

Pathological Changes in Intestine of Goats Naturally Affected with Cryptosporidiosis and Coccidiosis

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ABSTRACT

The study was aimed to know the pathomorphological changes in intestine of goats affected with *Cryptosporidium* spp. and coccidian parasites. The intestinal samples from 60 slaughtered goats less than 12 months old were collected. Of these, 2 (3.33%) cases were positive for *Cryptosporidium* spp. infection by modified Ziehl-Neelsen staining method and 4 (6.66%) cases were positive for coccidiosis on direct faecal smear examination. Small, oval-shaped oocysts of *cryptosporidium* spp. were seen in intestinal contents smear. Microscopically the intestines of goats infected with *Cryptosporidium* spp. exhibited exfoliation and clubbing of the villi, with infiltration of mononuclear cells in the lamina propria. Several microabscesses were also observed in the jejunum and ileum. In cases positive for coccidiosis, histopathological examination revealed necrosis of the crypt epithelial lining cells, formation of crypt abscesses containing necrotic debris, and infiltration of inflammatory cells. Several developmental stages of *coccidia* were present within the epithelial cells of the mucosa, and lymphoid depletion was noted in the Payer's patches of the ileocaecal region.

Key words: Coccidian, Cryptosporidium, Goats, Histopathology, Intestine, Z-N staining.

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INTRODUCTION

Nowadays, goats are the most popular and vital source of livelihood for those with limited resources. Small-scale goat production is of substantial advantage to families and communities globally, in a variety of geographical conditions. The young goats in India is estimated around 148.88 million (Senapati, 2022). Unfortunately, the enormous number of goat population succumbed due to digestive diseases at the early stage of their lives, resulting greater economic burden on animal husbandry. Several protozoan parasites, particularly *Cryptosporidium* spp. and other *coccidia*, are recognized as significant etiological agents of diarrhea in goats. The prevalence of these infections varies widely across farms and geographic regions, largely influenced by herd size, management practices, and environmental hygiene. *Cryptosporidium* infection, or cryptosporidiosis, in both small and large ruminants poses a notable zoonotic risk, as the shedding of environmentally resistant oocysts can lead to contamination of water and food sources, thereby facilitating transmission to humans (Delafosse *et al.*, 2006; Ceballos *et al.*, 2009; Squire *et al.*, 2017).

Cryptosporidium is an obligate intracellular but extra cytoplasmic apicomplexan enteric protozoan parasite (Fayer and Xiao, 2008). The life cycle is monoxenous and infection is acquired mainly through the faecal-oral route and rarely by inhalation or aspiration (Shirley *et al.*, 2012). *Cryptosporidium* may possess a potential public health problem due to infective oocysts in the environment (Fayer and Xiao, 2008).

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Cryptosporidiosis causes decrease production of animals, decreased feed utilization, infertility and financial distress in the form of treatment of sick animals (Paraud and Chartier, 2012). The important histopathological changes consisted mainly in desquamation of intestinal epithelial cells and atrophy of the villi (Kalkanov *et al.*, 2015). Effective disease prevention requires to understanding of the environmental factors which predispose animal to *Cryptosporidium* spp. infection (Collinet-Adler *et al.*, 2015). In small ruminants coccidian is a important parasite that cause severe diarrhea in animals. It is a single-celled parasite, transmitted through

fecal-oral route and its multiplication take place in intestinal tract. It is contagious protozoan disease of small ruminants (Georgi and Georgi, 1990). It destroys the intestinal epithelial cells and produce characteristic lesions that can be seen at post mortem examination of the animals. This study was aimed to evaluate the pathological changes in intestine of goats naturally affected with cryptosporidiosis and coccidiosis.

MATERIALS AND METHODS

Collection of Samples

A total of 60 young goats up to 12 months of age that were slaughtered in slaughter houses in and around Rewa district of Madhya Pradesh (India) were observed for any gross pathological lesions in the intestine and mesenteric lymph nodes. Intestinal samples showing lesions, along with its fecal content were collected and transported to the laboratory on ice. Tissues for histopathological examinations were collected and preserved in 10 % neutral buffered formalin solution. *Coccidian* parasites were screened by direct faecal smear examination under microscope

Modified Ziehl-Neelsen Staining for Screening of *Cryptosporidium* Oocysts

The smears were prepared directly from the faecal samples/ intestinal contents on clean glass slides and air dried. They were fixed in methanol for 3 min and stained with carbol-fuchsin for 15-20 min, followed by rinsing in tap water, then decolorized in acid-alcohol (1% HCl in methanol) for 20 sec, followed by rinsing in tap water. After that the smears were counterstained with methylene blue for 1 min, followed by rinsing and air drying (Henriksen and Pohlenz, 2021). Smears were examined using 100 X objectives under the light microscope for identification of *Cryptosporidium* spp. In MZN staining cryptosporidium oocysts were seen as red, round or oval, acid-fast bodies.

