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Prevalence, species composition, and patterns of ixodid tick infestations in theileriosis suspected cattle in Southern Tamil Nadu

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Abstract

Tick infestation remains a major constraint to cattle health and productivity in southern India, particularly in the context of emerging Oriental theileriosis. The present study investigated the prevalence, species composition, and patterns of single and mixed ixodid tick infestations in theileriosis suspected cattle across four southern districts of Tamil Nadu between January 2024 and August 2025. Of the 525 cattle examined, 410 were infested with ticks, yielding an overall prevalence of 78.10%, with no significant district wise variation. Single species infestations predominated (61.71%), while mixed infestations accounted for 16.38% of cases. *Rhipicephalus microplus* was the dominant species across all districts, followed by *Haemaphysalis bispinosa* and *Hyalomma marginatum*, exhibiting strong ecological clustering. Several district specific mixed tick infestations were observed, indicating localized ecological influences on tick coexistence. The findings highlight a stable yet heterogeneous tick fauna in southern Tamil Nadu and emphasize the need for region wide integrated tick management strategies to mitigate tick borne disease transmission, including *Theileria orientalis*.

Keywords: Tamil Nadu, *Rhipicephalus microplus*, mixed tick infestations, prevalence, tick ecology, cattle, southern

1. Introduction

In India theileriosis is an important tick-borne haemoprotozoan disease of cattle, caused by intracellular apicomplexan parasites of the genus *Theileria*. While *Theileria annulata* is well known for its high pathogenicity, *Theileria orientalis*, traditionally considered a benign species, has recently emerged as a major pathogen in several regions. Field outbreaks have documented that clinical Oriental theileriosis causes substantial economic losses due to reduced milk production in dairy herds, with an average loss of 624 litres per animal at 305 days of lactation compared to uninfected cows in Victoria, Australia (Perera *et al.*, 2014) [1]. Lew-Tabor and Rodriguez Valle (2016) [2] reported that worldwide tick and tick borne diseases (TTDS) caused an estimated loss of 22-30 billion USD/annum. Singh *et al.* (2022) [3] evaluated the economic impact of TTDS, including theileriosis, on Indian dairy production systems and reported a significantly high cumulative loss of 787.63 million USD due to TTDS in India. This shift in disease significance is largely attributed to evolving parasite genotypes and expanding tick vector populations across diverse agro-climatic zones (Watts *et al.*, 2016; Jacob *et al.*, 2024) [4, 5]. Globally, for *T. orientalis*, *Haemaphysalis longicornis* is the principal vector, while in India, *Rhipicephalus microplus* and *Haemaphysalis bispinosa* have been reported as vectors under natural field conditions, especially in crossbred cattle with lower innate tick resistance (Aparna *et al.*, 2011; Kakati *et al.*, 2015; Vinodkumar *et al.*, 2016) [6-8]. Transmission of *T. orientalis* occurs mainly through transstadial passage, but recent studies demonstrated parasite DNA in eggs supporting the possibility of limited transovarial transmission (Kakati *et al.*, 2015) [7]. These mechanisms contribute to persistent environmental contamination and silent spread of pathogenic genotypes.

Several ixodid tick genera, including *Haemaphysalis*, *Rhipicephalus*, and *Amblyomma*, are implicated in the epidemiology of bovine theileriosis worldwide (Baghel *et al.*, 2023) [9]. India remains a major endemic region, with *R. microplus*, *H. bispinosa*, and *H. anaticum* playing key roles in bovine *Theileria* transmission. Notably, *R. microplus* in northern India was found to harbor *T. orientalis* in 21.7% of examined ticks, confirming its vector competence. Similarly, the invasive *H. longicornis* in the United States has been shown to carry and transmit the highly pathogenic Ikeda genotype (Dinkel *et al.*, 2021) [10].

