Occurrence of helminth parasites and their developmental stages in snails from different regions of Bangladesh

Ph D Thesis



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Registration No. 57

Session: 2011-12 (Re)

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Occurrence of helminth parasites and their developmental stages in snails from different regions of Bangladesh

(Revised)

A dissertation submitted to the University of Dhaka in partial fulfillment of the requirements for the degree of Doctor of Philosophy in Zoology (Parasitology)



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Dedicated to my beloved brother	in law Late Biswajit Roy

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I hereby declare that this dissertation submitted to the University of Dhaka for

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Farhana Zaman, Department of Zoology, University of Dhaka.

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CERTIFICATE

This is to certify that the dissertation entitled 'Occurrence of helminth parasites and their developmental stages in snails from different regions of Bangladesh' submitted by Shuvra Roy for the degree of Doctor of Philosophy in Zoology (Parasitology branch), University of Dhaka, Bangladesh embodies the record of original investigation carried out by her under our supervision.

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ABSTRCT

Helminth infection study of freshwater molluscs in and around Dhaka, Khulna and Kishoreganj region was conducted at twenty three locations which include eleven from Dhaka region, six from Khulna region and another six from Kishoreganj region from different ponds during July 2011 to June 2013 on a once in a month basis. A total of three species of freshwater molluscs belonging to two families that comprises of Viviparidae, Ampullariidae and three genera (*Bellamya*, *Pila*, *Brotia*) were found in the pond eco-systems. Physico-chemical parameters like pH, temperature, dissolved oxygen, electrical conductivity and its variation at all the collection sites were analyzed. As the molluscs are intermediatory hosts between the parasites and animals, an overview study was also carried out on the known molluscs' various parasites and their diseases.

Bangladesh has been virtually unexplored with respect to its trematode fauna, the sampled freshwater snails from temporary and permanent ponds were collected. Helminth infections of 3 snails species, viz., *Bellamya bengalensis*, *Brotia costula* and *Pila globosa*. Samplings were done primarily on monthly basis. 31,717 snails (*B. bengalensis*, 10,915; *B. costula*, 9,961; *P. globosa*, 10,841) were autopsied, of which 4,099 were infected by helminthes (12.92%). The infection was eight types of cercariae (belonging seventeen species), one nematode adult morpho-type, two Cestodes, one Pentastomida and one Turbellaria. Among the hosts, prevalence of trematode infection was highest in *B.bengalensis* (14.7%), followed by *P. globosa* (12.95%) and *B. costula* (10.95%). Contrary to the general knowledge, infection of snails by nematode parasites, though rather poor in diversity, was higher (1.24-4.55%) than that by the DST (12.92%). Seasonally, DST was more prevalent in the rainy season, whereas nematodes were during the summer. With respect to parasites harboured, *Pila globosa* was the most diversified refusing all the 3 infector groups.

Altogether screened 31,717 individuals of three freshwater snail species (*Bellamya bengalensis*, *Brotia costula and Pila globosa*) for shedding cercariae. Collectively, eight morphologically distinguishable types of trematode cercariae were found:

Parapleurophocercous cercariae; Pleurophocercous cercariae; Xiphidiocercariae cercariae; Furcocercous cercariae; Echinostome cercariae; Amphistome cercariae; Renicolid cercariae and Cotylomicrocercous cercariae. Out of 23 locations the parasitics collected from *Pila globosa* other than trematode and nematode were two Cestodes, one Turballaria and one Pentastomida. Overall prevalence was found in August to September (1.03-2.26%) in 2011-2013.

In the present study monthly fluctuations in the cercarial prevalence were observed due to the variations in the temperature, rainfall and dissolve oxygen of each month. The occurrences of cercariae are maximum in August to September (18.18% - 28.57%) and minimum if February (3.45-8.33%) which is dealt with elevated temperature, rainfall and DO level in the Dhaka region. Whereas, In Khulna and Kishoreganj region the maximum prevalence was in August to Semtember (14.29% - 37.5%) and no infection was found in some months. It is made clear from the results that more number of species related with low DO content. It can be inferred from the above points that DO content plays a vital role in affecting the population dynamics of a particular species than the other physico-chemical factors like pH, temperature and EC. Moreover the molluscan population is abundant in a place where the aquatic plants are available in diverse forms. Since there is not much variation in pH, temperature, EC and salinity in the collection sites, they do not have much influence in the availability of specimens in this present study. Through this analysis, it is inferred that the possibility of variation in pH, temperature, EC can play a vital role in the life cycle of any plant or animal group and when it is moderate or in low, it cannot have much influence on the population dynamics.

In 2011-12, occurrence of trematode parasite in *Bellamya* was significantly higher than that of *Brotia* and *Pila* (p<0.05). Moreover, in 2012-13 occurrence of trematode parasite in *Bellamya* was significantly higher than that of *Brotia* and *Pila*. On the other hand, occurrence of this parasite in *Bellamya* found in 2012-13 was significantly higher than that of 2011-12. No significant different was found in *Brotia* and *Pila* in this two years (p>0.05).

The prevalence of trematode parasites in *Bellamya*, *Brotia* and *Pila* in 2011-2012 and 2012-2013 sampled from 23 different regions were determined. Each year was divided into three different seasons (Rainy, winter and summer) where the incidence of the parasites in rainy season in most of the cases was significantly higher than that of other seasons. However, the prevalence in summer (in most of the regions) was significantly higher than that of winter.

The overall natural environmental parameters varied within overlapping ranges among the ponds. Water temperature ranged between 4.7° C and 38° C depending on the day and time of collection. The hydrogen ion concentration was within the range providing protection for the aquatic life (7.2 to 9.8). The concentration of dissolved oxygen showed generally low in some locations due to pollution load (Dhaka region) and higher (> 7.0 mg/l) levels and exhibited a strong negative association with water conductivity (r_s = -0.665, p < 0.05). The data identified the correlation between temperature, pH, EC, DO, rainfall and prevalence of parasites in snails of three different regions in 2011-12 and 2012-13 academic years. Sometimes positive correlation with significant level (p>0.05) and without significant level were observed. Significantly positive correlation was found between temperature and prevalence of parasites in snails of Dhaka regions in 2011-12 (p<0.01).Positive correlation was also found between % occurrence and pH, EC, DO and rainfall, but was not significant (p>0.05).

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Positive correlation was found between temperature, EC (p<0.05), rainfall and prevalence of parasitesin snails of Kishoreganj regions in 2012-13 (p<0.01). Positive correlation was also found between % occurrence and pH, but was not significant (p>0.05). Also the positive correlation was found between temperature, EC (p<0.05), rainfall and prevalence of parasites in snails of Kishoreganj regions in 2012-13 (p<0.01). Positive correlation was also found between % occurrence and pH, but was not significant (p>0.05).

CONTENTS

	Page no.
Abstract	i-iii
Chapter 1: Introduction	1-26
Chapter 2: Review of Literature	27-51
Chapter 3: Materials and Methods	52-68
Chapter 4: Systematic and Taxonomy of infected cercaria	69-92
Chapter 5: Results and Observation	93-157
Chapter 6: Discussion	158-171
Chapter 7: Summary and Conclusion	172-177
Chapter 8: References	178-210
APPENDIX	211-255

INTRODUCTION

Mollusca are the second largest phylum of the animal kingdom, forming a major part of the world fauna. The Gastropoda is the only class of mollusks which have successfully invaded land. They are one of the most diverse groups of animals, both in shape and habit. The Phylum Mollusca is probably the third most important animal group after the arthropods and vertebrates (South, 1992). Snails and slugs belong to the class Gastropoda. Snails and slugs are mollusks, a group of invertebrate animals with soft unsegmented bodies. Slugs are often described as snails without a shell, while snail bodies are enclosed in calcareous shells (Barker, 2001; Ramzy, 2009).

Snails belong to a large and highly diverse group of invertebrate to the phylum - *Mollusca*, class - *Gastropoda* and order - *Stylommatophpra*. It acts as intermediate host of different trematode parasites, in which several developing larval stages such as sporocysts, rediae and cercariae are set up (Elsheikha and Elshazly, 2008). The proportion of snails that release cercariae (prevalence of infection) and the number of cercariae released from each infected snail (intensity of infection) play important roles in the transmission of trematodes from the snail host. Only *Lymnaea* group of snails is involved in establishing of life cycle in at least 71 species of trematodes (Soldanova *et al.*, 2010). Other species of snails also transmit various trematode parasites of livestock and birds. For example, *Indoplanorbis exustus* is responsible for the transmission of *Schistosoma nasale*, *Schistosoma spindale* and *Schistosoma indicum* as well as other trematodes such as *Echinostoma* spp. and some spirorchids (Liu *et al.*, 2010). Age and size of snails, light conditions, temperature ranges, depth of water are some of the factors that appear to affect the prevalence and intensity of digenetic trematode infections in the snail intermediate hosts (Fingerut *et al.*, 2003; Graham, 2003).

Trematode parasites share an intimate relationship with their gastropod intermediate hosts, which act as the vehicle for their development and transmission. They represent an enormous economic and medical burden in developing countries, stimulating much study of snail-trematode interactions. Laboratory- maintained snail-trematode systems and in vitro cell cultures are being

used to investigate the molecular dialogue between host and parasite. These dynamic and finally balanced antagonistic relationships, in which parasites strongly influence the physiology of the host, are highly specific and may occasionally demonstrate co-speciation. We consider the mechanisms and responses developed by trematodes and snails that result in compatibility or rejection of the parasite, and the macro evolutionary implication that they may effect. Although for gastropods the fossil record gives some insight into evolutionary history, elucidation of trematode evolution must rely largely upon molecular approaches and for both, such techniques have provided fresh and often surprising evidence of their origins and dispersal over time. Co-evolution of snails and trematodes is becoming increasingly apparent at both cellular and population levels; the implications of which are only beginning to be understood for disease control. Untangling the complex interactions of trematodes and snails promise opportunities for intervention to relieve the burden of parasitic disease (Lockyer, et al., 2003).

Parasites influence the life-history of the hosts. Parasite transmission determines the distribution of parasites in host populations. Trematodes are ubiquitous parasitic worms that infect a wide range of vertebrate and invertebrate animal and almost always use a mollusc as the first intermediate host. The adult parasite resides in a vertebrate host and releases eggs into the aquatic habitat. The first larval stage, miracidium, hatches from the egg and infects a mollusc. The 1st intermediate host or primary infection is where the parasite undergoes a series of asexual cycles eventually producing embryos that develop into cercariae. Cercariae are free –swimming stages which emerge from the mollusc to infect another vertebrate or invertebrate, which may act either as the definitive host (vertebrates only) where the adult parasite develops or as a second intermediate host where an intermediary stage, metacercaria, resides until transmission, usually via predation, to the definitive vertebrate host occurs (Graham, 2003).

Trematodes in mollusks are known to have wide-ranging effects on the physiology, immunology, ecology and endocrinology of their molluscan hosts (Thompson, 1997; Morley, 2006). It is also common for mollusc to act as second intermediate hosts for trematode metacercariae, which are typically less host specific and can cause increased mortality (Schneck and Fried, 2004), reduced growth (Theiltges, 2006), and altered behaviour (Babirat *et al.*, 2004) in infected individuals. The extensive effects trematode parasitism can cause to their molluscan hosts makes them an important natural stressor that of growth, loss of weight, emaciation and can impact on

molluscan communities, especially those exposed to polluted conditions (Morley *et al.*, 2003). As such, their status in field populations needs to be properly assessed so that ecological laboratory studies on molluscan model species may more accurately reflect natural conditions. Heavy infections with either larvae or adult trematodes are usually highly damaging to the host, resulting in retardation and unhealthy condition often leading to death (Hyman 1951, Theron and Mone, 1984).

In Bangladesh, very few studies have been conducted on the occurrence of helminth infections in snails. The works of Rahman and Jahan (1999, 2001, 2002, 2004 and 2006) dealt with cercarial types and their prevalence, abundance and intensity, seasonal influence on prevalence, abundance and intensity and biology of amphistome cercaria and effects on different organs and the fecundities of some freshwater gastropods in Bangladesh. Most of the early investigations on the growth of digeneans pertain to the growth of adult flukes from metacercariae either in experimental hosts or in culture media (Fried, 1962; Nollen, 1971; Hechinger and Lafferty, 2005; Franco *et al.*, 1986 and Ismail *et al.*, 2003). A few invastigations have also been undertaken on the growth of metacercariae from cercariae in the intermediate host (Benjamin and James, 1987).

Trematodes are of extreme economic importance, especially in tropical developing countries. There is considerable economic loss in the form of mortality, reduced weight gain, decreased meat and milk production, low fertility, reduction in draft power and poor hide quality in Bangladesh due to fascioliasis in ruminants (Begum, 1993). An estimation of annual economic loss of about 0.6 million taka from liver damage due to fascioliasis in goats was reported by Bhuiyan (1970). Besides, some trematodes cercariae produce "swimmer's itch" (cercarial dermatitis) in man (Sandosham, 1954). The freshwater snails are involved in the transmission of trematode species belonging to the superfamilies Schistosomatoidea, Fascioloidea, Clinostomo idea, Paramphistomoidea, Echinostomatoidea, Diplostomoidea and Pronocephaloidea.

Freshwater snails play a key role in the life cycle of trematodes. They serve not only as a source of food and a place of reproduction for these parasites but also as a means of transport which the parasite can get to its next host (Lockyer *et al.* 2004). The uniqueness of the relationship between these organisms derives from the fact that the majority of the 18,000 known species of trematodes exploit snails as the mandatory first intermediate host (Cribbs, *et al.*, 2001).

Many researchers have no doubt that these parasites were originally linked only with molluscs, and only as a consequence of evolutionary development did they include in their life cycle a further group of hosts (Pojmańska and Grabda-Kazubska, 1985; Cribb *et al.*, 2001). The intramolluscan development of trematodes begins with the swallowing of the eggs of the parasite or the active penetration by the miracidium. In both cases, the ciliated larva casts off the ciliated epithelium in the body of the snail, metamorphosing into the next stadium—mother sporocyst or redia. The remaining larvae can generate daughter stage sporocysts in which tens or even hundreds of thousands of cercariae are ultimately formed, successively leaving the snail's body, with the duration of the release of the cercariae depending not only on the species of parasite but also the health of the host (Wright, 1971).

Cercariae, depending on the species, transform into the next stage, metacercariae, or penetrate directly into the definitive host. Metacercariae encyst in the external environment or inside the second intermediate host, which can be either vertebrates or invertebrates—including snails (Furmaga, 1968; Wright, 1971). Cercariae of some species of trematodes can transform into metacercariae in the snails in which they were formed (Styczyńska-Jurewicz, 1962) or after penetrating into other individuals of the same or another species, usually locating themselves on the lining of the mantle cavity, pericardial cavity and kidney lumen (Adam and Lewis, 1992; Skirnisson *et al.*, 2004). Host snails in which cercariae or metacercariae appear are an important link in the distribution of trematodes in vertebrate populations, including farm animals and humans. This fact was decisive in initiating research on the occurrence of trematodes in snail populations in the early years of the twentieth century. Later, veterinary and epidemiological aspects of trematode invasion of snails were joined by further aspects, including the impact of parasites on the biology of the hosts and the dynamics of their populations.

Cercariae, the larva stage of flukes may be found in fresh water and terrestrial gastropods (Ito, 1980). There are reports showing that snails in the family Thiaridae serve as the first intermediate host for trematodes. For instance, *Tarebia granifera*. *Thiara toucheana*, *Brotiaasperata*, *B. costula episcopalism and B. c. peninsularis* are the first intermediate hosts for lung flukes (Tang, 1940; Tubangui *et al.*, 1950; Davis, 1971; Brandt, 1974).

The growth of a digenean is followed from cercaria to adult via the metacercaria employing cercariae released by a single infected snail. The large numbers of adult flukes collected from natural and experimental infections of frogs has also permitted a study of allometry and intraspecific variations of the flukes and an examination of the validity of species described in the genus.

Most cercariae of *Fasciola hepatica* swim several minutes after leaving their snail host and then attach to a plant, where they transform into metacercariae (Leuckart, 1882; Thomas, 1883). However, live cercariae are present in the mollusc when it dies after the last cercarial shedding. Histological examination demonstrated the presence of different stages of degenerating cercariae. They were rather numerous and were sometimes found to have fused protective layers. Internal metacercarial cysts were also found in rather low numbers. There is no mention of these latter cysts' characteristics in the literature.

Many species of freshwater snails have been incriminated as intermediate hosts of parasitic diseases such as fascioliasis, amphistomiasis, schistosomiasis etc. (Cheesbrough, 1987; Schmidt, 1990). Indiscriminate use of molluscicides for the control of freshwater snails leads to persistence of the molluscicides in the aquatic habitat and toxicity to non-target organisms, which ultimately leads to the disturbance of the ecosystem. Various biological control agents such as the competitor snails- *Marisa cornuaritis* (L.) and *Helisoma durai* whether by and various species of sciomyzid flies have been recommended for the control of vector snails as a part of integrated pest management (WHO, 1993).

The larval trematode stages developed a series of physiological adaptations as a result of living in the hostile environment of their intermediate hosts. In this restricted milieu, the parasite depends on the host for nutrient acquisition and survival, resulting in a close physiological and metabolic relationship between both parasite and host. Graczyk, *et al.*, (2001) stated that the parasitism is exclusively dependent on the degree of physiological integration between these organisms and physiological synchronization of their life cycle.

The trematode larvae (miracidium stage) must gain access into the snail host while evading its defense mechanisms (Becker, 1994). If the miracidia succeed to establish infection, the miracidium transforms into a mother sporocyst (Bayne and Yoshino, 1989). Intense growth and asexual reproduction are observed, and the host organism must provide all nutrients needed to sustain these processes, resulting in consumption of its own deposits. As a consequence, the infected snail must evolve adaptations to allow it to maintain the intense output of larval stages and its own demand on the nutrients. The presence of parasite derived excretory–secretory products released in the body of the snail brings also creates new pressures on the host, which must degrade and eliminate these highly toxic molecules.

Life cycle and developmental stages of Trematoda

The trematodes are parasitic flatworms of great medical and veterinary importance. An understanding of the evolution of trematodes depends on an interpretation of their complex and diverse life cycles. It is the life cycles in general and the stages that comprise these cycles that are the focus of the detailed analysis presented herein. It contains a broad scope of modern information on digenetic trematodes, from descriptions of their morphology and development to their behaviour and the structure of their population groups.

Host-parasite systems often are considered to be interesting but unusual examples of evolutionary processes. However, Price (1980) recently argued that parasites could be good model systems for studying general evolutionary principles. A currently contentious general evolutionary principle is the unified theory of evolution (Brooks and Wiley, 1988).

Price (1980) invoked a widespread view of evolution in his studies of parasite evolution when he asserted that the evolutionary "play" took place on a "stage" organized by the environment (an "ecological stage"). Under this view, phylogeny (evolutionary history) is the passive accumulation of the effects of environmental selection over time.

The life cycles of most trematode groups are known but developmental stages of numerous taxa still have not been elucidated. This is valid mainly for tropical regions where data on biology of trematodes are scarce (Yamaguti 1966; Williams and Jones 1994). Almost all trematodes use a mollusc (often a gastropod) as their first intermediate host, in which asexual reproduction takes

place (Yamaguti 1966; Ginetsinskaya 1988). Therefore, studies on the life histories of trematodes usually include examination of gastropods or bivalves for larval stages (i.e. sporocysts, rediae, cercariae or metacercariae). This examination may provide useful information about the occurrence of trematodes in a locality under consideration but also makes it possible to obtain developmental stages for experimental infections.

Parasites may be directly or indirectly involved in the ecology and evolution of a broad range of phenomena: host population dynamics and extinctions, maintenance of genetic diversity, sexual selection, evolution of genetic systems, and evolution of sexual recombination, to name just a few. Certainly, parasites possess features that make them very attractive as explanatory factors in the evolution and ecology of their hosts. These features include their high abundance in nearly every ecosystem, their typically narrow host range (compared with typical predators), their adverse effects on their hosts (e.g., reduced fecundity and survival), and density dependence during horizontal transmission (Price, 1980).

On the other hand, hosts are the environment for the parasites and thus define their niche. Most parasites are not viable outside of their hosts for extended periods (not considering resting stages) and therefore from the parasite's point of view-parasite and host form an inseparable biological unit. Thus, parasite ecology is closely linked to the ecology of its hosts, and the parasite's natural history is best seen in the light of its host's biology.

The life cycles of *Fasciola* sp, *Fasciolopsis* sp, *andEchinostoma* sp species are complex, requiring more than one intermediate host. Adult worms inhabit the liver or bile ducts of the definitive host (human), where they lay many eggs which are deposited into the environment in the feces. They are immature when passed; if they are passed into water they become mature in 9 to 15 days at the optimum temperature of 22-25°C.

