continental to subtropical which offers a niche for several ixodid tick species. Consequently, Pakistan has many endemic tick-borne infections that affect humans and animals, including livestock and pets. Ticks are reported to cause equine granulocytic anaplasmosis, equine piroplasmosis (EP) and Lyme borreliosis in horses (Laus *et al.*, 2013). Predominant ticks reported infesting equine in Pakistan belong to the genus *Hyalomma* (Javed, 2013). Among different diseases, EP is prevalent in Pakistan (Afridi *et al.*, 2017; Ali *et al.*, 2019) while horses of neighbouring countries were found seropositive for Crimean Congo hemorrhagic fever (CCHF) (Shanmugam *et al.*, 1976). Recent increased number of CCHF cases in Pakistan may possibly be linked to ticks infesting horses.

That is why, the current study was designed as a survey of the horse population in Sargodha and Lahore districts, among two high horse populated districts of the province (Anonymous, 2006), to characterize equine tick infestation, recognize its risk factors, and investigate climatic and environmental factors associated with various tick species.

## MATERIALS AND METHODS

**Study site:** District Sargodha largely contains flat and fertile plains, though a limited number of small hills are evident on the way connecting the city to Faisalabad. It is situated in the northeast of Pakistan and surrounded by Jehlum district on the north-west, Khushab district on the west, Jhang district on south, Chiniot district on south-east, Hafizabad district on east and Mandi Bahauddin district on north-east. Chenab River flows on east side, while Jhelum River lies on west and north sides of Sargodha. It has a local steppe climate featured by extreme heat in summer and moderate cold in winter. District Lahore lies on a flat alluvial plain, which is situated in northeast of Pakistan and surrounded by Sheikhupura district on north and west sides, Kasur district on south, and India on east. River Ravi lies on north side of Lahore. It has a local steppe climate featured by a rainy, long and extremely hot summer, a monsoon, a warm and dry winter, and dust storms (Ali *et al.*, 2016). Coordinates, elevations from sea level, areas, and horse populations of study districts are given in Table 1. Average temperature, humidity, pressure and precipitation of individual month of study districts are given in Table 2.

**Study design:** The study was designed to identify ticks infesting horses and associated risk factors in study districts from January to December 2017. The sample size was calculated using the formula given below assuming 50% prevalence, 95% confidence level and 5% desired absolute precision which resulted in 384 horses to be sampled in two districts (Thrusfield, 2005). This was increased to 500 horses for convenience of risk factor analysis. A convenience sampling technique was adopted to collect samples from hospitals, clinics, fields, stud farms, racecourses and polo clubs.

$$n = \frac{1.96^2 P_{exp} (1 - P_{exp})}{d^2}$$

Where;  $n$  = required sample size,  $P_{exp}$  = expected prevalence,  $d$  = desired absolute precision

**Ticks collection, identification and risk factor analysis:** Ticks were collected from horses after obtaining owners' consent. A fine-toothed forceps were used for tick collection. Utmost care was given to not damage mouth parts of ticks during collection. Tick specimens were shifted to tubes containing 70% ethyl alcohol and labelled. Specimens were brought to Entomology Laboratory, Department of Parasitology, UVAS Lahore and identified to species level under a stereo microscope using taxonomy covered in Multi-Key version 2.1 computer package (Walker *et al.*, 2005). Their identifications were also validated from original descriptions and re-descriptions of relevant tick species (Walker *et al.*, 2003). A questionnaire containing 13 close-ended questions was utilized to investigate the association of risk factors with tick prevalence. The questionnaire was divided into (A) temporospatial, (B) host, (C) husbandry practices.

