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Epidemiological Analysis of Theileriosis in Goats and Its Effect on Hematological Parameters in Southern Districts of Khyber Pakhtunkhwa, Pakistan

Original Article

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Abstract

Theileriosis is among the emerging issues of the wild ungulates, especially in the tropical and sub-tropical parts of the globe. The study carried out was aimed at looking into the goats having the presence of Theleria infection by identifying the presence of the infection with the use of the microscope and to see the disease impact on the hematological points. 300 blood samples from asymptomatic goats were to be taken using a convenient sampling technique at random. The simple microscopic examination was done in the Clinical Medicine and Parasitology laboratory on all the blood samples. This has seen the percentage of positive outcomes being found 34 % in the three hundred total samples. The highest prevalence was recorded in female goats (55.88%) than male (44.1%) and statistically significant (p<.04) / non-significant (P< .002) difference was observed Area wise study was also observed where the highest prevalence was recorded in the District of Karak (25.49%), followed by Bannu (20.58%), Lakki Marwat (16.66%), DIKhan (13.72%), while the lowest prevalence was recorded in the District of Tank (11.76%). Statistically, a significant (p<.04) / (P< .002) non-significant difference was observed, where the p value (p<.0000). The alterations in hematological parameters were also observed, where the highest increase was observed in the District Karak (KC) 26/103×100. while the number of DIKhan (DIK) 11/103×100 was seriously decreased.

Keywords: Epidemiology, Theileriosis, Goats, Hematological Parameters, Theileriosis.

Introduction

In this developing country, Pakistani natives have adopted meat as an essential part of their food culture to meet additional nutrient requirements. Buffaloes compose about 64 per cent of meat consumption in Pakistan, followed by goat meat (27 per cent), chicken (6 per cent), and sheep (3 per cent) (Joshi, Singh, & Giroti, 2001). Goat skin may also be used to produce various products, such as boots, gloves, among other products that require a soft hide. Cashmere goats produce the finest cashmere wool in the world, as the hair on their bodies beneath the tufts of guard hair is super soft and extremely fine. The hair of dolls and sweaters, among others, is made with Mohair. The pashmina has its origin in the Persian word pashm, which connotes something as being soft. In South Asia, where the pashmina word is used about cashmere, these goats are known as pashmina goats (Rizal, 2010).

The livestock industry is the most important in the economy of Pakistan (Qudus *et al*., 2013). Smallholders have been using livestock as a major source of food, protein, nutrition, and a source of financial income. Among the chief factors through which dairy farming reduces the rate of unemployment is by assigning workers who are both educated and non-educated to suitable both indoor and outdoor field employment opportunities (Hagmann, 2012). Livestock is a major tool for poverty reduction and improving the socioeconomic status of people residing in the countryside.

Historically, it was found out that ticks have existed as ectoparasites of animals as far back as 400 B.C. Historia Animalium is one of the most popular works by Aristotle, in which he refers to ticks as disgusting ectoparasites that are raised by grass (Hussain & Durrani, 2009). Ticks induce stress, immunodepression, weight loss, degradation of skin values, hypersensitivity, and toxicosis in bovines (Lorusso *et al*., 2013). One of the most significant issues that afflict the world, tick infestation affects about 80 percent of the livestock population (Kemal, Muktar, & Alemu, 2016). It is estimated that infestation of gnawers causes economic losses of 14,00018,000 million per year on average across the whole world and that a total of 498.7 million yearly in India was caused by tick-borne diseases (TBDs) (Minjauw & McLeod, 2003). The global costs of tick-borne diseases amount to between 13.9 and 18.7 billion US dollars per annum (Leger *et al*., 2013).

Literature Review

The most common clinical manifestations include the presence of petechial hemorrhages in the conjunctival mucosa, paler lymph nodes, as a consequence of intra-erythrocytic parasitism containing piroplasms, temperature rise, phlegm, malaise, and loss of weight (Horbar *et al.*, 2001; Van Rooij *et al.*, 2007). The heileriosis illness can cause the death of animals, but with ample attention, a rapid rate of recovery can also be achieved. This is based on the immunity and tolerance of the animal, along with the strains of the parasite. The severity of the disease, the diagnosis, and the prognosis could be estimated in terms of hematological parameters, clinical parameters, or changes in these parameters (Col & Uslu, 2007; Saeb, Baghshani, Nazifi, & Saeb, 2010). Consequently, the present study was predetermined to achieve the following objectives: to establish the impact of theileriosis on goats in Karak, urban, and peri urban of Pakistan.

