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Epidemiological study of bovine theileriosis in erstwhile west Godavari district, Andhra Pradesh: A five year retrospective case study (2020-2025)

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Abstract

A retrospective epidemiological study was conducted to determine the prevalence and temporal trends of bovine theileriosis in cattle and buffalo over a five-year period (2020–2025). A total of 20, 889 blood samples were examined, including 4, 940 from cattle and 15, 949 from buffalo. Overall, 2, 238 animals tested positive, with a cumulative prevalence of 10.71%. Species-wise prevalence was 8.54% in cattle and 11.25% in buffalo. Annual prevalence increased significantly from 2.88% in 2020–2021 to 14.36% in 2024–2025. Chi-square analysis showed a significant association between species and infection status ($\chi^2 = 32.84$, $p < 0.001$). Trend analysis also demonstrated a significant increase over the study period (χ^2 trend = 412.67, $p < 0.001$). Polymerase chain reaction (PCR) was performed on selected samples to confirm the presence of *Theileria annulata*. The study highlights an increasing burden of bovine theileriosis in the region and emphasizes the need for effective tick control and routine surveillance.

Keywords: Bovine theileriosis, retrospective study, prevalence, epidemiology, haemoprotozoan disease, West Godavari, tick-borne disease

1. Introduction

Bovine theileriosis is an economically important tick-borne haemoprotozoan disease caused primarily by *Theileria annulata* and transmitted by Hyalomma ticks [1, 3, 4]. The disease is characterized by fever, anemia, lymphadenopathy, reduced milk yield, and mortality [1, 2]. West Godavari district of Andhra Pradesh is a major dairy-producing region with climatic conditions favorable for tick proliferation [4, 8, 9]. Despite its economic significance, comprehensive epidemiological data from this region remain limited. Therefore, the present study was undertaken to analyze the temporal trends and species-wise prevalence of bovine theileriosis over a five-year period.

2. Materials and Methods

Blood samples were collected from suspected animals in EDTA vials. All samples were screened microscopically for intra-erythrocytic piroplasm forms of *Theileria* and other haemoparasites. Thin blood smears were prepared, air-dried, fixed, and stained using Leishman's stain. Identification was based on the characteristic morphology of the parasite (comma-shaped piroplasms).

Molecular Diagnosis

PCR was performed on selected microscopically positive samples to confirm the presence of *Theileria annulata*. Genomic DNA was extracted from 200 μ L of whole blood using a commercial DNA extraction kit according to the manufacturer's instructions. The cytochrome b gene was amplified using specific primers Cyt b1 (5'-ACTTTGGCCGTAATGTAAAC-3') and Cyt b2 (5'-CTCTGGACCAACTGTTTGG-3'), as described in previous studies [7].

2.1 Clinical examination and sample collection

Blood samples were collected from the suspected animals in EDTA vials and All collected blood samples were microscopically screened to detect intra-erythrocytic piroplasms form of *Theileria* and other haemoparasites. For each sample, a thin blood smear was drawn, air-dried, and leishmans stain was applied for 30 minutes. the identification of organism was based on the characteristic appearance of the organism (comma shaped).

Molecular diagnosis: The PCR assay was limited to microscopic positive samples for *Theileria* organism to identify the species through species-specific primers pair (Cyt b1/Cyt b2). Genomic DNA was extracted from a 200 μ L whole-blood sample using a Blood DNA Isolation Kit according to the manufacturer's protocol. In the present study, PCR analysis was carried out to target the Cytochrome b gene of *T. annulata* by utilizing the forward primer Cyt b1 (5'-ACTTTGGCCGTAATGTTAAAC-3') and reverse primer Cyt b2 (5'-CTCTGGACCAACTGTTTGG-3') based on many experimental studies which were already been performed and were told the results according to this primer are accurate and taken into consideration.

2.2 Study Design and Data Collection

A laboratory-based retrospective study was carried out using recorded diagnostic data from 2020–2021 to 2024–2025. Data included species, number of samples examined, and number of samples positive for theileriosis.

2.3 Statistical Analysis

Prevalence (%) was calculated as (Number Positive / Number Examined) $\times$ 100. Chi-square (χ^2) test was applied to determine association between species and infection. Trend analysis was performed to assess year-wise variation. Statistical significance was considered at $p < 0.05$.

3. Results

A total of 20, 889 samples were examined, out of which 2, 238 were positive with an overall prevalence of 10.71%.