Processing of Tissue for Histopathology

After proper fixation in 10 % neutral formalin solution, tissues were cut into small sections with a thickness of 2-3 mm and kept under tap water overnight for washing. The tissues were then kept in increasing grades of acetone, cleared in benzene and paraffin embedded (60-62°C melting point). Sections of 4-6 micron thickness were cut through a rotary microtome and stained with Haematoxylin and Eosin as per the standard procedures (Luna, 1972).

RESULTS AND DISCUSSION

On examination of intestinal contents of 60 slaughtered goats, two cases (3.33%) were found positive for *Cryptosporidium* infection. Dark red, small, round or oval-shaped oocysts of *cryptosporidium* were seen in faecal smears stained by

modified Ziehl-Neelsen staining method (Fig. 1A). In these cases, grossly the whole small intestinal tract was congested and the lumen was filled with watery, foul-smelling faecal material. Mesenteric lymph nodes (MLNs) were enlarged, edematous and showed subcapsular edema with infiltration of mononuclear cells. Microscopically in these cases, exfoliation of necrosed villi, and infiltration of mononuclear cells in the lamina propria of the jejunum was observed (Fig. 1B). In one more case microscopically, small intestine showed clubbing of the villi and formation of the several microabscesses in small intestine mainly in the jejunum and ileum (Fig. 1C). These microscopic changes in tissue were similar with the observations of others (Kalkanov *et al.*, 2015; Singh *et al.*, 2018). Unhygienic conditions, and insufficient and poor quality of colostrum may be the factors for the manifestation of the disease.

Out of 60 goats, 4 cases (6.66%) were positive for coccidian oocyst in the direct fecal smear examination (Fig. 2A). Grossly, congestion and hemorrhages of the intestinal tract were seen in one case. In another case thickening of the intestinal mucosa with multiple raised nodules was seen in the distal part of the small intestine (Fig. 2B). Microscopically these cases showed necrosis of the cryptic lining cells and cryptic abscess were seen in the distal part of small intestine (Fig. 3A), with lymphoid depletion of Payer's patches in ileocaecal area of intestine. Necrosis of muscle fibers with several developmental stages of coccidian was present in the epithelium of the mucosa (Fig. 3B). In the present study, gross lesions observed in coccidia-positive samples were in accordance with the findings reported earlier (Hussain *et al.*, 2017).

The study reports pathology of intestine of goats affected with *Cryptosporidium* spp. and coccidian parasites. Diarrhea is the prevalent disease of goats and causes acute dehydration and malabsorption leading to malnutrition in the affected animals. Exfoliation, clubbing of the villi and formation of the several microabscesses in the small intestine were the important changes that were observed. Young goats less than 12 months of age suffer more from coccidiosis and cryptosporidiosis due to underdeveloped immune system and stress conditions. Good hygiene and segregation of sick animals may prevent the transmission of these diseases. Effective prevention and control measures are needed to prevent the both parasite. Further study with large number of samples needed to study the actual status of *Cryptosporidium* spp. and coccidian parasite.

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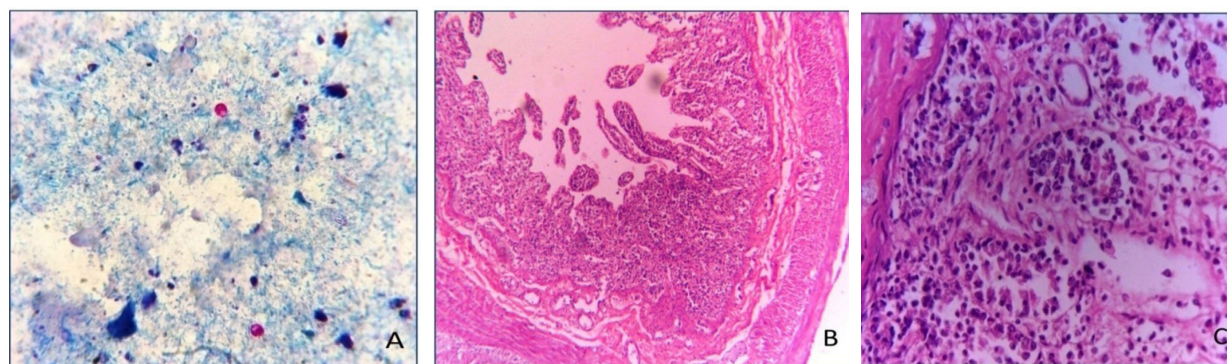


Fig. 1: (A) *Cryptosporidium* spp. oocysts in direct faecal smear of goats stained by mZN. Dark red small, round or oval oocysts (100X). (B) Histopathology of small intestine showing exfoliation of villi, infiltration of mononuclear cells in the lamina propria of jejunum (H&E, 10X). (C) Microphotographs showing severe infiltration of mononuclear cells and microabscess in crypt (H&E, 40X).