Materials and Methods

Study Area

This was done in the southern part of Khyber Pakhtunkhwa (KPK), in Pakistan; this includes districts of Dera Ismail Khan, Tank, Bannu, Lakki Marwat, and Karak. Area-wise, the place is bounded by latitudes of 31 33 North and 70 72 East. Southern KPK has an arid climate and enjoys severe summers and moderate winters, and the rainfall is minimal. The biodiversity transition between the Indus plains and the Waziristan hills is ecologically significant to this area. Further, the region is mainly rural, and the livelihood activities comprise mainly farming and keeping livestock. Southern KPK offers a good location to study, given its special environmental and socioeconomic nature.

Figure 1
Map of Khyber Pakhtunkhwa Showing Southern Regions

Selection of Animals

The microscopy and hematological analysis on the prevalence of Theileriosis in District Karak were done on a variety of caprine breeds. The 300 (n=300) samples were selected in 8 different localities where goats were randomly sampled and checked to screen for caprine theileriosis. In each region, 50 goats were examined to determine whether they were males or females. The regions are all found in Karak Tehsil. In the sample collection, special attention was paid to the equality of the ratio of numbers and gender.

Blood Samples

Three hundred asymptomatic goats were used to collect their blood samples. Blood samples, after being collected in EDTA tubes, are then placed in ice boxes and taken to the laboratory. Data regarding different animal attributes that were put into consideration during sampling, like age, feeding methods, breed, tick infection, sex, season of the year, and the size of the herd.

Processing of Samples

The samples of blood were collected with 20 ml syringes and deposited with EDTA solution. The test was done, and analysis was made in the blood samples in the CVS & AH AWKUM.

Microscopic View of Blood Samples

Labeling of the blood samples was done through the examination of all blood samples through the microscope in Chemical & Life Sciences, QUSIT, Laboratory, DIKhan. An ordinary technique of conducting microscopy in blood samples was adopted. The process that followed was applied (Shahnawaz *et al.*, 2011).

Preparation of Geimsa Stain

The Giemsa stain, which is part of the Romanowsky class of stains, was used to stain blood smears. Looking at stained slides and finding parasites: Souls by (1982) described how to use keys to find parasites.

Table. 1

Preparation and Staining of Blood Smear

Step 1	The glass slides were cleaned effectively to do away with any contamination or debris.
Step 2	A drop of blood sample was put in the slide held in one hand, and with the edge of the other slide held at an angle of 450 with the other hand, the blood was smeared over the slide.
Step 3	Smears were subsequently air dried and fixed in absolute methyl alcohol for 1- 2 minutes
Step 4	Smears were finally stained with Giemsa stain at 10 % using 5 minutes.
Step 5	Use running tap water over the slides to wash out excessive stains and air dry.
Step 6	The previously stained smears were covered with the cover slip and viewed under an oil immersion lens (100X)

Hematological Examination

All the positive and negative blood samples were examined under a hematological analyzer. This was useful in establishing how the disease influences the various crevasses of the goat, such as the amount of red blood cells (RBCs), the level of hemoglobin (Hb), the volumes of packed cells (PCV), and the lymphocytes.

Data Analysis

It was done by using IBM-SPSS version 24.0 to calculate the descriptive statistics. The level of confidence, which was 95 percent, was considered statistically significant; the given p-value of 0.05 or a lower value was considered regarding the shift in the prevalence of *theleria* in risk variables.

Results and Findings

The study was implemented to investigate about the festival of Caprine Theileriosis in eight regions of Southern Area of Khyber Pakhtunkhwa and 400 samples of animals were taken to test the presence of disease, and out of these 400 samples of animals 155/400 (50%) of the animals were male goats and 155/400 (50%) of the animals were female goats. The processing of the samples and their acquisition happened in the Laboratory of Chemical & Life Sciences (Zoology), Qurtuba University of Science & Information Technology, DIKhan, Khyber Pakhtunkhwa.

