Prevalence of Tick Infestation in Farm Animals from Punjab, Pakistan

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ABSTRACT
Ticks are the second to mosquitoes as vectors of a number of pathogens like viruses, spirochetes, bacteria, rickettsia, protozoa and filarial nematodes etc. that cause mortality in humans and animals. So, this study was carried out to check the prevalence of ticks in Punjab, Pakistan. The total 120 livestock farms were randomly selected from 12 districts of Punjab, 10 farms (05 urban and 05 rural) from each district. Tick species were collected systematically from head to tail directions with the help of small steel forceps. Then tick samples were taken to the research laboratory and were preserved in 70% methanol for identification. On the basis of morphology, the collected ticks were distinguished microscopically with the help of the dichotomous key. The prevalence of ticks was significantly less in the Northern zone (33.47%) as compared to the Southern (36.33%), Western (35.83%) and Central zones (40.43%). The total nine tick species i.e. Hylomma (Hy.) anatolicum (26.39%), Hy. marginatum (14.51%), Hy. dromedarii (5.62%), Hy. truncatum (2.45%), Hy. rufipes (1.81%), Rhipicephalus (Rh.) sanguineus (17.24%), Rh. appendiculatus (12.39%), Rh. microplus (14.24%) and Rh. decoloratus (5.35%) were identified. In all selected zones, Hy. anatolicum was the most abundant tick’s species. The overall prevalence of ticks infestation in all animals were 36.52% and it was significantly different in all animal species, like buffaloes (37.53%), cows (42.41%), goats (36.14%) and sheep (29.00%). It was concluded that there is wider variety of ticks in Pakistan.

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INTRODUCTION

Pakistan is an agricultural country with 75% of its population involved directly or indirectly with the agriculture sector. It is the 2nd larger sector that provides 21.2% contribution as GDP (Gross Domestic Production) by providing employment to 45% of the workforce. Majority of its population (>70%) lives in villages and generally depends on the rearing of animals for their livelihood (Mather and Abdullah, 2015). Different types of domesticated animals such as buffaloes, cattle, goats, sheep, poultry, horses, donkeys and camel are included in livestock (Khan, 2004). Parasitic diseases are the major issues to the poor farming community in tropical and subtropical areas in particular Pakistan (Jonsson, 2006; Khan et al., 2017; Mehmood et al., 2017; Naqvi et al., 2017; Zaman et al., 2017; Abbas et al., 2018; Khater et al., 2018). Among ectoparasites, ticks are blood-sucking parasites associated with mammals, birds and reptiles (Aslam et al., 2015; Ali et al., 2016) which, also transmit diseases in its host.

The distribution of ticks is cosmopolitan (Durrani et al., 2008). Being a sub-tropical country, Pakistan provides optimum conditions for ticks growth and multiplication. But, very little research was done on tick taxonomy and tick infestation (Sajid et al., 2008, 2009; Ali et al., 2016). Several factors such as species, age, sex, season, breed, photoperiod and management are responsible to influence the receptiveness and resistance of animals to tick infestation (Asmaa et al., 2014). Cracks and crevices in the floor and walls of animal shed, animal tethering and grazing were found bonded with a greater level of tick incidence (Iqbal et al., 2013). Generally, ticks damage hidden parts of the animals and cause lower productivity and mortality. Over the past two decades, the occurrence of tick borne diseases have increased and caused health problems (Kaur et al., 2015). Detrimental effects of ticks to animals are loss of blood, irritation, stress and despair
of the immune system. Due to the direct transmission of diseases into the host, ticks are highly responsible for economic losses in terms of decreasing skin quality of cattle up to 20-30% (Sultana et al., 2015).

Today, zoonotic pathogens are responsible for the most emerging infectious diseases, which are transmitted by tick vectors making ticks second to mosquitoes as vectors (Kaur et al., 2015). Out of 700 species, 269 species of hard ticks are found to cause health-related problems in human beings (Otranto et al., 2014) and Crimean-Congo hemorrhagic fever is one of them. This disease is transmitted to human beings through the bite of an infected adult *Hyalomma* ticks. This disease is increasing day by day in Pakistan, a total of 196 cases and 48 deaths were reported during 2012-2014 (WHO, 2014). Ticks also encompass a very strong veterinary problem as they spread diseases e.g. anaplasmosis, theileriosis, babesiosis, and trypanosomiasis and toxicosis (Kaur et al., 2015). Contagious diseases are important contributors to mortality and disease in animals and humans (Jabbar et al., 2015).

