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Research Paper

Gross and histopathological changes in colon of dairy calf infected with nodular worm

Z. A. Wani[1], R. A. Shahardar[1], S. A. Shah*[2], K.H. Bulbul[1], A. A. Shah[3], I. M. Allaie [1], H. M. Khan[3], M. Shafi[2] and A. A. Dar[3]

[1]Division of Veterinary Parasitology, F.V. Sc. & AH, Shuhama, Sher-e-Kashmir University of Agricultural Sciences and Technology of Kashmir; [2] Division of Vety. Pathology, FVSc & AH, SKUAST-K, Shuhama; [3]Mountain Livestock Research Institute, Manasbal
*e-mail: *vetshowkat@skuastkashmir.ac.in*

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ABSTRACT

A postmortem was conducted on the dairy calf at Mountain Livestock Research Institute, Manasbal to study the gross and histopathological changes in the colon infected with nodular worm (*Oesophagostomum radiatum*). Gross changes included thickened, oedematous mucosal surface with nodules prominent on its surface. Histological changes included granulomatous changes in the intestine along with infiltration of different types of cells. Further study is needed to report the pathological changes in detail and understand molecular and immunological mechanism lying behind the causation of disease so that appropriate control measures could be developed against the parasite.

Key words: Calf, colon, histopathology, nodular worm

Cattle rearing is an integral part of Indian economy including union territory of J&K. Cattle in J&K (18th All India livestock census of 2007). Cattle are being reared for their milk, meat, hides, cow dung, etc. Inspite of these advantages, livestock production is still low owing to their susceptibility to bacteria, viruses, rickettsia and parasites. These diseases not only cause heavy mortality in livestock species but also high morbidity which poses threat to profitable livestock production. The parasitic infections especially the gastrointestinal parasitism is present more often as subclinical infection which causes very huge economic losses without actually been noticed clearly. Among gastrointestinal nematodiosis, heamonchosis, mecistocirrosis, ostertagiosis, cooperiosis, paracooperiosis, oesophagostomosis, bunostomosis, ascariosis, etc. inflict colossal losses to animal husbandry sector. *Oesophagostomum radiatum* (nodular worm) is the most frequently encountered large bowel parasite of cattle (Kommu *et al*., 2017). These are usually 14-22 mm long, whitish and found in thick mucus in the caecum and proximal colon and cause anemia, protein-losing enteropathy, hypoproteinaemia and death (Chiejina, 1987). *Oesophagostomum* spp. also causes knotty gut or pimply gut in ruminants (Nwosu *et al.* 2007; Mbaya *et al.* 2009) which results in partial or total condemnation of intestines (Ahmed *et al.* 1994; Nwosu et al. 1996). Disease is more severe in debilitated and immunocompromised animals, while most of the infections are asymptomatic. The disease has been reported to be severe in younger stock less than 2 years of age. Oesophagostomosis infections result in thickened and oedematous intestine with small to large, round to irregular, hard, fibrotic, raised nodules about 5 mm in diameter, spreading to whole of the large intestine. Nodules are sometimes seen projecting into the lumen. Histopathology reveals granulomatous change in the intestine along with different types of cellular infiltration (Satish *et al*., 2018). The parasitic infection in cattle has been reported by various authors like Yadav *et al*. (2005); Mir *et al.,* 2003; Pandit *et al*., 2004; Bushra *et al*., 2013 and Aiman *et al*., 2017 from Jammu and Kashmir but the gross and histopathological changes associated with these parasites and *Oesophagostomum radiatum* in particular has not been studied. Therefore, this present work attempts to figure out the gross and histopathological changes in colon associated with *Oesopahagostomum* in cattle.

