ORIGINAL ARTICLE



Distribution and prevalence of ticks on livestock population in endemic area of Kyasanur forest disease in Western Ghats of Kerala, South India

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Abstract Tick borne zoonotic diseases are one of the major emerging threats to live stock and public health in India, especially in Western Ghats of south India. Since livestock and wild animals share habitats and grasslands, it is important to know the species composition of major tick parasitism on live stock as well as their geographical distribution for effective control of tick and tick borne diseases. This study provides basic knowledge that is necessary to initiate Kyasanur Forest Disease (KFD) prevention programs in these areas. Ticks were sampled from Wayanad districts of Kerala from domestic animals and identified morphologically. A total of 195 cattle searched, in which 168 (86.15%) cattle were infested with ticks and a total of 3633 ticks comprising three genera and seven species were collected, Rhipicephalus microplus (52.71%) was prevalent species followed by Haemaphysalis bispinosa (16.9%), Rhipicephalus decoloratus (15.77%), Haemaphysalis turturis (11.42%), Rhipicephalus sanguineus (1.32%), Amblyomma integrum (1.15%) and Haemaphysalis spinigera (0.71%) were identified based on their morphological characters. As R. microplus was the prevalent species, the risk of transmission of babesiosis and anaplasmosis to cattle increases and the presence of Haemaphysalis sp. point out the risk of KFD in among the tribal colony people and it can be reduced by applying with acaricides on domestic animals.

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Introduction

Ticks have become competent vectors, efficient distributers and amplifying agents of diverse pathogens within animal and human populations. The rate of effective contact between vector ticks with their host and the obtaining of blood meals are the driving forces of the rapid distribution of tick borne diseases within and among populations. (Peter et al. 2005; Razmi et al. 2007; Ghosh and Nagar 2014). In India, 106 tick species were recorded to be infesting domestic and wild animals (Geevarghese et al. 1997). These ticks are of very high veterinary and medical importance because of its major effect on the animal husbandry and productivity of livestock, weight gain, milk production and quality of hide (De Castro et al. 1997) and zoonotic pathogen transmission to humans. The global economic losses due to tick infestation have been estimated as US \$14,000-18,000 million annually and in India it was approximately US \$ 498.7 million (Singh and Rath 2013). In addition these animals are also serving as reproductive host for KFD vector ticks.

In India, the studies on ticks received importance after the discovery of KFD, transmitted by *Haemaphysalis* sp. in Western Ghats (Mourya et al. 2014). In human variety of tick species involved in the transmission of different pathogens which cause diseases like borreliosis or lyme diseases which is transmitted by Ixodid ticks (Maria et al. 2012), Rocky Mountain spotted fever is transmitted by *Dermacentor* sp. (Socolovschi et al. 2009a, b), Rickettsiosis and Boutonneuse fever transmitted by *Rhipicephalus sanguineus* (Socolovschi et al. 2009a, b; Levin

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et al. 2009). Hyalomma sp. and Haemaphysalis sp. which are responsible for viral diseases of Crimean-Congo haemorrhagic fever and Kyasanur forest disease (Mourya et al. 2014). The epidemic of Indian tick typhus was reported from Himachal Pradesh (Kumar et al. 2011) and serologically positive cases of Lyme disease and rickettsiosis were reported from Tamil Nadu and Pondicherry (Sharma et al. 1992; Stephen et al. 2018). The first instance of Crimean-Congo haemorrhagic fever was detected from Gujarat (Kumar et al. 2014). Since livestock and wild animals share habitats and grassland in these regions, studies are necessary to determine if ticks can transmit pathogens between livestock and wildlife, and even to humans in the region. This study provides very important information and basic knowledge that is necessary to initiate KFD prevention programs in these areas. In Kerala KFD is endemic in Wayanad district, however, only a few studies were done about the tick fauna in different hosts (Prakasan and Ramani 2007; Shyma et al. 2013). It is important to know the prevalence of the tick species parasitism on cattle as well as their geographical distribution for the control of tick and tick borne diseases. The aim of present study is to reveal the common tick fauna present in domestic animals of KFD endemic area in tribal settlements of Wayanad district, Kerala.

