Original Article

Prevalence, Distribution And Risk Factors Of Hard Ticks In Large Ruminants Of District Gujrat, Punjab, Pakistan

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Abstract

Background: Ticks are blood sucking ectoparasites that act as host for various pathogens like bacteria, viruses and protozoans that cause a number of diseases in humans and animals. They pose a serious threat to livestock industry because their major hosts are cattle and buffaloes. Present study was conducted in this scenario to find out prevalence and distribution of ticks in specified areas.

Materials and Methods: This research was conducted from January 2021 to July 2021. Two hundred large ruminants were examined on monthly basis. In order to find out prevalence area, age, gender, and seasonal parameters were observed. Total one hundred and ninety ticks were collected from different parts of their body.

Results:

Identified Species: Five species belonging to two genra (Hyalomma and Rhipicephalus) and one sub genus (Boophilus) were identified including Hyalomma marginatum, Hyalomma excavatum, Rhipicephalus (Boophilus) microplus, Rhipicephalus (Boophilus) decolorates and Rhipicephalus camicasi. Hyalomma excavatum was the most prevalent specie.
Percentage Prevalence: Tehsil Gujrat was the most affected by tick infestation (67%) followed by Tehsil Kharian (55%) and Tehsil Sarai Alamgir (39%). This study suggested that cattle were more susceptible to tick infestation (58%) than buffaloes (50%).
Effect of Seasonal Variations on Tick Infestation: Seasonal variations has major influence on tick infestation. They were more prevalent in summer followed by spring and winter. Temperature, humidity and rainfall has significant effect on tick infestation ($p<0.05$).
Gender: Males were more susceptible to tick infestation (77%) as compared to the females (40%). **Age**: Large ruminants at younger age (4<years) were more susceptible than adults (4>years). **Predilection Sites**: Major predilection sites of ticks were external genitals (42%), followed by the tail (14%), dewlap (11%), udder (9%), thighs (8%) face (5%) and flanks (3%).

Breeds: Australian Friesian breed among cows (38%) and Neeli Ravi breed among buffaloes (25%) was more likely to be infested with ticks.

Conclusion: Keeping in view prevalence rate and factors influencing spread of ticks further studies should be planned to design management systems in order to minimize the economic losses.

Key Words Ixodidae; Tick infestation; Prevalence; Hyalomma; Rhipicephalus; Boophilus

INTRODUCTION

Ticks are small blood sucking ectoparasites that belong to sub phylum chelicerata due to presence of pair of fangs like structure known as Chelicerae at anterior side of mouth. They are closely related to spiders and scorpions rather than insects hence they are placed in class Arachnida. There are about 899 species of ticks that are present all around the world. 10% of these species are vector of about two hundred diseases [1]. Two major families of ticks i.e., Ixodidae that are identified as hard ticks as they possess a firm dorsal shield and Argasidae which are recognized as soft ticks because they lack dorsal scutum. Majority of tick species belong to family ixodidae [2]. They have cosmopolitan distribution but they are more abundant in the region of sub-tropical and tropical [3].

Ticks are vector of various pathogens like viruses, bacteria and protozoans that causes many infections in animals and humans. The common tick-borne infection /diseases are babesiosis anaplasmosis, theileriosis, and heart water. They also cause anaemia, paralysis and dermatitis in large ruminants [4] also they are responsible for about one hundred thousand human diseases all around the world [5]. Their bites also decreases the skin and hides value of infested animals. Tick species having even with short hypostome like Boophilus are crucial for large ruminants when they are present in large numbers. However, tick species with extensive hypostome like Amblyomma are more dangerous and they cause inflammations in skin of large ruminants due to secondary bacterial infections. Amblyomma species may also cause loss of teats depending upon their predilection site. Hence calf mortality is increased due to teat loss. The saliva of some tick species is toxic because it contains paralyzing toxins. For example, Dermacentor andersoni cause death even in adult cattle [6]. Another example of tick borne disease in bovine is known as sweating sickness which is eczema like condition of calves in African region [7]. These disease are economically important because they not only cause immense economic loss by affecting milk and meat production but also by generating significant cost on management of ticks and inducing abortions and hence increasing mortality rate in animals. As a result, they obstruct the development of livestock industry in numerous countries of the world [8].

They also cause immense economic loss worldwide [9]. Tick borne pathogens effect approximately 1.3 billion cattle globally and in 1996 it has caused annual loss of 22-30 billion US$. Tick borne diseases cause production losses of about 13.9-18.7 billion US$ annually inflicting 80% cattle population have the possibility of tick infections [10]. It is estimated that skin and hide defile cause the economic loss of about 500000US$ due to infestation of ticks [11]. They are widely distributed throughout the world and different studies are reported from different countries. Tick borne diseases have marked high in some countries of world having poor management. These regions include Africa, America and Asia where demand of livestock products has raised [4].