Immature eggs are discharged in the biliary ducts and in the stool. Eggs become embryonated in water, eggs release miracidia, which invade a suitable snail intermediate host, including many species of the genus *Lymnae*. In the snail the parasites undergo several developmental stages (sporocysts, rediae, and cercariae). The cercariae are released from the snail and encyst as metacercariae on aquatic vegetation or other surfaces. Mammals acquire the infection by eating

vegetation containing metacercariae. Humans can become infected by ingesting metacercariae containing freshwater plants, especially watercress. After ingestion, the metacercariae excyst in the duodenum and migrate through the intestinal wall, the peritoneal cavity, and the liver parenchyma into the biliary ducts, where they develop into adults. In humans, maturation from metacercariae into adult flukes takes approximately 3 to 4 months. The adult flukes (*Fasciolahepatica*: up to 30 μm by 13 μm; *F. gigantica*: up to 75 μm) reside in the large biliary ducts of the mammalian host. *Fasciola hepaticas* infect various animal species, mostly herbivores.

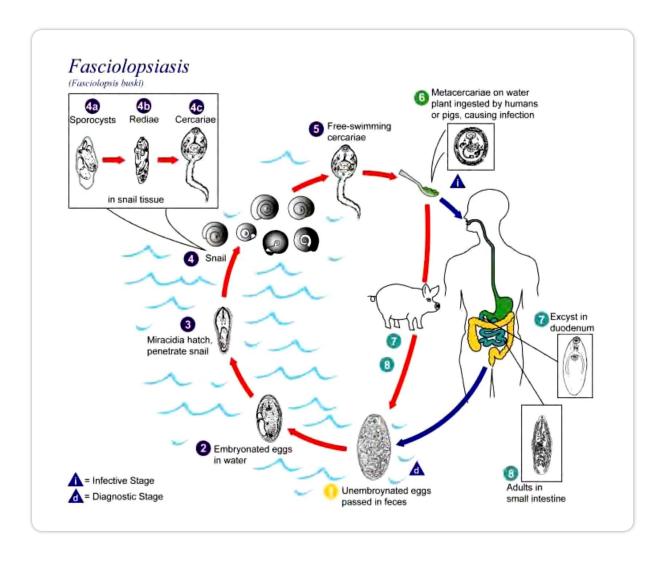


Figure 1.1: The life cycle of *Fasciolopsis buski* (WHO, 1965).

Developmental stages of Trematoda (DST)

Freshwater snails play a key role in the life cycle of trematodes. They serve not only as a source of food and a place of reproduction for these parasites but also as a means of transport (Lockyer et al. 2004). The uniqueness of the relationship between these organisms derives from the fact that the majority of the 18,000 known species of trematodes exploit snails as the mandatory first intermediate host (Littlewood and Bray 2001).

Many researchers have no doubt that these parasites were originally linked only with molluscs, and only as a consequence of evolutionary development did they include in their life cycle a further group of hosts (Pojmańska and Grabda-Kazubska 1985; Cribb et al. 2001). The intramolluscan development of trematodes begins with the swallowing of the eggs of the parasite or the active penetration by the miracidium. In both cases, the ciliated larva casts off the ciliated epithelium in the body of the snail, metamorphosing into the next stadium—mother sporocyst or redia. The remaining larvae can generate daughter stage sporocysts in which tens or even hundreds of thousands of cercariae are ultimately formed, successively leaving the snail's body, with the duration of the release of the cercariae depending not only on the species of parasite but also the health of the host (Wright 1971).

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metacercariae in the snails in which they were formed (Styczyńska-Jurewicz 1962) or after penetrating into other individuals of the same or another species, usually locating themselves on the lining of the mantle cavity, pericardial cavity and kidney lumen (Adam and Lewis 1992; Skirnisson et al. 2004). Host snails in which cercariae or metacercariae appear are an important link in the distribution of trematodes in vertebrate populations, including farm animals and humans. This fact was decisive in initiating research on the occurrence of trematodes in snail populations in the early years of the twentieth century. Later, veterinary and epidemiological aspects of trematode invasion of snails were joined by further aspects, including the impact of parasites on the biology of the hosts and the dynamics of their populations.

Environmental factors

Various environmental factors influence the behavior and physiology of parasites, including nematodes.

Temperature

It is well known that temperature influences the rate of development during embryogenesis and of free-living larval stages of parasitic nematodes (Ciordia and Bizzell, 1963; Gautam, 1982; Silverman and Campbell, 1959). However, the effects on the development of nematodes in intermediate hosts have not necessarily been clarified although the development of *Angiostrongylus cantonensis* has been briefly reported (Mackerras and Sandars, 1955).

Various environmental factors may impact the emergence of larval trematodes (cercariae) from their molluscan first intermediate hosts. For example, the effects of temperature on cercarial emergence are well documented. Many trematode species display increased emergence in response to increased temperature (Lo and Lin, 1991; El-Deeb *et al.*, 2003a; Abramson, 2004), however, decreased emergence in response to increased temperature has also been observed for *Renicola roscovita* and *Maritrema novaezealandensis*, using the molluscs *Littorina littorea* and *Zeacumantus subcarinatus*, respectively (Poulin, 2005). Temperature-mediated cercarial emergence has largely been studied in freshwater systems, with relatively little known regarding temperature effects on the emergence of marine trematodes. However, abiotic conditions in

marine environments, particularly intertidal systems, fluctuate in other ways that could affect cercarial emergence. A field study by Fingerut *et al.*, (2003) determined that tidal currents are a predictable signal that determines both submersion time of the first intermediate hosts (*Cerithidea californica*) as well as the emergence and aquatic transport of trematode larvae in a Californian intertidal estuary.

Salinity

Laboratory studies using the same host and trematode species (Himasthla rhigedana, Renicola buchanani, Euhaplorchis californensis, and Microphallid sp.) showed that cercariae emerged only if snails were totally submerged (Fingerut et al., 2003). In contrast, Mouritsen (2002a) found that water level had no effect on the emergence of Maritrema subdolum from its molluscan host (Hydrobia ulvae). Salinity is one of the most important marine environmental factors as it varies considerably in intertidal zones and estuaries while being relatively constant in open seas (Berger and Kharazova, 1997). Most marine systems have a salinity level of approximately 35 parts per thousand (Knauss, 1978). In contrast, the salinity levels of intertidal zones and estuaries can fluctuate in response to tidal levels and freshwater input. For example, intertidal mudflats often have pools of water that can experience greatly elevated salinity as water evaporates at low tide or, alternatively, decreased salinity in areas of high rainfall as freshwater input increases during low tide. While salinity clearly has the potential to influence cercarial emergence in marine environments, little is known about its importance. Mouritsen (2002a) found greater cercarial emergence at higher salinity levels but only at elevated temperatures. However, it appears that cercarial emergence generally increases with increasing salinity within a range of naturally occurring values (Rees, 1948; Sindermann, 1960; Sindermann and Farrin, 1962).

As relatively little is known about the effects of temperature on the emergence of marine cercariae and even less about the importance of water level and salinity, it is important to determine the effects of these environmental factors, particularly given that all three are likely to be impacted by global climate change (Marcogliese and Price, 1997). They examined the effects of temperature, water level, and salinity on the emergence of two marine trematode species from

a New Zealand intertidal mud snail in order to determine their relative importance both within and between trematode species.

Climate change

Global climate change is predicted to influence disease dynamics particularly for pathogens with complex life cycles (Harvell *et al.*, 2002). Many pathogens with complex life cycles have free-living infectious stages that are exposed to environmental conditions and are thus subject to the same changes experienced by their hosts (Pietrock and Marcogliese, 2003). For example, free-living trematode parasite cercariae have been shown to be vulnerable to a variety of abiotic factors (Pietrock and Marcogliese, 2003). With respect to parasitic disease in aquatic organisms, climate change is expected to have effects via alterations in temperature, water level, salinity, and pH (Marcogliese, 2001). Such changes could indirectly impact trematode transmission via host effects such as alterations in the population dynamics of molluscan first intermediate hosts (Hakalahti *et al.*, 2006) or modifications of host susceptibility to infection (Harvell *et al.*, 2002), but could also directly affect free-living cercariae. For parasites with complex life cycles involving multiplication within and transmission among various hosts, it is critical to understand the survival and rate of transmission of free-living stages in order to estimate the basic reproduction ratio (RO) of the parasite (Karvonen *et al.*, 2003).

Changes in temperature, salinity, UV light, and oxygen level can all affect the viability of nematode infective larvae in water (Thurston *et al.*, 2000; Grewal *et al.*, 2002). The level of infectivity of these larvae varies considerably among species, possibly related to foraging strategies, which incur different metabolic demands. Survival is related to declining energy reserves. Metabolic rates increase dramatically after a few weeks in water (Morley, 2010).

Natural variations and fluctuations have been shaping ecosystems over evolutionary timescales. However, changes in abiotic and biotic factors are occurring in this period of global change at unprecedented rates, scales and combinations (Vitousek, et al., 1994). These changes include a whole range of variables, well beyond climatic aspects only, which affect individual species as well as species interactions. The consideration of multiple environmental factors is of great

importance in order to more accurately address ecologically relevant consequences of these factors in general and of global change in particular.

Tides

In intertidal ecosystems, natural fluctuations of environmental factors are particularly pronounced, occurring at different spatial scales as well as various time scales. Compared to high tide, conditions at low tide may include a substantial warming of shallow water bodies, a concomitant increase in salinity due to evaporation and a direct exposure to ambient solar irradiation. Organisms in these ecosystems therefore experience a variety of challenging conditions including thermal, osmotic and ultraviolet radiation (UVR; 280-400 nm) stress (Przeslawski et al., 2005). A range of studies have investigated the effects of multiple environmental factors on intertidal or other marine organisms (Fredersdorf et al., 2009; Lenihan et al., 1999; Lotze and Worm, 2002; Przeslawski, 2005; Przeslawski et al., 2005; Russell and Phillips, 2009a; Russell and Phillips, 2009), describing complex interactions not predictable from single factor experiments. Conditions in intertidal habitats may become more extreme or stochastic due to on-going global changes, including global warming and stratospheric ozone depletion. Therefore, a better understanding of the effects of multiple environmental factors is an important step towards accounting for natural complexity, thus limiting the risk of underestimating or overestimating the ecological impacts of environmental conditions. In intertidal ecosystems, trematodes are an important and ubiquitous component linking several members of an intertidal community through their complex life cycles (Lauckner, 1984; Mouritsen and Poulin, 2002; Mouritsen and Poulin, 2010; Sousa, 1991).

Ambient condition

The transmission process of trematodes and other endohelminth parasites is dependent on usually more than one free-living larval stage, which are directly exposed to and influenced by ambient conditions (Pietrock and Marcogliese, 2003). Cercariae are the infective transmission stage leaving the first intermediate host in order to infect a second intermediate host. These cercariae are short-lived; because they do not feed, conditions encountered will determine the amount of time they stay alive until their energy reserves are used up (Lawson and Wilson, 1980).

For marine trematodes, environmental factors found to affect the survival of cercariae include temperature (Mouritsen, 2002; Studer *et al.*, 2010; Thieltges and Rick, 2006), salinity (Lei and Poulin, 2011; Studer and Poulin, 2012) and ultraviolet radiation (Studer *et al.*, 2012). A range of other environmental factors such as pH, oxygen, water depth, light or pollutants have also been shown to affect the survival and/or infectivity of the free-living stages of endohelminth parasites in aquatic ecosystems (Pietrock and Marcogliese, 2003). However, parasite transmission has received little attention in the context of more ecologically relevant experimental approaches, including also regarding the importance of biotic factors (Thieltges *etal.*, 2008). Mouritsen (2002), Koprivnikar and Poulin (2009) and Koprivnikar *et al.* (2010) have attempted to conduct experiments on the output and survival of cercariae in multi factorial designs in order to better account for the complexity of factors occurring in nature. Their research has shown that there are indeed factor interactions which could be of importance under natural settings.

UV rays

In previous studies investigating the separate effects of temperature, salinity and UVR on the transmission of the intertidal trematode *Maritrema novaezealandensis* from its first intermediate snail host (*Zeacumantus subcarinatus*) to one of its second intermediate crustacean hosts (*Paracalliope novizealandiae*), the survival of cercariae (approximately 170 µm in length including tail, Martorelli *et al.* 2004) was identified as the only step that was significantly affected by all three factors (Studer *et al.*, 2012; Studer and Poulin, 2012; Studer *et al.*, 2010). Results from these single-factor experiments indicated that cercariae died faster when exposed to either high temperatures or high UVR, and that survival of cercariae was not compromised at normal to increased salinities, but was reduced at lower salinities.

The aim of the present study was to investigate the survival of the cercariae of *M.novaezealandensis* with a multifactorial experiment in order to identify factor interactions between temperature, salinity and UVR. Cercarial survival is a crucial step in the overall transmission success of a trematode. The number of cercariae successfully transmitting has important ramifications for infection levels (prevalence and infection intensities) in second intermediate crustacean hosts and will influence survival of these hosts through intensity-

dependent mortality (Fredensborg *et al.*, 2004). To date, most studies on the survival of cercarial transmission stages have focused on single environmental factors which allow investigating a broader range of factor levels, but which do not account for potential factor interactions affecting organisms in nature. The experiment described here is one of few attempting to include such environmental complexity, as well as a factor rarely considered as an environmental component in this context, namely UVR.

Toxicology

Aquatic molluses are considered ideal invertebrate model systems for environmental monitoring and toxicology (Rittschof and McClellan-Green, 2005). In particular, they have been suggested as good test organisms for the assessment of Endocrine Disrupting Chemicals (EDCs). However, molluscs are often infected with digenetic trematodes; commonly between 4% and 65% of a population can carry an infection (Morley, 2006), which, because of their extensive pathological effects on the host, may be a potential major source of distortion for ecotoxicological studies. Trematodes are ubiquitous parasitic worms that infect a wide range of vertebrate and invertebrate animals, and almost always use a mollusc as the first intermediate host. The adult parasite resides in a vertebrate host and releases eggs into the aquatic habitat. The first larval stage, miracidium, hatches from the egg and infects a mollusc. This first intermediate host or primary infection is where the parasite undergoes a series of asexual cycles eventually producing embryos that develop into cercariae. Cercariae are free-living stages which emerge from the mollusc to infect another vertebrate or invertebrate, which may act either as the definitive host (vertebrates only) where the adult parasite develops or as a second intermediate host where an intermediary stage, metacercaria, resides until transmission, usually via predation, to the definitive vertebrate host occurs.

Self-defense factors

Gastropods have an effective internal defence system consisting of cellular (for review see Sminia 1981) and Humoral (e.g. Acton and Weinheimer, 1974; Renwrantz *et al.*, 1981) defense factors. However, in very specific, genetically determined combinations of species or strains, trematodes and snails are compatible (Richards, 1975). This implies that the parasite finds in its

snail host not only a physiologically suitable environment, but also an internal defense system that fails to destroy it.

One reason immune reactivity may not take place could be non-recognition of the parasite by the snail's defence system due to the presence of snail-like or snail-derived molecules on the surface membranes of the trematode (Benex and Tribouley, 1974; Yoshino and Bayne, 1983). This sharing of antigens also occurs in the schistosomesnail combination *Trichobilharzia ocellata Lymnaeastagnalis*; miracidia appear to mimic snail molecules and cercariae adopt substances from the snail's haemolymph (Van der Knaap *et al.*, 1981).

A second possible cause of the absence of effective anti-parasite immune-reactivity is interference: the trematode suppresses anti-parasite defense reactions in the snail. This mode of immune evasion, which may occur concomitantly with sharing of 9 antigens, was documented in an array of studies by Lie and co-workers and has been reviewed by Lokma (2007). Most of the data in support of the interference theory have been gathered in studies with the snail host *Biomphalaria glabrata*. However, there are indications that trematodes also interfere with host defences in lymnaeid snails. In field-collected *Lymnaea stagnalis appressa*, Bourns (1963) found a strikingly high incidence of double infections with certain combinations of trematode species. Lie *et al.*, (1983) succeeded in inducing much higher than normal rates of infection with *Echinostoma hystricosum* in *Lymnaea rubiginosa* when the snails had been infected in advance with *Trichobilharzia brevis*. One of the explanations for these observations may be that the trematode that entered the snail first suppressed immune reactions, thus enhancing the chances of survival of the subsequent species.

Related works around the World

India

The rust reference of Indian trematodes in modern times was made by Gilchrist (1841-1846) in his book "A practical treatise on the diseases of elephants, camels and homed cattle" Later on Cobbold (1859,1862,1869-1882) in a series of papers describe l'parasites of man, elephant,

cattle, gangetic dolphin and Orcaella brevirostris etc, thus laying the foundation of a systematic study of tematode fauna of the Indian region.

Fishoeder (1901-1903) reported a number of amphistomes collected from cattle, horses and elephants in India and Sri Lanka. Shipley and and Hornell (1904, 1905) described *Problitrema richiardii*, *Distoma palleniscum*, *Gymnophallus sumateriae* and *Aspidogaster margratifera*. The year 1906 was significant for the discovery of several species of Schistosomes viz. *Schistosoma spindalis*, *S. indicum*, *S. bovis*, *Ornithobelharzia bomfordi* (= *Orientobilharzia bomfordi*) from the blood vascular system of domestic animals. Trematodes of fishes of Bengal were reported by Southwell and Prashad (1918). Sewel (1920, 1922) redescribed *Mesocoelium sociable* and recorded *Leucochloridium assamense* respectively.

All these studies were made by foreign workers on material sent to them or collected by them during their stay in India. By now Indians started coming in the field. Bhalerao (1924) gave the description of *Eurytrema daji* and *Testifrondosa cristata* from the food mammals of Rangoon. He (1925) identified *Isoparorchis hypselobagri* from man sent to him by Chandler who was working on the parasites of cats of Calcutta. Bhalcrao (1935) produced a valuable monograph of helminth parasites of domesticated animals and described some new monostome trematodes from avaian hosts and reported the parasites collected-from elephants of Andaman and Burma. In 1936 Bhalerao published three parts of 'Trematode parasites of India.' Bhalerao in 1937 continue his researches on the Helminth parasites of India and published his fourth part on Indian trematodes. He also studied the trematode parasites collected from Calcutta Zoo and proposed new genus Pneumotrema. Srivastava (1944) studied the trematodes of Indian marine fishes and also elucidated the life history of *Paramphistomum explanatum*, *Gastrothylax crumenifer*, *Dicrocoelium dendriticum* and *Fasciola hepatica* and recorded a new intermediate hosts of *F. gigantica*.

Ganapati and Rao (1962) reported metacercaria in freshwater fishes of Waltair. By mid-sixties many workers came in the field and studies on trematodes got a boost. Many centres for helminth studies developed. Hafeczullah (1971), Hafeezullah and Siddiqi (1970) worked out the trematode parasites of marine fishes of India. Trematodes have been studied by Rai and Agarwal (1963) Fotedar and his collaborators (1959-1984) Gupta (1965, 1983), Pandey, (1962-1990) Srivastava, (1963-1990), Mukherjee and Ghosh (1967-1969), Gupta (1956-1986), Mukherjee,

and Srivastava (1988, 1989), Ghosh (1979-1989) and many others. Mehra (1970) gave a detailed classification of Trematoda recognizing five orders. No volume on Trematode Fauna of India was available till Mehra submitted his first volume dealing with the order Fasciolatoidea.

This was published under the Fauna of India series in 1980. Since the volume included species described till about 1963, Srivastava (1982) provided a supplement to this volume updating the volume. Chauhan and Ghosh (1975) reviewed the work done by the ZSI scientists in the last 50 years. Periodic review of the work done in India has been published by Bhalerao (1947, 1948) Thapar (1937, 1947) Srivastava (1960) Chauhan (1952).

Some species of sarcophagids are known to feed on snails (Beaver, 1980, Coupland and Baker, 1994). Using serological techniques Parashar and Rao (1988) reported *Sarcophaga*

(*Parasarcophaga*) *misera* (F.) as a predator of the snail *Indoplanorbis exustus* Deshayes which is a vector of animal schistosomiasis in India (Malek and Cheng, 1974). This snail is preferably distributed in ponds and pools of temporary nature, which remain dry for 3-6 months each year, during which period it undergoes dormancy and its populations are exposed to predator/sarcophagid attack. In India, its reproductive period in natural habitats ranges from July to November when ambient temperature varies from 25-35°c which is reported to be a favourable temperature range for its reproduction (Parashar and Rao, 1985). Depending upon the nature of habitat in relation to quantity of water available, the snails become dormant from December to June (Parashar and Rao, 1996). Therefore, *S. misera* has to operate in the field for different time periods depending upon the state of the habitat.

Bangladesh

The climatic condition of Bangladesh is favorable to the ecological conditions and suitable for survival and breeding of snail intermediate hosts. Long standing summer and rain in the country facilitate least stagnation of water in various pools where fresh water snails shelter and reproduce in abundance. Due to availability of scanty pasture lands in Bangladesh, animals are to eat infected grass in marshy areas inhabited by snails. So, control of important snails is very essential in certain areas as one of the sufficient measures to reduce the economic loss impaired by the parasites which they harbor.

