



## First recorded outbreak of *Paramphistomum cervi* in Andaman local goats (*Capra aegagrus hircus*) from Bay island of India: A brief communication

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### ABSTRACT

This communication reports an outbreak of immature paramphistomiasis in Andaman local goats (*Capra aegagrus hircus*) along with report of mortality and gross as well as histopathological changes. During the month of September, 2021 there was report of 38.57% mortality in goats which was investigated in depth. Outbreak occurred at South Andaman district where goats were reared under semi intensive managerial system and allowed to graze in the morning in surrounding marshy land. The affected animals showed the symptoms of immature amphistomiasis; it was further confirmed on necropsy and identification of causative organism as *Paramphistomum cervi* based on its typical morphological features. Histopathological examination showed damaged structure of rumen with typical degenerative changes and initiation of necrotic changes of cells with infiltration of inflammatory cells. We report the outbreak of immature amphistomiasis for the first time from Andaman and Nicobar archipelago along with the causative agent.

**Keywords:** Andaman and Nicobar Islands, Goats, Immature amphistomiasis, Outbreak

Immature paramphistomiasis is yet a very poorly understood disease of ruminants and falls under the category of neglected disease (Hajipour *et al.* 2021) and often remains undiagnosed (Phiri *et al.* 2007). It results in high death rates, reduced weight gain and milk production (Chaudhry *et al.* 2017). The disease is rampant in most parts of the world especially in Africa, Asia, Europe and Australia (Dube and Aisien 2010, Huson *et al.* 2017). Moreover, it has been declared as emerging problem in UK and Ireland (Murphy *et al.* 2008, Gordon *et al.* 2013). These parasites are commonly known as rumen fluke; there are four genera of rumen fluke: *Paramphistomum*, *Cotylophoron*, *Calicophoron* and *Ceylonocotyle*. Different species of rumen fluke are seen in different parts of the world. The most common species, *Calicophoron calicophorum* is mostly reported from Australia (Rengel *et al.* 2003). Another predominant species *Paramphistomum cervi* is seen throughout the globe. Likewise *Cotylophoron cotylophorum* is also cosmopolitan in distribution except in northern temperate zone. *Ceylonocotyle* is restricted in India and Australia (Soulsby 1982, Hotessa and Kalo 2020).

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Immature paramphistomiasis is a major problem in Indian subcontinent. The fluke causes havoc mortality in small domestic ruminants. Mortality rate may reach up to 75-88% (Dutt 1980, Choudhury 1994, Agrawal 2003). Prevalence and outbreak of immature amphistomiasis has been reported from Northern (Vohra *et al.* 2016, Jaiswal *et al.* 2018) and also from Southern part of India (Pavan Kumar *et al.* 2014). A perusal of available literature suggests that, till date species of amphistome has not been reported in domestic ruminants from ANI except in elephants in 1970s (Mukherjee and Chauhan 1972). We describe here the first recorded outbreak of immature amphistomiasis in Andaman local goats and first report of the species of rumen fluke from Bay island of India.

### MATERIALS AND METHODS

*Study area and samples:* Andaman and Nicobar Islands (ANI) is a union territory of India consisting of 572 islands; only 38 are inhabited and comprise of tropical rainforest canopy. These groups of islands are separated from the rest of the world by natural geographical barrier.

The outbreak occurred in Mithakhari village of South Andaman district, Andaman and Nicobar Islands in September, 2021. In this village, a total of 70 Andaman local goats were reared under semi intensive management system. During day time, animals were allowed to graze in

the nearby water logged marshy land since the weather was wet throughout the month of September, 2021 with 554 mm rainfall over 30 days. Affected animals exhibited symptoms of weakness, cachexia, distension of the abdomen, bottle jaw, profuse diarrhoea and ultimately death. Post mortem examination of dead animals was done and lesions were documented.

**Processing of samples:** Portion of rumen infested with worms were collected and fixed in Bouin's fluid at room temperature. After fixation of tissue samples, routine histopathological sections were prepared. In brief, samples were dehydrated by ascending grades of alcohol (70-100%). Following dehydration, the samples were cleaned in benzene and finally impregnated in paraffin. After preparation of paraffin block, sections of 5  $\mu$ M thickness were made and finally stained with haematoxylin and eosin. Digital photomicrographs were taken using Leica DM 2000 microscope.

Isolated immature worms were further flattened in between two glass slides and fixed in Bouin's fluid for 24 h at room temperature. Further, samples were stained by conventional borax-carmin stain and identified following standard methodology.

