



NOTE

Parasitology

Trematode infections in farm animals and their vector snails in Saint Martin's Island, the southeastern offshore area of Bangladesh in the Bay of Bengal

M. Golam YASIN¹⁾, Md. Abdul ALIM^{1)*}, Anisuzzaman¹⁾, Syed Ali AHASAN²⁾, Md. Nuruzzaman MUNSI³⁾, Emdadul Haque CHOWDHURY²⁾, Takeshi HATTA⁴⁾, Naotoshi TSUJI^{4)*} and Md. Motahar Hussain MONDAL¹⁾

¹⁾Department of Parasitology, Faculty of Veterinary Science, Bangladesh Agricultural University, Mymensingh-2202, Bangladesh

²⁾Department of Pathology, Faculty of Veterinary Science, Bangladesh Agricultural University, Mymensingh-2202, Bangladesh

³⁾Goat and Sheep Production Research Division, Bangladesh Livestock Research Institute, Savar, Dhaka-1341, Bangladesh

⁴⁾Departemnt of Parasitology, Kitasato University School of Medicine, Kitasato, Sagamihara, Kanagawa 252-0374, Japan

ABSTRACT. The prevalence of snail-borne trematode (SBT) infections in farm animals on the offshore Saint (St.) Martin's Island of Bangladesh were 68.9% for cattle, 76.7% for buffaloes, 56.3% for goats, respectively. Examination of fecal samples showed that paramphistomes infection was the most common at 50.5% followed by schistosomes at 23.7% and *Fasciola* at 2.3%. *Fasciola* infection was found in cattle (1.9%) and buffaloes (16.7%) but not in goats. Schistosome infection in cattle, buffaloes and goats were 31.1, 6.7 and 17.5%, respectively. Prevalence of SBTs was higher in older animals. *Thiara tuberculata* (*Melanoides tuberculata*) were found to serve as vector for paramphistomes and *Indoplanorbis exustus* for schistosomes and paramphistomes, respectively. Our results suggest that SBT and their vector snails are highly endemic on St. Martin's Island of Bangladesh, and proper attention is needed to control these infections.

KEY WORDS: farm animal, prevalence, snail, St. Martin's Island, trematodes

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Snail-borne trematodes (SBTs) (Trematoda: Digenea) infect humans and animals since pre-historic time and exert deleterious effects. They may also induce life threatening cancer leading to death. SBT diseases namely fascioliasis, paramphistomiasis and schistosomiasis greatly hamper profitable livestock production in many countries of the world, especially in the humid tropics and sub-tropics, including Bangladesh [2, 12, 18, 28, 33]. The losses are in terms of reduced milk and meat production, poor weight gain, infertility, loss of vigor, and increased susceptibility to many diseases and mortality [26].

Considerable attention has been paid to clinical parasitism due to SBTs worldwide because of its severity [12, 18, 26]. Several studies have been conducted to describe the trematode infections in livestock in the mainland of Bangladesh [2, 4, 24, 28, 31]. The southeastern Saint (St.) Martin's Island in the Bay of Bengal is geo-climatically distinct and different from the mainland of Bangladesh. Human settlement started there about two hundred and fifty years ago [7]. There are 350 cattle heads, 30 buffaloes, 669 goats on the Island which are reared for draft power, milk and meat purposes. It is assumed that the animals of this Island are also suffering from different parasitic diseases. However, no attention had been paid in investigating the parasitic infections in farm animals on this Island. Therefore, this cross-sectional study was performed from November, 2014 to January, 2015 to determine the prevalence of SBT infections (fascioliasis, paramphistomiasis and schistosomiasis) in cattle, buffaloes and goats as well as to detect the vector snails on the St. Martin's Island.

We calculated sample size using the formula, $n = \frac{Z^2 P(1-n)}{d^2}$

Where, n=sample size, Z=Z statistics for level of confidence, P=expected prevalence, d=precession [10]. The sample size was estimated as 385 at 95% level of confidence and 5% precession. However, we randomly collected and examined a total of 396 fecal samples of farm animals from the Island that included 206 cattle (*Bos indicus*), 160 goats (*Capra hircus*) and all 30 buffaloes

*Correspondence to: Tsuji, N.: tsujin@med.kitasato-u.ac.jp, Alim, M. A.: alim20471@yahoo.com

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(*Bubalus bubalis*). The prevalence of SBT infections in relation to the age and sex of animals was investigated and for that purpose only the cattle samples were taken into consideration. Cattle were also grouped into calves (≤ 1 year of age, $n=28$), yearlings (>1 year to 2 years, $n=60$) and adults (>2 years, $n=118$). The age of the cattle was determined by examining the teeth and questioning the farmers or from the record book.