Studies from Vietnam and Myanmar also reported frequent detection of *T. orientalis* in *Haemaphysalis* and *Rhipicephalus* spp., indicating active cocirculation among ticks and livestock (Khukhuu *et al.*, 2011; Bawm *et al.*, 2014) [11, 12]. In India, repeated identification of the *H. bispinosa* and *R. microplus* from *T. orientalis* affected animals accentuates the risk of genotype driven pathogenicity (Aparna *et al.*, 2011; Baghel *et al.*, 2023) [6, 9]. Furthermore, the detection of *T. orientalis* DNA in ticks collected from wildlife-livestock interfaces in Kerala highlights the broader ecological involvement of reservoir hosts and the potential for silent enzootic maintenance (Nimisha *et al.*, 2019) [13].

The rapid ecological expansion of *H. longicornis*, aided by parthenogenetic reproduction and climatic adaptability, further increases the risk of dissemination in new geographical zones (Thompson *et al.*, 2020; Dinkel *et al.*, 2021) [14, 10]. Collectively, global evidence indicates that multiple tick genera are capable vectors of *T. orientalis*, although the dominant species vary regionally, *H. longicornis* in Asia-Pacific and Australasia, *R. microplus* and *H. bispinosa* in the Indian subcontinent, and the emerging role of invasive *H. longicornis* in the United States. This regional dependence reinforces the need for localized vector surveillance to understand transmission dynamics more accurately.

In this context, the present study was undertaken to assess the prevalence and species composition of hard ticks infesting theileriosis suspected cattle in the southern districts of Tamil Nadu and to generate baseline data essential for regional tick control strategies and molecular epidemiology of *T. orientalis*.

2. Materials and Methods

2.1. Study Area and Experimental Design

The study was conducted in the southern agro-climatic belt of Tamil Nadu, covering four districts, which represent diverse ecological zones ranging from semi-arid plains (Tirunelveli), high rainfall foothills (Tenkasi), dry coastal belts (Thoothukudi), to humid coastal cum highland regions (Kanyakumari). Mean annual temperatures range from 25-34 °C, with relative humidity often exceeding 70% in coastal localities. The study spanned from January 2024 to August 2025. Cattle from organized farms, smallholder units, and free grazing rural herds were included, representing varied

management systems with differential tick exposure. Animals showing clinical signs such as pyrexia, anaemia, jaundice, lymphadenopathy, reduced milk yield, and visible tick infestation were sampled as theileriosis suspected cases. The sample size of cattle was determined by power analysis using a 95% confidence level and 5% allowable error based on an expected prevalence of 20% as reported in previous studies from India (Velusamy *et al.*, 2014; Krishnamoorthy *et al.*, 2021) [15, 16].

2.2 Tick Collection and Preservation

Ticks were collected from infested cattle during clinical examinations across the study districts. Ticks were carefully removed using blunt forceps by grasping near the mouthparts to avoid damage to diagnostic structures. Individual ticks were placed into sterile, labelled screw cap vials containing 70% ethanol and transported to the Department of Veterinary Parasitology, Veterinary College and Research Institute, Tirunelveli, Tamil Nadu.

2.3 Gross and Microscopic identification of tick species

Collected ticks were briefly air dried to remove excess ethanol and subjected to detailed gross and microscopic examination (Lynx LM-52-3611) following ixodid tick identification guides (Walker *et al.*, 2003) [17].

2.4 Statistical Analysis

Tick infestation prevalence was calculated as the proportion of tick infested cattle out of total examined and the confidence intervals (95%) were calculated using the Wilson score method (OpenEpi Version 3.01), as described in standard epidemiological methods (Thrusfield, 2018) [18]. Pearson's Chi-square (χ^2) test was applied to compare overall prevalence among districts, and to assess district wise variation in single species infestations, mixed species infestations, and the distribution of individual tick species and their combinations (Ghosh *et al.*, 2019; Yenew *et al.*, 2025) [19, 20]. Statistical significance was set at $p < 0.05$.