Livestock industry plays a significant role in economy of Pakistan. This sector helps to elevate the socioeconomic conditions of backward areas where small farms are used to meet the societal needs. 11.8% GDP was contributed by livestock in financial year of 2013-2014. The population of cattle and buffaloes is estimated as 35-40 million which provides 18000-31000 million tons milk [12]. Despite all this significance, this sector also faces a hideous issue i.e. parasitism. Pakistan has many physiographic parameters that support the tick infestation in animals and humans [13].

Different researches have reported the prevalence and identification of tick species in Pakistan but no study has been done in district Gujrat to check out distribution of different tick species affecting large ruminants in study area. The hypothesis of this research was to ascertain the prevalent tick species in selected localities of district Gujrat, their identification based on available identification keys, distribution pattern and different factors affecting their distribution.

Objectives: This research was conducted to find out prevalence and distribution by identifyig various species of hard ticks (Ixodidae) in three tehsils of district Gujrat namely Gujrat, Kharian and Sarai Alamgir.

MATERIALS AND METHODS

1 Study Area

Study was conducted during the month of January 2021 to July 2021 to collect the ticks belonging to family Ixodidae from district Gujrat, Punjab, Pakistan. Gujrat is located between two rivers Jehlum and Chenab. This district is about 229 m above sea level at 32.57ºN latitude and 74.09ºE longitude. Average temperature is around 25ºC with 32% humidity. The plains of Gujrat are hot and dry in summer while cold and dry in winters [14]. Different localities were selected from three tehsils of Gujrat for random sampling.

2 Selection of Sample

A cross sectional study was conducted on two hundred large ruminants of both sexes with different age groups from district Gujrat. Random sampling method was used to collect data.

3 Collection and Preservation of Samples

Hard ticks were taken from large ruminants of both sexes and different age groups on monthly basis after they were caste down and restrained completely. The distribution of ticks on large ruminants was determined by conducting survey dealing with different factors such as age, gender, breed, infestation sites, locality, temperature, humidity rainfall, season and tick species.

Ixodid ticks were searched from animal coat and were collected with the help of brush and forceps so that their body parts remain intact. After collection, specimen were shifted in small plastic tubes filled with 70% ethanol as preservative and marked properly with age, sex, date and area of collection. Appropriate precautionary measures were taken while collection of specimens. Collected samples were transported to the laboratory of department of Zoology, University of Gujrat for identification. Ticks were removed from plastic tubes and placed in petri dishes covered with filter paper so that excess preservative will be absorbed.

4 Identification of Ticks

Ticks were observed under dissecting microscope (LABOMED CSM2) and then identified by running through dichotomous key by [15].

5 Mounting of Ticks

Permanent mounting method was used to preserve the hard ticks. After identification, hard ticks were dehydrated through series of alcohol i.e. 30%, 50%, 70%, 90% and 100% respectively for 20 minutes. After dehydration specimens were treated with xylene and mounted in Canada balsam [16].

6 Statistical Analysis

Prevalence of ticks was calculated by using following formula:

Infestation Rate= (No on infested animals/ Total no of animals) × 100

Mean values of different parameters were obtained by SPSS (Statistical Package for Social Sciences) version 21.0. Relationship of tick infestation with age, gender and breed of host animal was assessed by Chi Square (X^2) test. Association of temperature, humidity and rainfall with tick infestation was calculated by using linear regression model. The p value was deliberated as statistically significant ($p < 0.05$).

RESULTS

Identified Species

Current study was conducted in three tehsils of district Gujrat from January 2021 to July 2021. During this time period two hundred bovines were examined from different localities of district Gujrat and one hundred and ninety ticks belonging to family ixodidae were collected from them. Out of 190 collected specimens 67, 62 and 61 were collected Gujrat, Kharian and Sarai Alamgir tehsils respectively (Table 1). Overall infestation rate was recorded as 54% (Table 2). Total three genera and five species were identified during this research. In general, 51.5 % ticks were identified as females and 48.4% as males (Table 1).The prevalent genera of ticks in district Gujrat were Hyalomma, Rhipicephalus (Boophilus) and Rhipicephalus. Two species Hyalomma excavatum (Figure 1), Hyalomma marginatum (Figure 2) were from Hyalomma genus. Two species Rhipicephalus (Boophilus) microplus , Rhipicephalus (Boophilus) decoloratus were from Boophilus genus and only one specie Rhipicephalus camicasi (Figure 3,4) was from Rhipicephalus genus. Tehsil Gujrat was the most affected by tick infestation (67%) followed by Tehsil Kharian (55%) and Tehsil Sarai Alamgir (39%).