Occurrence

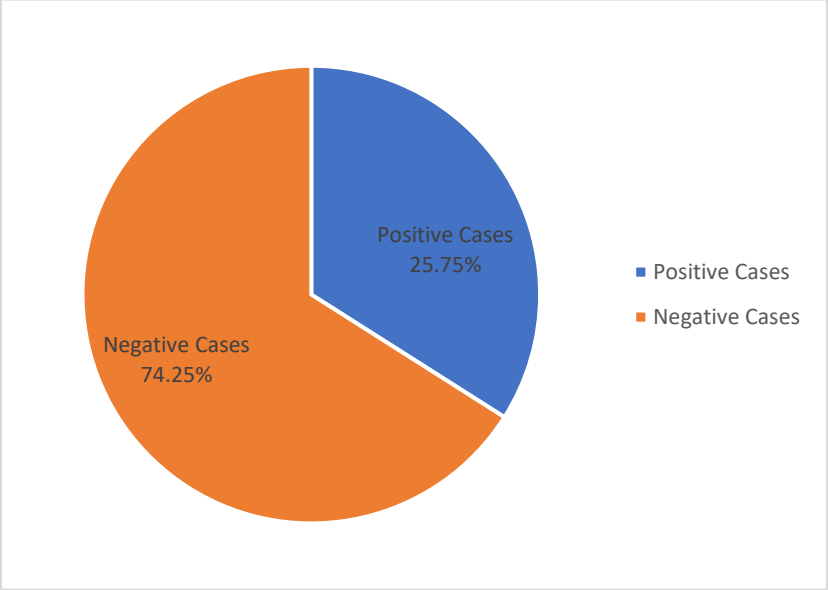
All the positive and negative blood samples were examined by using a hematological analyzer. This assisted us in deducing the impact the disease had on the various dimensions of physiology of the goat, such as the red blood cells (RBCs) count, hemoglobin (Hb) content, packed cell volume (PCV), and lymphocytes.

Overall Occurrence in Goats

Data from several sites showed that 103 out of 400 cases were positive (25.75%). Out of 25.75%, 44.18% (45 out of 103) were male positive cases, and 55.82% (57 out of 103) were female cases.

Figure 2

Overall Prevalence in Goats



Area-wise Prevalence

During the examination, prevalence in the region of Tank (TNK) was found to be 26/103x100, and the lowest prevalence was screened in the region of D. I. Khan (DIK), 11/103x100. The T-test results computed, and differences in how prevalent the disease was in each of the two regions were non-significant, i.e., ($p>0.05$).

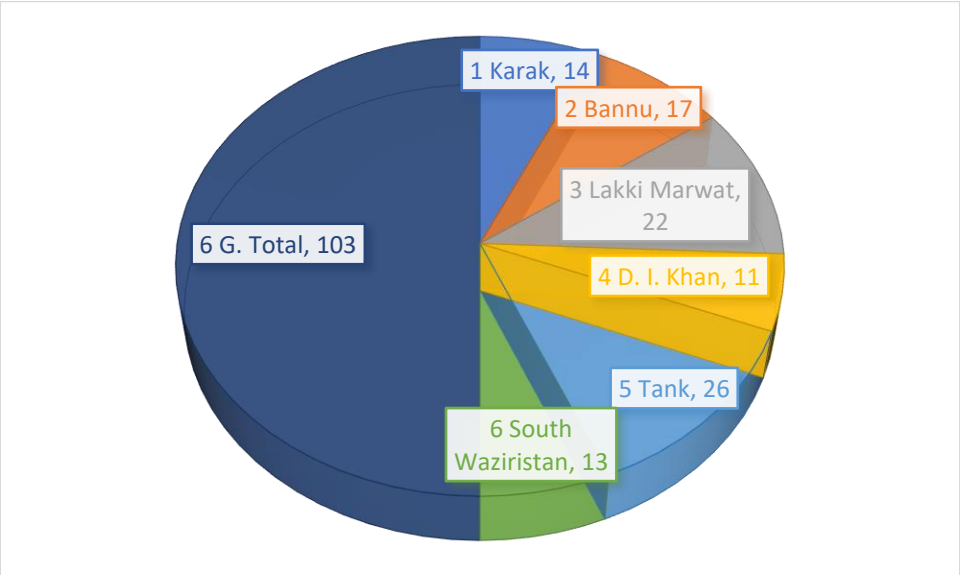
Table 2

District-wise Prevalence

No	Area	Sample Tag	Prevalence	%age	P-value
1	Karak	KC-001-52	14/103 ×100	13.59%	0.04
2	Bannu	BU-001-51	17/103×100	16.50%	0.04
3	Lakki Marwat	LM-001-52	22/103×100	21.35%	0.04
4	DIKhan	DIK-001-51	11/103×100	10.67%	0.04
5	Tank	TNK-001-52	26/103×100	25.24%	0.04
6	South Waziristan	SW-001-52	13/103×100	12.62%	0.04
	G. Total	400	103	25.75%	