Trematodes are of extreme economic importance especially in tropical developing countries. There is considerable economic loss in the form of mortality reduced weight gain decreased meat and milk production low fertility reduction in draft power and poor hide quality in Bangladesh due to fascioliasis in ruminants (Begum, 1993). An estimation of annual economic loss of about 0.6 million taka from liver damage due to fascioliasis in goats was reported by Bhuiyan (1970). Besides some trematodes cercariae produce swimmers itch (cercarial dermatitis) in man (Sandosham, 1951). The freshwater snails are involved in the transmission of trematode species belonging to the super families Schistosomatoidea, Fascioloidea, Climostomoidea, Paramplustomoidea.

The larval trematodes have received great attention in order to establish the biological relationship of the species with the intermediate hosts they utilize for the completion of their life cycles. The climatic condition of Bangladesh is favorable to the ecological conditions and suitable for survival and breeding of snail intermediate hosts. Long standing summer and rain in the country facilitate least stagnation of water in various pools where fresh water snails shelter and reproduce in abundance. Due to availability of scanty pasture lands in marshy areas inhabited by snails. So, control of important snails is very essential in certain areas as one of the feasible measures to reduce the economic loss impaired by the parasites which they harbor.

A number of studies have been made on the snail intermediate hosts of harmful trematodes in Bangladesh (Qadir, 1982: Begum, 1993: Rahman *et al.* 1997: Mondel *et al.* 2003: Alam, 2010). However, these investigations were kept limited within plain lands and hilly areas of the country. Mymensingh Sadar is relatively a low land of which many parts goes under water during rainy season. Again there are many canals, ponds, drains, paddy field with irrigation channels and marshy lands. The river Bhramamapotra is also flowing through the areas. As such identification of snail yectors and their role in the transmission of harmful trematodes are acknowledged as a prelude to address the anticipated problems.

Mediterranean region

Melanopsis praemorsa (L. 1758, Buccinum) is a conically elongated, dark brown to black prosobranch snail which may exceed 2 cm in length. The sculpture of the shell is highly varied, being either smooth or ribbed to a greater or lesser extent. However, no differences were

detected in the radulae of smooth and ribbed snails (Tchernov, 1975). This snail is widely prevalent in freshwater bodies of the Mediterranean region (Tchernov, 1971, 1975; Brown and Wright, 1980). In Jordan, it is one of the most abundant snails present in ponds, springs, streams and rivers. It is very common on the edges, especially mud banks, of the Yarmouk River, which runs in a deep valley that separates Jordan and Syria and connects westerly with the Jordan River. Therefore, it is to be expected that it may act as an intermediate host for many digenetic trematodes of various vertebrates in the area.

Ullman (1954) reported nine different types of cercariae found in *M. praemorsa* collected from Yarkon River, Palestine. These included two types of *Cercaria microcotylae*, two types of *C. virgulae*, one *Cotylocercous* cercariae, one xiphidiocercaria, one *Cercariaeum* cercaria, *C. vivax*, and *C. orospinosa*. However, the latter was the only one described. Additionally three types of cercariae were encountered in snails from Megiddo springs in the Yizre'el Valley: two xiphidiocercariae and one furcocercous cercaria (Lengy and Stark, 1971).

Pirenella conica Blainville (1826), the vector of heterophyasis of humans, other mammals and birds, is a prosobranch gastropod found in several biotopes around the Mediterranean Sea, the Red Sea and the Persian Gulf. The distribution is reviewed by Taraschewski and Paperna (1981). In the Middle East it is the first intermediate host of the trematode genus *Heterophyes* (Khalil, 1937; Kuntz, 1957), *Stictodora* spp. (Martin and Kuntz, 1955; Martin, 1959) and of several other unidentified trematodes (Demian *et al.*, 1963; E1 Gindy and Hanna, 1963; Yousif, 1970).

The ecological characteristics of several *P. conica* habitats on the coasts of Sinai were recently described in relation to trematode infections of the snails and the transmission of these trematodes (Taraschewski and Paperna, 1981).

Europe

In investigations of helminth parasites of British limpets (*Patella vulgata*, *Patella intermedia* and *Patelladepressa*), three larval trematodes (*Cercaria patellae* Lebour, 1911, an unidentified xiphidiocercaria and an unidentified tailless cercaria) and one larval cestode were found by Crewe (1951). This author also described a metacercaria in *Rvulgata and P intermedia* and

suggested that this metacercaria developed from the encysted xiphidiocercaria. *C. patellae in the* digestive gland and the gonads of *P. vulgata* was first observed at Loch Ryan in Scotland (Lebour, 1907, 1911). Later, Rees (1934) described the cercaria and gave an account of its pathological effects on *P vulgata.The* ecological aspects of the prevalence and also the effects of infectionon limpets from the Inner Fame (Northumberland) were studied by Thomas (1965). *C.patellae* was also recorded in a survey of the occurrence of larval digenea in littoral molluscs in Cardigan Bay (Wales) between 1958 and 1960 (James, 1968). In Northern Ireland the prevalence of *C. patellae* in *P vulgata* varied between the collections sites on the shore from 0 to 90% (Copeland *et al.*, 1987).

At several other localities on the British coast the prevalence of *P. vulgata* harbouring *C. patellae* was consistently lessthan 14% (Lebour, 1911; Rees, 1934; Crewe, 1947, 1951; Thomas, 1965; James, 1968). In the region of St. Andrews investigated in the present study, it was about 5% (data of Arnold from 1958, reviewed by Laverack and Blackler, 1978). Only James (1968) found infected specimens restricted towards the exposed clifftops and suggested that this was due to nesting and feeding of the final host. Infected limpets were also found in intertidal rockpools, indicating that the source of infection could be a shore bird feeding in the intertidal pools, where the miracidia hatch, and that the lower prevalence at low-tide levels may be correlated with the habitat of birds, which frequently alight on the rocks near the low-water mark (Crewe, 1951; Thomas, 1965; Copeland *et al.*, 1987).

According to Lebour (1911), Nicoll suggested that the cercariae may be the larval stages of *Echinostephilla virgula* Lebour, 1909 from the tumstone *Arenaria interpres*, but so far no investigations of the life-cycle have been carved out and modem taxonomic techniques have not been applied to the problem.

The study describes the life-cycle and the morphology of *Cercaria patellae* from the coast of St. Andrews and its subsequent developmental stages. The second intermediate host, in which encystment takes place, was found by considering factors relating to habitat, and by comparing measurements of metacercariae of natural infections with those of experimental infections. *In vitro* excystation experiments and a comparison of young adult trematodes, obtained after

experimental infection of young chicks, with published data on adult trematodes from the oystercatcher *Haematopus ostralegus* indicate that *C. patellae* is the cercaria *of Echinostephilla patellae*. This paper presents new data on the prevalence on the coast at St. Andrews, including differences between specific habitats and specific groups of limpets. In addition, it also reports investigations of the life-cycle, i.e. *in vit*ro excystation and experimental infection of young chicks, and the application of different taxonomic techniques, including measurement of developmental stages, chaetotaxy and scanning electron microscopy.

Contrary to redial development, little information is available on the quantity of cercariae, which may develop within an infected snail. Swales (1935) reported a total of 687 metacercariae for a naturally infected specimen of *Fossaria parva* but only found a mean of 125.5 cysts per snail when this species was experimentally infected. In the same way, Erhardová-Kotrlá (1971) noted 350 metacercariae for a naturally infected specimen of *G. truncatula*. Under these conditions, it was interesting to study the cercarial productivity of *F. magna* by addressing the following two questions: How many free rediae of *F. magna* may a natural snail host, such as *G. truncatula*, harbor within its body? How many cercariae of *F. magna* does a snail contain within its body? To answer these two questions, single-miracidium infections of French *G. truncatula* with a Czech isolate of *F. magna* were carried out under laboratory conditions.

Examination of molluscs for larval trematodes is the keystone for the determination of the autochthonous fauna in the region under study (Bock, 1982a). Trematodes (Digenea) from freshwater molluscs were studied for a long time with many new species described from Germany. Odening (1978) overviewed the species of the Trematoda found as adults in East Germany and listed 177 species with a freshwater life cycle that can potentially be completed in Germany (cercariae with unknown life cycles were omitted).

The first faunistic survey of cercariae in Germany was made by Nitzsch (1817) who studied cercariae near Lutherstadt Wittenberg; 100 years later, Lühe (1909) already attempted to summarize the known species. In the 1920s, Scheuring (1922) elucidated the life cycle of Sanguinicola inermis and Wunder (1924) studied the morphology of cercariae near Regensburg (Oberpfalz). The first systematic studies on the occurrence of cercariae started since the 1960s,

together with attempts to complete their life cycles. Odening and his co-workers, 2004 made the most outstanding contributions to the research of cercariae in Berlin and its surroundings.

Although a significant part of Finland is covered by lakes, the knowledge on the freshwater larval trematodes is relatively limited. The first study on the trematodes with freshwater life cycles was published by Wikgren (1956) who summarized the data on larval trematodes (28 species in total) occurring in southwestern Finland including the scattered data of previous Finnish investigations. After a long gap, ecological aspects of trematode communities of five species occurring in bivalves and their host-parasite relationships were studied by the group of Valtonen (Taskinen *et al.*, 1991, 1994, 1997; Gibson *et al.*, 1992; Holopainen *et al.*, 1997; Rantanen *et al.*, 1998). The investigations on the cercarial fauna of *Lymnaea* spp. by Niewiadomska *et al.*, (1997) in Central Finland and Väyrynen *et al.*, (2000) in a more northern area revealed another five species, which were new records for Finland.

Recently, during ecological studies focused on *Diplostomum* spp., another species was found (Karvonen *et al.*, 2004, 2006a, b). Based on previous studies, so far cercariae of 39 trematode species of 16 families are known to infect molluscs in Finland. In 2005, trematodes of three pulmonate snails (*Lymnaeastagnalis*, *Bathyomphalus contortus*, *Planorbarius corneus*) and the prosobranchiate *Valvata macrostoma*, previously not examined in Finland, were studied in Lake Konnevesi (Central Finland). This study presents eight furcocercous cercariae with descriptions and illustrations of new species records and new records of cercariae in Finland.

North America

Mexico, situated in a transition area between the Nearctic and Neotropical zoogeographic regions, possesses a rich fauna of trematodes (Scholz *et al.*, 2000). Despite this, in Mexico, larval stages of trematodes in gastropods have been scarcely studied (Caballero and Larios, 1940; Caballero *et al.*, 1961a, b; Gómez-Agudelo *et al.*, 1978; Lamothe-Argumedo *et al.*, 1983; Ito *et al.*, 1985; Pérez-Reyes *et al.*, 1985; Rangel-Ruiz and Lamonthe-Argumedo 1986; Castro-Trejo *et al.*, 1990; Salgado-Maldonado *et al.*, 1995; Ditrich *et al.*, 1996, 1997; Scholz *et al.*, 1994, 1995, 1996, 1997, 2000).

In particular, for the Lerma River marsh, there are few records in reference to parasite species present in freshwater gastropods; especially for the Basommatophora order, there are only a few records: *Echinostoma revolutum* Looss, 1899 in *Physa osculans* and *Lymnaea attenuata* (Caballero and Larios, 1940). Due to the scarcity of records and the importance of providing information on parasites in the marsh of the Lerma River, the objective of this work was to determine the larval stages of digenetic trematodes present in molluscs of the Basommatophora Order in Lake Chicnahuapan.

Justification of the present study

The study of parasites and their relationship to the host requires a multidimensional approach in order to understand the nature of parasitism and the pathological effects on the hosts. Such studies include phylogenetic relationships, morphological and ecological aspects, physiology and biochemistry of the parasites, host parasite relationship and aerobiological and immunological studies. In recent years, attention has been paid to the study of morphology, physiology, biochemistry, ecology of the parasites and the work on serological and immunological studies have been accelerated.

In the present work host parasite relationship is studied and full attention is paid to study the effects of larval stages of trematodes on the first intermediate host i.e. gastropods. The study of relationship between a trematode parasite, its intermediate host and final host brings about the ecological and histopathological studies of molluscs as well as vertebrate hosts. The study of molluscs harbouring the cercariae of various trematodes involves in the study of environment in which the molluscs live. Freshwater harbors a wide variety of animals depending upon its physico-chemical nature. There is a definite relation between the chemical composition, presence of molluscs and infection of cercariae.

There are only minimal studies carried out in Dhaka, Khulna, Kishoreganj and its surrounding areas on biodiversity with reference to freshwater samples, availability of specimens and physico-chemical parameters. Therefore, this present study on biodiversity of gastropods on these particular lentic eco-systems does not have much of previous influence. According to literature from 1902 till date, most of the gastropod-molluscan studies carried out were for a

mere purpose of medicinal values than anything else. Apart from this inference, few research analysts have also worked on this aspect with reference to parasites. However, there are no detailed taxonomic and seasonal studies available to survey the species diversity, behavioural expression etc. that would highlight the importance of preserving gastropod diversity. Although, molluscs are common components of the benthic community, their role in the dynamics of aquatic eco-system and their contribution to biomass production is not very clear (Supian and Ikhwanuddin, 2002). The freshwater habitats are taxonomically impoverished in comparison to the marine habitats. Marine molluscs have received more attention because of their aesthetic and gastronomic appeals (Subba Rao 1993) than the fresh water molluscs. In comparison, freshwater molluscs are drab coloured than the marine molluscs and are known to play significant roles in the public and veterinary health hence, needs to be significantly explored.

Since the study area is urban in nature, it becomes duly necessary to find out the number of species available with their systematic position is more. Along with the classification study, the different parameters such as pH, salinity, alkalinity and dissolved oxygen of each pond were also discussed in detail regarding the availability of parasites in each species. Therefore, this particular study was important to understand the species diversity and distribution of the gastropod molluscan population in and around the study areas and the effect of habitat on their distribution. We hope that the results of this present study will ensure the safety and sustainable surveillance of these molluscs and their medical importance.

Objectives of the study

This snail is of immense importance from the viewpoint of economy. They are the source of animal protein and act as the intermediate hosts of trematode parasites, the causative agent of helminth diseases. Moreover, they play an important part in the aquatic ecosystems. Like all other animals in the ecosystem, the distribution and abundance of freshwater snails are also influenced by the environmental components, such as, water qualities, aquatic vegetation, soil substratum, depth of water, temperature, turbidity, pH, Dissolved Oxygen (DO), hardness, etc. (Mukherjee, 1986; Raut, 1986; Rahman and Jahan, 1996).

Specific objective

The specific objective of the research is to investigate developmental stages of helminthes, its diversification with ecological parameters.

General objectives

- To investigate the developmental stages of helminthes, particularly trematodes in the body of snails in different ecological areas in Bangladesh.
- To determine the prevalence of infection of host snails, the developmental stages and the basic epidemiological parameter- the prevalence of infection by their parents.
- To determine the basic conditions of the habitat of the snails in water, like temperature, colour, pH and Dissolved Oxygen (DO) of the investigated bodies.
- To investigate whether there was any seasonal effect on the parasitization aspects of snails (in Bangladesh practically there are 3 seasons, summer (March-June), rainy season (July- October) and winter (November- February) (Ahmad, 1968).
- To identify the ecological relations of snails and definitive hosts.

REVIEW OF LITERATURE

General considerations

A large number of studies have been carried out on the developmental stages of trematode in snail's intermediate hosts. Most of them are based on the cercarial types and their prevalence. Some of them are given below:

Yoder and Coggins (1998) observed the larval trematode assemblages in the snail *Lymnaea* stagnalis from Southeastern Wisconsin. Their observations of cercarial shedding and subsequent dissection of snails revealed a relatively depauperate digenean component community consisting primarily of 3 common species. Among them the metacercariae of *Cotylurus flabelliformis* was the most frequent one. Sporocysts and cercariae of this species were rarely present. Snails were also reported to harbor sporocysts and shed cercariae of *Schistosomatium douthitti* and a plagiorchid.

In case of trematode parasites, molluscs are almost obligatory first intermediate hosts (Esch *et al.*, 2001). About 100 species of snails are reported to act as intermediate hosts for the trematode parasites (Subba Rao and Mitra, 1989). They serve not only as a source of food and a place of reproduction for those parasites, but also as a means of transport for getting to their next hosts (Lockyer *et al.*, 2004). The uniqueness of the relationship between these organisms derives from the fact that majority of the 18000 known species of trematodes exploit snail as the mandatory first intermediate host (Littlewood and Bray, 2001). Many researchers have no doubt that these parasites were originally linked with molluscs, and only as a consequence of evolutionary development did they include in their life cycle a further group of hosts (Pojmanska and Grabda-Kazubska, 1985; Cribb *et al.*, 2001). Snail-Trematode assemblages have recently attracted considerable interest from evolutionary ecologists studying the evolution of sexual reproduction (Lively 1996), host-parasite co-evolution (Lively, 1996; Dybdahl and Lively, 1998), the effect of parasitism on life-history evolution of host (Jokela and Lively, 1995) and host resistance (Webster and Woolhouse, 1998).

Sharif *et al.*, (2010) carried out a faunistic survey of cercariae isolated from Lymnaeid snails in Central areas of Mazandaran, Iran and found 3.6% snails to be infected. *Lymnaea gedrosiana*

were found to be infected with the furcocercariae of Diplostomidae, Echinostomidae and also cercariae of Plagiorchiidae. The latest infection was found to be in *L. palustris*.

In Bangladesh, Rahman and Jahan (1999, 2001, 2002, 2004 and 2006), published works are available on the occurrence of helminth infections in snails. The works of Rahman and Jahan dealt with cercarial types, their prevalence, abundance and intensity, seasonal influence on prevalence, abundance and intensity and biology of amphistome cercaria and their effects on the different organs and the fecundities of some freshwater Gastropods in Bangladesh. Hasnat, M (2006) carried out MS dissertation on 'Helminth infections of four snail species of three freshwater bodies of the Chittagong University campus and adjoining areas'. Islam et al., (2012) studied on different species of vector snails carry larval stages of different trematodes. Studies were performed to investigate different snail species and parthinate of trematodes carried by them, in some selected areas of Mymensingh Sadar, Mymensingh.

Investigations on the larval trematodes of fresh water snails and their host-parasite relationships from 11 districts of West Bengal were performed by Das Mahapatra *et al.*, (1982) and Majumdar *et al.*, (1983).Extensive works on Amphistome cercariae were reported from India by several workers (Sewell, 1922; Chatterjee, 1931; Pande, 1935; Bhalerao, 1945; Iyar, 1949; Peter, 1958; Sharma and Malik, 1960; Jain, 1972 and Mukherjee, 1986).

Hopkins (1937) reported a new type of anallocreadiid cercariae that included cercariae of an allocreadium and microcreadium in the snail *Amnicola peracutta* in Texas.

Johnston and Cleland (1938) reported a new larval trematode, *Cercaria murrayensis* (furcocercaria) from Lymnaea lesson collected from the River Murray near Tailem Bend, Australia.

Johnston and Angel (1942) recovered *Cercaria metadena n sp.* from Tailem Bend, Murray River, South Australia, from 15 *Planorbis isingi* specimens amongst 2,821 examined (1.17%).

Banks (1951) reported a new megalura cercaria from snails, *Goniobasis livescens*, collected from Big Darb Creek, Ohio and an incidence of infection of 2%. He found the redia as large sac-like which almost completely eroded the hosts digestive gland (liver). The bodies of the larvae were

of oval shape and white in appearance due to the abundance of mucoid material in their cystogenous cells.

Pike (1967) recovered some stylet cercariae and a microphalid type from British freshwater molluscs. Etges *et al.*, (1969) recovered two new species of xiphidiocercariae from Mountain Lake, Virginia. These were polyadenous xiphidiocercariae species from the planorbid snail, *Gyraulus parvus* (Say), and a virgulate xiphidiocercariae from the prosobranch snail, *Anaplocamus dilatatus* (Conrad). Murty (1976) studied on the Indian cercariae and reported 3 new species of virgulate plagiorchioid cercariae viz, *Cercariae indica*.

Chung et al., (1980) studied the larval trematode infections of snail Parafossarulus manchouricus in river Kumho, Kyungpook Province, Korea. They recovered 4 species of cercariae of digenetic trematodes, Clonorchis sinensis, Cyathocotyle orientalis, Exorchis oviformis and Loxogenes liberum. The Loxogenes jiberum was the most frequent and the rate was 43.3%.

Siddiqui (1980) studied the systematic and biolory of larval trematodes infecting freshwater snails in Panjab (Punjab), Pakistan and recovered 54 species of cercariae. These included one monostome, three amphistome, eight gymnocephalus, four echinostome, thirteen xiphidiocercous and twenty five furcocercous. Out of these, thirty six were new to science and ten were reported for the first time from Pakistan. Ten species of six genera of snails were collected and examined for trematode infection. *Bellamya bengalensis* was never found to harbor any trematode infection. The overall rate of infection of snails was noted to be 2.33%.