**Statistical analyses:** Prevalence of equine tick infestation was calculated using the formula given below (Thrusfield, 2005). Data regarding the prevalence of tick infestation and its risk factors were analyzed with descriptive statistics, Pearson's Chi-squared test (Khosravi *et al.*, 2012) and univariable model, using statistical software, IBM SPSS Statistics for Windows, Version 20.0. Variables presenting  $P < 0.20$  at univariable analysis were then analyzed by the final regression model at 95% confidence interval (Ali *et al.*, 2019).

$$P (\%) = \frac{\text{number of horses having tick infestation}}{\text{number of horses in the population examined}} \times 100$$

## RESULTS

**Tick prevalence:** Totally 500 horses were examined in two different districts: Sargodha ( $n=267$ ) and Lahore ( $n=233$ ). Majority of horses (89.2%) were males. Most horses (65.4%) examined were less than 6 years old and breed of most horses (94.6%) was nondescriptive. Overall tick prevalence in horses was 7% (35/500), which was lower in Sargodha district (5.99%) than Lahore district (8.15%) but the difference was non-significant ( $P > 0.05$ ). In total, 95 ixodid ticks [male: 63 (66.32%); female: 32 (33.68%)] were collected from 35 tick infested horses (Table 3). All ticks were identified as adults belonging to genus *Hyalomma*. Among identified ticks, *H. impeltatum* ( $n=52$ , 54.74%) was predominant species followed by *H. impressum* ( $n=25$ , 26.32%), *H. excavatum* ( $n=9$ , 9.47%), *H. anatomicum*, *H. scupense*, and *H. dromedarii* ( $n=3$ , 3.16% each). Out of all infested horses, 91.43% ( $n=32$ ) were infested with *H. impeltatum*, 62.86% ( $n=22$ ) with *H. impressum*, 25.71% ( $n=9$ ) with *H. excavatum*, and 8.57% ( $n=3$ ) with *H. anatomicum*, *H. scupense*, and *H. dromedarii* each. All tick positive horses had infestation of more than one tick species, but *H. scupense* and *H. dromedarii* were identified in Sargodha district, while *H. anatomicum* was present in Lahore district only.

**Temporal effects on tick prevalence:** A significant difference ( $P<0.05$ ) was found in the frequency of equine tick infestation by months of the year when compared with the surveyed population (Table 4). Similarly, the difference in frequency of equine tick infestation by the month of year in Sargodha and Lahore districts was found significant ( $P<0.05$ ). In summer, the percentage of equine tick infestation was high in Sargodha district. But in Lahore district, it was high in fall. The season was also revealed a significant risk factor ( $P<0.05$ ) associated with equine tick infestation. When horse populations of both districts were analyzed individually, the season remained a significant risk factor ( $P<0.05$ ) for both districts.

**Effects of host characteristics on tick prevalence:** Of infested horses, 34 were males and 1 was female. No significant difference ( $P>0.05$ ) was detected in the frequency of tick infestation by equine gender. Horses belong to the age group of  $>12$  years had less tick infestation compared to other two groups. However, no significant difference ( $P>0.05$ ) was found in the frequency of tick infestation by age of horses. Nondescriptive horses had high tick infestation when compared to exotic and crossbred horses. However, the breed was found non-significant risk factor ( $P>0.05$ ) (Table 5).

**Effects of husbandry practices on tick prevalence:** Horses having  $>3$  body condition score (BCS) were significantly ( $P<0.05$ ) less tick infested than those with  $\leq 3$  BCS. Horses with unsatisfactory nutritional status had significantly ( $P<0.05$ ) less frequency of tick infestation than their opposites. Horses managed in boxes contained a

high count of tick infestation than grazing horses. But this risk factor was found non-significant ( $P>0.05$ ). Rearing same breed horses had significantly ( $P<0.05$ ) high tick infestation compared to rearing different breeds of horses, and horses with other species. Horses reared with dogs had a high percentage of tick infestation compared to those without dogs. Presence of dogs in horse rearing area was found significant risk factor ( $P<0.05$ ). However, it was non-significant ( $P>0.05$ ) in district Sargodha. Presence of ruminants in horse keeping area was also linked to the high frequency of tick infestation but found non-significant ( $P>0.05$ ). Horse rearing with birds had less frequency of tick infestation compared to those reared without birds. Presence of birds in equine rearing place was revealed a significant risk factor ( $P<0.05$ ). However, it was not significant ( $P>0.05$ ) in district Sargodha. Horses kept for work had significantly ( $P<0.05$ ) high percent of tick infestation than those kept for sports, and as pets (Table 6).