3. Results

3.1 District wise Prevalence of Tick Infestation in Cattle

A total of 410 out of 525 cattle examined across the four southern districts of Tamil Nadu were found infested with ixodid ticks, yielding an overall prevalence of 78.10% (95% CI: 74.36-81.42). District wise, the highest prevalence of tick infestation was recorded in Kanyakumari district (80.00%; 95% CI: 71.77-86.29), while Thoothukudi district showed the lowest prevalence (74.67%; 95% CI: 63.79-83.14). However, a comparable prevalence was observed in Tenkasi (78.47%; 95% CI: 71.07-84.40) and Tirunelveli (78.01%; 95% CI: 71.62-83.30) (Table 1). The Chi-square test revealed that there was no statistically significant association between district and tick infestation status in cattle ($\chi^2=0.77$, $P=0.856$), indicating a uniformly high prevalence of tick infestation across all study districts (Table 2).

Table 1: Prevalence (%) of ticks of cattle in southern districts of Tamil Nadu

District	No. of cattle examined	No of cattle infested	Prevalence (%)	95% Confidence Interval
Tirunelveli	191	149	78.01	71.62-83.30
Tenkasi	144	113	78.47	71.07-84.40
Thoothukudi	75	56	74.67	63.79-83.14
Kanyakumari	115	92	80.00	71.77-86.29
Overall	525	410	78.10	74.36-81.42

Prevalence was calculated as the proportion of tick-infested cattle out of total examined.

95% confidence intervals were calculated using the Wilson score method (OpenEpi Version 3.01)

Table 2: Association between district and tick infestation status in cattle

District	No of cattle infested	No of cattle not infested	Total	Chi-square value (χ^2)	P-Value
Kanyakumari	92	23	115	0.772 ^{NS}	0.856
Tenkasi	113	31	144		
Thoothukudi	56	19	75		
Tirunelveli	149	42	191		
Total	410	115	525		

3.2 Pattern of single and mixed tick species infestations across districts

Out of 525 cattle examined, 324 cattle (61.71%) harboured single species tick infestations, while 86 animals (16.38%) exhibited mixed species infestations (Table 3). District wise prevalence of single infestations ranged between 58.67% (Thoothukudi) and 63.48% (Kanyakumari), whereas mixed

infestations remained consistently uniform across districts (16.00-16.67%). Chi-square analysis revealed no statistically significant association between district and tick infestation pattern (single vs mixed) ($\chi^2=0.02$, $P=0.999$), shows that the distribution of single and mixed tick infestations was similar across all districts. Indeed, single tick infestation was more common than mixed infestation in all districts.

**Fig 1:** *Rhipicephalus microplus***Fig 2:** *Rhipicephalus annulatus***Fig 3:** *Rhipicephalus haemaphysaloides***Fig 4:** *Rhipicephalus decoloratus***Fig 5:** *Haemaphysalis bispinosa***Fig 6:** *Hyalomma marginatum***Fig 7:** *Hyalomma marginatum* and *Hyalomma hussaini***Fig 8:** *Amblyomma spp*

3.3 Tick species composition in cattle across southern Districts of Tamil Nadu

A total of eight ixodid tick species, *Rhipicephalus microplus*, *R. annulatus*, *R. haemaphysaloides*, *Rhipicephalus decoloratus*, *Haemaphysalis bispinosa*, *Hyalomma marginatum*, *Hyalomma hussaini* and *Amblyomma spp.* were identified in the study area (Figure 1 to 8).

3.3.1 Single Species Tick Infestations

Single species tick infestations varied significantly across districts ($\chi^2=61.87$, $p<0.001$) (Table 4). *Rhipicephalus microplus* was the predominant species in all districts, with

highest prevalence in Tenkasi (45.83%) and Thoothukudi (40.00%). *Haemaphysalis bispinosa* showed marked focal distribution, with high prevalence in Kanyakumari (31.30%) compared to other districts (4.86-10.47%). *Hyalomma marginatum* was most prevalent in Tirunelveli (13.09%), occurred at moderate levels in Thoothukudi (9.33%) and Tenkasi (9.03%), and was absent in Kanyakumari. *Rhipicephalus haemaphysaloides* was rare, detected only in Tirunelveli (0.52%). Notably, *Rhipicephalus decoloratus* was recorded for the first time in the study area as a single species infestation, occurring at low prevalence across districts (0.87-2.09%), with higher occurrence in Tirunelveli and Tenkasi.