Khan and Haseeb (1981) studied the larval trematodes infecting freshwater snails in Pakistan and reported a new pleurolophocercous cercaria named *Cercaria bilaterolophocauda*.

Loker *et al.*, (1981) observed the trematode-gastropod associations in nine non-lacustrine habitats in the Mwanza region of Tanzania and recovered 38 morphologically distinguishable species of cercariae (13 furcocercous, 10 xiphidiocercous, 6 echinostome, 4 cystophorous, 3 gymnocephalus and 2 amphistome). The majority (63.8%) of all mature infections were xiphidiocercous. *Biomphalaria pfeifferi*, *B. sudanica and Ceratophallus natalensis* each yielded 11 species of cercariae. *Lymnaea natalensis* had the highest overall prevalence of infection (36.9%). *Cercaria gluttera* from *L. natalensis* accounted for 20.4% of all recovered trematode

infections and *C.blukua* from *Biomphalaria* accounted for 18.4% of all infections; the high prevalence of these two xiphidiocercariae may alter the transmission patterns of *Fasciola gigantica* and *Schistosoma mansoni*, respectively. *S. mansoni* was recovered from both *B. sudanica* (22 of 2393 infected) and *B. pfeifferi* (79 of 1913 infected); *S. haematobium* (or related species) was recovered from *Bulinus* (*Physopsis*) *nasutus* (50 of 1503 infected) and to a lesser extent from *B.(P.) africanus* (6 of 186 infected).

Loy and Hass (2001) carried out a long time study (from 1980-2000) on the prevalence of cercariae from *Lymnaea stagnalis* snails in a pond system in Southern Germany. They reported at least 18 species of cercariae and found high (44.9%) cercarial prevalence.

Radev *et al.*, (2004) worked on the cercarial fauna and recovered five different types of cercaria (gymnocephalata, amphistomata, monostomata, echinostomata and xiphidiocercaria) from *Lymnaea truncatula* (Muller, 1774) snails collected from hilly- pasture biotopes in Bulgaria.

Ghobadi and Farahnak (2004) carried out a faunistic survey on the cercariae of *Bellamya* (*Viviparus*) bengalensis snails and their zoonotic importance and found 0.4% of *Bellamya* infected with xiphidiocercariae belonging to Plagiorchiidae helminth parasites.

Choubisa (2008) focused on pathogenic trematode cercariae infecting fresh water snails (Mollusca: Gastropoda) on tribal region of Southern Rajasthan, India and reported six different types of cercaria, viz., amphistome, echinostome, furcocercous, gymnocephslous, monostome and xiphidiocercariae.

Sharif *et al.*, (2010) carried out a faunistic survey of cercariae isolated from Lymnaeid snails in Central areas of Mazandaran, Iran and found 3.6% snails to be infected. *Lymnaea gedrosiana* were found to be infected with the furcocercariae of Diplostomidae, Echinostomidae and also cercariae of Plagiorchiidae. The latest infected was found to be in *L. palustris*.

Sewell (1922) has been a valuable contribution to the knowledge of Molluscs and Cercariae from various regions of this subcontinent. Wesenberg-Lund (1934), working on the freshwater trematodes of Denmark. Probert (1966) studied ecology of British freshwater larval trematodes and their first intermediate hosts, Mollusca. Digenetic trematodes commonly known as flukes are responsible for a number of disease conditions in humans and many other Vertebrates. They

have a heterogeneous life cycle with freshwater snail as their first intermediate host. The adult stages are found in different vertebrate definitive hosts including amphibians, fishes, reptiles, birds and mammals. Disease characteristics of fluke infection vary with the parasite species and the site or organ of infection and are linked with the life cycle events like larval penetration, egg laying etc. Many species of freshwater snails serve as intermediate hosts for digenetic trematodes. Consequently, the distribution of freshwater snails accounts for the occurrence of different trematode taxa in a particular region. As the parasites are mostly host specific, higher heterogeneity of the parasites (Hechinger and Lafferty, 2005). Similarly, higher snail diversity leads to higher trematode diversity; trematodes show distinct and direct relationship with the temperature in their transmission process (Smyth, 1962, Poulin, 2005).

Ecological studies on larval trematodes

Ecological studies on larval trematodes and their first intermediate host have been carried out by several workers.

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Probert (1966) studied ecology of British freshwater larval trematodes and their first intermediate hosts, Mollusca. Digenetic trematodes commonly known as flukes are responsible for a number of disease conditions in humans and many other Vertebrates. They have a heterogeneous life cycle with freshwater snail as their first intermediate host. The adult stages are found in different vertebrate definitive hosts including amphibians, fishes, reptiles, birds and mammals. Disease characteristics of fluke infection vary with the parasite species and the site or organ of infection and are linked with the life cycle events like larval penetration, egg laying etc. Many species of freshwater snails serve as intermediate hosts for digenetic trematodes.

Poulin (2005) studied on the cervical output is directly influenced by the temperature due to both stimulating effect of temperature increasing the emergence from the snail and the acceleration of

cercarial production within the snail host. Lafferty (1993) examined on trematodes that can affect snail populations by directly reducing the egg production or by increasing snail mortality rates. Shostak and Esch (1990) proposed several hypotheses to explain the functional significance of these daily cycles. Cercarial species in which the target hosts do not regularly cohabit with the molluscan host producing the cercariae (Combes *et al.*, 1994).

Margolis *et al.*, (1982); Anderson and May (1991) worked on prevalence and intensity of infection are the major determinant factors of the rate of transmission of digenetic trematodes from the snail intermediate host to the next host in their life cycle. Mendis and Fernando (2002) and Naggs and Raheem (2000) worked on release of cercariae out of their snail intermediate host which is a measure of intensity of that particular trematodes infection.

Farrynkova *et al.*, (2008) identified snails according to the morphological features of the shell using keys. Cercariae released by the snails were isolated and slide mounted using single-staining procedure in Borax-Carmine. Identification of cercariae was carried out using descriptions and keys.

Fernandez and Esch (1991); Sausa, (1994) highlighted on spatial variation in infection which can easily arise as a consequence of the distribution of second characteristics that affect the risk of infection.

Gardner and Camphell (1992) focused on diversity of digenean fauna that is dependent on conditions that are conductive for transmission such as the presence of final and intermediate host. Studies have showed a consistent relationship between the density and heterogeneity of the snail populations with the trematodes density and heterogeneity (Hechinger and Lafferty, 2005). However, as a consequence of the direct interference of trematodes on snail egg production and their mortality rates, trematodes prevalence is hot directly related to the snail population density (Lafferty, 1993).

Granovitch et al., (2000) reported microphallus piriformes infection causing more than half of the periwinkle population, devoid of reproduction. Similarly snail populations in the dry zone

might have a castration effect of the existing trematode infections in the area resulting very low population density.

Faizal (2000) has reported nine types of cercariae collected from a site in *Kalametiya oya* in Matale District. Faloon *et al.*, 1996 proposed on the sedimentation that can interfere the trematode life cycle by preventing miracidia from finding the host, reducing egg supply and the survival of host snail. Environmental changes following irrigation development are likely to modify the pattern of distribution of snails and trematodes. Consequently the cercariae shed by snails can be used to assess the environment impact (Lafferty, 1997) as well.

Iwagamia *et al.*, (2009) reported that *Paragonimus westermani* utilizes *Paludomus sp*, as the molluscan intermediate host in the wet zone of Srilanka. Gymnophallus reported in this study closely resembled the cercariae of Paragonimus, Xiphidiocercariae reported in this study closely resembled the *Cercariae vesiculosa* described by Wesenberg Lund (1931).

Identification of cercariae to species level using fine characteristics like nervous system elements, sensory apparatus, body papillae such cheatotaxy is possible (Hrusanov and Radev, 2005). The diversity of infection in the dry zone was very low with only two morphotypes. Jaywardena, 2010 found that the cercariae diversity was very high in the wet zone and the prevalence of infection was very low compared to the dry and intermediate zones.

Islam *et al.*, (2012) studied on different species of vector snails carry larval stages of different trematodes. Studies were performed to investigate different snail species and parthinate of trematodes carried by them, in some selected areas of Mymensingh Sadar, Mymensingh.

Seasonal Variations in Snail Infection

As snail population is dependent on rainfall and temperature, it is no surprise to record change in snail population as per season which is highest during rainy season and lowest in summer. Accordingly, the percentage of positive snails is highest in monsoon or just after that, with the

lowest number in summer. Almost all year-round surveys have recorded seasonal variation in snail population and positivity for schistosomes.

Soparkar (1921) reported negativeness of *I. exustus* for *S. spindale* between February and April, with the highest infection rate between September and October in a 2 years research. In a survey near Bareilly, Uttar Pradesh, the population of *L. luteola* var *australis* was high during March to May and October to February, low in June and September, and rare in July and August; "natural infection of *S. incognitum* could be detected in these snails mostly during March, April and May" (Sinha and Srivastava, 1960).

Singh (1959) reported the highest positiveness of *I. exustus* for schistosome cercariae in the months of May and December, while Raut (1983) observed the maximum positive snails in the months of July and August. Year-round surveys have been made for *S. nasale* infection in the snails at four centers under All India project (Alwar, 1974) and for four mammalian schistosomes at Jabalpur (Kohli, 1991).

Muraleedharan *et al.*, (1976a) reported in the two enzootic areas (Dhanayakanapura and Hunchipura) of Karnataka state, a 3-year survey of positive snails was made by collecting 5,286 *I. exustus* snails of which only 25 (0.47%) were found positive for *S. nasale* cercariae. Interestingly, these positive snails could be observed only in the months of February, June, July, August, and October, in tandem with higher population during these months; they were negative in other months. A similar survey in Bhandara district of Maharashtra state between January 1995 and January 1996 revealed low snail population during late summer and early rainy season (April to August without finding any *I. exustus* in July and August), with its increase reaching a maximum in November.

Thakre and Bhilegaonkar (1998) recorded highest positiveness between November and January (maximum of 14.8% in January), whereas it was nil between April and August-when snail population was also low. A 3-year (1987-1989) survey of *I. exustus* at Jabalpur revealed 3.27% positivity for mammalian schistosome cercariae which was influenced by the season with

recording of no positive snail in May and June but highest prevalence of 7.9% in January and 12.6% in April (Kohli, 1991).

Mishra (1991) did a similar survey during the same years (1987–1989) on *I. exustus* at Jabalpur revealed 0.08–0.98% prevalence between April and July with the absence of any positive snails in the month of May; in other months, it varied between 0.37% and 11.35%, with the highest prevalence in October. Another survey on *L. luteola* at Jabalpur revealed overall prevalence of 3.35% of *S. incognitum* with observations of seasonal variation in the prevalence rate (Agrawal *et al.*, 2000).

Nevertheless, the prevalence is not influenced by season alone, but other factors are also responsible as shown by Agrawal, 2000 when all the 74 *L. luteola* snails, collected from Marhai locality of Jabalpur, were found negative, but 100 *L. luteola*, collected from the same pond just after 2 days, showed 13% being positive for *S. incognitum* infection. Likewise, variations in positiveness in other months have also been recorded suggesting that factors other than season are also influencing positiveness—perhaps a true assessment is difficult with present methodology.

Climatic variations in Snail Infection

Chandore (2011) found infection rate increased with the rise in heat. In the month of January infection in all the snail species was very lowest amount but in February and March it increases comparatively. Ultimately in the month of April and May it shows the maximum rate of infection. Some of these influencing factors might be size and genetics of snail population, spot, time, weather, and number of snails collected. Simultaneously, these observations are indicative of how wrong it will be to declare a geographical area free from schistosomiasis merely on one-time survey of local snail population—that too during adverse season.

However, a general inference may be drawn by analyzing above data that positiveness of the snails is in tandem with their population in the environment—higher population, higher

infection, and vice versa. This percentage of positive snails does not relate only with its population but also on miracidia population.

Muraleedharan et al., (1976b); Kalwaghe et al., (2003) highlighted during rainy season, there is increase in snail population, and a proportional increase should also be in miracidia number, thereby increasing not only percentage positivity but also absolute number of positive snails. As the absolute number of snails are decreasing post winter and summer, if the number of miracidia remains constant to the rainy season level, this would result in a higher percentage of positive snails due to the reduction in snail population (though not an absolute number), which is not happening. Indeed, a reverse is happening, decelerating positiveness of snails. This clearly hints that a lower number of snails are infected during winter and summer seasons, perhaps, because a lower number of miracidia are available in nature during these seasons. Or are some other factors responsible for lower positivity despite presence of constant number of miracidia? Indeed, reduction in miracidia number as per season is supported by the observations of nasal scrapping and fecal examinations of domestic animals where schistosomiasis prevalence is shown to be influenced by the season—maximum during monsoon and minimum in summer. As schistosomes are long lived (2–5 years), the only inference can be made is that egg extrusion is also influenced by the season, though there is no season when animals completely stop excreting schistosome eggs. Therefore, this cannot be the sole reason for diminishing positivity in the snails as there are observations when in certain months no snail is found positive for schistosome cercariae. For instance, May and June were negative for *I. exustus* in the survey made by Kohli (1991). As larval development in the snails takes about 30 days, it may be presumed that no I. exustus got infected during April/May.

Thus, there is a situation in certain seasons of the year when snails are present but none became infected. It is important to work out the reasons for this negativity as it may greatly help in our control programs of schistosomiasis. What can be the reasons for this refractoriness of the snails, despite presence of miracidia in the environs? To get a possible answer for the above refractoriness, it is important to note that with the change of the season, there are also changes in the snail's population dynamics, its habitats, and microclimates.

Upadhyay (2007) focused on the abiotic factors like dissolved oxygen, biochemical oxygen demand, pH, and mineral concentration of calcium, copper, iron, and zinc of soil and water of snail habitats differed as per season. Change in type of aquatic weed, its quantity, pH, calcium, and total solid concentration as per season were also observed in the ponds of endemic foci of *S. nasale* in Karnataka (Muraleedharan and Kumar, 1974). The water sources start receding and small ditches and pools may completely dry in summer season with changes in microclimate in all these snail habitats. There is a clear change in "biochemical ecology" as per season, and how the snail species are adjusting to these changes is not well understood.

Though the snails are considered being more flexible in their requirements, the same may not be true for schistosome miracidia or its activity. There is even a change in the proportion of snail population of gastropods with different percentage of *I. exustus* and *L. luteola* as per season (Upadhyay, 2007). Whether there is also a change in genetic makeup of these snail populations to withstand adverse environs (and so susceptibility to schistosomes) is not worked out, though it is well known that during pre-monsoons, snails have greater reproductive potentials. Obviously this phenomenon supports change in physiological and biochemical activities of the snails as per season; some might not be favorable for larval development resulting in encapsulation of miracidia that penetrate the snail during adverse seasons. There is also the possibility of changes related to the miracidia. Change in hatchability of eggs as per season is the first possibility—the prime factor will be change in chloride ions in the water or salinity of water. There may be change in excretory products of snails which are responsible for miracidia attraction.

Jourdane and Theron (1987) reported there is evidence that all miracidia are not capable of infecting its compatible host. A study of *S. mansoni* and *B. glabrata* revealed that 30–60% miracidia do not penetrate the snail, and 15% miracidia which are penetrating the snails degenerate in the snail host in absence of hemocytic reaction—thus, this is beside encapsulation in the presence of an existing trematode infection. There is all the possibility that both infectivity and developing capability of miracidia are affected by the season also. This seasonal effect may vary as per sex of miracidia being more favorable to male miracidia during adverse season (e.g., summer), thereby resulting in more shedding of male cercariae. This assumption gets support from the recovery of only male *S. incognitum* flukes from rabbits and dogs which were infected

subcutaneously with 390–1,200 *Schistosoma incognitum* cercariae, pooled from six naturally infected *Lymnaea luteola* collected in March and April from in and around Jabalpur (Agrawal *et al.*, 1984).

Environmental Influences

Temperature

Several authors like Van der Schalie and Berry (1973); El-Hassan (1974); Harris and Charleston (1977); Parashar *et al.*, (1983) found that temperature affects the growth and reproduction of fresh water snails. *Indoplanorbis exustus* Deshayes (1984), a common fresh-water snail inhabiting south and southeast Asian countries, is an intermediate host of *Schistosoma nasalis* Rao (1932), *S. spindale* Montgomery (1906) and *S. indicum* Montgomery (1906) (Malek and Cheng, 1974). In tropical countries like India, populations of this snail are exposed to low temperatures from December to February, but the effects of this are not known. The present study examines the effect and after-effect of low temperature exposure on growth, reproduction and survival of two age groups of *I. exustus* under laboratory conditions.

The temperature at which the host lives can influence the likelihood of being parasitized. Most helminths have transmission stages that must survive and in some cases, develop outside a host. Clearly, ambient conditions would be of great importance to these life history stages.

Sankurathri and Holmes (1976) reported that high temperatures associated with thermal effluents had a negative effect on two subspecies of *Chaetogaster limnaei*, an oligochaete associated with the gastropod Phyla Gyrina. These annelids were observed to eat larval stages of the trematode *Echinoparyphium recurvatum*, and a decrease in annelids coincided with an increase in *E. recurvatum* infection. Temperatures external to the host can affect both the state of the parasite and its effect on the host, even where homeothermic hosts are concerned.

Novak (1979) showed that the cestode *Mesocestoides corti* in mice exhibited greater biomass at colder temperatures. Also in mice, the cestode *Hymenolepis microstoma* was heavier, produced more eggs, and reached patency (egg production) sooner at colder temperatures (Evans and

Novak 1983). This tapeworm also influenced the weight of the liver (as did temperature and, to some degree, host sex) and the weight of the bile duct (Evans *et al.*, 1985) for effects of temperature and *Hymenolepis nana* infection on mouse small intestine).

In a field study of habitat variables and parasitism, Pence and Windberg (1984) found extrinsic factors such as seasonality to be influential in populations of coyote parasites.

Holmes (1979) examined the effects that biotic influences on hosts might have on their parasites. He noted that not all hosts were of equal quality where a parasite was concerned and delineated the following three types of hosts: 1) required - the parasite is well-adapted to this host, which is necessary for its survival; 2) suitable- the parasite can mature in this host, but cannot maintain its population with this host alone; and 3) unsuitable- the parasite can establish, but cannot reproduce.

Holmes (1979) observed that because parasites can often be exchanged among host, the host community composition should influence that of the parasites, and that such current ecological conditions might obscure historical host-parasite associations (Holmes, 1983).

Clearly, external conditions - both biotic and abiotic - and their effect on trophic interactions can influence parasites tucked away and unseen within their hosts. The castration of a host by its parasite is one of the more dramatic ecological effects that parasites can have and its impact has been considered in the contexts of individual evolutionary strategy (e.g. Baudoin 1975, Sousa 1983, Minchella, 1985) and population influences (Obrebski, 1975).

Kuris (1974) considered the trophic role of a parasitic castrator and stated it was similar to that of a parasitoid, among other things, "killing" one host per parasite.

Baudoin (1975) noted that parasitic castration could enhance parasite fitness through the increase in host growth and survivorship that resulted from reduced reproductive effort. In addition, parasitic castration has a variety of effects other than reduced reproduction. These can include

alterations in internal chemistry, secondary sexual characteristics, internal organs, and sexual/parental behavior.

Minchella (1985) discussed possible molluscan responses to parasitism and asked why resistance to infection was not more common. Minchella and LoVerde (1983) determined that in the presence of susceptible snails and schistosome miracidia (the infective stage), resistant snails exhibited lower reproductive success. Minchella (1985) then noted that the evolution of resistance depended not only on the presence of appropriate genotypes, the effect of the parasite and the probability of encountering it, but also on the cost of resistance as compared to those factors. Other responses are available to molluscs, and these include fecundity compensation (early reproduction prior to castration) and gigantism, which he expected to encounter as a host adaptation primarily in long-lived snails with some possibility of outliving the infection.

Water quality

Because many factors can alter water quality, it is unlikely that a generalization about the interaction between water quality and parasitism will emerge (Khan, 1991; Khan and Thulin, 1991; Poulin, 1992; MacKenzie *et al.*, 1995; Lafferty, 1997).

However, McDowell *et al.*, (1999) predicted that pollutants may reduce the immunological capabilities of hosts, rendering them more susceptible to some parasites. Gill ciliates of fish are a good example of this effect. Prevalence of ciliates increase with oil pollution (Khan, 1990), pulp mill effluent (Lehtinen, 1989; Axelsson and Norrgren, 1991; Khan *et al.*, 1994), industrial effluent (Dabrowska, 1974; Overstreet and Howse, 1977; Vladimirov and Flerov, 1975), and thermal effluent (Esch *et al.*, 1976; Nilsen, 1995). This appears to be due to an increase in susceptibility because toxic conditions compromise a fish's immune system (Khan, 1990).

