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Pathology of Natural Infections of *Schistosoma bovis* in Cattlein Ethiopia

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Abstract: Shistosomiasis is an infection of the vascular system of animals caused by *Schistosoma*species. AdultSchistosomaparasite counts and pathological lesions elicited by *Schistosoma*in the liver, intestine, mesenteric lymph nodes and mesenteric veinsof cattle slaughteredat abattoir were studied... Adult *Schistosoma bovis* (*S. bovis*) were detected in the mesenteric veins in 103 of 366 cattle (28.1%). Of 103 cattle with *S. bovis* in their mesenteric veins, 51 (49.5%), 42 (40.8%) and 10 (9.7%) cattle were heavily, moderately and lightly infested, respectively. Firm and pale liver was observed in infected cattle. The mucosa of the intestine was thickened; mesenteric lymph nodes were also atrophic and calcified. Microscopic evaluation of H and E stained sections of liver, intestine and mesenteric lymph nodes revealed granulomatous inflammation characterized by aggregates of mononuclear cells around the eggs. van Gieson's staining also showed excess collagen deposition in the liver. The mesenteric veins revealed marked medial hypertrophy. Statistically significant ($p \leq 0.05$) association was found betweenthe extent of microscopic liver lesions and worm counts in the veins. The study disclosed that lesions associated with *S.bovis* were not restricted to a single organ rather many organs were involved.

Key words: Cattle · Granuloma · Pathology · *Schistosoma*

INTRODUCTION

Shistosomiasis is an infection of the vascular system of mammals and birds due to trematodes of the genus *Schistosoma*. It is endemic in the tropics and subtropics [1-2]. Bovine Schistosomiasis was reported in fogera district, Northwest, Ethiopia with prevalence of 13.7% and 10.17% by Mersha *et al*. [3] and Mengistu *et al*. [4]. To date several *Schistosoma* speciesof veterinary and medical significance have been identified [5-8] *S. bovis* is the only species identified from domestic animals in Ethiopia [9]. The vectors, which are the snails, belong to the genus *Bulinus*, *Indoplanorbis* and *Planorbarius* [9].

Pathological lesions induced by *Schistosoma* consist of a granulomatous inflammation around the eggs trapped whiletraversingvia tissues [10-12] despitethe fact that Ethiopia has many wetlands that could favor snailmultiplication, studies on animal schistosomosis are scanty [9, 13]. In particular, pathologies associated with *Schistosoma* infection are untouched. Thus the purpose of the present study was to characterize pathological lesions associated with *S. bovis* infection in naturally infected, but apparently healthy cattle slaughtered at Bahir Dar abattoir, northwest Ethiopia.

MATERIALS AND METHODS

Study Area: The study was conducted atBahir Dar, which is located at 11° 29' latitude north and 37° 29' longitude east, northwest Ethiopia. The area has an altitude of 1802 masl with warm humid climate with an average annual rainfall of 700 mm. The annual temperature of the area ranges from 12.4-27°C. At the rainy seasons floods from Lake Tana and Abay River leave pockets of water bodies in adjacent areas.

Animals: The sampling units of the study were local breeds of cattle. During ante-mortem inspection, study animals were identified and the same animals were examined after slaughter. Three hundred and sixty six cattle were examined by systematic random sampling method.

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Sample Collection and Processing: The mesenteric veins of each animal were dissected and adult *Schistosoma* parasites were harvested from one third of the total length of mesenteric veins and counted [14]. Total worm counts were categorized as light (less than 30 *Schistosoma*), moderate (31-100 *Schistosoma*) and heavy (greater than 100 *Schistosoma*). The liver, intestine, mesenteric lymph nodes and mesenteric veins of cattle with adult *Schistosoma* parasites in their mesenteric veins were visually inspected, palpated and incised or dissected for identifying gross lesions. Tissue samples were fixed in 10% neutral-buffered formalin for histopathology. Tissue sections (4 μm thick) were stained with Hematoxylin and Eosin (H&E) and with van Gieson's [15]. Sections were examined under the microscope for the presence of egg-induced lesions. Microscopic liver lesions were categorized as mild, moderate and severe by considering the degree of inflammatory responses, states of *Schistosoma* eggs in the granuloma and distribution and intensity of lesions.

Statistical Analysis: The association of the extent of microscopic liver lesions and worm counts was tested by Chi-Square (χ^2) test. Values were considered statistically significant at $P \leq 0.05$.

RESULTS

Adult Worm Counts: Out of 366 cattle examined, 03 (28.1%) harbored adult *S. bovis*in their mesenteric veins and of these, 51 (49.5%), 42 (40.8%) and 10 (9.7%) cattle were heavily, moderately and lightly infested, respectively.