Table 3: Prevalence (%) of single and mixed tick species infestations in cattle in southern Tamil Nadu

District	No. of cattle examined	Single tick infestations		Mixed tick infestations		Chi-square value (χ^2)	P-Value
		No of cattle	%	No of cattle	%		
Tirunelveli	191	118	61.78	31	16.23	0.02 ^{NS}	0.999
Tenkasi	144	89	61.81	24	16.67		
Thoothukudi	75	44	58.67	12	16.00		
Kanyakumari	115	73	63.48	19	16.52		
Total	525	324	61.71	86	16.38		

3.3.2 Mixed Species Tick Infestations

Six mixed species tick combinations were recorded across the study districts. The distribution pattern and prevalence per

cent of mixed tick species infestation in cattle in southern Tamil Nadu are presented in Table 5.

Table 4: Distribution pattern and prevalence (%) of single tick species infestation in cattle in southern Tamil Nadu

Tick species	Tirunelveli (N=191)		Tenkasi (N=144)		Thoothukudi (N=75)		Kanyakumari (N=115)		Total (N=525)	
	No of Cattle	%	No of Cattle	%	No of Cattle	%	No of Cattle	%	No of Cattle	%
<i>Rhipicephalus microplus</i>	68	35.60	66	45.83	30	40.00	36	31.30	200	38.10
<i>Rhipicephalus haemaphysaloides</i>	1	0.52	0	0.00	0	0.00	0	0.00	1	0.19
<i>Rhipicephalus decoloratus</i>	4	2.09	3	2.08	1	1.33	1	0.87	9	1.71
<i>Haemaphysalis bispinosa</i>	20	10.47	7	4.86	6	8.00	36	31.30	69	13.14
<i>Hyalomma marginatum</i>	25	13.09	13	9.03	7	9.33	0	0.00	45	8.57
Total	118	61.78	89	61.81	44	58.67	73	63.48	324	61.71

Chi-square value (χ^2) 61.87**, p -value < 0.001

Table 5: Distribution pattern and prevalence (%) of mixed tick species infestation in cattle in southern Tamil Nadu

Tick species	Tirunelveli (N=191)		Tenkasi (N=144)		Thoothukudi (N=75)		Kanyakumari (N=115)		Total (N=525)	
	No of Cattle	%	No of Cattle	%	No of Cattle	%	No of Cattle	%	No of Cattle	%
<i>R. microplus</i> + <i>R. annulatus</i>	0	0.00	2	1.39	0	0.00	4	3.48	6	1.14
<i>R. microplus</i> + <i>R. haemaphysaloides</i>	4	2.09	5	3.47	0	0.00	0	0.00	9	1.71
<i>R. microplus</i> + <i>H. bispinosa</i>	7	3.66	6	4.17	4	5.33	15	13.04	32	6.10
<i>R. microplus</i> + <i>H. marginatum</i>	20	10.47	9	6.25	8	10.67	0	0.00	37	7.05
<i>R. microplus</i> + <i>Hyalomma hussaini</i>	0	0.00	1	0.69	0	0.00	0	0.00	1	0.19
<i>R. microplus</i> + <i>Amblyomma spp.</i>	0	0.00	1	0.69	0	0.00	0	0.00	1	0.19
Total	31	16.23	24	16.67	12	16.00	19	16.52	86	16.38

Frequency analysis revealed that mixed tick species infestation was observed in 86 out of 525 cattle, accounting for an overall prevalence of 16.38%. Among the mixed infestation patterns, *R. microplus* + *Hyalomma marginatum* was emerged as the most frequent combination (7.05%; N=37), followed by *R. microplus* + *Haemaphysalis bispinosa* (6.10%; N=32), together accounting for the majority of mixed infestations. Other combinations, including *R. microplus* + *R. annulatus* (N=6) and *R. microplus* + *R. haemaphysaloides* (N=9), occurred at lower frequencies and showed restricted geographical distribution. Rare mixed infestations such as *R. microplus* + *Hyalomma hussaini* (N=1) and *R. microplus* + *Amblyomma spp.* (N=1) were detected only in Tenkasi,

highlighting the presence of additional tick species circulating at very low levels within the local ecosystem, even when not detected as single species infestations.