Thus, an effective and well planned work to examine the incidence and prevalence of ticks in the province of Punjab, Pakistan is need of the time (Durrani et al., 2008). Most of the earlier research work was restricted to one district or a part of the district and no one consider agro-ecological zones and cropping pattern on a large scale that can also affect the ticks distribution and prevalence (Jabbar et al., 2015). So, this project was planned to investigate the prevalence of ticks across different geographical zones of the Punjab Province.

**MATERIALS AND METHODS**

**Study area:** These research activities were performed in four agro-ecological zones of Punjab (Rehman et al., 2017), Pakistan (Fig. 1), i.e., Southern zone (Muzaffargarh, Bahawalpur & Rajanpur), Western zone (Khushab, Bhakar & Layyah), Central zone (Jhang, Gujranwala & Faisalabad) and Northern zone (Rawalpindi, Attock & Chakwal) in four seasons, i.e., spring (February to April), summer (May to July), autumn (August to October) and winter (November to January) during 2016 & 17. In the study areas generally, the climate is dry and receives less than 200 mm of rainfall annually. There is a noticeable difference in the climate of Western and Central zone. Central and Northern zones are somewhat cooler than Southern and Western zones. Above cited four seasons are found in study areas and generally the temperature ranges from -2 to 45°C and can reach up to 50°C in summer and -5°C in winter. But during late summer, humidity increases that provide suitable conditions for tick infestation and multiplication. A total of 12 livestock farms were selected from 12 districts as mentioned above.

**Collection and preservation of ticks:** Ticks were collected from buffaloes, cows, goats and sheep. From each district, 10 livestock farms (05 urban and 05 rural) were randomly selected in such a way that each farm was at least 10km apart from the other farm in urban areas while in rural areas, each farm was selected from different villages that were at least 5km apart. The selected 5-10 animals were systematically observed from farms by close examination, parting the hairs against their natural direction for the recognition of ticks. Tick species were collected systematically from head to tail directions with the help of small steel forceps with blunt ends without damaging their mouth. Ticks were preserved in 70% methanol (Atif et al., 2012) and carried to the laboratory in the Department of Zoology, Government College University, Faisalabad for the identification.

**Identification of ticks:** Identification of ticks was carried out with the help of the anatomical and morphological characteristics in the laboratory using dissecting and compound microscopes according to the keys given by McCarthy (1967) and Estrada-Pena et al. (2006). Moreover, species level was identified under a stereomicroscope (OPTICA SZM-1: Italy) with 40-fold magnification (Apanaskevich and Horak, 2005).

**Statistical analysis:** The prevalence of ticks was determined by using logistic regressions (Rehman et al., 2017) and odd’s ratio (OR) at 95% confidence interval (CI) in all studied agro-ecological zones of Punjab, Pakistan by using SPSS software package (SPSS, 21).

**RESULTS**

Total nine tick species i.e. *Hyalomma* (*Hy.*) anatolicum, *Hy.* marginatum, *Hy.* dromedarii, *Hy.* truncatatum, *Hy.* rafipes, *Rhipicephalus* (*Rh.*) sanguineus, *Rh.* appendiculatus, *Rh.* Microplus and *Rh.* decorolatus were identified. In all selected zones, *Hy.* anatolicum and *Hy.* marginatum were the most abundant ticks species. The total prevalence of tick-infested animals was recorded at 36.52% (4382/12,000) in different districts of Punjab, Pakistan. The highest prevalence (40.43%) was observed from Central zone while the lowest (33.47%) from Northern zone and prevalence data for other zones was
shown in Table 1. The non-significant (P>0.05) differences were observed in Western zone while significant (P<0.05) differences in Southern zone and the highly significant (P<0.001) differences in Central zone. During the present study, a total of 12,000 animals i.e. 3200 buffaloes, 3200 cows, 2800 goats and 2800 sheep were randomly examined for tick collection. Out of 12,000 animals, 4382/12,000 (36.52%) animals were observed infested with ticks. The highly significant (P<0.001) differences were observed in the prevalence of all ruminants. The prevalence of ticks in buffaloes, cows, goats and sheep were detected (37.53, 42.41, 36.14 and 29.00%), respectively as described in Table 2.