Materials and methods

Ethical clearance

All aspects involving domestic animals use were conducted in accordance with the Indian Animal Welfare Act, 2011 (section 11), and recognized standards for the care and use of animals. Written consent from cattle owners were also taken as a prior permission for tick collection from respective cattle.

Study area

Wayanad district is a hill district in North-East of Kerala with an area of 2131 km², in the part of Western Ghats lie between Latitude N 11°37' and Longitude E 76°58' and situated at an altitude ranging from 2500 to 3000 ft above Sea Level. Wayanad district has the highest forest cover of 83.3%, agriculture and animal husbandry is the principal occupation of the people. The district has highest tribal population of about 1.25 lakh consisting 17% of the total population. This region experiences a moderate, semitropical climate with an average temperature of 18–30 °C in summer and winter with mean rainfall of 2786 mm and the annual average relative humidity is above 60%. The area receives most of its rainfall under the

influence of southwest monsoons and less rainfall under the influence of the northeast monsoons.

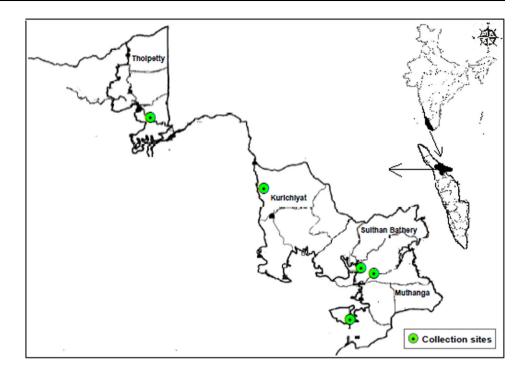
The abundance of vegetations and forest region provides favourable conditions for the survival of several tick species. The present study was performed in five different tribal villages (Tholpetty, Thottamoola, Cheeyambam, Kallumukku and Alathoor) in the northwest and southwest part of the district (Fig. 1) from January to May-2017. Cattle and goat are the main domestic livestock in the study area. Wild elephant, deer, giant squirrel, monkey, bison, boars and wolf are very abundant throughout the district, mostly in transition areas between forests and farmlands. In addition, small rodents and reptiles, which are major hosts for ticks, are also abundant in the district.

Tick collection and identification

Whole body of each domestic animal was searched thoroughly for the presence of ticks during day time. Precautions were taken before collecting ticks from cattle by wearing gloves, mask and applying the repellent Benzyl benzoate (I.P.25%) lotion to avoid the tick bites. The ticks were gently plucked up from the body of the host by hand with fingers were shield with rubber gloves or with the aid of blunt pointed tweezers. Ticks were grasped as close to the skin surface as possible and pulled upward with steady even pressure. Both nymph and adult ticks were collected and all were in fully blood fed condition. The specimens were kept in separate containers and preserved in 70% ethanol. Then the samples were labeled with the date, host, and locality on each container. These samples were transported to the laboratory of Medical entomology and Zoology, National Institute of Virology-Kerala unit, Alappuzha. The preserved specimens were washed, dehydrated and subjected for KOH treatment and mounted on a slide by using DPX (Distyrene, a plasticizer and Xylene) or Canada balsam for further reference. Adult ticks were identified up to species level following standard keys (Walker et al. 2013; Geevarghese and Mishra 2011) under stereomicroscope.