The occurrence of *R. microplus* + *H. bispinosa* mixed infestation differed significantly among districts ($\chi^2=12.68$, $P=0.005$), with highest prevalence in Kanyakumari (13.04%), followed by Thoothukudi (5.33%), Tenkasi (4.17%), and Tirunelveli (3.66%) (Table 6). Similarly, *R. microplus* + *H. marginatum* mixed infestation showed significant district wise variation ($\chi^2=13.78$, $P=0.003$), occurring in Tirunelveli (10.47%), Thoothukudi (10.67%), and Tenkasi (6.25%), but was absent in Kanyakumari (Table 7).

Table 6. Association between district and occurrence of *Rhipicephalus microplus* + *Haemaphysalis bispinosa* mixed infestation in cattle

District	No of cattle infected	No of cattle not infected	No of cattle examined	Prevalence (%)	Chi-square value (χ^2)	P-Value
Tirunelveli	7	184	191	3.66	12.68**	0.005
Tenkasi	6	138	144	4.17		
Thoothukudi	4	71	75	5.33		
Kanyakumari	15	100	115	13.04		
Total	32	493	525	6.10		

Table 7: Association between district and occurrence of *Rhipicephalus microplus* + *Hyalomma marginatum* mixed infestation in cattle

District	No of cattle infected	No of cattle not infected	No of cattle examined	Prevalence (%)	Chi-square value (χ^2)	P-Value
Tirunelveli	20	171	191	10.47	13.78**	0.003
Tenkasi	9	135	144	6.25		
Thoothukudi	8	67	75	10.67		
Kanyakumari	0	115	115	0.00		
Total	37	488	525	7.05		

4. Discussion

4.1 District wise Prevalence of Tick Infestation in Cattle

The present study recorded a uniformly high prevalence of ixodid tick infestation (78.10%) among cattle across the four southern districts of Tamil Nadu, with only marginal inter district variation (74.67-80.00%) and no statistically significant district wise association. This finding is consistent with reports from other humid tropical regions of India, including Mizoram and Kerala, where tick infestation prevalences exceeding 70% have been documented under field conditions (Ghosh *et al.*, 2019; Nimisha *et al.*, 2019) [19, 13]. Such consistently high prevalence indicates that tick infestation remains a persistent constraint in tropical cattle production systems.

The absence of significant spatial variation suggests that ecological and management factors governing tick survival are broadly similar across southern Tamil Nadu. Climatic conditions characterised by sustained warmth, high humidity, and moderate rainfall provide favourable microenvironments for tick development and off host survival throughout the year. Comparable uniform prevalence patterns have been reported from Ethiopia, where similar agro-ecological conditions and grazing practices were implicated (Yenew *et al.*, 2025) [20]. In the present region, extensive or semi-intensive rearing systems, communal grazing, and limited adoption of systematic acaricidal control likely contribute to sustained tick exposure across districts.

The high prevalence observed also parallels findings from Assam and other endemic regions, where tick infestation persists at elevated levels once enzootic stability is established (Khukhuu *et al.*, 2011; Bawm *et al.*, 2014; Kakati *et al.*, 2015) [11, 12, 7]. From an epidemiological standpoint, such widespread and persistent tick presence favours continuous circulation of haemoprotozoan pathogens, including *Theileria orientalis*, even in clinically inapparent cattle populations, as previously reported by Aktas *et al.* (2006) and Baghel *et al.* (2023) [21, 9]. The findings of the present study emphasize that tick infestation in southern Tamil Nadu represents a region wide challenge, necessitating coordinated, area based tick surveillance and control strategies rather than district specific interventions.

