Original Article

Identification of Babesia Parasite in Ruminants in District Dera Ismail Khan

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ABSTRACT

Background: Babesiosis, a significant tick-borne disease, poses a growing threat to small ruminants and public health due to the pathogenic Babesia parasite. This study focuses on the prevalence of Babesia in cows within the district of Dera Ismail Khan and its impact on red blood cells.

Objective: The primary objective of this research was to detect the presence of the Babesia parasite in cows and assess its effects on red blood cells.

Methods: Fifty blood samples were randomly collected from cows in Dera Ismail Khan. These samples underwent Giemsa staining and were subsequently examined under a light microscope to identify the presence of the Babesia parasite.

Results: Of the 50 blood samples analyzed, only 5 (10%) tested positive for Babesia. This suggests a lower prevalence of babesiosis in the region. The parasite was observed to cause significant destruction of red blood cells.

Conclusion: Although babesiosis is not widely prevalent in Dera Ismail Khan, its impact on infected cows is severe, leading to the destruction of red blood cells. Effective monitoring and control measures are essential to mitigate its spread and impact.

Keywords: Babesia, Babesiosis, Cows, Dera Ismail Khan, Giemsa Staining, Red Blood Cells, Tick-borne Disease.

INTRODUCTION

Babesiosis is a significant haemoparasitic disease transmitted by ticks, predominantly affecting a wide array of ruminants globally, particularly in tropical and subtropical regions (1). This disease is caused by protozoan parasites from the genus Babesia, with several species such as Babesia divergens, Babesia bigemina, Babesia ovis, and Babesia major known for their pathogenicity. These parasites infect a broad range of hosts, including cattle, dogs, cats, pigs, sheep, horses, and occasionally humans, with over 100 identified species, 18 of which are known to infect local animals and cause disease (2, 3).

The clinical manifestations of babesiosis vary greatly, ranging from asymptomatic infections to acute conditions characterized by circulatory shock, anemia, anorexia, jaundice, listlessness, and hemoglobinuria—a condition marked by the destruction of red blood cells (4). If left untreated, babesiosis can result in mortality rates exceeding 50%, although timely and effective treatment can reduce this to as low as 5% (5). The disease's transmission occurs through vectors such as ticks, particularly from the Ixodidae family, and can also spread via blood transfusion and mechanically through flies (2, 4).

The district of Dera Ismail Khan, with a substantial population of livestock including 120,000 sheep and 310,000 goats, serves as a critical study area for understanding the epidemiology of babesiosisThis region experiences a range of climatic conditions, with temperatures conducive to tick activity, which is crucial for the parasite's life cycle and transmission (6, 7).

Given the economic importance of livestock in the region, and the potential for significant losses due to babesiosis, this research aims to detect Babesia infection in large ruminants and assess its impact on red blood cells. The study's objectives are grounded in the necessity to address this neglected yet prevalent pathogen that, while causing only minor symptoms individually, can cumulatively lead to substantial economic losses in the agricultural sector(8, 9).

MATERIAL AND METHODS

The study was carried out in the district of Dera Ismail Khan, where the parasitology laboratory at Gomal University's veterinary lab served as the primary site for sample collection and analysis. Blood samples were systematically collected from the coccygeal vein of cows using anticoagulant-treated tubes to prevent clotting. These samples were then promptly transferred to petri dishes for further processing(10, 11).

For the preparation of thin-layer smears, a meticulous technique was employed where blood from the petri dishes was carefully placed on microscope slides. A second slide was then used to spread the blood at a 45-degree angle to create a uniform thin layer. This process was repeated to produce approximately 25 slides per session. Following the preparation of these slides, methanol was applied to each to fix the blood, ensuring that the cellular components were adequately preserved for microscopic examination(12, 13).

The staining of the slides was conducted using the Giemsa staining method, a choice dictated by both its effectiveness in highlighting the cellular details of blood parasites and the resources available in the laboratory. After the application of the Giemsa stain, the slides were left to air dry, preparing them for examination under a microscope(14, 15).

Microscopic analysis was performed using a standard light microscope. Initially, slides were examined at 40x magnification to identify and differentiate between normal and infected red blood cells. Following this preliminary examination, a 100x oil immersion lens was utilized to closely observe the morphological characteristics of Babesia parasites within the red blood cells. This detailed scrutiny under high magnification was critical for accurately diagnosing the presence of Babesia infections in the sampled cattle(16, 17).