Description of Gross Lesions: The liver was firm and pale in color in most cattle harboring adult *S. bovis* in their mesenteric veins. The dominant lesions in the intestine include thickeningof the mucosa with petechial hemorrhage. Mesenteric lymph nodes were also enlarged and edematous and in some cases atrophic. Gross lesions due to schistosomosis were not apparent in the mesenteric veins.

Description of Microscopic Lesions: Liver lesions. Two distinct types of granulomas induced by Schistosoma eggswere detected in the liver. Some of the granulomas contain Schistosoma eggs with intact miracidium and with dense infiltrates of mononuclear inflammatory cells around the eggs (Fig. 1a). Others contain partially deteriorated eggs. The degree of severity of liver lesions due to Schistosoma infection in cattle was categorized as mild, moderate and severe. Of the total 103 liver examined from Schistosoma infected cattle, 60 (58.3%) were found to be severely affected,whereas 33 (32%) were moderately affected and 10 (9.7%) were mildly affected. A strong association (χ^2=860, P = 0.0) has been observed between the severity of liver lesions and worm burdens in the mesenteric veins.

In mild forms of hepatic schistosomosis, few granulomas containing *Schistosoma* eggs were surrounded by variable number of inflammatory cells, but the hepatic parenchyma appeared unaffected. However, in moderate cases of hepatic schistosomosis, numerous granulomas were detected in the portal tracts and the limiting plates were distorted with inflammatory cells. The severe form of hepatic schistosomosis had variable lesions. Van Gieson's staining revealed the presence of dense collagen deposition around the intrahepatic and portal veins (Fig.1b) and nodular hyperplasia and bridging fibrosis that connects the portal area to the central area were also evident in the liver (Fig. 1c).

Intestinal, mesenteric lymph node and mesenteric vein lesions. *Schistosoma* egg granulomas were detected in the submucosa of the intestine. Mononuclear and epithelioid cell infiltrates were detected in the submucosa. In the mesenteric lymph nodes, *Schistosoma* egg granulomas were detected in the cortex and medulla. Abundant inflammatory cell infiltrates consisting of eosinophils and hypertrophy of the tunica media of veins in the lymph nodes characterized lesions caused by *Schistosoma* parasite in the mesenteric lymph nodes. The mesenteric veins exhibited marked medial hypertrophy (Fig. 1d).

DISCUSSION

On the basis of postmortem examination, 28.1% (103/366) of cattle were positive for *S. bovis*. Of this, 49.5% (51/103) of cattle were heavily infested with *S. bovis*. This clearly indicates the level of contamination of grazing areas with cercariae. Cattle with adult *Schistosoma* parasites in their mesenteric veins had gross lesions in the liver, intestine and mesenteric lymph nodes. Cattle infected with *S. bovis* had firm liver with diffuse gray to white nodules in the hepatic parenchyma. The intestinal wall was also thick and mesenteric lymph nodes also showed edematous to atrophic changes. These findings were in support of previous reports and mayoccur as a result of trapping of eggs while traversing via tissueswith consequent granulomatous inflammation [16].