4.2 Pattern of single and mixed tick species infestations across districts

The present study showed that single tick species infestations predominated among cattle across southern Tamil Nadu (61.71%), whereas mixed infestations were less frequent (16.38%) and uniformly distributed across districts, with no significant district-wise association. Similar dominance of single-species infestations has been reported from other parts

of Tamil Nadu and southern India, where environmental homogeneity and stable host-tick interactions favour infestation by a single dominant species at a given time (Nimisha *et al.*, 2019; Ranganathan *et al.*, 2025) [13, 22].

The predominance of single infestations is likely driven by ecological niche specialization and competitive exclusion among ixodid ticks. In particular, *Rhipicephalus (Boophilus) microplus*, with its strong host preference, rapid life cycle and efficient attachment behaviour, often achieves numerical dominance, limiting opportunities for co-infestation by other species. Seasonal and microhabitat influences further reinforce this dominance under favourable climatic and management conditions (Ranganathan *et al.*, 2025) [22].

Although less common, mixed tick infestations were consistently detected across all districts, suggesting stable ecological overlap rather than sporadic occurrence. Comparable low but persistent mixed infestations have been documented from endemic regions where repeated exposure through communal grazing facilitates contact with multiple tick species (Kakati *et al.*, 2015; Ghosh *et al.*, 2019) [7, 19]. Epidemiologically, such mixed infestations are important, as concurrent feeding by different tick species may increase transmission complexity and support the maintenance of multiple haemoprotozoan species within cattle populations.

From a disease transmission perspective, the coexistence of single and mixed infestations may influence the epidemiology of *Theileria orientalis*. Mixed infestations increase exposure to vectors with differing competence, potentially contributing to parasite genetic diversity and prolonged transmission cycles, as reported from India, Australia and the United States (Sivakumar *et al.*, 2014; Perera *et al.*, 2014; Oakes *et al.*, 2019; Jacob *et al.*, 2024) [23, 1, 24, 5]. Overall, the observed pattern reflects a stable tick host equilibrium in southern Tamil Nadu and supports region wide integrated tick management strategies targeting dominant species while accounting for secondary vectors involved in pathogen persistence.

4.3 Tick species composition in cattle across Southern Districts of Tamil Nadu

4.3.1 Single species tick infestations

The present study revealed significant district wise heterogeneity in single species tick infestations across southern Tamil Nadu ($\chi^2=61.87$, $p<0.001$). *Rhipicephalus microplus* was the predominant species in all districts, accounting for 38.10% of infestations, with the highest prevalence in Tenkasi (45.83%) and Thoothukudi (40.00%). This finding corroborates earlier reports identifying *R. microplus* as the principal cattle tick in tropical and subtropical India, attributed to its strong host specificity, high

reproductive capacity, and adaptability to semi intensive management systems (Ghosh *et al.*, 2019; Nimisha *et al.*, 2019; Ranganathan *et al.*, 2021) ^[19, 13, 25].

A pronounced focal distribution of *Haemaphysalis bispinosa* was observed in Kanyakumari (31.30%), contrasting with its lower prevalence elsewhere. This pattern likely reflects the district's humid coastal climate, dense vegetation, and wildlife-livestock interface, conditions favourable for *Haemaphysalis* spp., as reported from Kerala and Western Ghats regions (Nimisha *et al.*, 2019; Ranganathan *et al.*, 2025) ^[13, 22]. In contrast, *Hyalomma marginatum* predominated in Tirunelveli and was absent in Kanyakumari, consistent with its preference for drier, open grazing ecosystems (Aktas *et al.*, 2006; Ghosh *et al.*, 2019) ^[21, 19].