Site mapping and statistical analysis

Geographic coordinates of collection sites were marked using a handheld Global Positioning System (GPS). These coordinates were integrated to a GIS database using the software Arc GIS to map the distribution of ticks in domestic animals of the associated area. Spearman's Correlation analysis was used to assess the statistical significance among tick species using IBM-SPSS statistics viewer (version 20). Fig. 1 Spatial distribution map of study area inWayanad district



Result

The study was conducted from January to May from five sites such as Alathoor, Cheeyambam, Kallumukku, Tholpetty and Thottamoola in Western Ghats of Wayanad district, Kerala state. A total of 195 domestic animals were searched in which 168 (86.15%) animals were found positive for ectoparasitism. The details of cattle tick infestation are presented in Table 1. Among the total animals surveyed 68.20% of cattle and 17.94% goats were infested with one or more species of ticks. Among the five sites visited, maximum number (50.91%) of ticks was collected from Alathoor followed by Kallumukku 40.13% and in other three sites such as Tholpetty 4.03%, Thottamoola 3.89% and Cheeyambam 0.94% of ticks were collected (Table 2 and Fig. 2).

A total of 3633 ticks comprising adult 2607 (71.76%) and immature 1026 (28.24%) ticks belonging to three genera and 7 species were collected, namely Rhipicephalus, Haemaphysalis and Amblyomma are commonly prevalent in all sites studied. The highest prevalence rate was identified as Rhipicephalus microplus 1915 (52.71%) and it was the most predominant tick species, followed by R. decoloratus 573 (15.77%), Haemaphysalis bispinosa 614 (16.9%), H. turturis 415 (11.42%), R. sanguineus 48 (1.32%), Amblyomma integrum 42 (1.15%) and H. spinigera 26 (0.71%) (Table 2). Spearman's correlation analysis between the H. bispinosa and H. turturis were found as significant (P = 0.05). Among the *Rhipicephalus* sp., Rhipicephalus microplus was the most prevalent species followed by R. decoloratus and R. sanguineus. Three species of Haemaphysalis such as H. bispinosa, H. turturis and H. spinigera were collected (Table 3). These results

Sl. no.	Place	Cows		Goat		Total no. of cows and goat		% of infestation
		Searched	infested	Searched	infested	Searched	Infested	
1.	Alathoor	88	76	7	7	95	83	87.4
2.	Kallumukku	12	10	0	0	12	10	83.3
3.	Cheyambam	32	28	5	4	37	32	86.5
4.	Tholpetty	12	12	23	19	35	31	88.5
5.	Thottamoola	10	7	6	5	16	12	81.3
	Sub total	154	133	41	35	195	168	

Table 1 Rate of tick infestation on cows and goats of five sites

Table 2 Different species identified in cattle from five different sites with its sex differentiation

Sl. no.	Site	Species name	Number		Total	%
			Male	Female		
1	Alathoor	Rhipicephalus microplus	120	886	1006	27.69
	N 11°41′04.1″	R. decoloratus	55	295	350	9.63
	E ₀ 76°21'35.3"	R. sanguineus	6	13	19	0.52
	2860 ft	Haemaphysalis bispinosa	59	211	270	7.43
		H. turturis	18	177	195	5.37
		Amblyomma integrum	1	9	$ \begin{array}{r} 1006 \\ 350 \\ 19 \\ 270 \\ 195 \\ 10 \\ 1850 \\ 823 \\ 195 \\ 7 \\ 249 \\ 179 \\ 6 \\ 1459 \\ 5 \\ 5 \\ 7 \\ 18 \\ 35 \\ 51 \\ 7 \\ 13 \\ 62 \\ 6 \\ 8 \\ 147 \\ 30 \\ 21 \\ 4 \\ 26 \\ 35 \\ 26 \\ 142 \\ \end{array} $	0.27
Sub total			259	1591	1850	50.91
2	Kallumukku	R. microplus	80	743	823	22.65
	N 11°41′28.0″	R. decoloratus	46	149	195	5.36
	E ₀ 76°20'38.2"	R. sanguineus	1	6	7	0.19
	2967 ft	H. bispinosa	64	185	249	6.85
		H. turturis	14	165	179	4.92
		A. integrum	0	6	6	0.16
Sub total			755	704	1459	40.13
3	Cheeyambam	R. microplus	0	5	5	0.13
	N 11°47′27.8″	R. sanguineus	0	5	5	0.13
	E ₀ 76°13'21.0"	H. bispinosa	0	7	7	0.19
	2456 ft	A. integrum	6	12	18	0.49
Sub total			6	29	35	0.94
4	Tholpetty	R. microplus	11	40	51	1.40
	N 11°52′44.6″	R. decoloratus	0	7	7	0.19
	E ₀ 76°04'47.7"	R. sanguineus	0	13	13	0.36
	2451 ft	H. bispinosa	0	62	62	1.70
		H. turturis	0	6	6	0.16
		A. integrum	7	1	8	0.22
Sub total			18	129	147	4.03
5	Thottamoola	R. microplus	0	30	30	0.83
	N 11°37′37.2″	R. decoloratus	0	21	21	0.57
	E 76°19′47.9″	R. sanguineus	0	4	4	0.11
	2944 ft	H. bispinosa	0	26	26	0.71
		H. turturis	0	35	35	0.96
		H. spinigera	5	21	26	0.71
Sub total			11	131	142	3.89
Gross total			1649	1984	3633	