Several husbandry-related key risk factors including BCS, rearing system, and presence of dogs and birds in farms were identified by the final logistic regression model using elimination technique (backward manual stepwise process) removing the redundant and confounding variables (Table 7). Horses of Sargodha district having  $>3$  BCS were at high risk of tick infestation ( $OR=16.97$ ,  $P=0.001$ ) compared to others. Among rearing systems, raising horses of the same breed ( $OR=5.000$ ,  $P=0.000$ ) and different breeds ( $OR=4.683$ ,  $P=0.001$ ) were at more risk of tick infestation as compared to horses reared with different breeds. Farms having dogs had more risk ( $OR=2.572$ ,  $P=0.018$ ) of tick infestation.

**Table 1:** Coordinates, elevations from sea level, areas, and horse populations of Sargodha and Lahore districts

District	Coordinates		Area (km <sup>2</sup> )	Elevation (from sea level – m)	Horse population*
	Latitude (°N)	Longitude (°E)			
Sargodha	32°10'00"	72°30'00"	5,854	190	11,720
Lahore	31°25'00"	74°19'60"	1,772	224	8,196

\*Source: Anonymous, (2006).

**Table 2:** Average temperature, humidity, pressure and precipitation of individual month of 2017 in Sargodha and Lahore districts

Month	Sargodha				Lahore			
	Temperature (°C)	Humidity (%)	Pressure (mbar)	Precipitation (mm)	Temperature (°C)	Humidity (%)	Pressure (mbar)	Precipitation (mm)
Jan	12	79	1018	5.03	13	72	1018	5.67
Feb	16	62	1016	0.01	17	60	1016	0.07
Mar	20	59	1011	6.79	21	54	1012	8.53
Apr	26	51	1005	9.55	29	40	1007	2.91
May	32	43	1002	2.73	33	37	1004	0.88
Jun	31	58	998	11.86	32	54	1001	15.5
Jul	31	74	998	2.86	31	68	1000	12.38
Aug	31	77	999	5.78	31	69	1002	4.18
Sep	28	73	1004	10.68	30	63	1006	1.22
Oct	25	66	1008	0	28	56	1010	0
Nov	17	76	1014	0.04	18	72	1016	3.1
Dec	13	69	1018	0	15	63	1018	0.09

(Source: Time and Date, 2017; World Weather Online, 2017).

**Table 3:** Species, sex, count and prevalence of adult ticks infesting horses of Sargodha and Lahore districts

Tick species	Sargodha			Lahore			Overall		
	M:F	No. (%)	Prevalence (%)	M:F	No. (%)	Prevalence (%)	M:F	No. (%)	Prevalence (%)
<i>H. impeltatum</i>	22:00	22 (55.00)	15/16 (93.75)	28:02	30 (54.55)	17/19 (89.47)	50:02	52 (54.74)	32/35 (91.43)
<i>H. impressum</i>	01:09	10 (25.00)	09/16 (56.25)	00:15	15 (27.27)	13/19 (68.42)	01:24	25 (26.32)	22/35 (62.86)
<i>H. excavatum</i>	01:01	02 (05.00)	02/16 (12.50)	05:02	07 (12.73)	07/19 (36.84)	06:03	09 (09.47)	09/35 (25.71)
<i>H. anatolicum</i>	00:00	00 (00.00)	00/16 (00.00)	03:00	03 (05.45)	03/19 (15.79)	03:00	03 (03.16)	03/35 (08.57)
<i>H. scutense</i>	03:00	03 (07.50)	03/16 (18.75)	00:00	00 (00.00)	00/19 (00.00)	03:00	03 (03.16)	03/35 (08.57)
<i>H. dromedarii</i>	00:03	03 (07.50)	03/16 (18.75)	00:00	00 (00.00)	00/19 (00.00)	00:03	03 (03.16)	03/35 (08.57)
Total	27:13	40		36:19	55		63:32	95	