Rhipicephalus haemaphysaloides occurred rarely and was restricted to Tirunelveli, indicating its limited role in cattle infestation. Importantly, *Rhipicephalus decoloratus* was recorded for the first time as a single species infestation in the study area, albeit at low prevalence, suggesting possible range expansion linked to livestock movement or climatic shifts, as reported by Bawm *et al.* (2014) ^[12] and Baghel *et al.* (2023) ^[9]. These findings highlight the strong influence of local ecological and husbandry factors on tick species composition. The dominance of *R. microplus* signify its priority in control programmes, while the focal distribution of secondary species and emergence of *R. decoloratus* emphasize the need for region specific surveillance and adaptive tick management strategies.

4.3.2 Mixed Species Tick Infestations

Mixed tick species infestations were recorded in 16.38% of cattle across southern Tamil Nadu, comprising six distinct species combinations. The predominance of *Rhipicephalus microplus* associated combinations, particularly *R. microplus* + *Hyalomma marginatum* (7.05%) and *R. microplus* + *Haemaphysalis bispinosa* (6.10%), reflects the ecological dominance and broad host adaptability of *R. microplus*, which facilitates co-infestation with sympatric tick species. Similar patterns have been documented from Kerala and Assam, where high *R. microplus* densities increase the likelihood of co-attachment (Kakati *et al.*, 2015; Nimisha *et al.*, 2019) ^[7, 13]. The limited diversity of mixed infestations suggests ecological filtering driven by host preference, microclimatic suitability, and niche overlap. The association of *H. marginatum* with *R. microplus* in Tirunelveli, Thoothukudi, and Tenkasi aligns with the preference of *Hyalomma* spp. for drier agro-climatic zones, whereas its absence in Kanyakumari likely reflects unfavourable humid coastal conditions (Ghosh *et al.*, 2019; Ranganathan *et al.*, 2025) ^[19, 22]. Conversely, the significantly higher prevalence of *R. microplus* + *H. bispinosa* in Kanyakumari corresponds to the humid, vegetated environment favouring *Haemaphysalis* spp. (Nimisha *et al.*, 2019; Sivakumar *et al.*, 2014) ^[13, 23].

The detection of rare mixed infestations involving *H. hussaini* and *Amblyomma* spp., though sporadic, is epidemiologically important, indicating the presence of low abundance or emerging tick species that may contribute to pathogen introduction and maintenance. Mixed infestations are of particular concern as they can facilitate co-transmission of haemoparasites and enhance the circulation of *T. orientalis* genotypes, as reported in India and other endemic regions (Perera *et al.*, 2014; Oakes *et al.*, 2019; Jacob *et al.*, 2024) ^[1, 24, 5].

The district specific distribution of mixed tick infestations accentuates the influence of local ecological and climatic

factors on tick community structure and highlights the need for integrated tick control strategies that address both dominant and secondary species across agro-ecological zones.

5. Conclusion

The present study elucidates the epidemiology of ixodid tick infestations in cattle across southern Tamil Nadu, revealing a uniformly high tick burden with marked district wise variation in species composition. *Rhipicephalus microplus* predominated across all districts, confirming its central role in sustaining tick populations and facilitating endemic transmission of tick-borne haemoprotozoa, including *Theileria orientalis*. The spatial distribution of *Haemaphysalis bispinosa* and *Hyalomma marginatum* reflected their ecological preferences for humid and xeric environments, respectively, while the first record of *Rhipicephalus decoloratus* highlights the dynamic expansion of tick fauna in the region. Although single species infestations were most frequent, the consistent occurrence of mixed infestations particularly involving *R. microplus* with *H. marginatum* and *H. bispinosa* indicates complex tick-host-environment interactions that may enhance pathogen transmission and genetic diversity. These findings demonstrate that tick infestation is a region wide challenge rather than a district specific phenomenon and emphasize the necessity for integrated, area wide tick control strategies that prioritize dominant vectors while accounting for secondary and emerging tick species to effectively mitigate tick borne disease risks in southern India.

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Conflict of Interest

The authors declare that there are no conflicts of interest regarding the publication of this article.

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