RESULTS

Microscopic observation on 50 blood smears determined 10% samples were infected by babesia and theileria respectively. The mixed infections occurred in four samples. Morphology study of infected blood smears showed occurrence of parasite at different locations. Marginal, sub marginal and central respectively. Babesia had a great effect on red blood cells. Babesia parasite grow in red blood cells and cause hemolytic anemia.
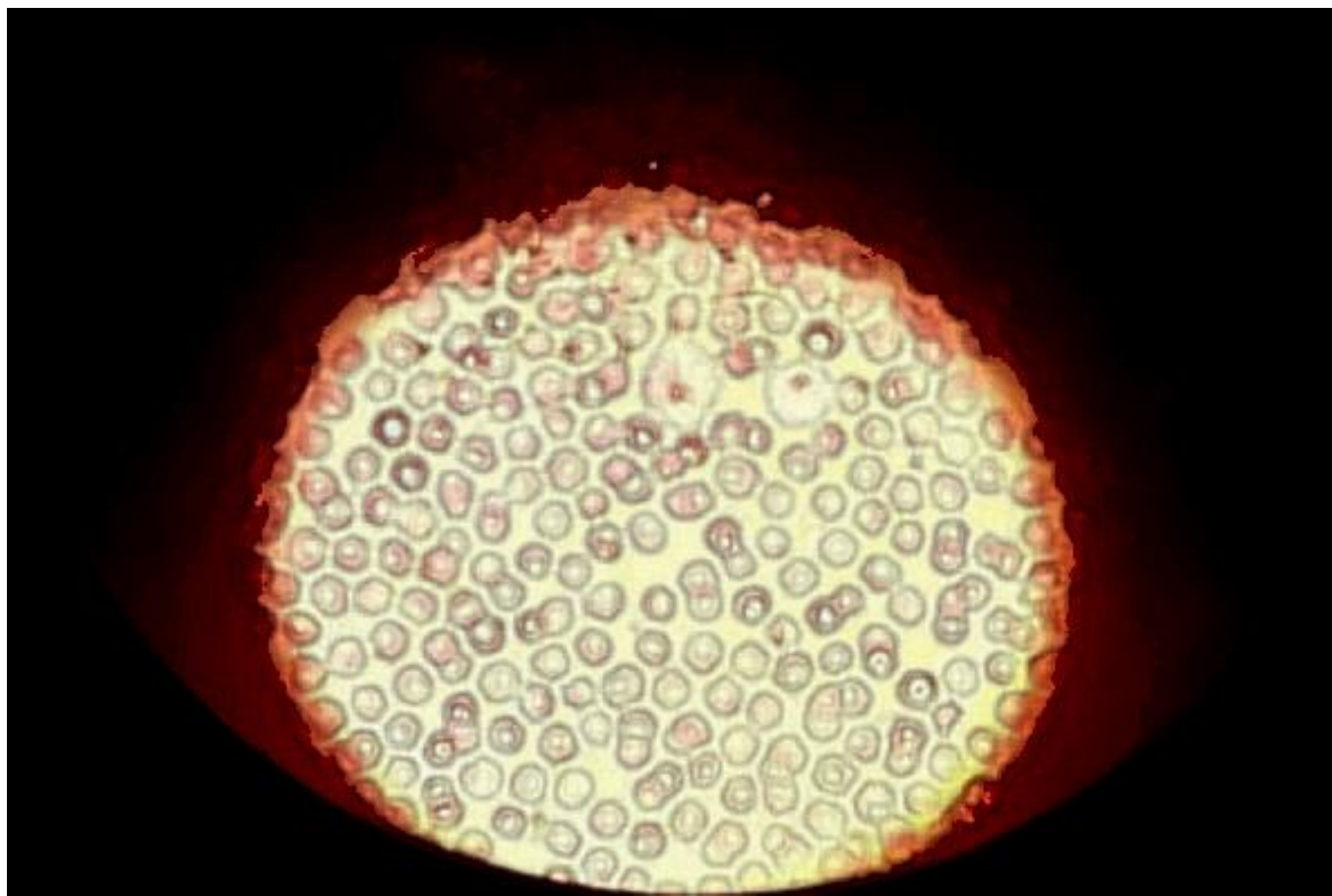
No	Disease Name	Blood Cell	Effect on Blood Cell	Numbers	Total	Percentage
1	Normal	RBC	No effect	39	50	78%
2	Babesia	RBC	Brusting	5	50	10%
3	Theileria	WBC	Brusting	6	50	12%
4	Mixed	RBC	No effect	7	50	14%

DISCUSSION

Babesiosis is a critical disease in livestock, causing substantial morbidity and mortality and leading to significant economic losses globally. The disease prominently affects red blood cells, leading to a range of severe clinical symptoms and often resulting in death if not treated timely. In our study conducted at the onset of the winter season, the primary diagnostic approach was the microscopic examination of Giemsa-stained blood smears, a method chosen for its accessibility despite its known limitations in sensitivity and the requirement for considerable expertise due to the morphological similarities among Babesia species[18, 19].