Fresh fecal samples were collected directly from the rectum of each animal using gloves. In some incidences, samples were collected from the top of freshly voided fecal mass. About 20–25 g of feces was collected in a polythene bag from each animal. Few drops of 10% formalin were added to each sample. During fecal sample collection the relevant information such as age, sex and deworming of the animals and molluscicide use schedule, were recorded carefully. The fecal samples were examined by simple sedimentation technique [34]. Briefly, 10 g of feces was weighed and taken into a beaker containing 100 ml of normal saline. A homogenous suspension was made and strained through a sieve (50 mesh). The filtrate was left to stand for 1 hr, and then the supernatant was discarded keeping ~1 ml sediment. After stirring the sediment, a few drops were taken on a glass slide, covered with cover slip and examined under compound microscope (LABOMED, Los Angeles, CA, U.S.A.) using 10 \times objective. The sediment was examined in triplicates. We analyzed data using SPSS program and level of significance were determined using χ^2 test [15].

Coprological examination showed that the SBT infections were prevalent in farm animals in the St. Martin's Island. Morphological identification of the eggs revealed infection with *Fasciola* sp., paramphistomes and *Schistosoma* spp. (*Schistosoma indicum* and *Schistosoma spindale*) (Fig. 1a–d). As this investigation was carried out on live animals and identification of the parasites was based on their eggs, it was not possible to distinguish the different species of the family Paramphistomatidae and the parasites were considered as paramphistomes. The study revealed that 64.4% farm animals were infected with SBTs (Fig. 2A). We observed that prevalence of paramphistomiasis was the highest (50.5%) while that of fascioliasis was the lowest (2.3%) (Fig. 2B). Among the three farm animal species examined, the highest prevalence of SBT infections was detected in buffaloes (76.7%) (Fig. 3). Great variations in distribution of infections with different SBT species were noticed in the farm animals. The highest prevalence of paramphistomiasis (73.3%) and fascioliasis (16.7%) were detected in buffaloes (Fig. 3). Higher prevalence of schistosomiasis (17.5%) was found in goats. Interestingly, in goats no *Fasciola* infection could be detected (Table 1) although fascioliasis in goats is endemic in the mainland of Bangladesh [16]. During this study, we detected mixed infections with *Fasciola* and paramphistomes in 1.5% of cattle and 6.7% of buffaloes, however, mixed infections with paramphistomes and *Schistosoma* spp. were detected in 11.5% of cattle, 10.0% of buffaloes and 5.0% of goats. It is worth mentioning that the farmers never use any chemical or herbal anthelmintic against parasitic diseases.

Detection of SBT infections in approximately two-third of the farm animals indicates a horrific scenario of health of the farm animals reared in the off shore St. Martin's Island. Previously, STB infections were found in 26.2–72.9% of ruminants [2, 30, 35] in the mainland of Bangladesh. Interestingly, in the present study very few animals (2.3%) were found infected with fascioliasis. However, the previous report showed that fascioliasis was found in 50.65 and 73.58% of buffaloes through fecal examination and post-mortem investigation, respectively from different areas of mainland of Bangladesh [2]. Other reports described fascioliasis in 26–39% of farm animals in Bangladesh [4, 24, 30, 31], in 11–15% of cattle in India [14, 36], in 25–27% of cattle in Pakistan [20] and in 25–48% of cattle in Indonesia [9]. On the St. Martin's Island, we could not find lymnaeid snails which are the established intermediate hosts of *F. gigantica* in Bangladesh and India [3, 6, 19]. Detection of the highest prevalence of paramphistomiasis in the farm animals is in accord with several previous reports (34.5–72.9%) in ruminants from the mainland of Bangladesh [4, 30, 31, 35]. Higher prevalence of paramphistomiasis has also been recorded in goats (51.8%) in neighboring West Bengal, India [25]. Infection in cattle with paramphistomes has been reported to be 8.88% in Andaman and Nicobar Island, India [18].