MATERIALS AND METHODS

Collection of the colon: A post-mortem was conducted on a cow calf of Manasbal dairy farm which died without exhibiting severe clinical signs. All internal organs were dissected out including large intestine in which colon was opened to look for any parasite or parasitic lesions. The portion after cleaning appeared thickened, congested and oedematous with numerous nodules studded on its surface along with presence of large number of adult worms. On opening few nodules caseous and calcified mass was seen, but no larval stage was observed. The tissue was fixed in 10% formalin for histomorphological studies and was further processed as per the procedure described below.

Processing of tissue for micro sectioning: Paraffin embedding technique was followed for micro-sectioning of tissues which were then cut into small portions of 2-3 mm thickness, washed in water for several hours followed by dehydration in ascending grades of alcohol. Clearing of tissues was performed in benzene and embedding in paraffin. Casting of blocks was carried out in L- molds (two L- shaped pieces) which facilitated the manipulation of size as per the requirement.

Micro sectioning: Rotary type microtome used for cutting paraffin sections of 5mm thickness. Continuous ribbons (6-7 inches long) of the material were cut and laid on the surface of constant temperature water bath (around 55^{o}C). The sections were separated with a heated scalpel after they spread completely. The cut sections were mounted on clean glass slides using Mayer's egg albumin as section adhesive. The mounted slides were marked for identification and dried in paraffin oven at 60^{0}C for one hour.

Staining of paraffin sections: The tissue sections were stained by Harris haematoxylin and eosin staining method. The paraffin sections were deparaffinised with Xylene before hydration through graded alcohol to distilled water. Staining was done as per the standard procedure followed by dehydration in ascending grades of alcohol. Clearing was performed in Xylene and mounting in DPX.

Mounting: A drop of DPX mountant was placed on a cover slip and the section on the slide pressed on it after taking out from Xylene. The slide was inverted and the cover slip pressed with a rod to remove air bubbles if any trapped.

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RESULTS AND DISCUSSION

The adult *Oesophagostomum radiatum* lives in the large intestine, primarily the caecum of cattle, but its larvae may be found from pylorus to anus in the gut mucosa. The parasite is considered as an important pathogen during cool and wet seasons as well as in cattle when present in large numbers (Craig, 2009). In acute infections, there is inflammation of small and large intestine with black foetid diarrhoea being passed out (Soulsby, 1982). In this infection, extensive nodule formation (pimply gut) occurs affecting whole of the intestine. The nodule formation occurs owing to previous sensitization or exposure to the larvae of *Oesophagostomum* spp. during which larvae pass to the submucosa and marked reaction in the form of localized inflammation around the larva takes place. Leucocytes especially eosinophils and foreign body giant cells collect around the parasite and the focus becomes encapsulated by fibroblasts resulting in the formation of nodule. They may encyst in the colonic wall and become mineralized sub serosal nodules or may mature to adults. These nodules generally are of no clinical significance, but they make the intestines unsuitable for use as sausage casings. Occasionally they are associated with, and can be the cause of intussusceptions (Gelberg, 2017). The parasites appeared whitish in colour, 1-2 cm in length, robust with anterior end bent spirally and possessing cephalic vesicles with rounded mouth collar and two rows of leaf crowns. These were identified as *Oesophagostomum radiatum* as per key described by Soulsby (1982). Representative portion of the tissue depicting the gross lesions was processed further to study the histopathological changes. Gross pathology included thickened, congested mucosa. Numerous nodules were found attached all along the surface (Fig 1& 2). Microscopic examination of the tissue section revealed a central necrotic mass surrounded by layers of inflammatory cells (Fig 3 & 4). The infilitrating cells were an admixture of various inflammatory cells and included eosinophils, plasma cells and lymphocytes, among which eosinophils were more clearly distinct (Fig 5 &6). Similar changes reported by Shelton and Grifth (1967) and Sharma *et al*., (1997) included observed thickening of mucosa, nodule and creamy exudates material from the incised nodules in sheep. Kommu et al., (2017) reported nodules in the intestine of calves from which fourth stage larvae of *Oesophagostomum radiatum* was recovered. Satish *et al*., (2018) reported multiple small to large, round to irregular, hard, fibrotic, raised nodules that projected into the lumen of large intestine in sheep. On histopathological examination, large intestine showed parasitic nodules with central baso-eosinophilic necrotic area surrounded by layers of inflammatory cells.