Table 3 is showing season-wise (prevalence %) for overall data. The highly significant (P<0.001) differences were observed in the prevalence of ticks in different seasons. During this study, the highest prevalence was observed in summer (55.87%) followed by spring (41.30%), autumn (29.57%) and lowest during winter (19.33%).

Ruminants including buffaloes, cows, sheep and goats were observed infested with different tick species, from four agro-ecological zones of Punjab. From all genera, Hy. anatolicum (26.39%) was the most common species in all four zones followed by Rh. sanguineus (17.24%) and Rh. microplus (14.24%) while the least common was Hy. rufipes (8.11%). In all zones, Hy.Marginatum was the second common tick species in ruminants except sheep while Hy. Dromedarii were present in all zones except Northern zone. It was observed that Hy. truncatum and Hy. rufipes were present only in Western zone and absent in Southern, Central and Northern zones of Punjab, Pakistan (Table 1). Similarly, Rh. sanguineus and Rh. microplus both were present in all regions (central, western, southern and northern regions) while Rh. appendiculatus and Rh. decoloratus were observed in three zones and were not observed in Northern zone.

DISCUSSION

Ticks are the most important pest of livestock in tropical and subtropical regions of the world (Admassu et al., 2015). The results of the current research showed the presence of one or multiple tick species from all the observed livestock farms. Variations were present in the prevalence of ticks infestation within farms of same study areas. The prevalence of ticks in Western, Central, Southern and Northern zones was recorded 35.83, 40.43, 36.33 and 33.47%, respectively. These variations in tick prevalence were due to the geographical condition, temperature and climatic conditions of different study area of Province, Punjab (Iqbal et al., 2014). The previous studies on tick prevalence were centred on a specific area or different parts of the same region (Iqbal et al., 2013) only instead of agro-ecological regions (Jabbar et al., 2015). The data regarding the prevalence of ticks in various species of animals were observed during different seasons of the year. The results of the current study revealed the highest prevalence of tick infestation in summer because in summer the weather was hot and humid which supported the survival of tick infestation.

Table 1: Prevalence of identified tick species in different zones from Punjab, Pakistan

| Ticks species | Central Zone NAE/NAI/NTC | Western Zone NAE/NAI/NTC | Southern Zone NAE/NAI/NTC | Northern Zone NAE/NAI/NTC |
|---------------|--------------------------|--------------------------|---------------------------|---------------------------|
| Hy. anatolicum | 3000/1213/5220 | 3000/1075/8682 | 3000/1090/5120 | 3000/1004/2716 |
| Rh. appendiculatus | 21.63% (1241) | 36.64% (2102) | 24.23% (1390) | 17.50% (1003) |
| Hy. marginatum | 21.68% (684) | 28.81% (909) | 30.18% (952) | 19.33% (609) |
| Hy. dromedarii | 32.81% (401) | 38.21% (467) | 28.96% (354) | 0 |
| Hy. truncatum | 0 | 100% (332) | 0 | 0 |
| Hy. rufipes | 0 | 100% (393) | 0 | 0 |
| Rh. sanguineus | 26.19% (982) | 43.43% (1628) | 21.28% (797) | 9.10% (341) |
| Rh. microplus | 29.84% (804) | 47.99% (1293) | 22.16% (597) | 0 |
| Rh. decoloratus | 22.97% (711) | 29.43% (912) | 22.95% (710) | 24.65% (763) |

Table 2: Prevalence of identified tick species in different farm animals in Punjab, Pakistan

| Ticks species | Buffaloes NAE/NAI/NTC | Cows NAE/NAI/NTC | Goats NAE/NAI/NTC | Sheep NAE/NAI/NTC |
|---------------|------------------------|-----------------|------------------|------------------|
| Hy. anatolicum | 1183 | 2109 | 138 | 926 |
| Hy. marginatum | 812 | 1318 | 1024 | 0 |
| Hy. dromedarii | 333 | 310 | 579 | 0 |
| Hy. truncatum | 178 | 354 | 0 | 0 |
| Hy. rufipes | 193 | 200 | 0 | 0 |
| Rh. sanguineus | 799 | 1214 | 989 | 746 |
| Rh. microplus | 626 | 846 | 838 | 384 |
| Rh. decoloratus | 590 | 657 | 944 | 905 |