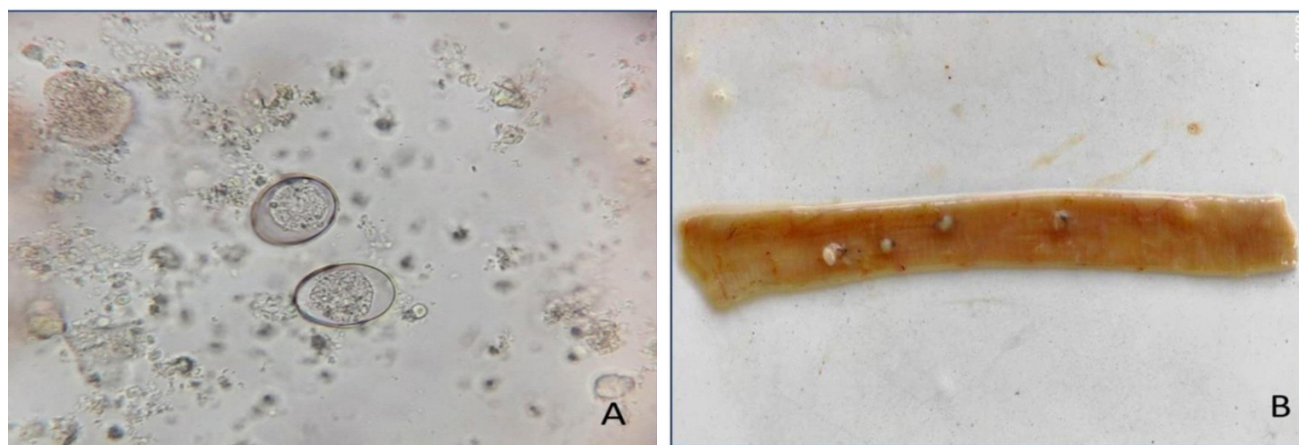


Fig. 2: (A) Coccidian oocysts in direct fecal smear of goat (40X). (B) Intestine of goat showing thickening of intestinal mucosa with several nodules.

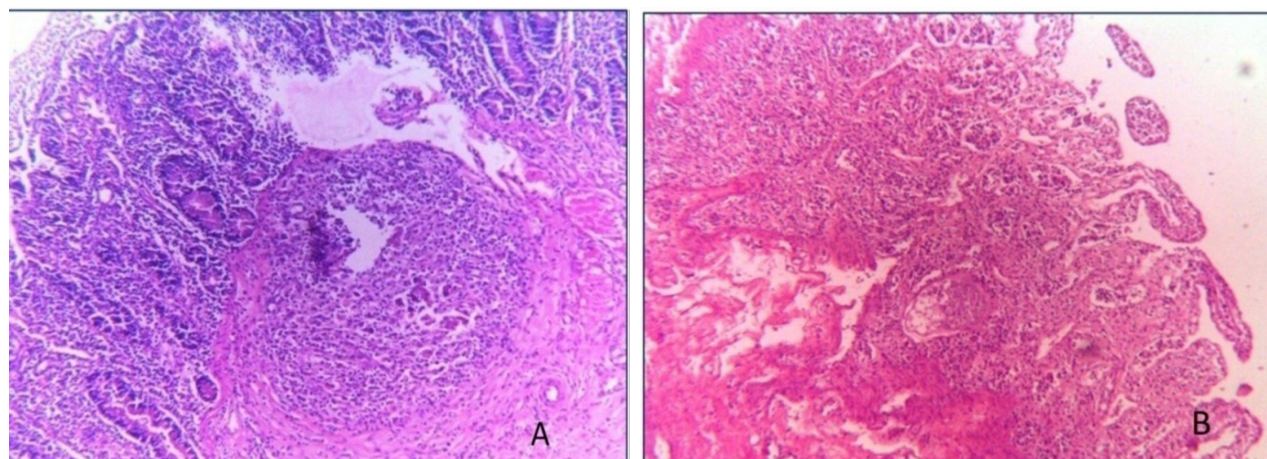


Fig. 3: (A) Microphotograph of distal part of small intestine of goat showing necrosis of the cryptic lining cells and cryptic abscess (H&E, 20X). (B) Microphotograph showing necrotic muscle fibers and developmental stage of *Eimeria* spp. (H&E, 10X)

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