The utilization of Giemsa staining, while influenced by limited resources, remains a standard practice. However, the method's low sensitivity is a notable limitation, particularly when distinguishing between different Babesia species, which can present similarly under microscopic examination. Despite these challenges, the study identified the presence of Babesia in 10% of the samples tested, indicating an active infection among the livestock examined. This finding underscores the persistence of the parasite within the local cattle population and highlights the continuous threat it poses to livestock health and economic stability[2, 20].

The life cycle and transmission dynamics of Babesia involve complex interactions with vector ticks, particularly Ixodes ricinus, which plays a critical role in the disease's epidemiology. The success of Babesia transmission is dependent upon several factors, including the vector's infection at its larval stages and its survival and activity across seasonal variations. Notably, disease occurrences are biphasic, peaking during the late spring and early autumn, which aligns with the active periods of the tick vectors[2, 21].

In cattle, immunity to Babesia can be temporary and influenced by various factors, including age and continuous exposure to infected ticks. Calves typically show immunity until about nine months of age, but this can wane if there is no subsequent exposure to the parasite, leading to susceptibility in older animals. This aspect of the disease highlights the importance of managing tick populations and monitoring cattle for signs of infection regularly, particularly in areas known for rough grazing practices where naive animals may be exposed to ticks carrying Babesia[1, 21].

Clinically, babesiosis manifests with symptoms ranging from fever and anorexia to more severe neurological signs and haemoglobinuria, which can lead to death if not managed promptly. The impact of the disease on livestock behavior, health, and productivity underscores the need for effective control measures and diagnostic capabilities, particularly in rural areas where access to veterinary services may be limited[4, 22].

The findings of this study contribute to the broader understanding of babesiosis in cattle, emphasizing the need for enhanced diagnostic techniques and the potential benefits of developing more sensitive and specific testing methods to better manage and control this disease. While the study achieved its aim of detecting Babesia in cattle and provided valuable insights into its prevalence and impact, the reliance on less sensitive diagnostic methods and the limited geographic scope of the sampling are recognized as constraints. Future research should focus on incorporating more advanced molecular techniques to improve detection rates and on expanding the study to include a wider range of environmental and climatic conditions to better understand the dynamics of Babesia transmission[6, 23].

CONCLUSION

Effective control and management of babesiosis in livestock hinge on comprehensive strategies that address both the tick vectors and the disease itself. Integrating chemical and biological control methods, along with the strategic use of vaccines, can significantly reduce the incidence of babesiosis. Recommendations for future actions include enhancing waste management to disrupt tick

habitats, using targeted chemical treatments responsibly to minimize environmental impact, and improving overall animal care standards. Government intervention is crucial in facilitating early disease detection and in implementing sustainable disease management practices that not only prevent acute cases but also mitigate the substantial economic losses associated with babesiosis in the agricultural sector.

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REFERENCES

1. Michel AO, Mathis A, Ryser-Degiorgis M-P. Babesia spp. in European wild ruminant species: parasite diversity and risk factors for infection. Veterinary research. 2014;45:1-11.

2. Alemu Hamsho AH, Gebregergs Tesfamarym GT, Gurara Megersa GM, Mulisa Megersa MM. A cross-sectional study of bovine babesiosis in Teltele District, Borena Zone, Southern Ethiopia. 2015.

3. Ulucesme MC, Ozubek S, Karoglu A, Turk ZI, Olmus I, Irehan B, et al. Small Ruminant Piroplasmosis: high prevalence of Babesia aktasi n. sp. in goats in Türkiye. Pathogens. 2023;12(4):514.

4. Ijaz M, Rehman A, Ali M, Umair M, Khalid S, Mehmood K, et al. Clinico-epidemiology and therapeutical trials on babesiosis in sheep and goats in Lahore, Pakistan. 2013.

5. Niazi N, Khan M, Avais M, Khan J, Pervez K, Ijaz M. A study on babesiosis in calves at livestock experimental station Qadirabad and adjacent areas, Sahiwal (Pakistan). Pak J Agri Sci. 2008;45(2):209-11.

6. Khattak R, Rabib M, Khan Z, Ishaq M, Hameed H, Taqddus A, et al. A comparison of two different techniques for the detection of blood parasite, Theileria annulata, in cattle from two districts in Khyber Pukhtoon Khwa Province (Pakistan). Parasite: journal de la Société Française de Parasitologie. 2012;19(1):91.