Year-wise prevalence showed a progressive increase from 2.88% (2020–2021) to 14.36% (2024–2025), indicating a significant upward trend (χ^2 trend = 412.67, $p < 0.001$).

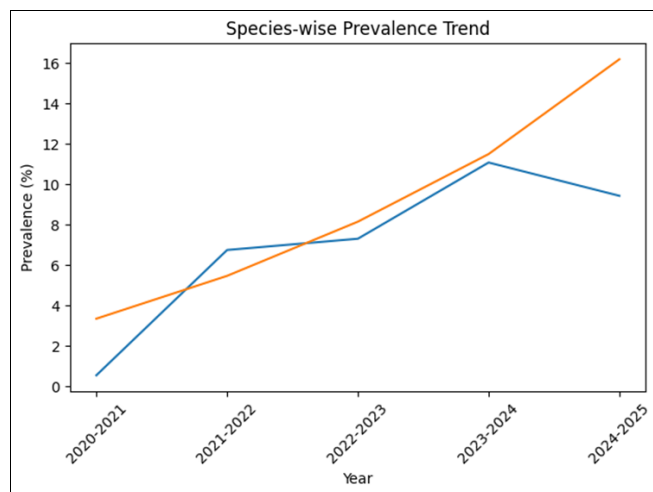
Year	Total Examined	Total Positive	Prevalence (%)
2020–2021	1, 076	31	2.88
2021–2022	1, 295	75	5.79
2022–2023	5, 169	412	7.97
2023–2024	6, 651	758	11.40
2024–2025	6, 698	962	14.36

Species-wise analysis showed that buffalo had a higher prevalence (11.25%) compared to cattle (8.54%). The difference was statistically significant ($\chi^2 = 32.84$, $p < 0.001$).

Cattle		Buffalo	
Year	Prevalence	Year	Prevalence
2020–2021	0.55%	2020–2021	3.35%
2021–2022	6.75%	2021–2022	5.47%
2022–2023	7.31%	2022–2023	8.15%
2023–2024	11.07%	2023–2024	11.49%
2024–2025	9.43%	2024–2025	16.18%

Peak prevalence in cattle was recorded in 2023–2024.

Buffalo showed a marked and steady increase, reaching highest prevalence in 2024–2025



4. Discussion

The present study revealed a significant increasing trend in the prevalence of bovine theileriosis over the five-year period. The overall prevalence of 10.71% indicates a considerable disease burden in the study region. The progressive increase in prevalence from 2.88% in 2020–2021 to 14.36% in 2024–2025 suggests enhanced vector activity, increased exposure of animals to tick infestation, and favorable climatic conditions for tick survival and propagation.

Species-wise analysis demonstrated a significantly higher prevalence in buffalo (11.25%) compared to cattle (8.54%). This difference may be attributed to variations in management practices, grazing patterns, and increased exposure of buffalo to tick-infested environments such as wallowing areas. Additionally, differences in host susceptibility and immune response between cattle and buffalo may contribute to the observed variation.

The increasing trend observed in the present study is consistent with previous reports on tick-borne haemoprotozoan diseases in tropical and subtropical regions^[5, 8, 9], where climatic factors such as temperature, humidity, and rainfall play a crucial role in tick population dynamics and disease transmission. Similar epidemiological patterns have been reported in other parts of India, indicating a rising burden of bovine theileriosis.

Microscopic examination remains a practical and widely used diagnostic tool under field conditions; however, it may have limitations in detecting low parasitemia or subclinical infections. In the present study, PCR was performed on selected samples to confirm the presence of *Theileria annulata*, thereby improving diagnostic accuracy and supporting microscopic findings^[6, 7]. Molecular techniques provide higher sensitivity and specificity and are valuable in confirming infection, particularly in early or carrier stages.

The findings of this study emphasize the need for strengthened tick control strategies, regular surveillance, and improved farm management practices. Strategic acaricide use, awareness among farmers, and early diagnosis can significantly reduce the disease burden and associated economic losses.

Host susceptibility and variation in immune response among different species may influence disease occurrence and progression, as reported in previous studies^[10, 11].

5. Conclusion

Bovine theileriosis prevalence increased significantly over the five-year period. Buffalo were more affected than cattle.

Strengthened tick control measures, regular screening, and improved farm management practices are recommended.

6. Acknowledgement

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Conflict of Interest

Not available

Financial Support

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