We detected fairly higher prevalence of schistosomiasis in cattle in the Island, which is in accord with the results reported by Sardar *et al.* [31] at Mymensingh (29.44%) in Bangladesh. However, Chowdhury *et al.* [8] reported much lower prevalence of schistosomiasis in cattle (5.9%) in Savar, Bangladesh. Schistosome cercariae penetrate the skin of animals while they graze in water. Because of their grazing habit and wallowing nature, the buffaloes are likely to be more exposed to *Schistosoma* infections. However, we detected the lowest prevalence of schistosomiasis in buffaloes and, our results are substantiated by the previous findings [4, 24, 30]. The results indicate that buffaloes have inherent genetic properties to combat schistosomiasis or resistance to schistosomiasis develops in buffaloes following initial exposure. A protective immune response has been described in water buffaloes against *Schistosoma japonicum* in experimentally infected cases or immunized with DNA and recombinant vaccines [21, 27].

The results revealed that overall SBT infection was a little higher in female cattle (70.1%) than in the males (65.4%) but the difference was not statistically significant. The female cattle were found to be more susceptible to infection with paramphistomes and *Schistosoma* spp. than the male. However, no difference in the prevalence of fascioliasis was detected among the sex groups (Table 2).

Although sex of the hosts did not significantly influenced the occurrence of SBTs but age had a great impact on the prevalence of

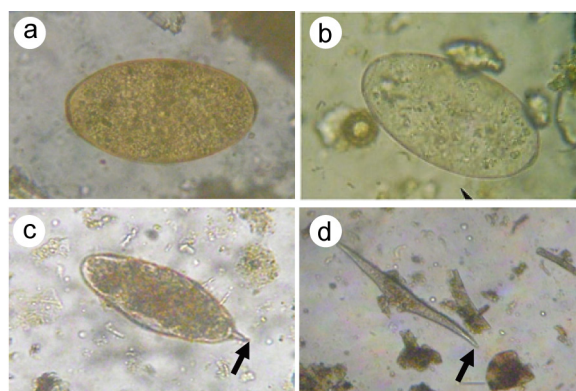


Fig. 1. Eggs of SBTs identified. Fresh fecal samples were collected and examined using simple sedimentation method and the SBT eggs were identified. Egg of (a) *Fasciola* sp.; (b) Paramphistomes; (c) *Schistosoma indicum*, arrow indicates the terminal spine; (d) *S. spindale*, arrow indicates the terminal spine.

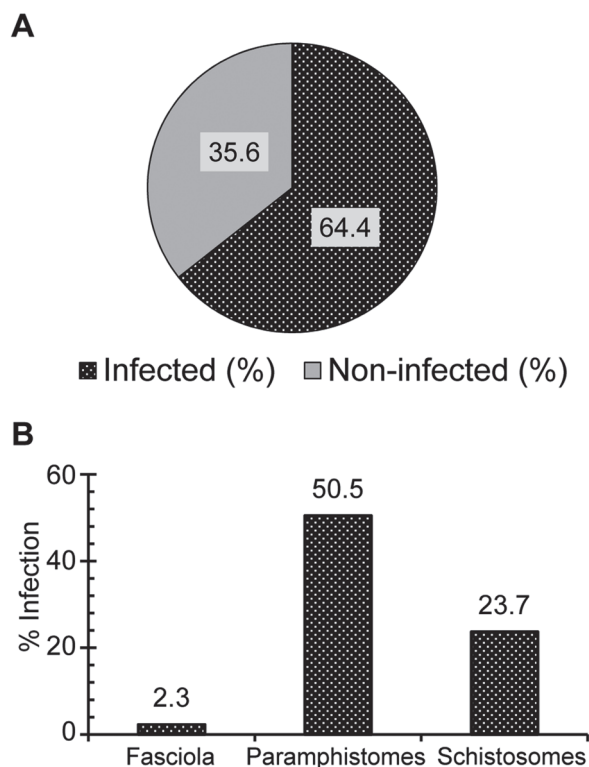


Fig. 2. SBT infections in St. Martin's Island. Fresh fecal samples were collected, examined and eggs were identified. (A) Overall prevalence of SBTs (B) Prevalence of different SBTs.