Figure 3

District-wise Prevalence



Sex-wise Prevalence

When the blood was examined under a microscope, we realized that the hemocrit blood count of the infected was distinct compared to that of the normal blood. It was confirmed that the chances of a female goat getting infected were higher than compared of male goats. The number of female goats infected by Theileria (57 out of 102) was higher than that of males or 45 out of 102. At the end of the test, the results show that the RBC, Hg, and Hct decreased significantly ($P<0.01$). The infected young people also had significantly higher WBC ($P<0.01$) compared to those who were not.

Table 3

Area-wise Prevalence

No	Region	Male	%age	Female	%age	P-Value
1	Karak	05/103×100	4.9%	09/103×100	8.82%	0.0000
2	Bannu	09/103×100	8.82%	08/103×100	7.84%	0.0002
3	Lakki Marwat	08/103×100	7.84%	13/103×100	12.74%	0.004
4	DIKhan	12/103×100	11.76%	14/103×100	13.72%	0.315
5	Tank	06/103×100	5.88%	06/103×100	5.88%	0.012
6	South Waziristan	05/103×100	4.9%	07/103×100	6.86 %	0.219
	G. Total	45/103×100	44.18%	57/103×100	55.82%	

Figure 4

Sex wise Prevalence

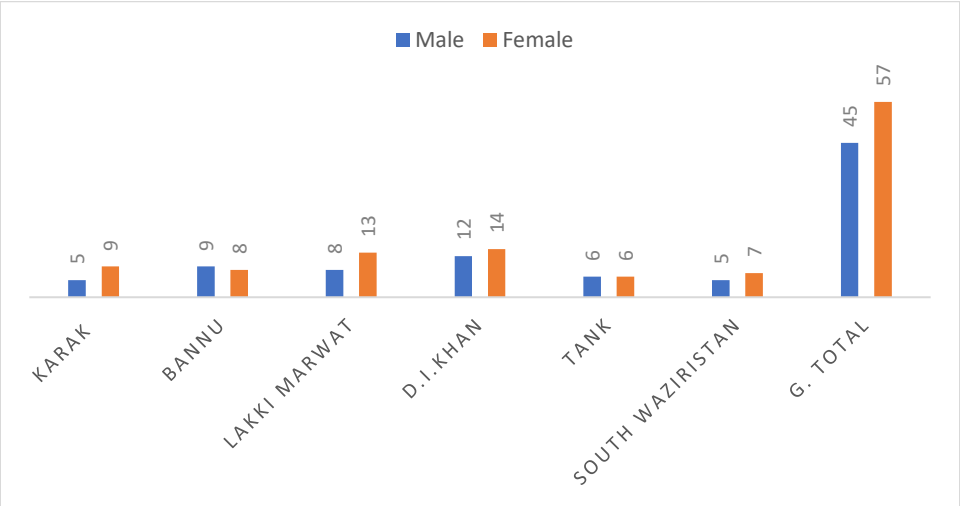


Table 4
Effect on Hematological Parameters

No	Parameter	Healthy Goat	Infected Goats	P-Value
1	Total WBC ($x10^3$/µl)	4-14	18.2	0.0000
2	Total RBC ($x10^6$/ µl)	8.02 -18.02	6.44	0.0002
3	Hgb (g/dL)	9-13	7.1	0.004
4	Lymphocyte (%)	22,00-10,000	12,500	0.315
5	Monocyte (%)	0-660	930	0.012
7	Granulocyte (%)	1300-7300	36,50	0.219
8	Platelets ($x10^3$/ µl)	399.48	580.5	0.00001