**Table 4:** Association of temporal risk factors with tick infestation in horses of Sargodha and Lahore districts

Variable	Category	Sargodha Positive/Total (%)	P-value	Lahore Positive/Total (%)	P-value	Overall Positive/Total (%)	P-value
Month	Jan	00/29 (00.00)		00/29 (00.00)		00/58 (00.00)	
	Feb	00/15 (00.00)		00/15 (00.00)		00/30 (00.00)	
	Mar	00/37 (00.00)		00/31 (00.00)		00/68 (00.00)	
	Apr	02/12 (16.67)		02/12 (16.67)		04/24 (16.67)	
	May	01/02 (50.00)		01/02 (50.00)		02/04 (50.00)	
	Jun	00/17 (00.00)	<0.01*	00/13 (00.00)	<0.01*	00/30 (00.00)	<0.01*
	Jul	06/45 (13.33)		02/35 (05.71)		08/80 (10.00)	
	Aug	07/32 (21.88)		06/30 (20.00)		13/62 (20.97)	
	Sep	00/30 (00.00)		00/31 (00.00)		00/61 (00.00)	
	Oct	00/30 (00.00)		08/20 (40.00)		08/50 (16.00)	
	Nov	00/16 (00.00)		00/14 (00.00)		00/30 (00.00)	
	Dec	00/02 (00.00)		00/01 (00.00)		00/03 (00.00)	
Season	Winter (Nov-Feb)	00/62 (00.00)		00/59 (00.00)		00/121 (00.00)	
	Spring (Mar-Apr)	02/49 (04.08)	<0.01*	02/43 (04.65)	0.01*	04/92 (04.35)	<0.01*
	Summer (May-Aug)	14/96 (15.58)		09/80 (11.25)		23/176 (13.07)	
	Fall (Sep-Oct)	00/60 (00.00)		08/51 (15.69)		08/111 (07.21)	

\*significant difference P&lt;0.05.

**Table 5:** Association of host-related risk factors with tick infestation in horses of Sargodha and Lahore districts

Variable	Category	Sargodha Positive/Total (%)	P-value	Lahore Positive/Total (%)	p-value	Overall Positive/Total (%)	P-value
Sex	Male	15/242 (06.20)	0.66	19/204 (09.31)	0.09	34/446 (07.62)	0.12
	Female	01/25 (04.00)		00/29 (00.00)		01/54 (01.85)	
Age	≤ 6 years	11/185 (05.95)	0.21	13/142 (09.15)	0.58	24/327 (07.34)	0.79
	7-12 years	05/52 (09.62)		03/60 (05.00)		08/112 (07.14)	
Breed	> 12 years	00/30 (00.00)		03/31 (09.68)		03/61 (04.92)	
	Non-descriptive	16/250 (06.40)		19/223 (08.52)		35/473 (07.40)	
Breed	Exotic	NA	0.28	00/03 (00.00)	0.63	00/03 (00.00)	0.34
	Crossbred	00/17 (00.00)		00/07 (00.00)		00/24 (00.00)	

**Table 6:** Association of husbandry practices-related risk factors with tick infestation in horses of Sargodha and Lahore districts

Variable	Category	Sargodha positive/Total (%)	P-value	Lahore positive/Total (%)	P-value	Overall positive/Total (%)	P-value
BCS	≤ 3	07/18 (38.89)	<0.01*	03/11 (27.27)	0.02*	10/29 (34.48)	<0.01*
	> 3	09/249 (03.61)		16/222 (07.21)		25/471 (05.31)	
Nutritional status	Unsatisfactory	09/249 (03.61)	<0.01*	16/222 (07.21)	0.02*	25/471 (05.31)	<0.01*
	Satisfactory	07/18 (38.89)		03/11 (27.27)		10/29 (34.48)	
Management	Boxed	16/263 (06.08)	0.61	19/231 (08.23)	0.67	35/494 (07.09)	0.5
	Grazing	00/04 (00.00)		00/02 (00.00)		00/06 (00.00)	
Rearing system	Same breed	09/62 (14.52)	<0.01*	04/20 (20.00)	<0.01*	13/82 (15.85)	<0.01*
	Different breeds	00/12 (00.00)		09/48 (18.75)		09/60 (15.00)	
Dogs in farms	Different species	07/193 (03.63)		06/165 (03.64)		13/358 (03.63)	
	No	08/150 (05.33)		01/78 (01.28)		09/228 (03.95)	
Ruminants in farms	Yes	08/117 (06.84)	0.61	18/155 (11.61)	0.01*	26/272 (09.56)	0.01*
	No	01/32 (03.13)		00/09 (00.00)		01/41 (02.44)	
Birds in farms	Yes	15/235 (06.38)	0.47	19/224 (08.48)	0.36	34/459 (07.41)	0.23
	No	01/46 (02.17)		13/63 (20.63)		14/109 (12.84)	
Activity	Yes	15/221 (06.79)	0.23	06/170 (03.53)	<0.01*	21/391 (05.37)	0.01*
	Work	07/09 (77.78)		07/15 (46.67)		14/24 (58.33)	
Activity	Sport	09/258 (03.49)	<0.01*	11/214 (05.14)	<0.01*	20/472 (04.24)	<0.01*
	Pet	NA		01/04 (25.00)		01/04 (25.00)	