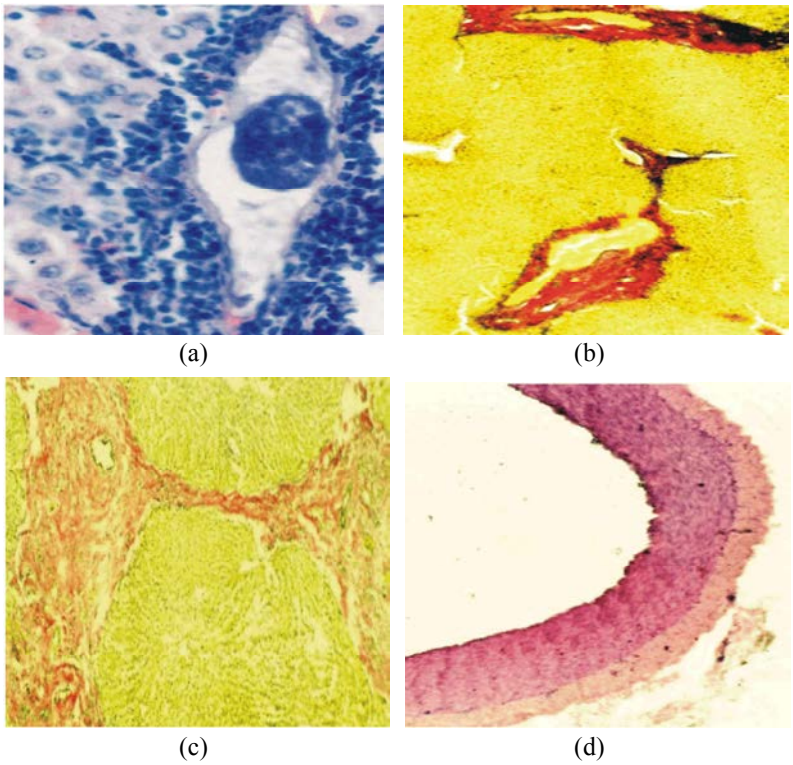

Fig. 1: Liver and mesenteric veins from cattle affected with *Schistosoma* (a) *Schistosoma* egg granuloma containing intact miracidium (arrow) and encircled by mononuclear cells (H and E, 400x) (b) dense collagen deposition (red) around congested intrahepatic vein (arrow) (van Gieson's, 100x) (c) bridging fibrosis; linking portal area to central area with nodular hyperplasia of the liver parenchyma (arrow) (van Gieson's, 100x) and (d) mesenteric vein showing medial hypertrophy (arrow) (H and E, 40x). Total magnification is given in parenthesis.

Histopathological lesions associated with *Schistosoma* infection were prominent in the liver. According to the stages of deterioration of *Schistosoma* eggs in the granuloma, type I eggs appeared morphologically intact and contain miracidium. Type II eggs were partially deteriorated in the granuloma. In rodents experimentally infected with *S. mansoni*, eggs with varied stages of deterioration were detected in the granuloma [17]. This may be due to host immune response mounted against eggs antigens. A strong association was observed between the severity of microscopic liver lesions and worm burdens. As the worm burden increases, liver lesions were more severe. In experimental study in cattle exposed to cercariae, the extent of liver lesions were found to be directly proportional to the number of cercariae administered and the number of eggs trapped in the liver [16]. The mild form of hepatic schistosomosis was characterized by few granulomascontainingepithelioid cells, lymphocytes and macrophages in the liver. Hussien [11] observed comparable hepatic lesions in animals infected with *Schistosoma*. Many granulomas containing *S. bovis* eggs and variable number of mononuclear cells were also detected in the hepatic parenchyma and in portal areas. These lesions characterize the moderate form of hepatic schistosomosis and were consistent with the reports of Hussien [11] and Andrade [18]. Severe hepatic schistosomosis characterized by numerous granulomatous inflammatory foci were observed in the portal tract and in hepatic parenchyma. Collagen deposition around the portal and intrahepatic veins and also nodular hyperplasia with bridging fibrosis explain the chronicity of the problem. Several authors have demonstrated chronic hepatitis as a result of eggs trapped in the liver, which may result in cirrhosis and nodular hyperplasia distorting the hepatic vasculature [11, 18-19].

Acute, but mild enteritis due to *S. bovis*was marked by the presence offew infiltrates of eosinophils and lymphocytes in the lamina propria and in the submucosa of the intestine. The chronic intestinal lesion was characterized by few granulomatous fociwith aggregates of macrophages, epithelioid cells and lymphocytes in the

vicinity of the eggs. Perivascular cuffing of mononuclear cells and frank vasculitis have been observed in the intestinal submucosa of goats infected with *Schistosoma* [20, 21]. Intestinal granuloma associated with *S. bovis* has been reported to contain smaller number of lymphocytes than liver granuloma [17]. Weinstock and Boros [22] also demonstrated in mice infected with *S. mansoni*, less intense granulomas in the colon and ileum than in the liver. Similar lesions described for the liver, also appeared in the mesenteric lymph nodes. Smaller veins in the mesenteric lymph nodes also showed medial hypertrophy. Hussein [11] has demonstrated hyperplastic portal and mesenteric lymph nodes with the formation of granuloma due to *Schistosoma* infection. The mesenteric veins also showed medial hypertrophy. The reason for medial hypertrophy of veins may be due to intimaldamage andthrombi formation induced by the parasite. Adult *Schistosoma* worms may cause phlebitis and venous thrombosis with medial hypertrophy [11] Tit i*et al*. [23] stated that mature parasites in the mesenteric veins did not evoke any inflammatory reaction. The present study considered one of the most important parasitic diseases of domestic animals, whose pathologies remainuntouched in Ethiopia. This is the first pathological study conducted to demonstrate lesions caused by *Schistosoma* infection in naturally infected cattle in Ethiopia. Thus detailed pathological, immunological and molecular studies are recommended to be done in high-risk areas of Ethiopia.

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