suggest that the areas are under the risk of KFD virus transmission. Average tick load per cattle of *R. microplus* was 11.39, *R. decoloratus* 3.41, *R. sanguineus* 0.28, *Haemaphysalis bispinosa* 3.65, *H. turturis* 2.47, *H. spinigera* 0.15 and *Amblyomma integrum* 0.25.

Discussion

The Wayanad district provides favorable ecological conditions for the propagation of haematophagous arthropods including ticks due to its richness in vegetation, domestic and wild animal fauna. The present study shows that cattle are highly infested with ticks, which are responsible for transmitting harmful pathogens to both animals and human. More than 85% of domestic animals were found as infested with ticks, suggested that the area is highly under the risk of tick borne diseases. The uncontrolled movements of

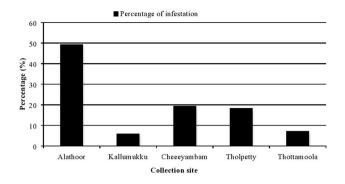


Fig. 2 Percentage of infestation in five surveyed sites

domestic animals in forest remain the primary reason behind the large numbers of ticks that are introduced from their territories into far regions. Earlier study from Prakasan and Ramani (2007) reported that Ixodid tick fauna of 5 genera and 18 species from nine districts of Kerala.

The most prevalent species observed in the study was R. microplus and H. bispinosa which was found infesting majority of the cattle and goat. The highest prevalence of R. microplus in Nilgiri, the adjoining area of Wayanad district in Western Ghats and also the same result was observed in KFD endemic area of Karnataka (Rajagopalan and Sreenivasan 1981; Kumar et al. 2014). It is considered as the most important tick parasite of livestock in the world and carries many pathogens (Ghosh and Nagar 2014; Kumar et al. 2014; Manaswini et al. 2017). According to Rajagopalan and Sreenivasan (1981) these species did not show any definite seasonal pattern of infestation. H. bis*pinosa*, on the other hand, is a tick breeding principally in the cattle sheds of the KFD area of Karnataka. The infestation of nymphs and adults was observed throughout the year. Thus, the infestation of H. bispinosa on cattle and buffaloes seems to reflect their restricted breeding habits in the cattle sheds and the availability of the hosts throughout the year (Rajagopalan and Sreenivasan 1981; Bhat 1971).

Three species of Haemaphysalis such as *H. bispinosa*, *H. turturis* and *H. spinigera* were observed on cattle and in