\*significant difference P&lt;0.05.

**Table 7:** Multivariable analysis of husbandry practices-related risk factors of tick infestation in horses

Variable	Categories	B	Standard error	Wald statistics	Degree of freedom	P-value	Odds Ratios	Lower CI (95%)	Upper CI (95%)
Sargodha BCS	≤ 3	2.831	0.591	22.968	I	0.000	16.970	5.331	54.021
	> 3	Ref							
Total BCS	≤ 3	2.240	0.441	25.739	I	0.000	9.389	3.953	22.305
	> 3	Ref							
Rearing system	Same breed	1.609	0.414	15.127	I	0.000	5.000	2.222	11.251
	Different breeds	1.544	0.459	11.323	I	0.001	4.683	1.905	11.511
Dogs in farms	Yes	0.945	0.398	5.640	I	0.018	2.572	1.179	5.608
	No	Ref							
Birds in farms	Yes	-0.954	0.364	6.883	I	0.009	0.385	0.189	0.786
	No	Ref							

## DISCUSSION

**Tick species and prevalence:** In this study, 66:34 ratio of male:female ticks were found that is similar to a past study in which 59:41 ratio was reported (Tirosh-Levy *et*

*al.* 2018). All ticks collected were adults, which suggest that young ticks infest other animals (Tirosh-Levy *et al.*, 2018). All ticks identified belonged to *Hyalomma* genus. In the previous study, *Hyalomma* ticks were most common in tick infested horses, mules and donkeys of

district Lahore, followed by *Boophilus* and mixed infestation (Javed, 2013). *Hyalomma* has identified the predominant tick genus in working and riding horses in Ahwaz, Iran (Khosravi *et al.*, 2012). *Hyalomma* was also reported being the most prevalent genus (70% of ticks) infesting horses in Israel (Tirosh-Levy *et al.*, 2018). *Hyalomma* is predominant tick genus infesting livestock in arid and semi-arid climates of Asia, Africa and Europe (Estrada-Peña *et al.*, 2004).

Unluckily, no study has been conducted on equine tick species in Pakistan, and so it is not possible to compare results with findings of past studies. Tirosh-Levy *et al.* (2018) identified *H. excavatum* most abundant tick species infesting equine in Golan Heights. Cattle, sheep, goats, camels, horses and donkeys are hosts of adult *H. excavatum*. It can feed as two-host or three-host tick depending on availability of hosts (Walker *et al.*, 2003). Tick species, *H. dromedarii* was also detected which is most closely associated with camels. However, adult *H. dromedarii* can also parasitize other domestic animals (Walker *et al.*, 2003). Kleinerman *et al.* (2013) also collected one specimen of *H. dromedarii* from horse near Dead Sea, Israel.

Tick species, *H. anatomicum*, *H. excavatum*, *H. scupense* and *H. dromedarii* are known vectors of EP (Walker *et al.*, 2003; Scoles and Ueti, 2015), which is prevalent in Pakistan (Afridi *et al.*, 2017; Ali *et al.*, 2019). Other tick species, *H. impeltatum* and *H. impressum* are not known vectors of equine diseases, and their potential to transmit infectious agents between horses and other hosts should be needed to be further explored. *H. impeltatum* identified in this study has been reported as capable of transmitting the CCHF virus to humans (Walker *et al.*, 2003).