Khan (1987) agreed that the pathology that seems most directly causal to the success of these with ciliates is a toxicant's ability to impair mucus production—a fish's main defense against gill parasites. In contrast to the above example, altered water quality may improve conditions for parasites if their host's density increases.

Eutrophication and thermal effluent

It often raises rates of parasitism because the associated increased productivity can increase the abundance of intermediate hosts.

Beer and German (1993) described how eutrophication improved conditions for snails that, when combined with a thriving population of urban-adapted mallards (escaped from local farms), fueled the life cycle of the digene, *Trichobilharzia ocellata*. This avian blood fluke causes outbreaks of swimmer's itch (schistosome dermatitis).

Similarly, Valtonen *et al.*, (1997) described how eutrophication among lakes and over time was associated with greater overall parasite species richness in two fish species.

Eure and Esch (1974) found that acanthocephalans were more common near thermal effluent because intermediate hosts were able to retain high densities in winter.

Sankurathri and Holmes (1976) showed that thermal effluent altered the normal seasonal dynamics of a digenean community, essentially providing summer conditions so that transmission could continue uninterrupted year-round. Warm temperatures prevented icing over of the lake and fueled primary productivity.

The warm water and increased vegetation extended the breeding season and provided extra food and habitat for snail populations that served as intermediate hosts for several species of digenes. Because it was ice-free, the area attracted migrating birds—the definitive hosts of most of the digenes. In addition, the warm water was unfavorable to a normally abundant oligochaete worm that is commensal with the snail host and interferes with the free-living stages of the digene's life cycle. The main consequence of this was an increase in the total prevalence of digenes in snails and (presumably) other hosts.

Crude oil and heavy metals

Khan (1987); Lafferty (1997) recorded that the effect of certain pollutants varies strongly among parasites. Crude oil increases parasitism by some species of ciliates, nematodes, and monogenes,

yet decreases parasitism by acanthocephalans and digenes. Perhaps the negative effects of crude oil on some invertebrates that serve as intermediate hosts for these parasites reduced their abundance. The effects of sewage sludge are even more difficult to predict (Lafferty, 1997). Acid rain may also affect parasites and their hosts in complex ways (Leavitt *et al.*, 1999; Niyogi *et al.*, 1999).

Marcogliese and Cone (1996) found that parasite species richness in yellow eels (*Anguilla rostrata*) from Nova Scotia was 4.0 per eel at buffered sites, about 2.5 per eel at moderately acidified sites, and 2.0 per eel at the most acidified sites. Separating this effect by parasite group indicated that while the prevalence of monogenes and digenes (and their snail hosts) decreased with acidity, acanthocephalans may have increased with acidity and tapeworms and copepods were relatively unaffected (Lafferty, 1997).

Toxic chemicals and trace metals have a relatively consistent negative effect on intestinal helminths (Lafferty, 1997). These pollutants may be analogous to a drug that kills the parasite but does not kill the patient.

Selenium, for example, is more toxic to tapeworms than to their fish hosts (Riggs *et al.*, 1987). A pollutant may also kill sensitive free living stages of the parasite (Evans, 1982). For example, trace metals in sewage sludge reduced survival of free-living cercariae and miracidia, leading to a lower prevalence of the digene, *Zoogonoides viviparous*, in snails (Siddall and Des Clers, 1994). In other cases, parasitic infection may make the host more susceptible to toxins (Guth *et al.*, 1977; Stadnichenko *et al.*, 1995).

Brown and Pascoe (1989) found that exposure to cadmium killed 94% of the amphipods infected with larval acanthocephalans compared to a 14% mortality rate of uninfected amphipods. Although the effect of trace metals is often to reduce parasitism, this does not imply that trace metals provide a benefit for aquatic populations.

Habitat alteration

Heyneman (1979) found that altering habitats can favor vectors and intermediate hosts and lead to an increase in human diseases. Several environmental disasters ensued after Egypt built the

Aswan Dam in 1971 to provide inexpensive electricity for city dwellers and irrigation for greatly expanding farm land. The annual floods along the Nile ceased, starving the river farmlands of fertile sediments, the Mediterranean sardine fishery collapsed, and most segments of the river slowed and offered better habitat for the snails that serve as the intermediate host for the flukes that cause human schistosomiasis. The snails and their flukes spread along the river and around the newly formed Lake Nasser and its associated irrigation canals, causing serious human morbidity. This epidemiological disaster has occurred repeatedly including Lake Volta in Ghana and most recently and spectacularly at Richard Toll in Senegal in 1988 (Gryseels *et al.*, 1994), producing perhaps the most intense focus of transmission known. At some impoundments it is likely that the cold, clean running water at the dam outflow has created ideal conditions for blackfly larvae and attendant river blindness because the adult black- flies serve as filarial vectors.

Parasite-Host interactions

Behavioral Alterations

The emphasis of this research is on trophic interactions, and perhaps the most unusual way that parasites influence trophic interactions is by affecting the availability of their preyitem/intermediate hosts (hosts that harbor a juvenile parasite that must be eaten in order to reach adulthood in the predator/final host). In so doing, they may enhance their own probability of transmission to the final (definitive) host.

Holmes and Bethel (1972) delineated four ways in which parasites might take advantage of definitive host foraging behavior and thus enhance transmission: they may alter host stamina, increase host conspicuousness by changes in appearance, disorient the host as a result of neurological damage, and alter responses to environmental stimuli.

More recently, Radabaugh (1980a) has shown that social (schooling) behavior in minnows is affected by a trematode that encysts in the brain (Radabaugh, 1980b). Infected minnows may form less compact schools and may be more vulnerable to piscivorous birds. When responses to

environmental stimuli are altered, distributions of hosts may be affected, with infected animals moving into areas of increased predator-prey encounter (Holmes and Bethel, 1972).

Moore and Lasswell (1986) showed that when infected with an immature nematode (*Dispharynx nasuta*), the isopod *A. vulgare* exhibited altered behaviors other than those accompanying acanthocephalan infection. They tentatively concluded that in at least one of these host-parasite associations (acanthocephalan or nematode), the changed behavior was peculiar to the association and not a generalized isopod response to infection. Other examples of parasites that alter invertebrate host behavior include the larva of the trematode.

Rau (1983a, b, 1984a) discovered that mice infected with *Trichinella spira* are less active and more likely to exhibit subordinate behavior that could lead to predation and parasite transmission. Different predators may be involved in this parasite's life cycle. The cestode plerocercoid of Schistocephalus affects the respiration of its stickleback intermediate host. These fish require more oxygen, move to shallower water, and may be easier prey for piscivorous birds (Lester, 1971). The swimming abilities of some intermediate hosts are impaired by the trematode, *Nanophyetus salmineola*, with a similar possible result (Butler and Millemann, 1971).

Changes are not always specific, however, and Brassard *et al.*, (1982) found that Diplostomum increased guppy susceptibility to brook trout, which are not suitable hosts for the parasite. They speculated that such a risk may be part of the cost of lowering guppy activity levels and thus possibly enhancing general predation, including that from birds. Conspicuous intermediate host appearance is another way that predation risk may be increased (Holmes and Bethel, 1972).

Oetinger and Nickol (1982) have shown that there are differences in ommochrome fractions between uninfected *Asellus intermedius* and *A. intermedius* infected with *Acanthocephalus dirus*. These differences support the idea that the acanthocephalan may compete with the isopod for compounds used in ommochrome synthesis, thus resulting in pigment dystrophy.

Brattey (1983) has recently reported that *Asellus aquaticus* exhibits darkened respiratory opercula when infected with *Acanthocephalus lucii*. In his laboratory studies, perch ate a

disproportionately large number of infected *A. aquaticus*. The evolutionary history of behavioral changes associated with parasitism is unclear. Holmes and Bethel (1972) considered alteration of host behavior to be a parasite strategy.

Smith Trail (1980) hypothesized that under some conditions, behavioral changes could be a kinselected host adaptation. Through suicidal behavior, the host individual could eliminate the parasite from the host population. Parasite transmission might later be favored by such suicide. Moore (1984a) argued that this was unlikely in the case of acanthocephalans. She noted that in at least one case, (Bethel and Holmes, 1974) the behavioral switch does not occur until the acanthocephalan is infective for the final host, a trait that clearly benefits the parasite. In addition, many intermediate hosts that suffer altered behavior when infected with acanthocephalans are not thought to occur near their kin, and some of them are reproductively functional, thus increasing the cost of suicide considerably (Wilson, 1977).

Studying non-helminth parasites, Stamp (1981) found the behavior of gregarious caterpillars containing parasitoids to be inconsistent with the host suicide hypothesis.

Freeland (1976) has suggested that primate social behavior, including aspects such as xenophobia, group size, infant handling, and sexual fidelity, may have been influenced by correlates of pathogen transmission. Current news sources speculate that human social behavior may be undergoing modification in response to risk of parasite acquisition.

Curtis (1985) showed that after the breeding season the carrion response of the snail, *Jlyanassa obsoleta*, was affected by a variety of trematode larvae. This was not thought to be related to transmission efficiency, as the snails were first intermediate hosts for the parasites, which will actively leave the snails in search of the next host. Compared to uninfected conspecifics, *Helisoma trivolvis* infected with *Echinostoma revolutum* is less attracted to food (Boland and Fried, 1984), and exhibits reduced activity. Female grasshoppers infected with *Nosema locustae*, a protozoan, consume less food than conspecifics (Oma and Hewitt, 1984). For general reviews of the physiological aspects of such host parasite interactions (Vinson and Iwantsch, 1980; Thompson, 1983; and Beckage, 1985). Host Distribution Parasites can affect the distribution of

their hosts on a variety of scales, both locally and globally, and not all of these involve parasite transmission probabilities or host foraging.

Curtis and Hurd (1983) showed that trematode parasites were an important component of the local spatial heterogeneity exhibited by the snail, *Ilyanassa obsoleta*. Because of large spatial differences in prevalence of trematodes, there were spatial differences in Ilyanassa reproductive output. "Heavily infected snails are little more than 'sacks' of parasites with especially gonads, and sometimes digestive glands, virtually absent" (Curtis and Hurd, 1983). Parasites can clearly affect the distribution of their hosts on a global scale. The meningeal worm, *Parelaphostrongylus tenuis*, is benign in whitetail deer, but pathogenic for some other cervids (Karns, 1967; Anderson, 1972). Range expansion of whitetail deer has resulted in decreases in other cervid species.

Barbehenn (1969) hypothesized that parasites may give a host a competitive advantage against other animals for which the parasite is more pathogenic (Cornell, 1974). Freeland (1983) expanded upon this idea at the community level and suggested that the notable phylogenetic, morphological and ecological differences among coexisting animals that are often attributed to competition might result from the influence of parasites. Such differences could act as barriers to parasite transmission and thus allow coexistence. There is certainly evidence that parasites can modify the effects of host species on one another (Holmes 1979, 1982; Holmes and Price, 1986), and Park's (1948) classic work on *Tribolium castaneum* and *T. confusum* showed clearly that parasites can reverse the outcome of competition between two species.

Price *et al.*, (1986) have recently reviewed the literature concerning ecological interactions that are mediated by parasites (broadly defined). They observed that the results of such "germ warfare" were unlike those of more direct interaction, in that the effects of parasites could be far greater than their cost to the original host and could be observed over large distances.

Population Effects

In a series of papers, Anderson and May have explored host-parasite interactions at the population level (Anderson and May, 1978, 1979; May and Anderson, 1978, 1979; May, 1983,

1985a). In their models, Anderson and May assumed that host mortality rates increase with parasite levels. Using this and other assumptions about transmission rates and effects of parasite density and distribution, they emphasized the importance of threshold host densities (for parasite persistence), host mortality rates, and, m the case of macroparasites, parasite reproductive rates in the regulation of host populations. They found at least three stabilizing influences in host-parasite relationships. First, some types of functional relationships between parasite-induced death and parasite burden are stabilizing; others are not. Second, this relationship is linear and if parasites are clumped in distribution, they may regulate host population growth. Finally, density-dependent constraints on parasite populations acting within the host can have a stabilizing influence. May and Anderson also discovered destabilizing influences, which included a negative effect of parasites on host reproduction, temporal lags between transmission and reproduction of parasites, and within host parasite reproduction that enhances parasite population size.

Anderson and May (1978, 1979) listed at least four factors that influence disease patterns: 1) the host as parasite habitat, 2) host mortality, 3) immunity, and 4) transmission. They noted that because of changes in virulence, the ability of a parasite to regulate the population of its host may vary over time. Cyclic variation in prevalence itself can result from stochastic fluctuations in prevalence or temporal variation in transmission.

May and Anderson (1978) indicated that the results of these models may be useful for public health consideration of chemotherapy. They further explored the role of immunity in host populations (Anderson and May 1985a), mathematically modeling helminth population dynamics in a host population whose individuals are heterogeneous in their ability to acquire immunity.

Holmes (1982) agreed with Anderson and May on many points, but argued that parasites, in many instances, were probably a source of compensatory mortality, not additive mortality, and stressed the need for more field studies.

Getz and Pickering (1983) also urged increased study of individual host-parasite associations. Holmes et al. (1977) stated that there were two prerequisites for population regulation: 1) reproductive activity must have the potential to increase population size, and 2) a feedback mechanism that prevents such increase must exist.

They cited Bradley (1972, 1974) in emphasizing transmission and immunity as important limits to parasite population growth. To this they added the influence of parasite-parasite competition. In a modeling/simulation exercise, they showed that a parasite supra-population might be regulated by the occurrence of competitive interactions in only one host species, provided the other species were inadequate to maintain the parasite population. As has been recognized, the host may influence infra-population size by a complete or partial immune response. Parasitologists have discovered even more subtle and unexpected host influences on parasite levels. Possibly related to host immunity is the phenomenon of arrested development, a condition seen in some nematodes that temporarily cease development within the host until some future time.

Schad (1977) reviewed this literature, and while he noted that arrested development might be controlled by host resistance or be a parasite response to adverse conditions, he emphasized its role in parasite population regulation. In the case of mammals, populations of parasites may also appear where they're least expected - in neonates - as a result of transmammary transmission from infected mothers.

Stone and Smith (1973) reviewed this literature, which deals primarily with nematodes. Campbell et al. (1980) documented relationships between the foraging habits of some deep sea fish species and their parasite fauna. They also noted that differences in host habitat and agerelated dietary shifts influenced this fauna, as did host community diversity. In a study of flatfish, Scott (1982) noted the roles of host diet, age, sex, and geographic distribution in parasite community composition.

Recently, Kennedy et al. (1986) have compared intestinal helminth communities in fish and birds (plus one mammal). They found the ectotherms to be relatively depauperate in both richness and

abundance, and concluded that a variety of factors contribute to helminth community diversity. While these factors certainly include breadth of host diet, as well as inclusion of intermediate host species in that diet, they also encompass the diversity of habitats visited by the host and the exposure to penetration by parasites that enter through non-dietary means. The increased energy requirements and intestinal differentiation of endotherms are yet other factors that may favor rich communities. Coevolution The evolution of virulence/avirulence is an active area of research emerging from evolutionary theory that is closely related to the work described in the previous section. This is in part because virulence and parasite reproductive rate are used synonymously, a habit that may give a parasitologist pause, for the effect that a parasite has on its host is often determined by factors far more numerous and complex than the number of offspring it produces. The reasoning behind the equation of these two terms is nonetheless logical: In the case of two parasites that share nearly identical biology and host-parasite attributes, the one that produces more offspring will probably demand more host resources.

Holmes (1983) discussed the following three models of host-parasite coevolution: 1) The mutual aggression model, in which the parasite increases exploitation and the host increases defense; 2) the prudent parasite model, in which parasites are favored that limit damage to the host; and 3) the incipient mutualism model, in which both parasite and host are selected to benefit one another.

In a narrow sense, parasites influence and participate in trophic interactions when they alter the encounter rates between predators and their prey or host feeding patterns. Beyond such alterations, they can affect the abundance and distribution of their hosts through effects on host reproduction, direct pathogenicity, or mediation of interactions between free-living organisms. As Holmes and Price (1986) observed, parasites both respond to and engender patchy biotic environments). Because many parasites are transmitted through host diets, they can be indicators of host foraging habits. Hosts, by their dietary preferences, can influence parasite community characteristics. Although parasites are often considered a trophic level, they are diverse in their nutritional requirements. The parasite interactions that may result from such requirements in a limited environment can, along with other factors, influence the array of parasites available to a host or host community and, by extension, the ecology of the host (s).

Infection of snails by nematodes

Snails serve as natural and parasitic hosts of nematodes. A good number of reports are available on the occurrence of nematodes in snails. Some of them are:

Richards and Merritt (1967) studied on *Angiostrongylus cantonensis* in molluscan intermediate hosts. He found first- stage larvae of this species from rat feces developed to infective third stage larvae in 20-26 species of freshwater mollusks. Numbers of larvae up to 2,000 perbsnail and survival of larvae for a year were observed in *Biomphalaria glabrata*. First-stage larvae were susceptible to drying but survived at least 3 weeks in freshwater and were infective to snails after at least 2 weeks in either freshwater or seawater. *B. glabrata* was routinely infected by ingestion of infected rat feces and first-stage larvae were observed penetrating the wall of the intestine of snails. Isolated third-stage larvae were susceptible to drying but survived up to 11 days in B. glabrata removed from water, larvae also remained alive 4 days after death of the snails. Rats became infected by drinking water containing third-stage larvae which were observed active in freshwater for up to a week.

Bartlett and Anderson (1985) studied the 3rd stage larvae of *Spiroxys contortus*, *Falcaustra wardi* and *Serpinema trisinosus* in acquatic snails *Lymnaea stagnalis* and the prevalence were 79.8%, 22.5% and 1.6% respectively, in 110 snails. Khromovas suggestion that snails are parasitic hosts of *S. contortus* was confirmed by these two authors. Snails were also likely paratenic hosts of *S. trispinosus*.

Skorping (1985) reported the *Lymnaea stagnalis* as experimental intermediate host for the protostrongylid nematode *Elaphostrongylus rangiferi*, a neurotrophic parasite of reindeer, *Rangifer tarandus*. All juvenile snails exposed to first- stage larvae of *E. rangiferi* were reported to be infected.

Chao et al., (1993) studied on the prevalence of larval helminth in freshwater snails of the Kinmen Islands, Thaiwan, Republic of China and reported third-stage larvae of *Parastrongylus cantonensis* in *Ampullarius canaliculatus* (5/103), *Sinotaia quadrata* (20/141), *Hippeutis umbilicalis cantor* (1/70) and *Gyraulus spirillus* (2/87) *Segmentina hemisphaerula* were not infected.

Seehabutr (2005) recorded nematodes *Rhabditis* sp. In alimentary tracts of giant African snail (terrestrial) (*Achatina fulica*) in Thailand, the nematodes of this species were also found in terrestrial snails (*Hemiplecta distinct*) and slugs (*Psrmarion* sp.).

Farahnak *et al.*, (2006 b) studied the nematode association with *Bellamya bengalensis* snail and evaluated its medical and veterinary importance in Khouzestan province (South west of Iran). From the total of *Bellamya* snails examined for nematodes, 27 (2.36%) snails were found to be infected with *Oionchus* nematode parasite. These results have been recorded for the time and showed the importance of *Bellamya* snails in the region.

Komalamisra et al., (2009) reported Pila ampullacea and Pomacea canaliculata, as new paratenic hosts of Gnathostoma spinigerum. Aquatic snails, Pila ampullacea and Pomacea canaliculata were experimentally found to be suitable paratenic hosts for advanced third-stage larvae (L3) of the nematode Gnathostoma spinigerum, the causative parasite of gnathostomiasis in humans. G. spinigerum (L3) were found to be encapsulated in the tissue of the snails foot and its organs. This was the first evidence to reveal that not only vertebrates but also invertebrates (snails) could serve as paratenic hosts to this parasite.

MATERIALS AND METHODS

The main aim of the present work is to study the ecobiological conditions prevailing in water of the different ecosystem in Bangladesh, various molluscs living in and around and the cercariae they harbor. In the present work various species of cercariae from various snails found in and around the ecosystem. In the present work host parasite relationship is studied and full attention was paid to study the effects of larval stages of trematodes on the first intermediate host i.e. gastropods. The effects of larval trematodes on the molluscan host depend upon four important factors: the degree of infection, size and age of host, the development pattern of the larvae and the nature of organ invaded.