## RESULTS AND DISCUSSION

Immature amphistomiasis is a neglected trematode disease of ruminants and is responsible for economic loss (Hajipour *et al.* 2021). The incidence of the disease in goat varies from 18.26 to 91% (Singh *et al.* 2015). Apart from India (Swarnkar and Singh 2012, Pavan Kumar *et al.* 2014, Vohra *et al.* 2016, Jaiswal *et al.* 2018), the disease has been reported from Pakistan (Farooq *et al.* 2012), Bangladesh (Hossain *et al.* 2015), Thailand (Sangvaranond *et al.* 2010) and Malaysia (Tan *et al.* 2017). This parasitic infection is responsible for economic losses through morbidity,

mortality, reduced meat and milk production, reproductive disorders and expenditure on the purchase of anthelmintic (Mogdy *et al.* 2009). Production loss occurs since the parasite is responsible for lower nutrition conversion (Horak 1971). It has been estimated that more than 500 million ruminants worldwide are at risk (Ristic 1988). Further, the death rate due to immature paramphistomosis is very high and may go up to 80-90% in young ruminants (Shablh and Juyal 2006).

During the present study, the farmers reported mortality of the goats (average 2-3 per day) showing the symptoms of cachexia, abdominal pain, bloat/distension of abdomen, abnormal accumulation of clear oedema fluid under the skin of the lower jaw (bottle jaw), profuse water diarrhoea followed by death. Within 2 weeks, 27 goats of more than one year age died with mortality rate of 38.57%. The high incidence of immature paramphistomosis is attributed to presence of stagnant water bodies due to continuous rainfall. This was conducive for the dominance of snail population (*Indoplanorbis* spp., *Gyraulus* spp. and *Lymnaea* spp.), which in turn predisposes the release of number of cercaria into the water and nearby forage (Shablh and Juyal 2006, Pavan Kumar *et al.* 2016). During the present investigation, it was found that the animals were raised in semi intensive system and were also allowed to graze in the marshy land.

Examination of abdomen revealed accumulation of straw coloured fluid in the peritoneal cavity and distension of gall bladder. Numerous immature flukes and patches of haemorrhages were detected in the duodenum in the intestine; they were embedded in the mucosa and reported to be plug feeders by drawing pieces of mucosa in the suckers. In the abomasum, there were ulcerative changes with presence of few numbers of flukes. Numerous worms were isolated from rumen of the affected animals. Ingestion of large number of metacercaria settled at duodenum caused severe haemorrhagic areas and thickening of duodenal mucosa. Bottle jaw and ascites might be due to hypoproteinaemia caused by immature flukes (Radostits *et al.* 2010).

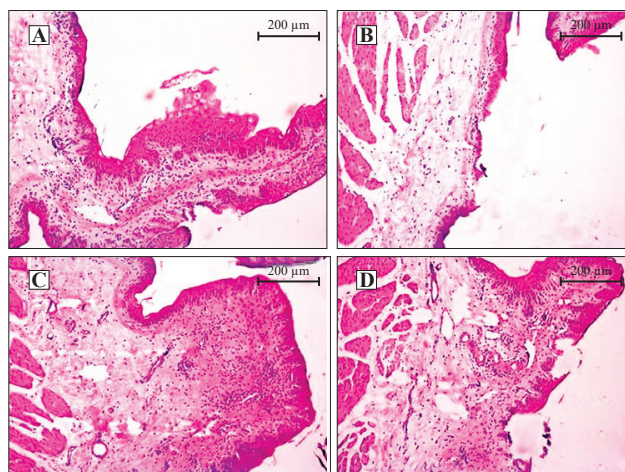


Fig. 1 (a-d). Histopathology of infected rumen: (a) Section of rumen papilla showing infiltration in propia-submucosa and necrosis in lining epithelium; (b) Section of rumen showing focal desquamation of tunica mucosal layer; (c) Section of rumen showing huge infiltration of polymorph into submucosa and degenerative changes; (d) Section of rumen showing sloughing tunica mucosa and vacuolar space in submucosa.

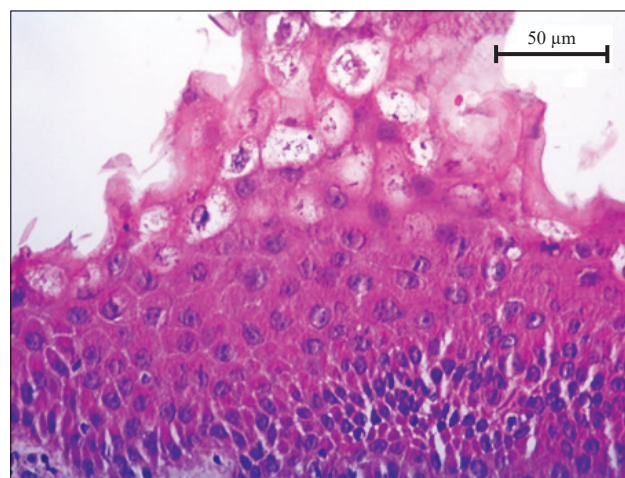


Fig. 2. Histopathology of rumen showing swollen, misshapen cells with birefringent having vacuolar degeneration and karyorrhexis.