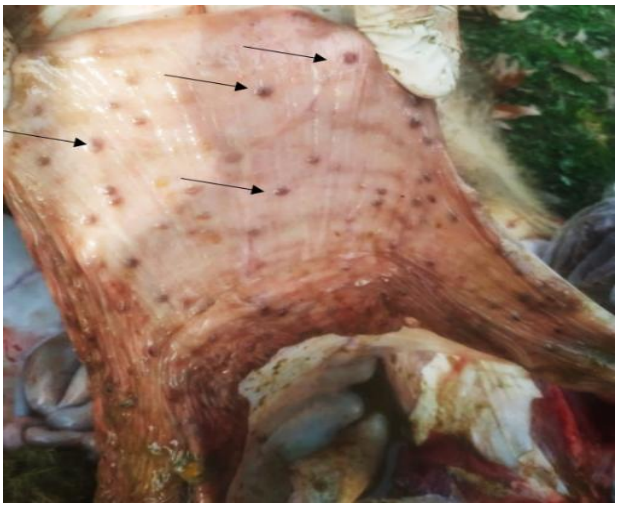


Fig. 1: Colon of calf revealing presence of nodules (arrows) on the mucosa

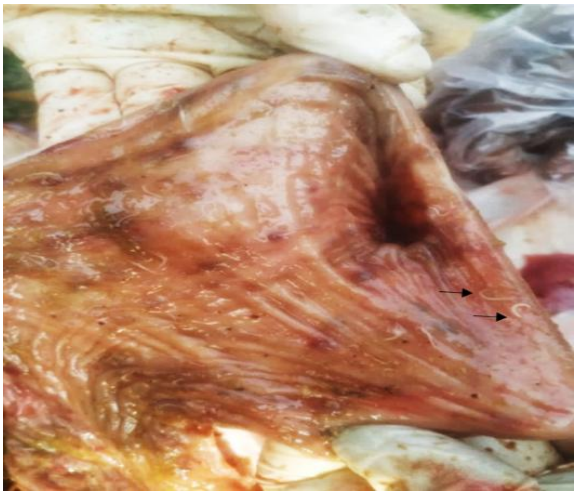


Fig. 2: Adult *Oesophagostomum radiatum* (arrow) lying attached to the mucosa

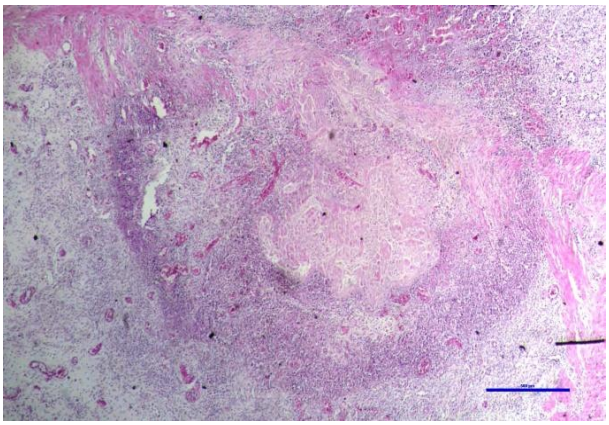


Fig. 3: Photomicrograph of a section of colon showing generalised necrosis of mucosa, a central necrotic mass surrounded by layers of inflammatory cells. *H.E. Scale bar 500 µm*