Table 1: Tehsil wise Detail of Caught Hard Ticks from District Gujrat

Hard Tick Species	**Gujrat**	**Kharian**	**Sarai Alamgir**	**Total**
Hyalomma excavatum	22	24	23	69
Rhipicephalus (Boophilus) microplus	16	0	18	34
Rhipicephalus (Boophilus) decoloratus	0	16	8	24
Rhipicehalus camicasi	6	4	0	10
Hyalomma marginatum	16	16	21	53
Total	**67**	**62**	**61**	**190**

Table 2: No of Animals Observed During Study Period

Host	**Observed Cases**	**Positive Cases**	**Infested %**
Female Buffaloes	62	21	33.8
Male Buffaloes	38	29	76.3
Female Cattle	61	28	45.9
Male Cattle	39	30	76.9
Total	200	108	54

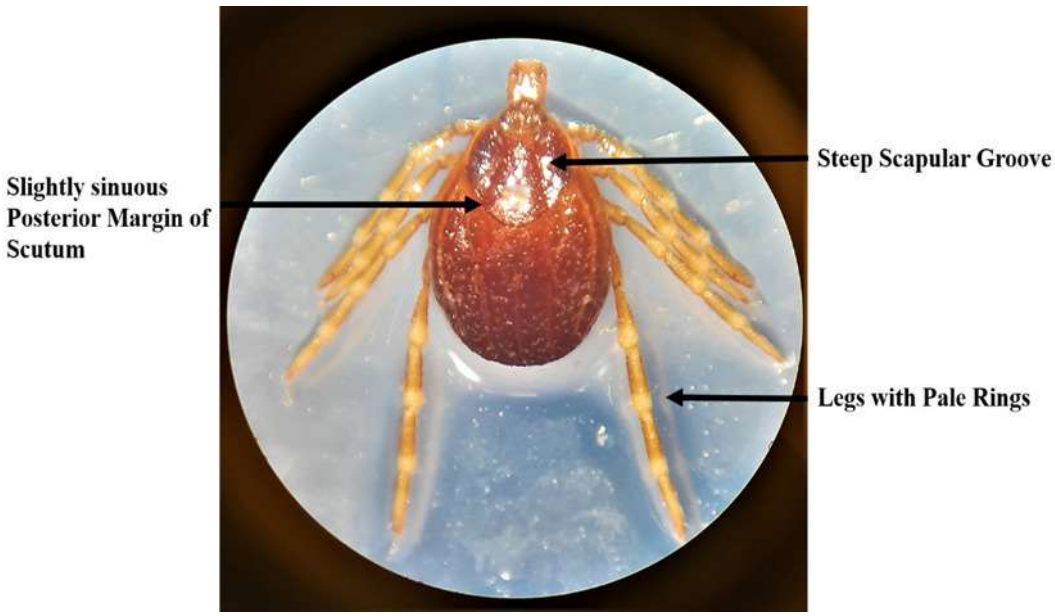

Figure 1: Dorsal Side of Female Hyalomma excavatum

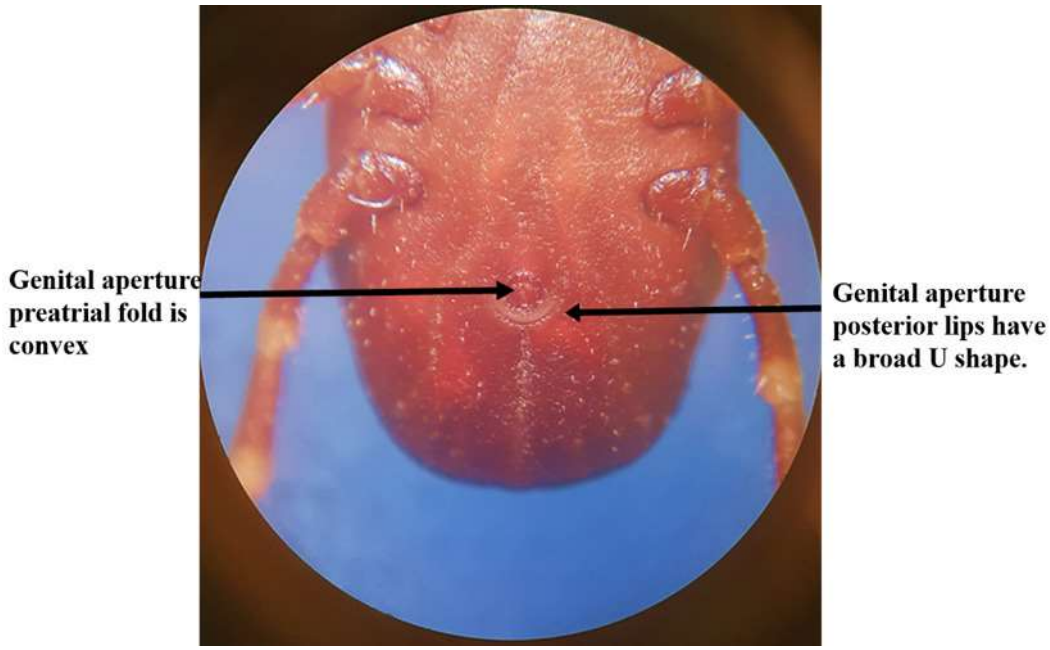

Figure 2: Ventral Side of Female Hyalomma marginatum

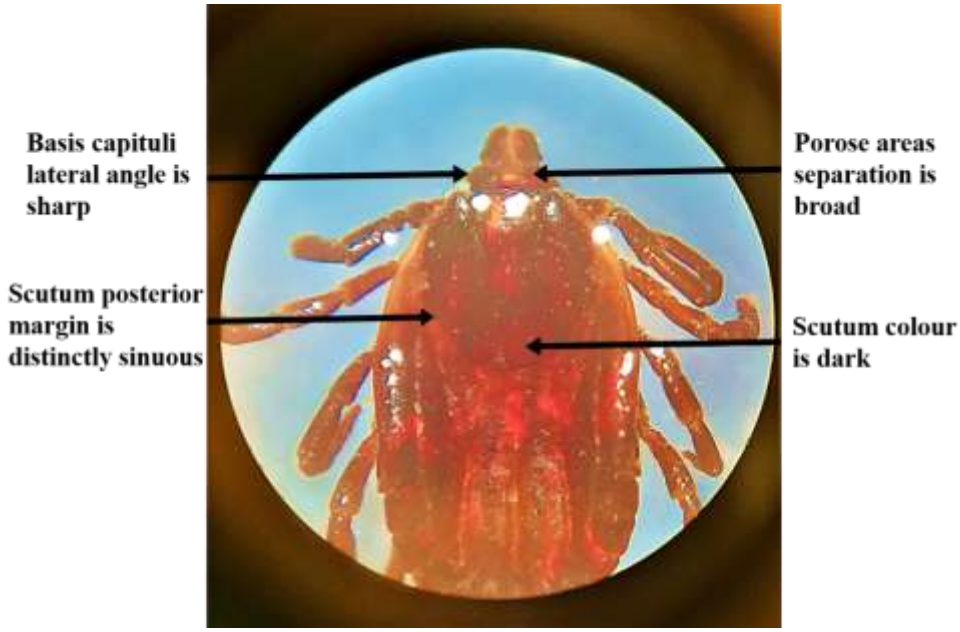

Figure 3: Dorsal Side of Female Rhipicephalus camicasi

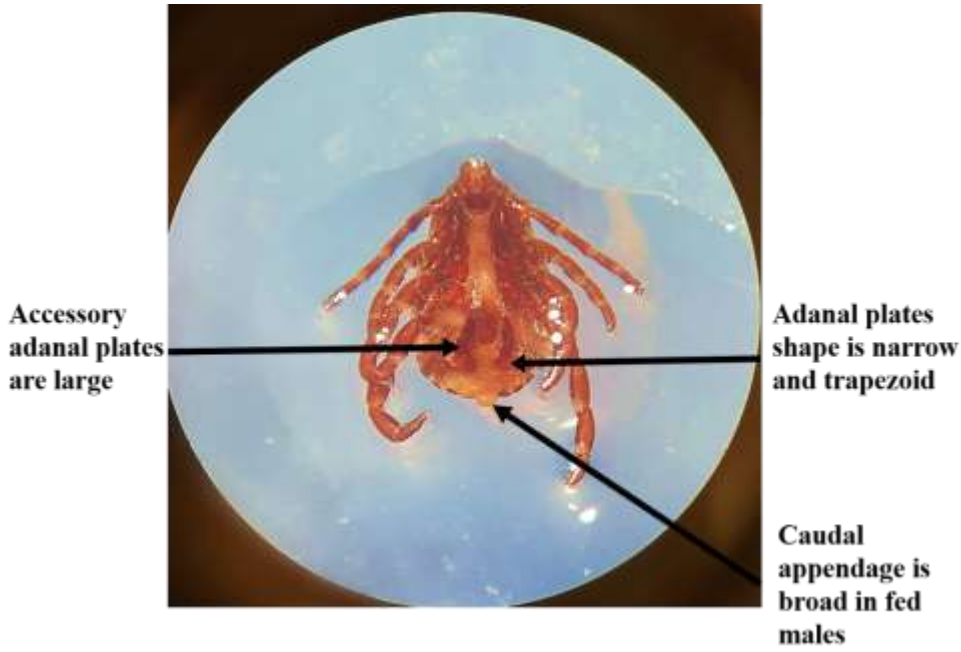

Figure 4: Ventral Side of Male Rhipicephalus camicasi

Effect of Temperature, Humidity and Rainfall with Tick Infestation

Temperature: This study revealed that temperature has significant effect on prevalence of ticks ($p<0.05$). Highest infestation was recorded in the month of July while lowest infestation was recorded in January (Figure 5). This observation has shown that temperature was the significant factor that control tick infestation ($p<0.05$). Optimum temperature for tick infestation was 36ºC-38ºC.

Humidity: According to this study, Humidity is positively linked with tick infestation ($p>0.05$). As humidity increased on monthly basis, number of ticks were increased. July was the most humid month of study duration in which highest number of ticks were collected from study area.

Rainfall: This study has covered four seasons including winter (January), spring (February-March), summer (May-June) and rainy season (July). Seasonal analysis of the tick infestation had suggested that maximum figure of ticks were collected during rainy season followed by spring, summer and lastly in winters (Figure 6). As maximum number of ticks were collected in July so it is deduced that rainfall and tick infestation are significantly correlated ($p<0.05$). Rainy season has significant effect on tick infestation ($p<0.05$) however, winter is least prevalent season for ticks.