The Study Area

Snails viz., *Bellamya bengalensis*, *Pila globosa* and *Brotia costula* were collected from different localities of Bangladesh. The collected samples were from different types of ecosystem:

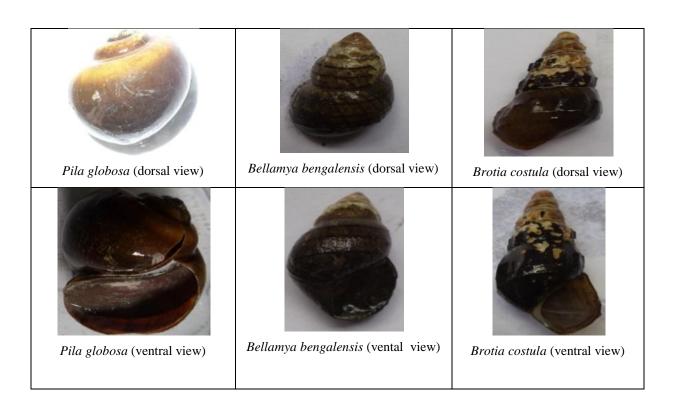


Figure 3.1 The studied snail samples.

Site 1: Dhaka and surrounding regions

(Ramna Park Pond, Dhaka; Curjon Hall Pond, Dhaka; Jagannath Hall Pond, Dhaka; Keranigonj Pond, Dhaka; Tongi Railway Station Pond, Dhaka; Jaydebpur Railway Station Pond, Gazipur; Bhawal-Gazipur Railway Station Pond; Salna BSMAU Pond, Gazipur; Mawna Bazar Pond, Gazipur; Tongi Bari Bazar Pond, Munshiganj; Lahajang Bazar Pond, Munshiganj).

Site 2: Khulna and surrounding regions

(Khulna University Pond, Khulna; Dacope Upazila Complex Pond, Khulna; Shyamnagar Upazila Complex Pond, Satkhira; Kaliganj Upazila Complex Pond, Satkhira; Bagerhat P.C College Pond; Fakirhat Bazar Pond, Bagerhat) and

Site 3: Kishoreganj and surrounding regions

(Solakia Bazar Pond, Kishorganj; Astagram Upazila Complex Pond; Kuliarchar Bazar Pond, Kishorganj; Mithamain Upazila Complex Pond; BAU Mymensingh Pond; Kabi Nazrul University Pond, Trishal).

The study period was belonging to July 2011 to June 2013 from the above mentioned locations.

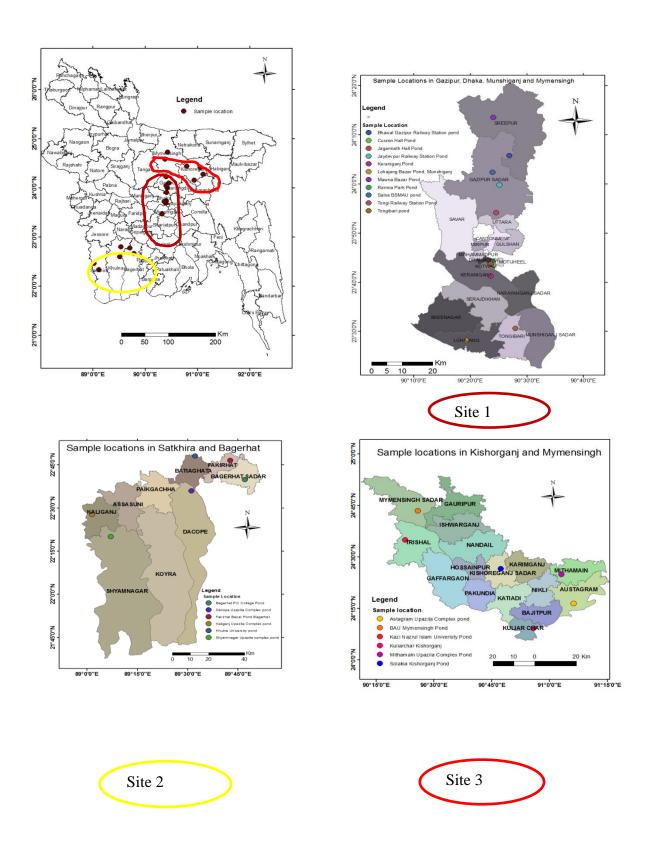


Figure 3.2 Location map of the study areas.

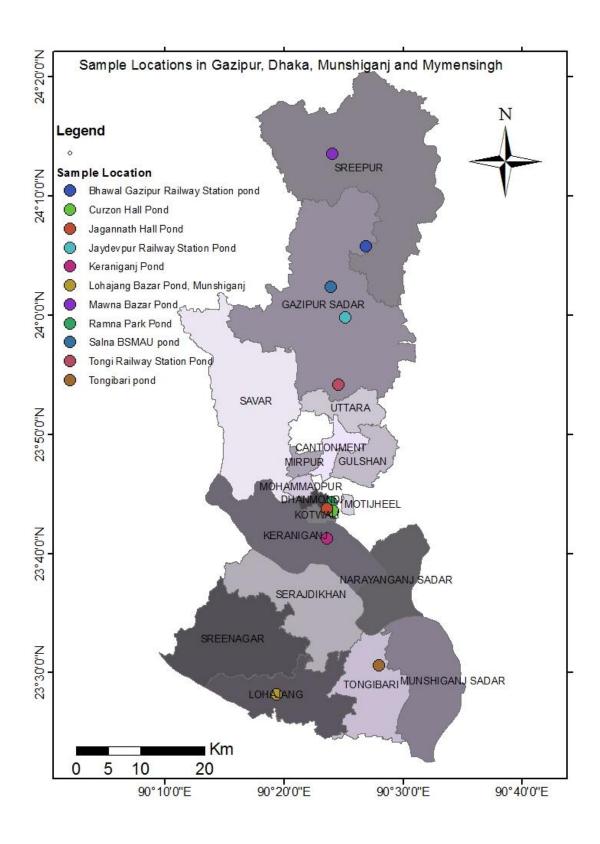


Figure 3.3 Location map of Site 1: Dhaka and surroundings.

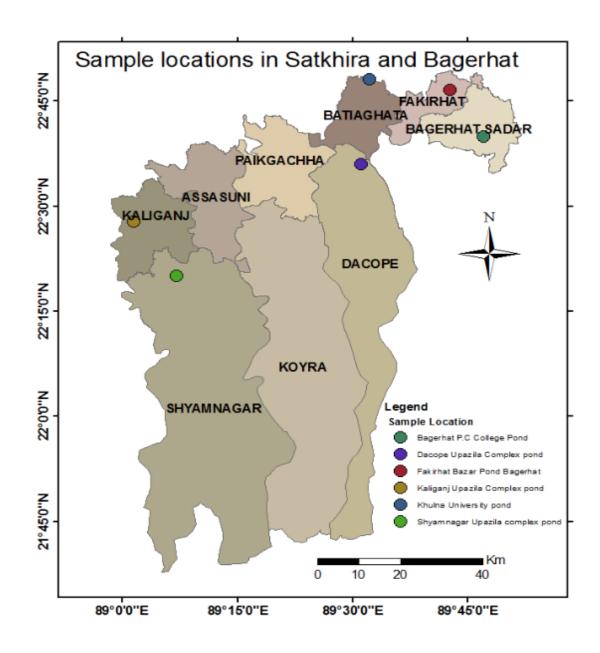


Figure 3.4 Location map of Site 2: Khulna and surroundings.

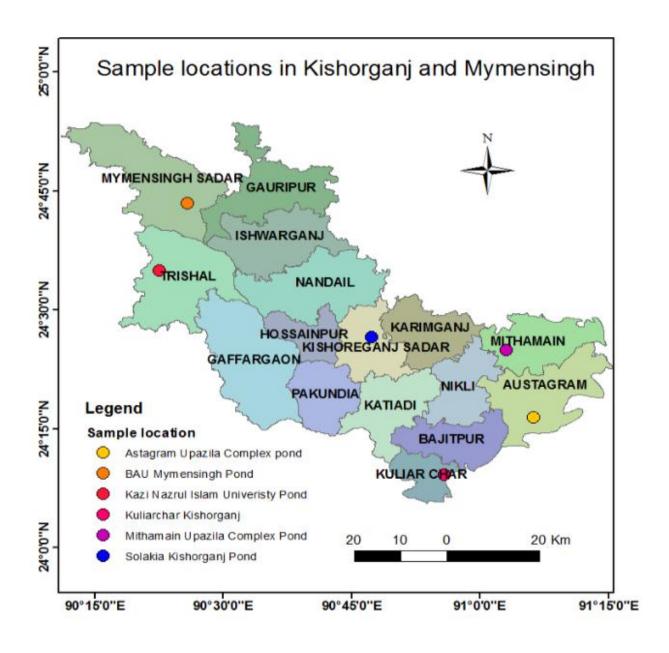


Figure 3.5 Location map of Site 3: Kishoreganj and surroundings

 Table 3.1 Geo-reference of the study areas.

Serial	Sampling Site	Latitude	Longitude
Site 1: Dhaka and surroundings			
1	Ramna Park Pond, Dhaka	23°73′75.3′′N	90°40′0.37′′E
2	Curjon Hall Pond, Dhaka	23°72′59.6′′N	90°40′26.4′′E
3	Jagannath Hall Pond, Dhaka	23°72′92.4′′N	90°39′35.1′′E
4	Keranigonj Pond, Dhaka	23°68′78.7′′N	90°39′32.4′′E
5	Tongi Railway Station Pond, Gazipur	23°90′24.4′′′N	90°40′93.1′′E
6	Jaydebpur Railway Station Pond, Gazipur	23°99′65.1′′N	90°41.87.4´´E
7	Bhawal-Gazipur Railway Station Pond	24°09′61.9′′N	90°44′80.3′′E
8	Salna BSMAU Pond, Gazipur	24°03′96.9′′N	90°39′84.4′′E
9	Mawna Bazar Pond, Gazipur	24°22′58.1′′N	90°40′12.5′′E
10	Tongi Bari Bazar Pond, Munshiganj	23°30'36.52"N	90°28'0.34"E
11	Lahajang Bazar Pond, Munshiganj	23°46′96.3′′N	90°32′35.4′′E
Site 2: Khulna and surroundings			
1	Khulna University Pond, Khulna	22°48'1.38"N	89°32'9.04"E
2	Dacope Upazila Complex Pond, Khulna	22°35'56.79"N	89°31'2.39"E
3	Shyamnagar Upazila Complex Pond, Satkhira	22°20'2.09"N	89° 7'5.35"E
4	Kaliganj Upazila Complex Pond, Satkhira	22°27'48.56"N	89° 1'29.21"E
5	Bagerhat P.C College Pond	22°39'51.86"N	89°46'56.84"E
6	Fakirhat Bazar Pond, Bagerhat	22°46'32.92"N	89°42'35.27"E
Site 3: Kishoreganj and surroundings			
1	Solakia Bazar Pond, Kishorganj	24°26'32.81"N	90°47'22.88"E
2	Astagram Upazila Complex Pond	24°16'24.31"N	91° 6'12.91"E
3	Kuliarchar Bazar Pond, Kishorganj	24° 9'8.53"N	90°55'50.73"E
4	Mithamain Upazila Complex Pond	24°24'53.93"N	91° 3'2.94"E
5	BAU Mymensingh Pond	24°43'22.23"N	90°25'47.46"E
6	Kabi Nazrul University Pond, Trishal	24°34'55.65"N	90°22'32.91"E

Sampling Design

Whether there was any seasonal effect on the parasitization aspects of snails, three seasonal practically, summer (March-June), rainy season (July-October) and winter (November-February) was as rule sampled once in a month. Monthly collections of snails were made from July 2011 to June 2012. Sampling was carried out using a wire net of aperture 33 cm in length and width and 8 cm in depth (WHO, 1965). At each site, sampling was carried out along a 200m length of the pond, using 10 dips per site.

The various physico-chemical factors of water were studied. The parameters of the water constituents are pH, temperature, Electrical conductivity (EC) and Dissolved oxygen (DO) and climatological factors as temperature and rainfall. The study is correlated with the occurrence of snails and cercariae in and around the three sites.

Depth of water, water quality parameters, nature of bottom and other ecological conditions were recorded details. Each time, water temperature, pH, dissolve oxygen (DO), electric conductivity were measured. pH by pH meter (HANNA, HI 98106), Temperature, EC and TDS by EC metre (p^{Hep}, H₁ 98107 by HANNA), Dissolved Oxygen by Digital Oxygen Meter (Lutron DO 5508) at site during sample collection of the water were determined.

The water samples were collected at each site at every collection in a two liter container to study the different parameters at *in vitro* conditions. Only the fresh water samples were used for the analysis of physico-chemical parameters analysis. This study was carried out within 2-4 hrs of sampling to get the original value without much variation. From the table, it is observed that the pH of all the sampling sites remain constant as 7-8 without any change. The temperature is also the same for all the sampling sites with slight variation ranging from 26°C to 28°C with a variation of 2°C. The reason for a low temperature is due to the collection timings i.e between 7 am to 8 am early morning.

The freshwater molluscan snails collected, their yearly seasonal occurrence and percentage of infection is observed throughout the year. The various digenetic larval stages such as metacercariae, rediae, sporocysts and cercariae were observed.

Sampling procedures

Collection and Maintenance of snails

Studies on cercariae commenced with collection of first intermediate laboratory. The snails transfer to glass water bowls and well aerated acquires already provided with a rich water plants. After a short period of acclimatization the snails transfer to individual Petridis in order to detect the cercariae. The collected snails sample either hand-picked or dragging a net through water and transported to the laboratory with the same pond water from which they were collected.

A rectangular grid of about 0.7m (2m from the margin) depth and of 5m length (that is horizontally) was selected on each water body. In each sampling time samples were drawn according to the following design: a dredge net was dragged in a zigzag fashion along that grid and the bottom samples were poured into big bucket. Grab water samples from middle point of each pond of the plug-flow and 50 centimeter below the surface will be collected. The sample will be collected in pre-sterilized (washing by 0.01N H₂SO₄ solution) one litter plastic bottles and preserved with 40mg HgCl₂/l.

Identification of the larval stages

This part deals with morphology, anatomy and systematic position of cercariae belonging to Parapleurophocercous, Pleurophocercous, Xiphidiocercariae, Echinostome, Amphistome, Renicolid, Cotylomicrocercous and Furcocercus cercariae. Matured molluscan snails worked as intermediate hosts are selected for observing the effect of larval stages of trematodes on the hepatopancreas, viscera, foot and shell. The identification was followed by published articles of home and abroad.

Procedures

The parasitic infections with trematode cercariae were studied by using shedding and crushing methods. The cercariae were collected in dechlorinated water and observed for their swimming behavior (Krailas *et al.*, 2003). The occurrences of sporocysts and /or rediae were examined under a dissecting microscope. Examination of snails for larval trematodes was carried out as described by Preston (1915); Saliba *et al.*, (1978) and Rao (1989). All the cercaria was morphologically identified with the help of standard sample and manual prepared by Dechruksa *et al.*, 2007. Observations on recovered larval trematodes were made on live, unstained or vitally stained (0.5% neutral red or 0.5% brilliant cresol blue) specimens as well as on specimens fixed either in formalin-acetic acid-alcohol (FAA) solution or in 70% ethanol, and subsequently stained in acetocarmine.

Measurements were taken on a minimum of 10 specimens of live and fixed larval trematodes. Drawings were made with the aid of a camera lucida or freehand, from preparations examined under a light microscope. The behavior of cercariae was observed using a dissecting microscope (Olympus BH2).

The host name, locality, date of collection and the incidence of infection are given at the beginning of each description. Routine methods for the collection and maintenance of snails, and the study of cercariae (Haseeb and Khan, 1984) were employed.

In brief, naturally emerged cercariae were examined in temporary mounts, with and without neutral red staining. Cercariae were fixed in 5% formalin (70~ 0 C) and stained with aceto-alum carmine (Southwell, 1930). Hand drawings of fixed specimens through sophisticated microscope were done from observations on living cercariae. Measurements from at least ten fixed specimens of each species are presented in millimeters as ranges followed by means in parentheses.

Collection of cercaria

The snails were carried in bucket to the Department of Zoology (Parasitology branch). The collected snails kept under observation at for some time. The snails which are fully grown showed larval infection while the young ones normally free from larval infection. For study of cercariae heavily infected snails were selected. Two methods were used for the morphological observations.

- 1. Natural emerging method: In natural emerging method, the snails were kept in separate test tubes. This was a constant source of living cercariae naturally emerging from the snails. The sunlight and artificial light play an important positive role in stimulating the emergence of cercariae.
- **2. Crushing method:** This method of investigation of cercariae collection found suitable for morphological observations on various developmental stages such as sporocysts and rediae. This quick method was useful for studying the seasonal percentage of infection of cercariae. The cercariae collected were subjected to various artificial methods for the study of various internal structures.

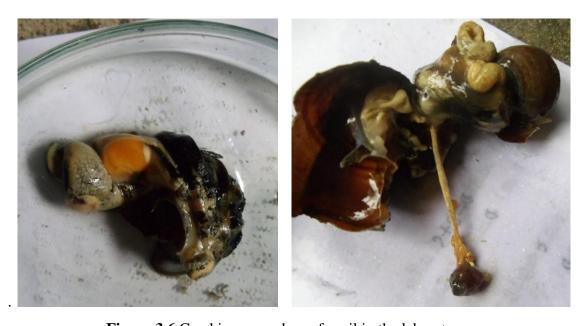


Figure 3.6 Crushing procedure of snail in the laboratory.

- **3. Movement relaxation:** Sometimes cercariae were found to be so active that observation under high power was impossible without some method interfering with or controlling their movements. Sometimes dilute solutions of gum, starch, gelatin were used to slow down their movements.
- **4. Vital stains:** For the study structural details in live condition, vital stains were used such as neutral red, methyl green, Nile blue, Asur II and Nile blue sulphate. For the preparation of permanent mounts, the cercariae were fixed in 1% formalin, stained in Delafield's haematoxylin, cleared in clove oil and mounted in D.P.X.
- **5. Measurement:** Most of the specimens were measured in live state. The diagrams have been made with the aid of a camera lucida. Sketches were drawn at different magnifications using oil immersion objective if necessary.

Precautions, Preservation and Laboratory investigation

All the molluscs thus collected were then examined for developmental stages. Snails were brought alive to the laboratory and were placed individually in 10 ml test tubes, each tube filled to its half volume with dechlorinated tap water. After the collection, the individuals were exposed to diffuse sunlight in glass beaker of 100 ml size. After a couple of hours the water of the beakers was examined carefully under microscope for the occurrence of cercariae.

Sometimes to liberate cercariae, the snails were exposed to artificial light for about two hours. Water in each tube was examined under the microscope for the presence of cercariae. Snails which did not shed cercariae were checked weekly for release of cercariae up to the end of the fourth week of collection. Snails that failed to shed cercariae were crushed between two slides and examined to confirm the absence of infection. Two indices were used to characterize the community in terms of species composition. The indices were used for species richness and Simpson diversity.

The snails, which shed cercariae, were isolated and the others were dissected at the end of the investigation to conform the absence of any helminth infections. The shell of individual snail was broken with tweezers; the mantle, the digestive gland and the viscera in general were then sliced under a binocular microscope into small fragments for examination. Each collected parasite was then fixed, in slightly warm glycerine alcohol, the medium in which it was also mounted.

Statistical analysis

The Prevalence was calculated as the percentage of snails harbouring trematode larvae according to Bush *et al.*, (1997). One way ANOVA and Kruskal-Wallis statistics were used for statistical comparison for normal distributed data and for nonparametric data, respectively, and results were considered significant at P<0.05. Spearman rank correlation coefficient was calculated to determine the correlation between trematode prevalence per season per host species and snail host abundance. Testing of effects of both individual and interacted factors (Site, season and infection) on snail abundance was statistically analyzed using the General Linear Interactive Model (GLIM). All the statistical tests were performed by using the software packages SPSS 12.0.0 (USA).

The following statistical parameters were used in the present research:

1) Sampling

Sampling is a procedure by which one or more members of a population are picked from the population.

Random Sample: There are several methods for constructing random samples—the present research considers only simple random samples. This process operates so that each member of the population has an equal chance of being selected into the sample.

The program outputs the sample sizes required to estimate the true value with the desired precision and confidence, for both an infinite population and for a population of the specified

size. If population size is left blank or zero, only the sample size for an infinite population is calculated.

Sample size is calculated using the formula:

$$n = (Z^2 \times P (1 - P))/e^2$$

Where Z = value from standard normal distribution corresponding to desired confidence level (Z=1.96 for 95% CI)

P is expected true proportion

e is desired precision (half desired CI width).

For small populations n can be adjusted so that $n(adj) = \frac{Nxn}{N+n}$

2) Prevalence

Prevalence is a term which means being widespread and it is distinct from incidence. Prevalence is a measurement of *all* individuals affected by the disease at a particular time, whereas incidence is a measurement of the number of *new* individuals who contract a disease during a particular period of time.

Prevalence=(Number of infected host)/(Total number of host examined)x100.

3) Mean intensity

This is the mean number of parasites found in the infected hosts (the zeros of uninfected hosts are excluded). Since sample size and prevalence are known, mean intensity defines the quantity of parasites found in the sample of hosts.