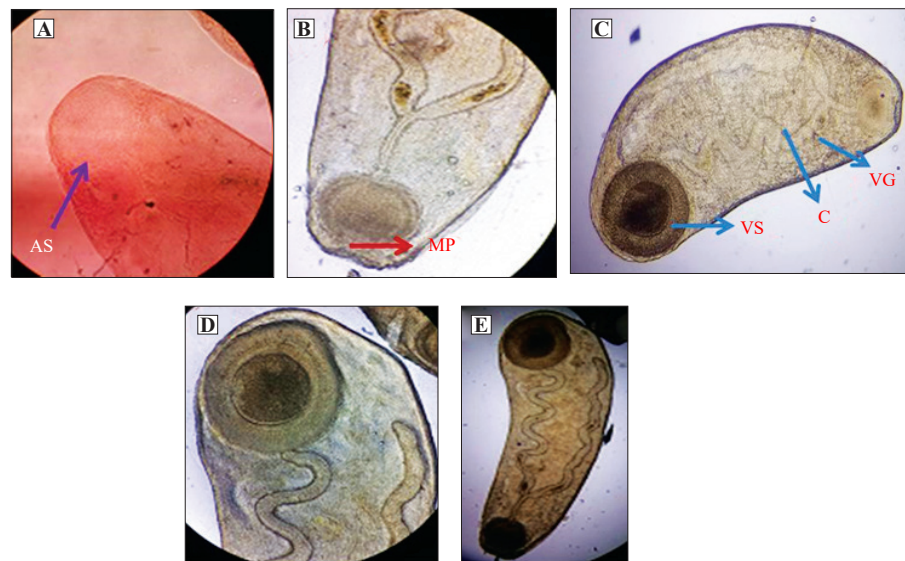


Fig. 3 (a-e). Morphology of *Paramphistomum cervi*: (a) Anterior sucker (AS) present at the tip of the cone (AS); (b) Mouth portion (MP) situated at the terminal end; (c) Caeca (C) serpentine in shape, clusters of vitelline glands (VG) extended from pharynx, reached up to posterior sucker (VS), lie between caeca and lateral margin of the body; (d) Intestinal caeca extends up to anterior portion of posterior sucker; (e) Worm convex dorsally and concave ventrally.

Rumen papillae were represented with keratinized stratified squamous epithelial cells. Rumen papillae showed infiltration in propria mucosa and there was zone of necrosis at the lining epithelium (Fig. 1a). Section of rumen exhibited focal degeneration of tunica mucosal layer (Fig. 1b). The deeper layer of rumen papillae was irregular, arranged around the central cores papillary processes, which extended into the epithelium and rumen cavity. Rumen showed massive infiltration polymorphonuclear leucocytes into submucosal layer along with degenerative changes (Fig. 1c). There was sloughing of tunica mucosa and formation of vacuolar space in submucosa (Fig. 1d). A zone of flattened cells was also found closer to the lumen. Rumen also showed misshapen cells with birefringent having signatures of vacuolar degeneration and karyorrhexis (Fig. 2).

This has been depicted in the past that, adult amphistomes in rumen do not cause any pathological changes and infected animals become lifelong carrier (Soulsby 1982). But the opinion was contradicted by Basak and Sinha (1978) who reported knob infiltrated with polymorphonuclear leucocytes and such knob blood vessels were highly congested. Subsequently, findings of Basak and Sinha were supported by Singh *et al.* (1984) who performed study through experimental infection. The workers observed pathological changes in the rumen 80 days post infection (pi) in goats. We could record histopathological changes in rumen which might indicate that, the infection was picked up from the pasture by the Andaman local goats not less than 80 days before. As the infection could be related with rainfall due to abundance of snail fauna, the animals might have picked up infection in the month of May since heavy rainfall started from May and ended in the month of September (range: 321.90-

649.20 mm) (<http://andssw1.and.nic.in/ecostat/2021/totalrainfall.pdf>). Pathological changes of rumen papillae and cellular infiltration, desquamation of epithelial cells of rumen papillae and infiltration of mononuclear cells were documented in the past (Cankovic and Batistic 1963, Singh *et al.* 1984) which are in line with our present findings.

The isolated worms from the rumen were light pink in colour. Anterior sucker was present at the tip of the cone (Fig. 3a). Mouth portion was situated at the terminal end and was funnel shaped (Fig. 3b). Caeca was serpentine in shape (Fig. 3c), clusters of vitelline glands extended from pharynx, reached up to posterior sucker, lie between caeca and lateral margin of the body (Fig. 3c), and intestinal caeca reached up to anterior end of ventral sucker (Fig. 3d). The intact worms were pear shaped, concave ventrally with a large sub-terminal sucker (Fig. 3e) and mean length and breadth were  $428.55 \pm 11.81 \mu\text{M}$  (range 380-530  $\mu\text{M}$ ) and  $126. \pm 2.14 \text{ } 25 \mu\text{M}$  (110-140  $\mu\text{M}$ ), respectively. These morphological features were indistinguishable from *P. cervi* (Soulsby 1982) which has been depicted as the most common amphistome species of goat reported from mainland of India (Dutt 1980).

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