In this study, an overall 7% tick infestation was recorded among screened horses. It is in association with findings of Afridi *et al.* (2017), who reported 9.77% tick infestation in equine tested for seroprevalence of *Theileria equi*. However, Tirosh-Levy *et al.* (2018) recorded high prevalence (25%) of tick infestation in horses in Israel. Environmental factors including temperature, rain and vegetation affect tick activity and life cycle (Estrada-Peña and Venzal, 2007; Estrada-Peña, 2008), and could be responsible for the difference of prevalence with the former study.

**Temporal effects on tick prevalence:** Season plays a vital role in the dynamics of tick population (Singh and Rath, 2013). A significant difference was detected in the frequency of tick infested horses in different seasons. Results of the current study stated that summer was the most favourable season for tick infestation except in Lahore district where it was fall. Hot and humid environmental conditions are most conducive for the development of various developmental stages of ticks. While cold and dry conditions of winter are unfavourable for tick survival. Engorged females, nymphs and larvae, and unfed adults hide into cracks and crevices (Chaudhuri, 1969) thus leading to low tick infestation levels in winter. Javed (2013) recorded similar seasonal variation in the frequency of tick infested horses. Likewise, Tirosh-Levy *et al.* (2018) recorded a high number of ticks in the spring

season (May-Jun), followed by summer (Jul-Aug), fall (Nov-Dec) and winter (Feb-Mar), respectively.

**Effects of host characteristics on tick prevalence:** Male horses had more tick infestation compared to their mates but the difference was not significant. The previous study described similar results (Khosravi *et al.*, 2012). Age of host animal plays a role in tick infestation pattern (Manan *et al.*, 2007). Result of the current study exposed that young animals are more susceptible to tick infestation. However, Khosravi *et al.* (2012) reported a high prevalence in horses having age >5 years. Provision of better animal husbandry practices to adult animals while least attention given to young animals in terms of food, management practices and acaricides may be possible reasons of different results (Singh and Rath, 2013). Young ones also possess weak immune status. Also, low tick infestation in adult and old animals is probably because of acquired resistance from repeated exposure in early life (Das, 1994).

**Effects of husbandry practices on tick prevalence:** Horses of  $\leq 3$  BCS were significantly high tick infested than those  $>3$  BCS. Horses with satisfactory nutritional status had a significantly high frequency of tick infestation than their opposites. Rearing same breed horses had significantly high tick infestation compared to rearing different breeds of horses, and horses with other species. Horses reared with dogs had a significantly high percentage of tick infestation compared to those without dogs. Grech-Angelini *et al.* (2016) reported tick species of domestic dogs i.e., *R. sanguineus* on cattle, mouflons, wild boars and more rarely on sheep and horses of French Mediterranean island of Corsica. Horses rearing with birds had significantly less frequency of tick infestation compared to those reared without birds. A previous study also recorded significantly low tick prevalence in livestock having rural poultry. Rearing birds on animal farms decrease tick infestation as birds act as natural predators and control ticks by picking from animal bodies and from their surroundings too (Rehman *et al.*, 2017). Horses kept for work had a significantly high prevalence of tick infestation than those kept for sport, and as pets. Khosravi *et al.* (2012) also recorded the high prevalence of tick infestation in working horses as compared to riding horses.

**Conclusions:** Tick species, *H. anatomicum*, *H. excavatum*, *H. scupense* and *H. dromedarii*, are known vectors of EP. Other tick species, *H. impeltatum* and *H. impressum* do not vector equine infections, and their potential to transfer pathogens between horses and other animals should be further inquired. *H. impeltatum* identified in this study has been reported as capable of transmitting CCHF virus to humans. Risk factors comprising month and season, BCS, nutritional status, rearing system, presence of dogs and birds at farms, and activity were proved key risk factors of equine tick infestation. Veterinarians and horse owners should think about the practice of tick control, particularly during summer and fall seasons.

**Acknowledgements:** Authors are thankful to Entomology Laboratory, Department of Parasitology, UVAS, Lahore

for provision of laboratory and technical support during the study.

**Authors contribution:** SA, MI, AZD and MIR designed study. SA, MI, MO and AM helped in study execution. SA, MO, AG and AM contributed in data analysis and interpretation. SA, MI, AG, AZD and MIR prepared manuscript. All authors gave final approval of manuscript.

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