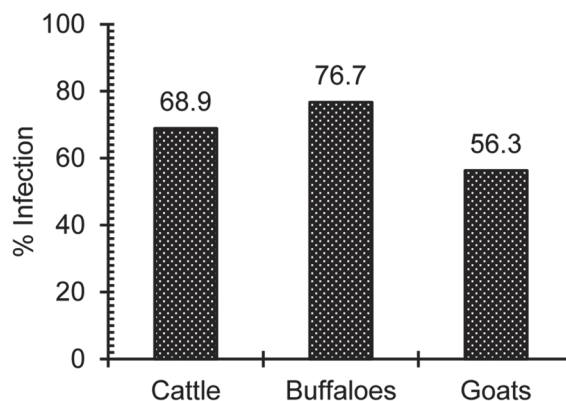


Fig. 3. Farm animals affected with SBTs. Fresh fecal samples were collected from different species of farm animals, examined and prevalence of SBT infections was identified.

the infections. The prevalence of SBT infections varied at different ages of cattle and the lowest overall prevalence was recorded in calves of ≤ 1 year of age (35.7%). It was observed that prevalence of the SBTs increased as the animals grew older with the highest prevalence in the adult cattle. Among the SBT species, the prevalence of paramphistomiasis was the highest in cattle of all age groups and no *Fasciola* infection could be detected in calves (Table 2). This can be explained by the fact of more exposure of the adult cattle to infection as the calves and the yearling are usually less grazed in the fields and marshy land compared to the adults.

The sources of freshwater on the Island such as man-made ponds and canals were extensively searched. The inhabitants of the Island never use any molluscicide in the water bodies. A total of 367 snails of different species were collected by hand picking or with the help of a scooped-net. The snails were washed thoroughly in running tap water for 15 min to remove the external derbies from their shells. The snails were brought to the laboratory and identified by their shell characteristics as described previously [5, 23]. For detection of cercarial infection, the snails were examined as described earlier [3, 23]. In brief, the snails were placed individually in test tubes or glass Petri dishes containing water and were exposed to artificial light (30°C) for emergence of cercariae. The snails, from which cercariae did not emerge following exposure to light, were individually crushed with the help of pestle and mortar. The entire alimentary tract and the associated glands were placed in normal saline, dissected and examined under a stereomicroscope (Leica, China). In positive cases, the cercariae were drawn with the help of a medicinal dropper and placed on glass slide. One drop of 1% methylene blue was added to the cercaria and studied under compound microscope using high power objective (40 \times). Cercariae were identified by their distinct morphological features as described previously [11, 29, 32].

The sources of freshwater on the Island such as

Of the 367 freshwater snails collected and examined from different man-made ponds (Table 3), 114 were *Indoplanorbis exustus*, 99 were *Thiara tuberculata* (*Melanooides tuberculata*), 90 were *Viviparous bengalensis*, and 64 were *Pila globosa* (Fig. 4). We could not detect any lymnaeid snails, on the Island which are the well established intermediate hosts of *Fasciola* spp. in Bangladesh and India [3, 6, 19]. On examination, amphistome cercariae (Fig. 5a), characterized by the presence of large ventral sucker which is very close to posterior extremity, were detected from *I. exustus* (15.8%) and *T. tuberculata* (*M. tuberculata*) (12.1%) snails. Furcocercus cercariae (Fig. 5b) were detected in *I. exustus* (2.6%) only. Importantly, no cercarial infection could be detected in *V. bengalensis* and *P. globosa*. It was found for the first time that *T. tuberculata* snails acted as intermediate hosts of amphistomes in Bangladesh. We observed that the snail habitats were the small stagnant water bodies (man-made ponds) with various aquatic vegetations predominated by water hyacinth (*Eichhornia crassipes*). The pH of water ranged from 6.8–7.2. Rahman *et al.* [29] detected furcocercus and amphistome cercariae from *I. exustus* and described this snail species as the vector of *Schistosoma* and amphistomes in mainland of Bangladesh. Liu *et al.* [22] reported that *I. exustus* is the most important intermediate host for *S. spindale*, as well as for *S. indicum* in certain regions of Asia. Farahnak *et al.* [13] described that *Schistosoma* dermatitis was transmitted by *T. tuberculata* snails in Iran. We detected very high prevalence of paramphistomiasis in animals which is obviously due to the presence of *I. exustus* and *T. tuberculata* intermediate hosts in the St. Martin's Island. The lymnaeid snails are very sensitive to water pH, turbidity, water current and soil quality. They prefer clear slow flowing water bodies in clay or in loamy clay or sandy clay types of soil with sparse vegetations [3]. Our investigation revealed that the soil quality of St. Martin's Island is sandy. Possibly the soil type, dense vegetation and stagnant condition of the water bodies has made the St. Martin's Island