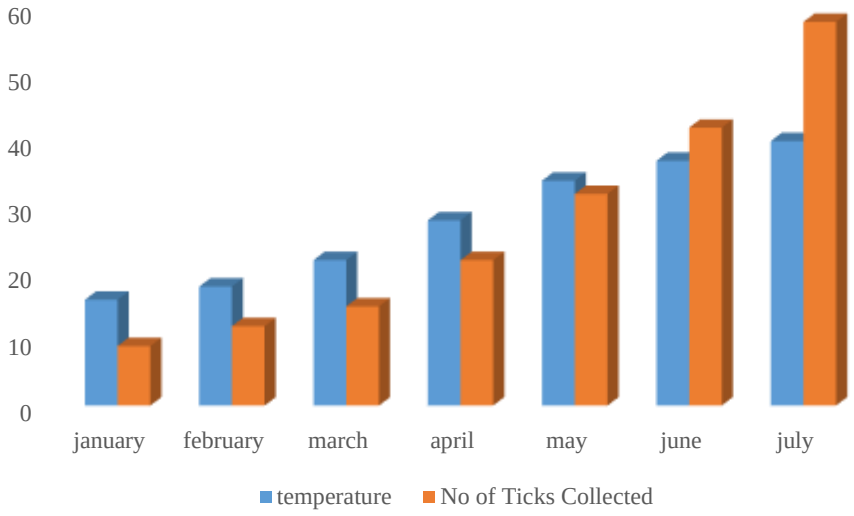

Figure 5: Effect of Temperature on Ixodid Tick Infestation

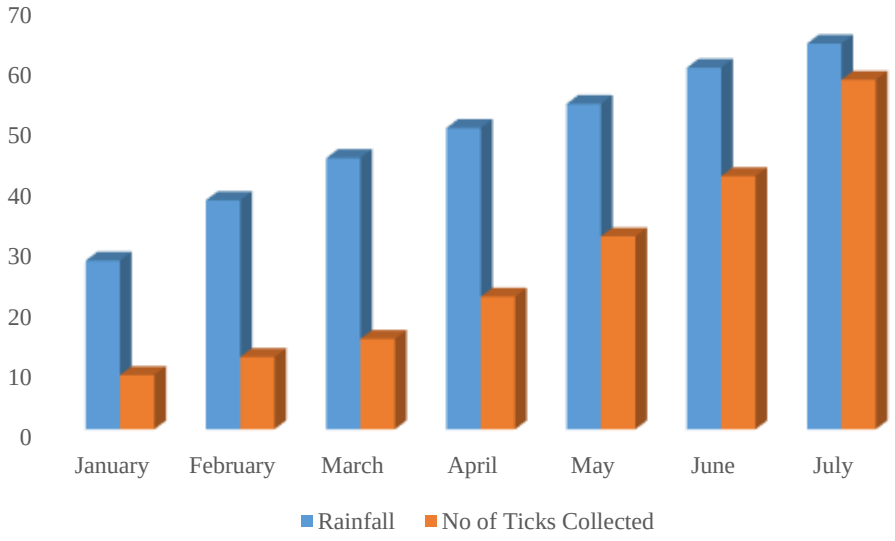

Figure 6: Effect of Rainfall on Ixodid Tick Infestation average %age vs. months

Effect of Seasonal Variation on Ixodid Tick Infestation

This study has covered four seasons including winter (January), spring (February-March), summer (May-June) and rainy season (July). Seasonal analysis of the tick infestation had suggested that highest number of ticks were collected during rainy season followed by spring, summer and lastly in winters. As highest number of ticks were collected in July (31%) while lowest in the January (4%). So it is deduced that rainfall and tick infestation are significantly correlated ($p<0.05$).

Association of Bovine Type, Age, Gender and Breed and Predilection Sites of Bovine with Tick Infestation

Bovine Type: This study suggested that cattle were extra sensitive to tick infestation (58%) than buffaloes (50%).

Gender: Current study has revealed that males are more prone to tick infestation than females. Infestation rate was recorded higher in males as compared to females (Figure 7). This study revealed that overall occurrence of tick invasion in large ruminants was 54%. Prevalence rate of hard ticks were higher in males (77%) as compared to females (40%).

Age: This study resulted that large ruminants at younger age are more susceptible to tick invasion as related to adults (Figure 7). About 56% large ruminants were infested upto age of 4≥ years. Infestation in adults were recorded as 46% upto age 10≥years. Age is a crucial determinant in prevalence of tick infestation. The current study showed that bovines at younger age were greater prone to ixodida attack as compared to adults.

Breed: This study has reported that Australian Friesian breed (38%) of cows is more likely to be infested with ticks as compared to Dhanni (8%) and Red Sindhi (10%). On the other hand, Neeli Ravi breed (25%) is more prone to tick infestation in buffaloes as compared to local (15%) and Sahiwal breed (12%). Most infested breed among cows was Australian Friesian and Neeli Ravi among buffaloes.