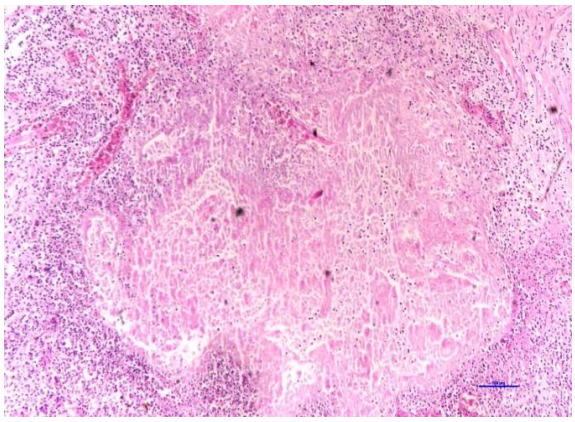


Fig. 4: Photomicrograph of a section of colon showing central eosinophilic necrotic area surrounded by admixture of inflammatory cells. *H.E. Scale bar 500 µm*

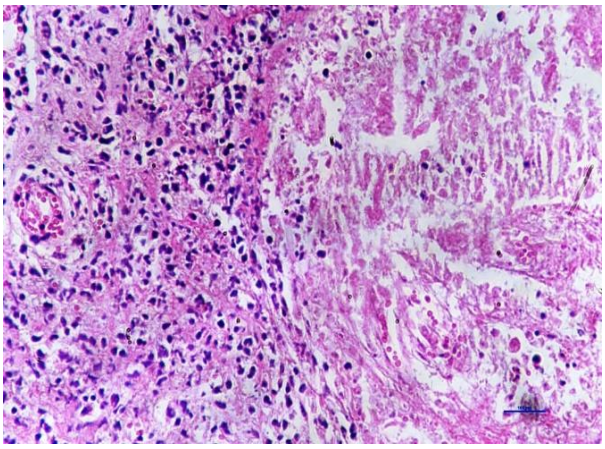


Fig. 5: Photomicrograph of a section of colon showing central eosinophilic necrotic area surrounded by inflammatory cells more clearly. *H.E. Scale bar 100 µm*

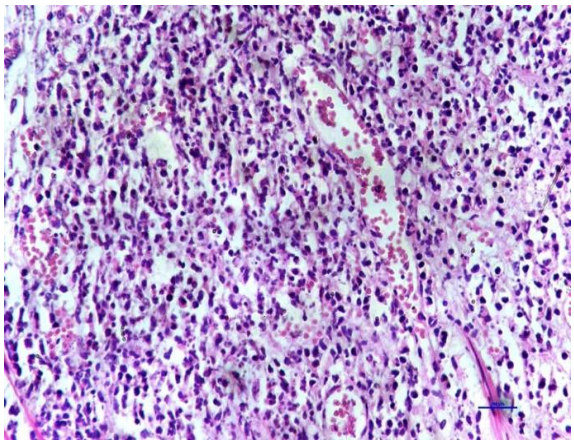


Fig. 6: Photomicrograph of a section of colon showing severe infiltration of eosinophils, plasma cells and lymphocytes. *H.E. Scale bar 100 µm*

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The lesion seen mostly on necropsy is a granulomatous nodule, 0.5 to 1.5 cm in diameter which appeared as big as pea was reported by Carlton and MCGavin (1995). Therefore, it is concluded that these worm infections need to be controlled to prevent economic losses to livestock industry as well as to the farmer community. But, effective worm control cannot always be achieved by drugs alone. Hygiene also plays an important role where the pen must be cleaned thoroughly and daily. Reducing pasture contamination by alternately grazing and cropping can help to minimize the parasite burden. It is better to adopt integrated parasite management strategy in letter and spirit.

CONCLUSION

This study highlights the significant pathological impact of *Oesophagostomum radiatum* infection in cattle, particularly during cool and wet seasons. The primary lesion observed was the formation of nodules in the intestinal mucosa, a result of larval migration and subsequent inflammatory reactions. Histopathological examination revealed central necrotic areas surrounded by eosinophils, plasma cells, lymphocytes, and fibroblasts, consistent with previous findings in both cattle and sheep. This study underscores the importance of recognizing and managing *O. radiatum* infections to prevent economic losses in the livestock industry and potential gastrointestinal complications in affected animals.

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