Table 1. Prevalence of snail-borne trematode infections in farm animals

Animal species	No. of animals examined	No. and (%) of animals infected with			
		<i>Fasciola</i> sp.	Paramphistomes	<i>Schistosoma</i> spp.	Subtotal
Cattle	206	4 (1.9)	108 (52.4)	64 (31.1)	142 (68.9)
Buffaloes	30	5 (16.7)	22 (73.3)	2 (6.7)	23 (76.7)
Goats	160	0 (0.0)	70 (43.8)	28 (17.5)	90 (56.3)

Table 2. Prevalence of snail-borne trematode infections in different sexes and age groups of cattle

Parasites	Sex group Infected (%)		Age group Infected (%)		
	Male (n=52)	Female (n=154)	Calves (n=28)	Yearlings (n=60)	Adults (n=118)
<i>Fasciola</i> sp.	1 (1.9)	3 (1.9)	- (-)	1 (1.7)	3 (2.5)
Paramphistomes	24 (46.2)	84 (54.5)	6 (21.4)	32 (53.3)	70 (59.3)
<i>Schistosoma</i> spp.	13 (25.0)	51 (33.1)	4 (14.3)	18 (30.0)	42 (35.6)
Subtotal	34 (65.4) ^{a)}	108 (70.1) ^{a)}	10 (35.7) ^{a)}	40 (66.7) ^{b)}	92 (78.0) ^{b)}

Different superscripts (a, b) in the same group (age group) are statistically significant ($P < 0.01$). The same superscripts (a, a or b, b) in the sex or age group (yearling and adults) are statistically insignificant.

Table 3. Prevalence of cercariae in freshwater snails in St. Martin's Island

Snail species	No. of snails examined	No. and (%) of snails infected with	
		Amphistomes cercariae	Furcocercus cercariae
<i>Indoplanorbis exustus</i>	114	18 (15.8)	2 (2.6)
<i>Thiara tuberculata</i>	99	12 (12.1)	0 (0)
<i>Vivipara bengalensis</i>	90	0 (0)	0 (0)
<i>Pila globosa</i>	64	0 (0)	0 (0)

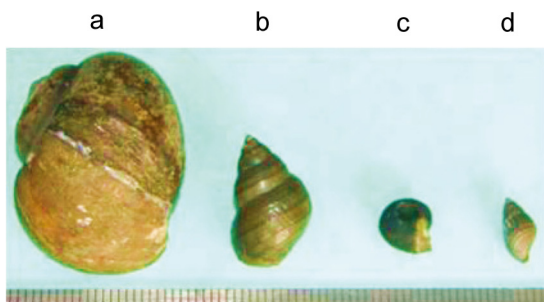


Fig. 4. Vector snails identified from St. Martin's Island. Snails of different species were collected, washed thoroughly and identified by their morphological features. (a) *P. globosa*, (b) *V. bengalensis*, (c) *I. exustus* and (d) *T. tuberculata*. One unit of the scale is 1.6 mm.



Fig. 5. Cercariae recovered from vector snails. Snails were collected, washed and exposed to artificial light (30°C) for emergence of cercariae. Released cercariae were identified at 40X objective adding one drop of 1% methylene blue. (a) Amphistome cercaria, (b) Furcocercus cercaria.

an unsuitable habitat for lymnaeid snails. Detection of fascioliasis in cattle and buffaloes in absence of lymnaeid snails on the St. Martin's Island is puzzling. Literatures suggest that *Fasciola* may also be transmitted by the *I. exustus* and *T. tuberculata* [1, 17]. Although we could not detect gymnocephalus cercariae in the snails during our investigation, but *I. exustus* or other snails might be the possible vectors for *Fasciola* in the Island which needs to be elucidated.

In conclusions, our study revealed that, like in the mainland, the SBT infections and their vector snails are endemic in the off shore St. Martin's Island of Bangladesh. This study also highlights that with the human settlement, the vector snails and the SBT infections move even in the off shore islands areas. Proper attention should be paid to control those parasites and vector snails.

CONFLICTS OF INTEREST. The authors declare that they have no conflicts of interest.

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