7. Ozubek S, Ulucesme MC, Aktas M. Discovery of a Novel Species Infecting Goats: Morphological and Molecular Characterization of Babesia aktasi n. sp. Pathogens. 2023;12(1):113.

8. NUGRAHENI YR, ARIYADI B, ROCHMADIYANTO R, KESUMANINGRUM N, IMRAN K, KARTIKO BP, et al. Molecular detection of Babesia infection in cattle in Yogyakarta, Indonesia. Biodiversitas Journal of Biological Diversity. 2023;24(7).

9. Sawitri D, Wardhana A, Ekawasti F, Dewi D, editors. Parasitological and molecular detection of babesiosis in cattle and buffalo in west and central Java. 9th International Seminar on Tropical Animal Production (ISTAP 2021); 2022: Atlantis Press.

10. Kose O, Bilgic HB, Bakirci S, Karagenc T, Adanir R, Yukari BA, et al. Prevalence of Theileria/Babesia species in ruminants in Burdur province of Turkey. Acta Parasitologica. 2022;67(2):723-31.

11. Githaka NW, Bishop RP, Šlapeta J, Emery D, Nguu EK, Kanduma EG. Molecular survey of Babesia parasites in Kenya: first detailed report on occurrence of Babesia bovis in cattle. Parasites & Vectors. 2022;15(1):161.

12. Ganzinelli S, Byaruhanga C, Primo ME, Lukanji Z, Sibeko K, Matjila T, et al. International interlaboratory validation of a nested PCR for molecular detection of Babesia bovis and Babesia bigemina, causative agents of bovine babesiosis. Veterinary Parasitology. 2022;304:109686.

13. Galon EM, Zafar I, Ji S, Li H, Ma Z, Xuan X. Molecular reports of ruminant Babesia in Southeast Asia. Pathogens. 2022;11(8):915.

14. Martínez-García G, Santamaría-Espinosa RM, Lira-Amaya JJ, Figueroa JV. Challenges in tick-borne pathogen detection: the case for Babesia spp. identification in the tick vector. Pathogens. 2021;10(2):92.

15. El-Sayed SAE-S, Mohamed A, Ali MO, Al-Araby M, Omar MA, El-Beskawy M, et al. Molecular detection and identification of Babesia bovis and Trypanosoma spp. in one-humped camel (Camelus dromedarius) breeds in Egypt. Veterinary world. 2021;14(3):625.

16. Tumwebaze MA, Byamukama B, Tayebwa DS, Byaruhanga J, Angwe MK, Galon EM, et al. First molecular detection of Babesia ovis, Theileria spp., Anaplasma spp., and Ehrlichia ruminantium in goats from western Uganda. Pathogens. 2020;9(11):895.

17. Otgonsuren D, Sivakumar T, Amgalanbaatar T, Enkhtaivan B, Narantsatsral S, Tuvshintulga B, et al. Molecular epidemiological survey of Babesia bovis, Babesia bigemina, and Babesia sp. Mymensingh infections in Mongolian cattle. Parasitology international. 2020;77:102107.

18. Gh H, Sepahvand-Mohammadi E, Afshari A, Bozorgi S. Molecular detection of Theileria spp. and Babesia ovis Infection in Sheep in Baneh, Iran. Archives of Razi Institute. 2020;75(2):289.

19. Rajabi S, Esmaeilnejad B, Tavassoli M, editors. A molecular study on Babesia spp. in cattle and ticks in West-Azerbaijan province, Iran. Veterinary research forum; 2017: Faculty of Veterinary Medicine, Urmia University, Urmia, Iran.

20. Hamsho A, Tesfamarym G, Megersa G, Megersa M. A cross-sectional study of bovine babesiosis in Teltele District, Borena Zone, Southern Ethiopia. J Veterinar Sci Technol. 2015;6(230):2.

21. Mohamed G, Ebied M. Epidemiological studies on bovine Babesiosis and Theileriosis in Qalubia governorate. BVMJ. 2014;27:36-48.

22. Esmaeilnejad B, Tavassoli M, Asri-Rezaei S, Dalir-Naghadeh B, Malekinejad H, Jalilzadeh-Amin G, et al. Evaluation of antioxidant status, oxidative stress and serum trace mineral levels associated with Babesia ovis parasitemia in sheep. Veterinary parasitology. 2014;205(1-2):38-45.

23. Ahmad S, Khan M, Khan M. Epidemilogy and seasonal abundance of canine babesiosis in Lahore, Pakistan. 2011.