Predilection Sites: Different ectoparasites prefer different attachment sites on their host body. In present study, major predilection sites of ticks were external genitals (42%), followed by the tail (14%), dewlap (11%), udder (9%), thighs (8%) face (5%) and flanks (3%). Boophilus decoloratus was mostly present on head, back and dewlap while Hyalomma marginatum prefer external genitals and tail region because these areas were highly supplied with blood. Boophilus larvae were mostly collected from flanks and dewlap while genus Hyalomma was present from ears, neck, thighs and lips of vulva.

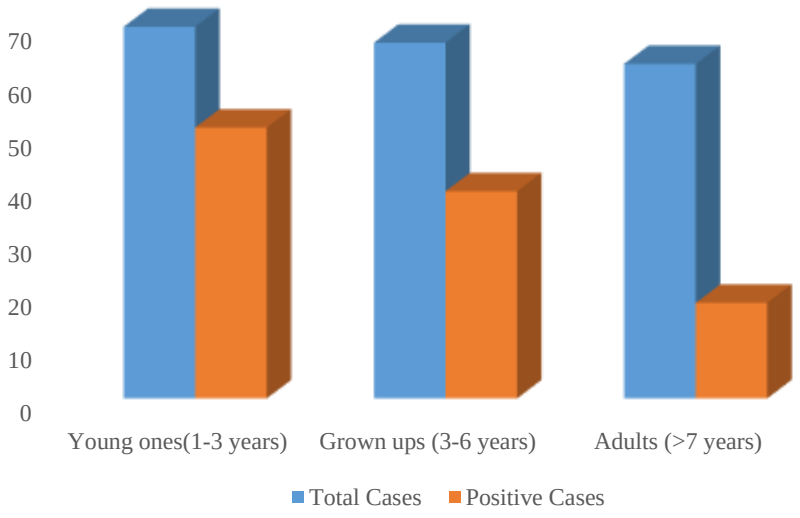

Figure 7: Association of Age with Ixodid Tick percentage Infestation Rate

DISCUSSION

In many countries it is serious threat of tick infestation specially in dairy industry including Pakistan. Current research was calculated to explore the diversity of hard ticks in district Gujrat. Two hundred large ruminants including buffaloes and cows were examined and one hundred and ninety hard ticks were collected from their body. This study revealed that overall prevalence of tick infestation in large ruminants was 54%. [17, 18] reported that Hyalomma anatolicum was widely distributed in Punjab which is contradictory to present study which stated that Hyalomma excavatum was more prevalent specie in district Gujrat. This study suggested that cattle were more susceptible to tick infestation (58%) than buffaloes (50%). This finding is in line with [19]. who reported that cattle are more prone to tick infestation than buffaloes. Similar studies were also conducted at Quetta, Dera Ghazi Khan with comparable results [20]. High infestation rate in cattle could be attributed to dense hair coat which help the ticks attach firmly with skin. Moreover, wallowing nature of buffaloes might help in dropping f ticks from their body resulting in lower rate of tick infestation [21].

Prevalence rate of hard ticks were higher in males (77%) as compared to females (40%). Similar results were reported by [18, 22]. This observation can be supported by fact that males are mainly used for breeding and meat purpose only and therefore are not properly maintained by their owners. Moreover, they have hair hide which is preferred by the hard ticks.

This study resulted that large ruminants at younger age are more susceptible to tick infestation as compared to adults. About 56% large ruminants were infested upto age of 4≥ years. Infestation in adults were recorded as 46% upto age 10≥years. Age is a crucial determinant in prevalence of tick infestation. The current study showed that bovines at younger age were more prone to ixodida attack as compared to adults. The reason can be explained by the fact that younger bovines has thinner skin and hairy hide which facilitate tick to cling and penetrate their mouthparts for blood sucking. Moreover, large ruminants at younger age has weak immune system which further increased the risk of tick attack [18, 22]. In adults infestation rate was minimum because they already had experienced repetitive tick attacks and developed strong immune system against tick [23, 24] reported that in Pakistan calves were 2.23 times more susceptible to tick infestation than adult cattle. However a contradictory study in which adult cattle were more infested than calves was reported by [25, 26]. This study has reported that Australian Friesian breed (38%) of cows is more likely to be infested with ticks as compared to Dhanni (8%) and Red Sindhi (10%). On the other hand, Neeli Ravi breed (25%) is more prone to tick infestation in buffaloes as compared to local (15%) and Sahiwal breed (12%). Most infested breed among cows was Australian Friesian and Neeli Ravi among buffaloes because they have thick hairs that help ticks to attach firmly with their host body for blood meal. This observation coincided with [27].

Different ectoparasites prefer different attachment sites on their host body. In present study, major predilection sites of ticks were external genitals (42%), followed by the tail (14%), dewlap (11%), udder (9%), thighs (8%) face (5%) and flanks (3%). Boophilus decoloratus was mostly present on head, back and dewlap while Hyalomma marginatum prefer external genitals and tail region because these areas were highly supplied with blood. Boophilus larvae were mostly collected from flanks and dewlap while genus Hyalomma was present from ears, neck, thighs and lips of vulva.