which *H. bispinosa* was the predominant species. This is a significant observation in the context of recently reported Kyasanur forest disease (KFD) incidence in the district. These species are playing an important role for the transmission of KFD in Karnataka, Kerala, Maharashtra and Goa (Rajagopalan and Sreenivasan 1981 and Mourya et al. 2014). Haemaphysalis spinigera, a major vector of KFD virus formed 0.71% of ticks on cattle. The immature of H. spinigera are known to parasitize certain species of wild mammal, small mammals and ground birds in the KFD area (Rajagopalan et al. 1968a, b, 1978; Rajagopalan and Anderson 1971; Rajagopalan 1972; Rajagopalan and Sreenivasan 1981). Cattle have become the most important hosts for the adults of H. spinigera and play a prominent role in the dissemination of larval population on the forest floor. The studies on the ixodid tick population in the forests of KFD area have shown a distinct seasonal appearance in the Western Ghats of Karnataka. The Haemaphysalis larvae start appearing on drags in September soon after the monsoon. They are abundant on the forest floor between November and December. This is followed by the appearance of nymphs in the drier months between December and January with a peak in February or March (Rajagopalan and Sreenivasan 1981). However, in our present study in Kerala (NIV Unpublished data) nymph peak density was occurred in the month of January thereafter gradually decreased and these ticks show the KFD virus prevalence during their peak months. The variation in the prevalence of ticks in this area may be due to climatic factors, climate can limit the distribution of ticks directly by affecting growth or survival. The presence of these species on cattle represents a twofold risk; cattle support their higher prevalence in the area and the bigger risk is the chance of potential vector contact/bite with the human and KFD amplifying host.

The movements and abundance of wild animals that serve as reservoirs of KFDV and hosts for *Haemaphysalis* spp., are profoundly affected by the climate, especially temperature. During summer season domestic animal home

Sl. no.	Species name	Nymph	Adult	Total	%	Average tick load/cattle
1	Rhipicephalus microplus	664	1251	1915	52.71	11.39
2	R. decoloratus	125	448	573	15.77	3.41
3	R. sanguineus	48	0	48	1.32	0.28
4	Haemaphysalis bispinosa	58	556	614	16.90	3.65
5	H. turturis	105	310	415	11.42	2.47
6	H. spinigera	26	0	26	0.71	0.15
7	Amblyomma integrum	0	42	42	1.15	0.25
Total		1026	2607	3633		

Table 3 Species of ticks recovered from cattle with respect to their genera and life stage

ranges (fringe area of forest) become wild animal refuges for food, water and shelter are common. As a result, there is a significant increase in the participation of cattle and buffaloes as hosts for ixodid ticks. The fringe area of forest is not only home range for domestic animals but also a potential KFDV circulating area between ticks and host animals. This network behavior enables the transmission of the KFD vector tick from domestic animal to wild animals vice versa. Among other species of ticks, the infestation of H. turturis, R. decoloratus and A. integrum on cattle suggests that these animals are also important hosts along with other mammals in the KFD area. R. sanguineus, the tropical dog tick, distributed globally throughout the warm countries; it is implicated as vector of B. motasi and B. ovis, and may play a role in transmission of B. equi (Ghosh and Nagar 2014). The pathogenic B. bigemina is transmitted by R. decoloratus, predominant with more than 15% in the present study, also responsible for the diseases in livestock such as Babesiosis or Redwater fever (Abdela and Jilo 2016).

There are currently many Haemaphyslis sp. capable of transmitting human and wildlife KFD virus in Western Ghats of India (Rajagopalan et al. 1968a, b, 1978; Rajagopalan and Anderson 1971; Rajagopalan 1972; Rajagopalan and Sreenivasan 1981). The presence of domestic animal host and their contributions to supporting different tick species can vary considerably across the forest. Previous studies in Kerala and Karnataka characterising ticks and their host have reported that domestic animal populations supporting vector ticks in Western Ghats of India (Rajagopalan and Sreenivasan 1981 and Kumar et al. 2002). The changing ecology, particularly deforestation, reforestation, plantation and associated increases in wildlife and domestic animal populations provide increased proximal host contact with the widely distributed bloodfeeding ectoparasites in these areas. Further studies of tickhost relationships are necessary for better understand tick species distributions, population dynamics prevalence of tick-borne pathogens, and their potential effects on human and animal health.

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Compliance with ethical standards

Conflict of interest The authors report no conflicts of interest.

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