Table 3: Season-wise prevalence (%) of ticks from Punjab

| Season | Total | Infected | Prevalence (%) | Odds Ratio | Confidence Interval 95% | P-value |
|--------|-------|----------|----------------|------------|-------------------------|---------|
|        |       |          |                |            |                         |         |
| Spring | 3000  | 1239     | 41.30          | 2.936      | 2.614 - 3.297           | <0.001  |
| Summer | 3000  | 1676     | 55.87          | 5.282      | 4.704 - 5.930           | <0.001  |
| Autumn | 3000  | 887      | 29.57          | 1.752      | 1.554 - 1.974           | <0.001  |
| Winter | 3000  | 580      | 19.33          | 1.340      | 1.150 - 1.540           | <0.001  |

(P<0.05) significant.
These results are at par with the results of Ghosh et al. (2007), Durrani and Shakoori (2009), Rony et al. (2010), Sultana et al. (2015) and Ali et al. (2016) who also noted high infestation of ticks in Summer. However, the animals remained infested with ticks all over the year. Variation in the infestation of the tick may be due to geographical positions and climatic conditions of different study areas. Different environmental factors including rainfall, temperature and moisture support the existence of tick in any area (Greenfield et al., 2011), some other factors like season, availability of host status of nutrition in animals (Alonso et al., 2007; Yacob et al., 2008) and farming practices (Sajid et al., 2011) also affect ticks infestation ratio. The results of our research were similar with that of Mustafa et al. (2014) who also noted high tick prevalence in the hot months (June to August) and Atif et al. (2012) who reported the highest infestation of ticks in the months of June and July in the study areas of Sargodha district Punjab, Pakistan. The findings of the present research were also in line with the findings of Kabir et al. (2011) who noted the higher prevalence of ticks in summer season (41.66%) followed by winter (31.5%) in Bangladesh.

The current study had been conducted in four seasons in different agro-ecological areas of Punjab to examine the prevalence of ticks in animals. The prevalence findings of our research were in line with the findings of Iqbal et al. (2013) who noted 31% prevalence of tick species from Pakistan. However, the prevalence results of this study were in contrast with the results of Mustafa et al. (2014) who reported 85% in Sargodha district Punjab, Pakistan. This difference in the prevalence of ticks may be due to the variation in climatic and geographical situations of the study zones, study periods, target populations and farming practices (Iqbal et al., 2014). Prevalence of ticks in animals was abundant in Africa and Asia. A higher infestation of tick in these continents is due to the warmer climate which provides favourable conditions for the development of ticks, variation in housing styles, husbandry practices and strategies for tick control. It was reported that in last few years, the ticks’ prevalence in the province Punjab had been increasing rapidly which could be due to the resistance of acaricides (Sajid et al., 2008, 2009; Ali et al., 2013; Mustafa et al., 2014). The prevalence was considerably higher in the Central region in the current study due to high temperature that offered optimum situations for the multiplication of tick than the Northern region where the temperature was low. The results showed that the prevalence of tick infestation was associated with animal species, season and study area. The highest prevalence was found in summer followed by spring, autumn and winter seasons.

During our research, we collected ticks from 120 livestock farms from 12 districts of Punjab, Pakistan. We found 9 tick species from three genera, i.e., two hard genera (Hyalomma & Rhipicephalus). Most of the ticks were found from buffalo during the summer season. Tick infestation was high in the central zone of Punjab, Pakistan. Hyalomma species are the vectors of most of the diseases found in animals and man. Most of the Hyalomma ticks were recorded from the Western zone, so here is more danger of diseases. Farmers should treat their animals to kill ticks before transporting their animals to other parts of the country to spread of transmission of ticks and tick borne diseases.

Authors contribution: SN, MB & IY designed the study and wrote the manuscript, AR and MY edited the manuscript.

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