[11] conducted a survey on ticks in Eastern Ethiopia to check their prevalence and infestation sites in relation with gender, age and breed of cattle. In this survey, five species were identified and each species has its own specific infestation site on host body [11]. Based on these facts it can be said that is in agreement with already determined facts in various regions of the world. This study has suggested that external genitals were the prime predilection site in bovines followed by tail, dewlap, udder, thighs, face and flanks. This observation is similar to [28].

CONCLUSION AND RECOMMENDATIONS

This study was conducted in three tehsils of district Gujrat from January 2021 to July 2021. During this time period two hundred bovines were examined from different localities of district Gujrat and one hundred and ninety ticks belonging to family ixodidae were collected from them. Total five species belonging to three genera were identified

during this research. The most prevalent genera of ticks in district Gujrat were Hyalomma, Rhipicephalus (Boophilus) and Rhipicephalus. Two species Hyalomma excavatum, Hyalomma marginatum were from Hyalomma genus. Two species Rhipicephalus (Boophilus) microplus, (Rhipicephalus) Boophilus decoloratus were from Boophilus genus and only one specie Rhipicephalus camicasi was from Rhipicephalus genus. Tehsil Gujrat was the most affected by tick infestation followed by Tehsil Kharian and Tehsil Sarai Alamgir. This study suggested that cattle are more susceptible to tick infestation (58%) than buffaloes (50%). Seasonal variations has major influence on tick infestation. They were more prevalent in rainy season followed by summer, spring and winter. Temperature and rainfall has significant effect on tick infestation ($p<0.05$). However, humidity has no significant ($p>0.05$) relationship with prevalence of ticks. Males were more susceptible to tick infestation (77%) as compared to the females (40%). Large ruminants at younger age (4>years) were more susceptible than adults (4<years). Major predilection sites of ticks were external genitals (42%), followed by the tail (14%), dewlap (11%), udder (9%), thighs (8%) face (5%) and flanks (3%). Australian Friesian breed among cows (40%) and Neeli Ravi breed among buffaloes (28%) was more likely to be infested with ticks.

Thus by considering above study it can be concluded that a detailed study should be conducted on ticks to plan and implement the management system. Acaricides should be used before the onset of tick infestation. It is recommended that resistant breeds should be chosen to avoid economic loss. In this way, cost of antiparasitic treatment can be significantly reduced. Moreover, further studies should be planned on epidemiology of different ectoparasites of large ruminants in district Gujrat to assess their diversity and to design management systems in order to minimize the economic losses.

Data Availability Statement

Data can be provided on reasonable request to the corresponding author.

Competing Interests

Authors have declared that no competing interests exists

Ethical Approval: The ethical issues is not applicable

Consent of Participate: Not applicable

Consent of Publication: Not applicable

REFERENCES

1. Gargili, A., Kar, S., Yilmazer, N., Cerit, C., Sonmez, G., Sahin, F., Vatansever, Z. (2010). Evaluation of ticks biting humans in Thrace Province, Turkey. Kafkas Universitesi Veteriner Fakultesi Dergisi, 16(Suppl-A), 141-146.
2. Estrada-Peña, A. (2015). Ticks as vectors: taxonomy, biology and ecology. Scientific and Technical Review, 34(1), 53-65.
3. Shemshad, K., Rafinejad, J., Kamali, K., Piazak, N., Sedaghat, M. M., Shemshad, M., Enayati, A. A. (2012). Species diversity and geographic distribution of hard ticks (Acari: Ixodoidea: Ixodidae) infesting domestic ruminants, in Qazvin Province, Iran. Parasitology research, 110(1), 373-380.
4. Delgado, C., Rosegrant, M., Steinfeld, H., Ehui, S., & Courbois, C. (2001). Livestock to 2020: The next food revolution. Outlook on Agriculture, 30(1), 27-29.
5. de la Fuente, J., Estrada-Pena, A., Venzal, J. M., Kocan, K. M., & Sonenshine, D. E. (2008). Overview: ticks as vectors of pathogens that cause disease in humans and animals. Frontiers in Bioscience, 13(13), 6938-6946.
6. Spickett, A. M., Heyne, I. H., & Williams, R. (2011). Survey of the livestock ticks of the North West province, South Africa. Onderstepoort Journal of Veterinary Research, 78(1), 1-12.
7. Ambrose, N., Lloyd, D., & Maillard, J. C. (1999). Immune responses to Dermatophilus congolensis infections. Parasitology Today, 15(7), 295-300.
8. Abah, S., Njangloga, A. M., Zoli, A. P., Mamoudou, A., Sevidzem, S. L., Souley, A. & Tchuinkam, T. (2017). Commercial acaricides in pour-on formulation react differently in reducing tick numbers on cattle in extensive and intensive management systems in Bockle, North Cameroon. Direct Research Journal of Agricultural and Food Science, 5, 232-238.