4) Standard Error of the Mean

The population that includes all possible samples of size n is a long list of numbers and the variance for these numbers can, in theory, be calculated.

The square root of this variance is called the standard error of the mean. It is simply the standard deviation for this population of means.

$$\sigma_{\bar{x}}^2 = \frac{\sigma^2}{n}, \sigma_{\bar{x}} = \frac{\sigma}{\sqrt{n}}$$

5) Pearson Correlation Coefficient

The Pearson correlation coefficient is used to measure the strength of a linear association between two variables, where the value r = 1 means a perfect positive correlation and the value r = -1 means a perfect negative correlation. So, for example, you could use this test to find out whether people's height and weight are correlated (they will be - the taller people are, the heavier they're likely to be).

Requirements for Pearson's correlation coefficient

- Scale of measurement should be interval or ratio
- Variables should be approximately normally distributed
- The association should be linear
- There should be no outliers in the data

Equation

$$r = \frac{\sum_{i} (x_i - \overline{x})(y_i - \overline{y})}{\sqrt{\sum_{i} (x_i - \overline{x})^2} \sqrt{\sum_{i} (y_i - \overline{y})^2}}$$

A One-Way Analysis of Variance is a way to test the equality of three or more means at one time by using variances.

6) Standard Deviation

The standard deviation is the square root of variance. Thus the way we calculate standard deviation is very similar to the way we calculate variance.

In fact, to calculate standard deviation, we first need to calculate the variance, and then take its square root.

But here we explain the formulas.

The symbol for Standard Deviation is σ (the Greek letter sigma).

This is the formula for Standard Deviation:

$$\sigma = \sqrt{\frac{1}{N} \sum_{i=1}^{N} (x_i - \mu)^2}$$

 σ = standard deviation

xi = each value of dataset

x (with a bar over it) = the arithmetic mean of the data (This symbol will be indicated as mean from now)

N = the total number of data points

 $\sum (xi - mean)^2 = The sum of (xi - mean)^2 for all datapoints$

Microphotography

Initial drawings of cercaria were made with the aid of freehand, from preparations examined under a light microscope. The behavior of cercariae was observed using a dissecting microscope (Olympus BH2). The final photographs were taken by a sophisticated microscope (Olympus BH 2).

Hypothesis

In Helminthes parasite, the trematode parasites complete their life cycle in two or three host. The first is vertebrate definitive host, second and third is invertebrate intermediate host. Here the Molluscan host is second intermediate host and their larval development and multiplication is in the hepatopancreas of the Molluscs.

Limitation of the study

This study was carried out to determine the prevalence of trematode parasites seasonally but the study doesn't reveal why some parasites were more predominant and others were not. This study is limited to certain parameters and some of the parts of the study were left untouched due to time and cost factors so that future researchers can elaborate this study by approaching the untouched portion.

SYSTEMATICS AND TAXONOMY OF INFECTIVE CERCARIA

The systematic description

The systematic description of the examined snail species are below:

a) Bellamya bengalensis

Kingdom	Phylum	Class	Order	Family
Animalia	Mollusca	Gastropoda	Architaenioglossa	Viviparidae

Scientific Name: Bellamya bengalensis Lamarck, 1882 (Fig.3.1).

Taxonomic note: Brandt (1974) recognized *Bellamya bengalensis* as *Filopaludina* (*Filopaludina*) bengalensis whilst Nesemann et al., (2007) recognize B. bengalensis as *Bellamya* (*Filopaludina*) bengalensis (Lamarck, 1822).

Based on the shell characteristics 22 forms have been recognized (Annandale, 1921) including *B. bengalensis* f. *balteata*; *B. bengalensis* f. *typica*; *B. engalensis* f. *annandalei*; *B. bengalensis* f. *mandiensis*; *B.bengalensis* f. *colairensis*; *B. bengalensis* f. *doliaris*; *B.bengalensis* f. *nepalensis* and *B. bengalensis* f. *eburnea*.

Bellamya bengalensis is a widely distributed species and it is used as food. Even though it is highly harvested in some parts of its range, it is a very common species and current levels of use cannot be considered as a threat. It is assessed at Least Concern.

Geographic range: The species is widely distributed all over the southern Asia region; Pakistan, Iran, Bangladesh, Nepal, Pakistan, Myanmar, Sri Lanka, and throughout India.

Habitat and ecology: This species is present in almost all types of lowland water bodies, mainly stagnant water and low saline water resources such as rivers, streams, lakes, ponds, wetlands, marshes, ditches, paddy fields, etc. According to (Ramakrishna and Dey, 2007), it can tolerate a maximum level of salinity of 0.2mg/l.

b) Pila globosa

Kingdom	Phylum	Class	Order	Family
Animalia	Mollusca	Gastropoda	Caenogastropoda	Ampullariidae

Scientific name: Pila globosa Swainson, 1822 (Fig.3.1).

There is a lot of confusion in the nomenclature of apple snails: the family name is often referred to as Ampullariidae, sometimes as Pilidae (invalid).

In 1758 Linnaeus, who regarded the Ampullariidae as terrestrial, referred *Pila ampullacea* to the genus *Helix*. His assumption was based on the fact that he never saw a living example; he only got a shell and thought it to be a terrestrial snail. In 1797, Humphrey attempted to erect the generic name *Pomus* and described 5 species in this genus. In 1798, Röding proposed the generic name *Pila* (with *Helix ampullacea* as the type species designated by Dall (1904).

This genus description did fulfil the requirements of the nomenclature rules, although it was later restricted to the Old World Ampullariidae, with the exception of the West African genera *Saulea* (Gray, 1867) and *Afropomus* (Pilsbry and Bequaert, 1927).

In 1799 Lamarck used the name *Ampullaria* for a single species (*Helix ampullacea*), making it the type species for *Ampullaria* by monotype. However, Dall (1904) designated *Helix ampullacea* as the type species of *Pila* rendering *Ampullaria* Lamarck, 1799 a junior objective synonym of *Pila* Röding, 1798, despite his designation of *Nerita urceus* Müller, 1774 as the type of *Ampullaria*.

Ecology and Habitat: Apple snails are tropical and sub-tropical freshwater snails from the family Ampullariidae (sometimes referred to as Pilidae). The Ampullariidae are divided in several genera. The genera *Asolene, Felipponea, Marisa,* and *Pomacea* are the New World genera (South America, Central America, the West Indies and the Southern U.S.A.), while the genera *Afropomus, Lanistes* and *Saulea* are found in Africa. The genus *Pila* is native in both Africa and Asia.

The shell is globose with an oval opening. In contrast with *Pila ampullacea*, *Pila globosa* has a large and deep umbilicus. The colour varies from olive green to grey green with a tinge of

red. A large number of variations are known. The interior of the shell is dull reddish with very faint spiral bands visible, white at the columella.

c) Brotia costula

Kingdom	Phylum	Class	Order	Family
Animalia	Mollusca	Gastropoda	Architaenioglossa	Ampullariidae

Scientific name: *Brotia costula* Rao, 1989 (Fig.3.1).

Brotia costula was described as Melania costula Rafomesque, 1823 and later assigned to Brotia (Antimelania) costula (Subba Rao, 1989).

The Basic Lifecycle of the Major Groups of the Digeneans

Most digeneans are hermaphroditic (the major exception being the schistosomes, and one other group). In the majority of these parasites self-fertilization may occur, but cross fertilization between different individuals is more generally the rule. The sperm enter the female system, either via the Laurers canal or more commonly through the common genital atrium, which opens into the uterus.

1) The Digenean Trematode Egg

The formation of the digenean egg follows that described for the platyhelminthes as a group. Briefly, as the egg enters the fluke it becomes surrounded by a predetermined number of vitelline cells, the number of which will be specific for different parasites, which form the food reserve of the egg. These vitelline cells produce globules of a mixture of proteins and phenols, which are extruded to the outer surface of the developing egg. Here the phenols oxidise to form quinone, which then coalesces with the protein, reacting to form scleratin, a hard inert yellowish substance, making up the egg shell. As the eggs of different species may vary in thickness, their colours may vary from yellow; to a dark brown. The digenean egg is usually operculate, in common with other platyhelminthes. Exceptions to this may occur however, the most important being with the schistosomes. Here the eggs are non-operculate, and are ornamented with spines, the appearance of which are characteristic for different species of schistosome.

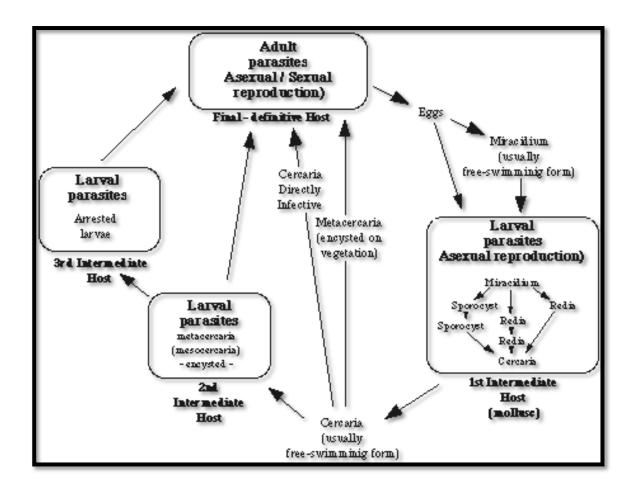
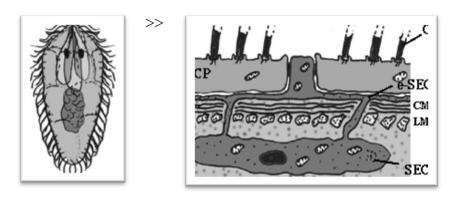


Figure 4.1 Adult parasites sexual/asexual reproduction.

2) The Larval Digeneans - the Miracidium

The miracidium is the name of the ciliated larval stage that is hatched from the digenean egg. In comparison with the other larval platyhelminthes it is very similar to the larvae of the monogeneans, (the oncomiracidium) and the larval cestodarian, or lycophore. In most cases the miracidium is usually a free swimming stage, that seeks out the primary, and in some cases only, intermediate hosts of these parasites. In all cases these primary, or 1st intermediate hosts are molluscs. In the few examples where the miracidium is not a free swimming stage the eggs are ingested, as with the lancet fluke *Dicrocoelium dendriticum*. Here the eggs hatch in the intestine of the mollusc liberating the miracidium, from where it immediately penetrates the intestinal wall to invade the molluscan tissues. In the free swimming miracidia the larval parasite exhibits distinct behavioural responses that enable it to enter the environment of, detect the presence of its hosts. These behavioural responses have principally been studied in the case of the schistosome miracidium. Morphologically the surface of the miracidium is covered with a series of ciliated plates, which may be clearly

seen using electron microscopy after removal of cilia. These ciliated epidermal plates (in some species the cilia being replaced by spines) are discontinuous, not being in contact with each other but being separated by extensions of the underlying subepidermal layer, the whole structure being illustrated below.



CP = Ciliated Plate
C = Cilia
SEC = Sub-epithelial cell
e-SEC = extension of
subepithelial cell
CM = Circular Muscle
LM = Longitudinal
Muscle

Figure 4.2 The internal morphology of Miracidium.

The plates themselves show a definite arrangement, being placed in four to five transverse rows, the exact arrangement of which may vary between different trematodes. Beneath the plates are layers of muscle fibres. At the anterior end of the larvae is a non-ciliated conical projection, the terebratorium, (or anterior papillae), bearing apertures of the apical and penetration glands. These are found at the anterior end of the body. Miracidia possess a number of sensory organ, the most important of which are the dorsaly situated eye spots, beneath which is found the cerebral mass. Other sensory organs are situated within folds of the terebratorium. Below all of the structures is found the miracidium's large rounded germinal cells, which often are often grouped in clusters called germ balls. Finally the miracidia possess a protonephridial excretory system, basically similar to that found in the adult parasites. On examination of eggs containing mature miracidia it is the clearly seen that flame cell activity that is the first sign of the initiation of hatching of the egg.

On invasion of the molluscan tissue the miracidium sheds its ciliated plates, in almost all cases rapidly transforming into an endoparasitic form, the sporocyst, although in a few unusual groups the miracidium may contain a fully developed redia.

3a) Larval Digeneans - the Sporocyst

The sporocyst develops within the molluscan host as a hollow fluid filled germinal sac, into which protrude germinal masses. At the conical anterior of the sporocyst body a birth pore is located, from which subsequent generations of larvae emerge. The germinal masses develop internally into either daughter sporocysts, which are essentially the same as their parent sporocysts, or into a second larval stage, the redia described below.

Different species of trematode will undergo different patterns of larval development, although the miracidium will always develop into a sporocyst to start with, and if daughter sporocysts are formed, redia does not develop. For example, in the schistosomes there are two generations of sporocyst in the snail host, but the redia stage is absent. In contrast in the lung fluke *Paragonimus* the parent sporocyst produces two generations of redia. In the organisms where there are two generations of sporocyst, these may be found in different locations within the body of their host, the locations differing between different species of trematode. For example in the trematode *Schistosomatium douthitti* mother sporocysts are found near the molluscan host's oesophagus and cerebral ganglia. The daughter sporocysts when they emerge from the parental sporocyst migrate through the host tissues, localising near the mollusc's digestive diverticulum. These cycles of asexual division within the mollusc result in an enormous increase in the reproductive potential of these organisms, unsurpassed within Metazoan organisms, whereby a single miracidium is capable of giving rise to many hundreds of thousands of cercariae. The sporocyst stage obtains nutrients by passage of soluble material across the sporocyst tegument.

3b) Larval Digeneans - the Redia

The redia are the second larval form to develop within the molluscan host (but may be absent in some groups, such as the schistosomes). They are similar to sporocysts, containing germinal masses within a fluid filled sac, which may develop into either second generation daughter redia, or more commonly into the final larval stage within the mollusc, the cercaria.

They differ from the sporocysts however, in that they are a much more active form, and importantly they possess simple gut. The tissue they feed on is predominantly molluscan in origin, but the redia of some groups (e.g. those of the echinostomes) may actively seek out the developmental stages of other trematodes (e.g. schistosome sporocysts) within the same

intermediate host. This was observed in a series of experiments carried out in the 1960's by Lie *et.al*. The gut itself consists of a mouth, opening into a large muscular pharynx, which in turn opens into a simple rhabdocoel like intestine. Externally, behind the mouth many redia have a ridge-like collar, below which the birth canal opens and from which either cercariae or daughter redia emerge. Further along the body there a lobe like extensions of the body, which are thought to aid the movement of the parasite within its host's tissues.

An interesting exception to the general rule that cercaria are produced by the redia is found in a few tremadodes where the redia produce progenetic metacercaria, fully capable of producing viable eggs. In these few very unusual cases the trematode may only have a single molluscan host, although the metacercaria may still be capable of developing in a second host as well. Exceptions such as these, and those described above involving miracidia containing fully developed redia is evidence of the evolutionary past of these organisms. It has been noted that the redia bears some resemblance to some of the more advanced turbellarians, and as described above, this stage is a very active form of the parasite, fully capable of actively ingesting host material, and in some cases even predation of competing parasites within their hosts. It has been postulated that the group as a whole emerged from an ancestral parasitic turbellarian, with a single molluscan host, after the development of internal division and asexual reproduction, later developing specialized forms to exploit the varying environments that these organisms have to cope with.

4) Larval Digeneans - the Cercaria

Some of the Types of Cercariae

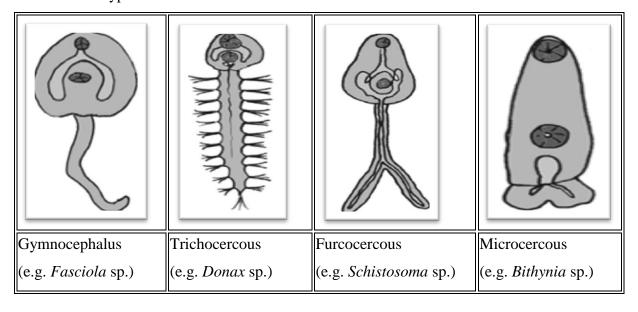


Figure 4.3 Types of cercaria.

In almost all species of trematode it is the cercarial stage that emerges from the mollusc, and is the infective form for the vertebrate host, although there may be exceptions to this general rule. For example in some cases a sporocyst, modified to have a thickened internal wall resistant to the environment, emerges, to be ingested by a second intermediate host, (e.g. as is the case in the trematode *Dicrocoeloides petiolatum*) Other exceptions, involving redia producing progenetic metacercaria, have already been described above.

The trematode cercaria exhibits considerable variations in structure, which is very important taxonomically, and reflects in many cases adaptations to the specific lifecycle of the parasite involved. Because of this great diversity of form, a system of cercarial classification has evolved, based on the gross morphology of these larval forms. Firstly cercariae may be divided into three major groups;

- i) Monostome Cercariae These lack a ventral sucker, and have simple tails. These forms develop within rediae
- **ii) Amphistome Cercariae** In these the large ventral sucker is situated at the base of a slender unbranched tail. These forms develop within rediae.
- **iii) Distome Cercariae** This is the commonest cercarial form, with the ventral sucker lying some distance from the posterior end, in roughly the anterior third of the body. These distome cercariae may themselves be divided into a large number of subgroups, based on other morphological features, particularly the form that the cercarial tail takes. Some of these forms are described below:
- a) *Leptocercous cercariae* These cercariae have straight slender tails, which are much narrower than the cercarial body. This form is further subdivided into;
- i) Gymnocephalous Cercariae In these the suckers are equal in size. This is a common form, represented within such species as *Fasciola hepatica*, and develop within rediae ii) Xiphidiocercariae These are similar to the gymnocephalous forms, but in these the oral sucker is equiped with a stylet, used in penetration of their next hosts, and they generally develop within sporocysts.

- iii) Echinostome *Cercariae* In these there is a ring of spines at the anterior end of the larvae, as in adult forms of these parasites. These are found within trematodes of the genus *Echinostoma*, and develop within rediae.
- **b)** *Trichocercous cercariae* These forms have long tails, equiped with rings of fine bristles. They are usually found in marine trematodes.
- c) Cystocercous cercariae In these the end of the tail is highly enlarged, with a cavity into which the larval body may be retracted. These usually develop within sporocysts.
- **d**) *Microcercous cercariae* Cercaria with vestigial tails and which may develop within both rediae and sporocysts.
- e) *Cercariaea cercariae* Cercaria with no tails, where the cercaria is not a free swimming form and may develop within both rediae and sporocysts.
- **f**) *Furcocercous cercariae* In these the tails are forked at the end. The cercarias of the most important group of trematodes, the schistosomes, have cercariae of this form. This form develops within sporocysts.

Otherwise, both externally and internally the structure of the body of the cercaria resembles that of the adult trematode into which they will grow. For example, the ring of spines found at the anterior end of echinostome cercariae is also present in the adult flukes.

The outer surface of the cercaria is a tegument, which may however differ from that found in the adult form in a number of ways. For example in the schistosomes the tegument is covered with a trilaminate plasma membrane, (as opposed to the two bi-lipid membranes found in the adult), on the outer surface of which there is a glycocalyx, (absent in the adult). However many other features of this tegument appear similar to that of the adult, the differences almost certainly being adaptations due to the differing environments that these two lifecycle stages experience. For example, spines are found on the surface of both forms of tegument, and the overall structure of a syncytium connected to subtegumental cells is the same. For more details on the structure of the tegument, go to the page devoted to the digenean tegument.

Within the cercarial body a number of different types of gland cells may be found, including cystogenous gland cells, used by the larvae to secrete a cyst wall during formation of the metacercarial stage, and penetration gland cells, used by the cercaria to penetrate its next host, either a second intermediate host, or in some groups the definitive host, (such as the schi stosomes), where the cercaria is the final larval stage.

The cercarias released from their molluscan intermediate host are usually a free swimming form. These must then locate either their next, and usually final intermediate host, their definitive host which they actively penetrate (e.g. in members of the family Schistosomatidae), or locate a suitable solid substrate to encyst upon, or be ingested by their definitive host.

To locate these various targets the cercariae are equipped with a variety of sensory organs. These commonly include two or more eye spots, as well as touch receptors, and allow specialized cercarial behavior, designed to bring the cercariae into an environment giving the maximum probability of infecting their next hosts. For example the cercariae of the schistosomes exhibit negative phototrophy (swimming to the surface of the water), and positive thermotrophy and thigmotrophy, being attracted to warm objects moving in the water. As well as these behavioral responses within the free swimming cercariae, the parasite exhibits definite circadian rhythms in terms of shedding from the molluscan host, again being shed at times optimal for bringing them into contact with their next host. For example the schistosome cercariae are generally shed during daylight, in the morning, whilst those of other species emerge only at night. For a more full treatment on the subject or circadian rhyth ms exhibited by parasites.