Discussion

The purpose of this study was to find out how common the dangers and effects of theileriosis are on blood parameters in different parts of Karak. It also kept track of how different types of housing, the time of year, the animals' age, and their gender affected their health. Theileriosis is a disease that spreads by ticks and affects some types of goats. Once an animal is sick, it may spread the disease to other species, which makes it a constant threat to animal populations since it can spread to humans. It was revealed that 102 out of 300 (34%) goats were positive cases once the laboratory diagnostic was finished. The right ecological conditions make it easier for *theileria* species to spread since their eggs may grow and there are a lot of vectors. The rise in the prevalence rate is due to a lack of effective health inspections and treatment. In the worst cases, it costs the goats money. A case study done by (Durrani *et al*., 2006) on 600 samples of animal blood displayed 17.8 percent of the disease. The clinical manifestations had such points as weakness, low blood pressure, abnormal gait, stiff neck muscles, abnormal posture, droopy eyes, infertility, dermatitis, and ticks in every part of the body. Gachohi, Skilton, Hansen, Ngumi, & Kitala (2012) reported that an epidemiological investigation found that a variety of things can affect the occurrence of theileriosis. These are the immunological

condition of the host, the infecting population of the vectors, the case fatality rate, and the animals affected by it. Other things that might increase the risk are where you live, how your house is set up, how the environment changes, how strong your immune system is, and how sensitive you are to illnesses. When you treat various genotypes as discrete units (species), it makes it much harder to look at parasite epidemiology research because it might hide the effects of adding or competing across genotypic populations. But they are quite important when it comes to transmitting the illness to ticks as a whole (Mans, Pienaar, & Latif, 2015). The weather conditions and the flora of the agro-agricultural regions in Karak make ideal conditions for the growth of Haylomma ticks that are the primary agent in transmitting tropical theileriosis in large parts of the Karak region. It is the basic explanation why the prevalence ratio is higher. The cross-sectional study of the caprine population in different geographical regions of the Karak district revealed that there were health issues due to theileriosis. Theileria species can infect goats because of dirty environmental conditions. *Theileria annulata* not only tears down blood cells, but if they aren't treated, they can also cause big financial losses. In goats that are nursing, it causes less milk to be produced. *Theileria* infection can also cause problems with reproduction, including abortion and lower fertility, since it affects the body's hormones. The study found that females were more likely to get theileriosis than males because their immune systems are weaker and they are exposed to a wider range of environments.

Conclusion

Theileriosis is becoming problematic for wild ungulates, particularly in tropical and subtropical regions globally. This study aimed to investigate *Theleria* infection in goats by microscopic identification and blood assays. During this experiment, we randomly collected 300 blood samples from healthy goats using a simple sampling approach. The blood samples were collected using disposable syringes and kept in EDTA tubes until they could be examined. Following appropriate staining procedures, the blood samples were examined under a microscope. Upon microscopic examination, it was determined that 102 out of the 300 samples, representing 34%, tested positive. A total of 45 male positive cases were identified, accounting for 44.1% of the overall cases, while 57 female positive cases represented 55.88% of the total. The area exhibiting the greatest prevalence was Lawaghar Chani Khel (LCK), accounting for 26 out of 102 cases, whereas the area with the least prevalence was Shaheedan Banda (SB), comprising 12 out of 102 cases. A total of 14 individuals from a group of 102 in Karak City (KKC), 17 from 92 in Mitha Khel (MK), 21 from 102 in Kamran Chowk (KC), 26 from 102 in Lawaghar Chani Khel (LCK), 12 from 102 in Ghundi Mirkhan Khel (GMK), and 12 from 102 in Shaheedan Banda (SB) were noted. The chi-square test revealed no significant difference ($p< 0.05$) in the prevalence of the disease between the two regions. A well-designed questionnaire that includes the owner's name and address, the season, the month the sample was obtained, the animal's age, sex, breed, tick status, and management (stall feeding or grazing). Blood parameters were determined by microscopy and auto-hematological analysis of the same blood samples. The t-test and P value for prevalence, risk variables, and hematological markers were all below 0.05 at the end of the study.

Declarations

Ethical Approval and Consent to Participate: This study strictly adhered to the Declaration of Helsinki and relevant national and institutional ethical guidelines. Informed consent was not required, as secondary data available on websites was obtained for analysis. All procedures performed in this study were by the ethical standards of the Helsinki Declaration.

Consent for Publication: Written informed consent was taken from the patient for the case report and publication. None of the personal information will be disclosed in the final publication.

Availability of Data and Materials: Data for this study will be made available upon request from the corresponding author.

Competing Interest: The authors do not have any conflicts of interest to declare.

Funding: Not Applicable

Authors' Contribution: All authors have an active role in the conduct, writing, and submission to the journal.

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