9. Tadesse, F., Abadfaji, G., Girma, S., & Jibat, T. (2012). Identification of tick species and their preferred site on cattles body in and around Mizan Teferi, Southwestern Ethiopia. Journal of Veterinary Medicine and Animal Health, 4(1), 1-5.
10. Ghosh,S., Patra, G., Borthakur, S.K., Behera, P., Tolenkhomba, T.C., Das, M., & Lalnunpuia, C. (2018). Prevalence of hard tick infestations in cattle of Mizoram, Indian Biological Research, 1-11.
11. Kassa, S., & Yalew, A. (2012). Identification of Ixodide ticks of cattle in and around Hararamaya district, Eastern Ethiopia. Scientific Journal of Crop Science, 1(1), 32-38.
12. Jabbar, A., Abbas, T., Sandhu, Z. U., Saddiqi, H. A., Qamar, M. F., & Gasser, R. B. (2015). Tick-borne diseases of bovines in Pakistan: major scope for future research and improved control. Parasites & vectors, 8, 28.
13. Khalil, M. I., Lashari, M. H., & Akhtar, M. S. (2018). Prevalence of ticks infesting buffaloes in and around Jampur district Ranjanpur, Pakistan. FUUAST Journal of Biology, 8(2), 327-330.
14. Hussain, M., Malik, M. F., Siddique, S., Umar, M., Zainab, T., & Zafar, F. (2018). Diversity and Distribution of Coccinellid Beetles in Irrigated and Rainfed Fields of Gujrat, Punjab, Pakistan. Punjab University Journal of Zoology, 33(1), 1-6
15. Walker, A. R. (2003). Ticks of domestic animals in Africa: a guide to identification of species . Edinburgh: Bioscience Reports, (3-210)
16. Ramzan, M., Naeem-Ullah, U., Saba, S., Iqbal, N., & Saeed, S. (2020). Prevalence and identification of tick species (Ixodidae) on domestic animals in district Multan, Punjab Pakistan. International Journal of Acarology, 46(2), 83-87.
17. Atif, F., A., Khan, M. S., Iqbal, H. J., Ali, Z., & Ullah, S. (2012). Prevalence of cattle tick infestation in three districts of the Punjab, Pakistan. Pakistan Journal of Science, 64(1), 49.
18. Tsai, Y. L., Chan, P., Chen, S., Hsieh, J., & Chuang, S. (2011). Survey of species of ticks infesting cattle in Taiwan. Taiwan Veterinary Journal, 37(2), 74-82.
19. Kakar, M. N., & Kakarsulemankhel, J. K. (2008). Prevalence of endo (trematodes) and ecto-parasites in cows and buffaloes of Quetta, Pakistan. Pakistan Veterinary Journal, 28(1), 34.
20. Khajuria, V., Godara, R., Yadav, A., & Katoch, R. (2015). Prevalence of ixodid ticks in dairy animals of Jammu region. Journal of parasitic diseases, 39(3), 418-421.
21. Bibi, S., Rafique, N., Kareem, A., Taj, M. K., Iqbl, K., Bibi, A., ... & Ijaz, A. (2020). 15. Prevalence and taxonomic identification of hard ticks (Ixodidea) found in livestock of Harnai District, Balochistan, Pakistan. Pure and Applied Biology (PAB), 9(4), 2330-2338.
22. Sajid, M. S., Iqbal, Z., Khan, M. N., Muhammad, G., & Khan, M. K. (2009). Prevalence and associated risk factors for bovine tick infestation in two districts of lower Punjab, Pakistan. Preventive veterinary medicine, 92(4), 386-391.
23. Manan, A., Khan, Z., Ahmad, B.,& Abdullah. (2007). Prevalence and identification of ixodid tick genera in frontier region Peshawar. Journal of Agriculture and Biological Sciences, 2(1):21–25
24. Kabir, M.H.B., Mondal, M.M.H., Eliyas, M., Mannan, M.A., Hashem, M.A., Debnath. N.C., Miazi.O.F., Mohiuddin, C., Kashem.M,A., Islam, M.R., Elah, M.F. (2011). An epidemiological survey on investigation of tick infestation in cattle at Chittagong district, Bangladesh. African Journal of Microbiology Research, 5(4):346–352
25. Razzak,A., Shaikh, H. (1969). A survey on the prevalence of ticks on cattle in East Pakistan. Pakistan Journal of Vetinary Sciences, 3(2):50–60
26. Yakhchali,M., & Hasanzadehzarza, H.S. (2004). Study on some ecological aspects and prevalence of different species of hard ticks (Acarina:Ixodidae) on cattle, buffalo and sheep in Oshnavieh suburb. Pajouhesh-va-Sazandegi. Animal Fish Science, 63:30.
27. Veríssimo, C. J., Giglioti, R., D'Agostino, S. M., de Toledo, L. M., Katiki, L. M., Duarte, K. M. R., & de Miranda Santos, I. K. F. (2019). Cattle herd shearing can help to control Rhipicephalus microplus ticks. Experimental and Applied Acarology, 79(1), 99-106.
28. Sajid, M.S., Iqbal, Z., Khan M,N., Muhammad, G. & Iqbal, M.U. (2007) Effect of Hyalomma ticks (Acari: Ixodidae) on milk production of dairy buffaloes (Bos bubalus bubalis) of Punjab Pakistan. Italian Journl of Animal Sciences. 6:939–941.