In a few groups, such as *Alaria* spp. However, the parasite employs three intermediate hosts. In these cases the cercaria penetrates the second intermediate host to form a resting stage, the mesocercaria described below. In these cases this second intermediate host is in turn ingested by a third intermediate host, where it encysts to form a metacercaria.

5) Larval Digeneans - the Mesocercaria

The mesocercaria is essentially a resting stage within the parasite lifecycle, employing a second intermediate host in a parasite lifecycle utilising four hosts. It is defined as follows (by Pearson 1956);

The mesocercaria is a definite prolonged stage in the adult generation of strigeate trematodes, which closely resembles the cercarial body, from which it develops in the second intermediate host, and which does not possess metacercarial features; it develops in turn into the metacercaria in another host.

In parasites having this larval stage the mesocercaria are capable of infecting and surviving within a very wide range of paratenic hosts which may ingest the second intermediate host, thus in effect increasing the number of hosts which the parasite may use in its lifecycle. For example amphibians infected with mesocercaria of *Alaria* may themselves infect a wide variety of other amphibians, reptiles, birds and mammals if they are ingested by these animals.

6) The Larval Digeneans - the Metacercaria

This is a much more common "resting" larval stage of the trematode parasitic lifecycle, formed either in a final intermediate host (when a mesocercaria, or more commonly a cercaria enters its body), or on a solid substrate in the external environment. The final intermediate host may be a fish (e.g. *Opisthorchis sinensis*), an arthropod (e.g. *Dicrocoelium dendtriticum*, employing an ant second intermediate host, and *Paragonimus westermani* employing a crustacean), or another mollusc, as with some of the echinostomes. As stated above, some trematodes however do not have second intermediate hosts, but either encyst as metacercariae on solid substrate's, such as aquatic vegetation or on shells of aquatic organisms, which will in turn be ingested by the parasites definitive host, or in some groups such as the schistosomes, as already described, the cercariae directly penetrate the skin of, and infect, the parasites definitive host. Although generally the metacercariae are inactive encysted forms, the metacercaria of some species do remain free and active. For example the metacercariae of trematodes belonging to the genus *Diplostomulum* where the larvae are found lens, humours and cranial ventricles of a wide range of hosts. In most other metacercariae however encystment does occur. The structure of the cyst wall itself varies

considerably, though generally it is a complex mixture of tanned proteins, lipids and polysaccharides. Within the cyst wall the morphology of the larva usually closely resembles that of the cercarial body, although as described above, in some groups sexual maturation may occur either fully or partially. To continue further the metacercaria must be ingested, either along with the body of the intermediate host it inhabits by a carnivorous definitive host, or along with the vegetation it has encysted on by a herbivorous or omnivorous host.

7) Larval Digeneans - the Juvenile Adult Stages

On ingestion the metacercaria (or cercariae) must transform into the adult form. The precise details of this process will vary considerably, depending on how the definitive host was infected. For example in some species the adult flukes are found within the alimentary tract. In these cases the metacercarial cyst wall is broken down to release what is essentially a young fluke, which only has to migrate a short distance to reach their preferred site within the hosts body. In other groups however the adult forms are located in other sites within the body. In these cases the liberated young fluke must penetrate the gut wall, or in the case of the schistosomes penetrate the host's skin. Then they must undergo a migration through the host's body. This is usually via the circulatory system, but again the precise details of the migratory path will vary considerably.

Taxonomy of Parasitic infections

A total of 31,717 snail species were collected and examined for trematode infections. The cercarial infections were examined using shedding and crushing methods. The overall infection rate was 12.92%. Eight types and sixteen species of cercariae were categorized. The types and species were identified following the published article of home and abroad. The figures and photographs in this chapter are considered as standard for identification (Krailas, 2003, 2014). They were:

Table 4.1 List of types and species of recovered Trematode cercariae in the present study.

Types of cercaria	Species
Parapleurophocercous	Haplorchis pumilio, Haplorchs taichui , Stictodora tridactyla
Pleurophocercous	Centrocestus fnusormosa

Xiphidiocercariae	Acanthatrium hitaense, Loxogenoides bicolor,	
	Haematoloechus similis, Cloacitrema philippinum	
Furcocercous	Cardicola alseae, Alaria mustelae,	
	Apatemon gracilis, Mesostephanus appendiculatus	
Echinostome	Echinochasmus pelecani	
Amphistome	Gastrothylax crumenifer	
Renicolid	Cercaria caribbea	
Cotylomicrocercous	Podocotyle lepomis	

Type 1. Parapleurophocercous cercariae

1. Haplorchis pumilio Looss, 1899 (Yamaguti, 1975) (Plate 2, fig 6)

Description: Haplorchis pumilio were found from Pila globosa. The infection rate was 4.16%. The average size ranges are body (105-50 mm), tail (30-480 mm). The body shape is oval, and its surface is covered with fine spines and sensory hairs. The pigment eyespots and pharynx are present. There are seven pairs of penetration glands, which are arranged in two longitudinal series with a ventral sucker and genital primordia. Their ducts are arranged in two bundles. Four of them were open through the dorsal wall, and four through the ventral wall of the oral sucker in two oblique symmetrical rows. The mouth aperture has transverse rows of spines. The tail is long, attached to the dorsal end of the body, with lateral finfolds nearby and a dorso-ventral finfold for the greater distal portion. Cercariae were produced within rediae.

Movement behavior: The cercaria moved by rolling up and springing the body back to move forward in a screwing motion for 2–4 seconds and then rested for 15–20 seconds on the surface of water. It survived up to 2.5–3 hours in the water after emergence.

2. Haplorchis taichui Nishigori, 1924 (Yamaguti, 1975) (Plate 2, fig 5)

Description: Haplorchis taichui were found from *Pila globosa*, the infection rate was 0.29%. Cercarial body is oval in shape, colored with orange yellow, and entirely covered with minute spines and sensory hairs. The average size ranges are body (130-245 mm), tail (35-485 mm). The oral sucker is situated ventrally in the head region. There are transverse rows of spines at

the mouth aperture. The pigment eyespots and a pharynx are present. Seven pairs of penetration glands extend from the pharynx to the end of the body. A long tail is attached to the dorsal end of the body. Cercariae were produced within the rediae.

Movement behavior: Cercariae were escaped from the rediae. In the water, they floated on the surface or in the middle. It survived up to 2–3 hours in the water after emergence.

3. Stictodora tridactyla Martin and Kuntz, 1955 (Yamaguti, 1975) (Plate 3, fig 5; Plate 2.fig 9).

Description: Stictodora tridactyla were found from Brotia costula. The infection rate was 6.81%. The body is oval in shape and yellowish brown in color. There are 3 rows of oral spines (4–6, 12–14, 22–24), and 7 pairs of penetration glands in 4 groups of 3:4:4:3. There is one pair of eye spots with coarse granules, with a small globular pharynx between the eye spots. The excretory bladder in the flattened V-shaped is situated at the end of the body. The tail is longer than the body with a bilaterial finfold and a dorso-ventral finfold. Both the dorsal and ventral finfolds arose at a short distance from the anterior and the posterior end of the lateral finfold. The average size ranges are body (112-275 mm), tail (45-585 mm). Cercariae were produced within the rediae.

Movement behavior: The cercaria floated on the surface or in the middle of the water.

Type 2. Pleurophocercous cercariae

4. Centrocestus fnusormosa Nishigori, 1924 (Yamaguti, 1975) (Plate 3, fig 3).

This parasite was found in *Bellamya bengalensis*. The infection rate was 0.23%. Cercarial body is oval in shape. The average size ranges are body (65-120 mm), tail (15-85 mm). A pair of eyespots lay at the level of the pharynx. The oral sucker has two rows of oral spines similar to hooks of the tapeworm. Cercariae were produced within the rediae.

Movement behavior: The cercariae moved by rolling up and springing the body back to move forward in a screwing motion for 8–10 seconds and then rested for about 45–50 seconds. It survived up to 3–4 hours in the water after emergence.

Type 3. Xiphidiocercariae

5. Acanthatrium hitaense Koga, 1953 (Yamaguti, 1975) (Plate 4, fig 2).

Description: This parasite was found in *Pila globosa*. The infection rate was 0.27%. The parasite is virgulate xiphidiocercaria. The body is oval in shape and white in color. The average size ranges are body (75-105 mm), tail (25-68 mm). The tail is shorter than the body, inserted to the posterior end of the body. Cercariae were produced within the sporocyst.

Movement behavior: The cercariae floated on the surface or in the middle of the water. They moved by rolling up and springing the body back to move forward in a screwing motion. It survived up to 2–4 hours in the water after emergence.

6. Loxogenoides bicolor Kaw, 1945 (Yamaguti, 1975) (Plate 4 fig 3)

Description: Loxogenoides bicolor was found from *Pila globosa*. The infection rate was 7.41%. The body of cercariae is spinose and oval in shape. The average size ranges are body (75-110 mm), tail (25-75 mm). Its entire body is dotted with granules. The ventral sucker is smaller than the oral sucker. The tail is spinose, with slightly longer spines at the tip. Cercariae were produced within the sporocyst.

Movement behavior: The cercariae moved by folding its tail and rolling up the body and moved from left to right quickly. In resting position, they floated on the surface or in the middle of the water.

7. Haematoloechus similis Looss, 1899 (Yamaguti, 1975) (Plate 3, fig 6)

Description: Haematoloechus similis was found in Brotia costula. The infection rate was 1.46%. This parasite was classified into Xiphidio cercariae. The body is ovate, and the surface is covered with spines. The average size ranges are body (95-150 mm), tail (30-105 mm). The tail is not finflod. Cercariae were produced within the sporocyst.

Movement behavior: The cercariae floated on the surface or in the middle of the water. The body sank lower than the tail. It moved by folding its tail back to the body and turning its body to roll quickly from left to right, darting forward for about 15–20 seconds, and resting for about 10–15 seconds. It survived up to 1–2 hours in the water after emergence.

8. Cloacitrema philippinum Velasquez, 1969 (Yamaguti, 1975) (Plate 3, fig 1).

Description: Cloacitrema philippinum was found in Bellamnya bengalensis. The infection rate was 0.24%. The body is elongate and muscular. The average size ranges are body (165-395 mm), tail (35-480 mm). There is no eye spot, and no spine on the body surface. There are numerous sensory papillae on the surface of body. The oral sucker has 12 opening ducts. There is an adhesive organ at the tip of the tail, with no lateral finfolds. Cercariae were produced within the rediae.

Movement behavior: The cercaria floated on the surface or in the middle of the water. It moved by stretching the body and rolling on the water surface. The adhesive gland anchored on the container surface. It survived up to 2–4 hours in the water after emergence.

Type 4. Furcocercous cercariae

9. Cardicola alseae Meade & Pratt, 1965 (Yamaguti, 1975) (Plate 2, fig 7)

Description: Cardicola alseae were found from *Pila globosa* which is equivalent to an infection rate of 0.1% of the total number of the collected snails. The average size ranges are body (25-95 mm). The small hook-liked body had an anterior organ, and was covered with minute spines. The longer spines were found in some parts of the body, the dorsal and ventral of the posterior end. The dorso-median finfold was observed in the middle part of the body. Many large granules were observed, with a penetration gland located at the middle part of the body. The excretory bladder was small. The tail is furcocercous; its furcae are shorter than the beginning of the tail. Minute spines and sensory hairs were observed.

Movement behavior: The cercaria floated on the surface of the water. The body and tail hanged rolling. It moved by folding its tail back to the body, moving forward around 6–10 seconds, and resting for 3–4 seconds.

10. Alaria mustelae Bosma, 1899 (Yamaguti, 1975) (Plate 2, fig 4)

Description: Alaria mustelae were found from Brotia costula which is equivalent to an infection rate of 0.15% of the total number of the collected snails. Cercarial body has a long shape (135-255). The body is covered entirely with spines and two irregular spines around the aperture of the ventral sucker. Cercaria developed within the sporocyst. The tail stem was

without spines; the furcae was irregularly spinose (long hair like), with no caudal bodies. The excretory pore was found at the fork tail tip.

Movement behavior: The cercariae moved by rolling up and springing back the body to swiftly move forward in a quick semi-circular motion.

11. Apatemon gracilis Szidat, 1928 (Yamaguti, 1975) (Plate 2, fig 2)

Description: Apatemon gracilis were found from *Pila globosa*, which is equivalent to an infection rate of 0.17% of the total number of the collected snails. The cercarial body is oval in shape, and the prepharynx is practically absent. The anterior region has a cuticle spine. It has unpigmented eyespots. The tail stem contains about 16 caudal bodies. The excretory bladder is very small. Cercariae were produced within the sporocyst.

Movement behavior: The cercariae floated on the surface or in the water.

12. Mesostephanus appendiculatus (Cicrea, 1916) Lutz, 1935 (Yamaguti, 1975) (Plate 2, fig 3).

Description: Mesostephanus appendiculatus were found Brotia costula, which is equivalent to an infection rate of 0.29% of the total number of the collected snails. The cercarial body is oval in shape and spinose. The average size ranges are body (108-220 mm), tail furcae (20-160 mm). There are many spines on the surface and the oral sucker. The pharynx is small and round. The tail is forked and longer than the body and the tail surface is covered with many spines. The tail stem is longer than the furca. The tail tubule opens at the tip of each tail furca in which no flame cell is found. Cercariae were produced within the sporocyst.

Movement behavior: The cercariae floated on the surface or in the water. The body sank lower than the spreading fork tail. It moved by rolling up and springing back the body to swiftly move forward in a semi-circular motion for about 2–4 seconds.

Type 5. Echinostome cercariae

13. Echinochasmus pelecani Johnston & Simpson, 1944 (Yamaguti, 1975) (Plate 4, fig 1)

Description: Echinochasmus pelecani were found from Pila globosa which is equivalent to an infection rate of 0.06% of the total number of the collected snails. Cercarial body is

elongate, white in color, no eye spot, oral sucker with 3 opening of duct. Collar spine is not appearance. Tail is the same length as body, flexible, vacuole appearance along the tail. Cercariae were produced within rediae.

Movement behavior: The cercaria floated on the surface or in the water. It moved very fast by rolling up and springing back about 5–10 seconds. It survived up to 3–4 hours in the water after emergence.

Type 6. Amphistome cercariae

14. Gastrothylax crumenifer (Creplin, 1847) Otto, 1896 (Yamaguti, 1975) (Plate 2, Fig 1)

Description: Gastrothylax crumenifer were found from Bellamnya bengalensis which is equivalent to an infection rate of 0.02% of the total number of the collected snails. Cercariae were liberated from the rediae. The body shape is ovate and large. The eye spots have conical lens with yellow pigment through the body with a smooth surface. The oral sucker is equal to the ventral sucker. The tail inserted to the posterior end of the body. There are various sizes of vacuole through the tail.

Movement behavior: The cercaria floated on the surface or in the water. It moved by wavering on the surface of the water for around 8–10 seconds, and then rolling up and springing back for about 5–10 seconds. It survived up to 3–4 hours in the water after emergence. The cercariae were photo-sensitive. They shrank rapidly in changing light conditions.

Type 7. Renicolid cercariae

15. Cercaria caribbea (Cable, 1963) (Yamaguti, 1975) (Plate 3, fig 2)

Description: Cercaria caribbea were found from Brotia costula which is equivalent to an infection rate of 0.14% of the total number of the collected snails. Cercariae developed in the sporocyst. Its body is flat with yellow pigment and numerous minute spines on the surface of the body, with no eye spot. The tail is straight, and is longer than the length of the body, with no lateral finfold.

Movement behavior: The cercariae moved slowly on the bottom of the container, and swam continuously. It survived up to 2–3 hours in the water after emergence.

Type 8. Cotylomicrocercous cercariae

16. Podocotyle (Podocotyle) lepomis Dobrovolny, 1939 (Yamaguti, 1975) (Plate 2, fig 8)

Description: Podocotyle lepomis were found from Bellamnya bengalensis which is equivalent to an infection rate of 0.19% of the total number of the collected snails. Cercariae developed in the sporocyst. The body is cylindrical in shape, clear white in color, with no sensory hair, spine, 6 papillae on the head, and rough granules present on the body. The stylet is present in oral sucker. There were 2 rows of sensory papillae around the oral sucker, with a long prepharynx. Pharynx is round. The tail is short (52-61 mm), only half the length of the body, cup-shaped, with an adhesive gland present at the end of the tail for attaching.

Movement behavior: The cercaria floated with the ventral upside. It moved by floating with its head and tail folded together, and then sprang up.

Table 4.1 List of types and species of recovered Trematode cercaria in the present study.

Types of cercaria	Species	Host
Parapleurophocercous	Haplorchis pumilio	Pila globosa
	Haplorchs taichui	Pila globosa
	Stictodora tridactyla	Bellamya bengalensis
Pleurophocercous	Centrocestus fnusormosa	Bellamya bengalensis
Xiphidiocercariae	Acanthatrium hitaense	Pila globosa
	Loxogenoides bicolor	Pila globosa
	Haematoloechus similis	Brotia costula
	Cloacitrema philippinum	Bellamnya bengalensis
Furcocercous	Cardicola alseae	Pila globosa
	Alaria mustelae	Bellamnya bengalensis
	Apatemon gracilis	Pila globosa
	Mesostephanus appendiculatus	Bellamnya bengalensis
Echinostome	Echinochasmus pelecani	Pila globosa
Amphistome	Gastrothylax crumenifer	Bellamnya bengalensis
Renicolid	Cercaria caribbea	Brotia costula
Cotylomicrocercous	Podocotyle lepomis	Bellamnya bengalensis



Plate 1 Different types of Cercaria found in the studied snail species.

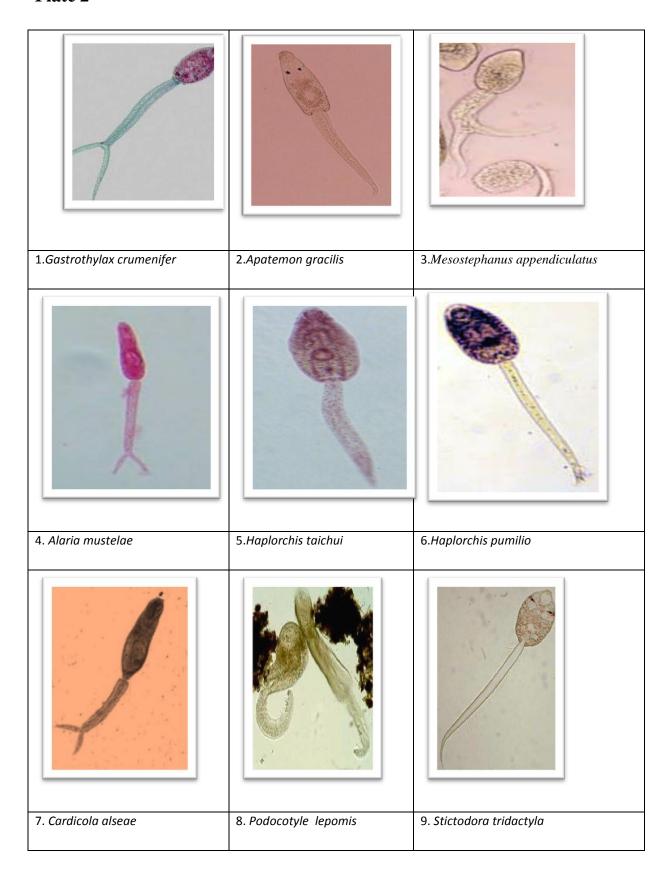


Plate 2: Different types of Cercaria found in the studied snail species.

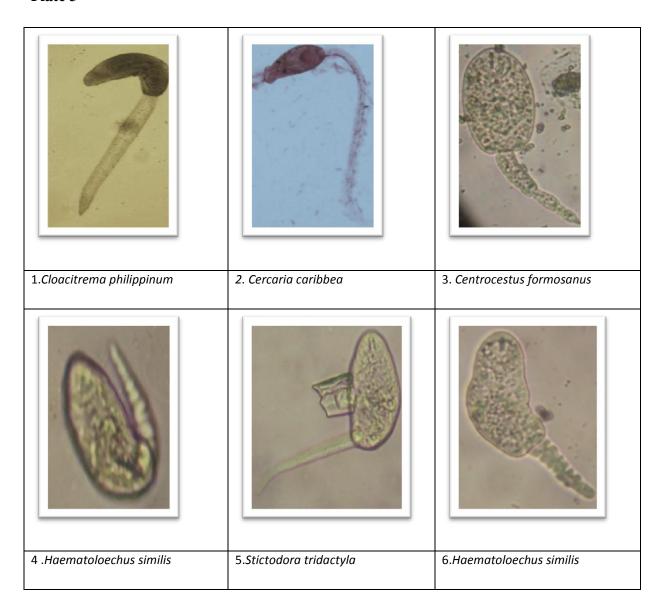


Plate 3 Different types of Cercaria found in the studied snail species.

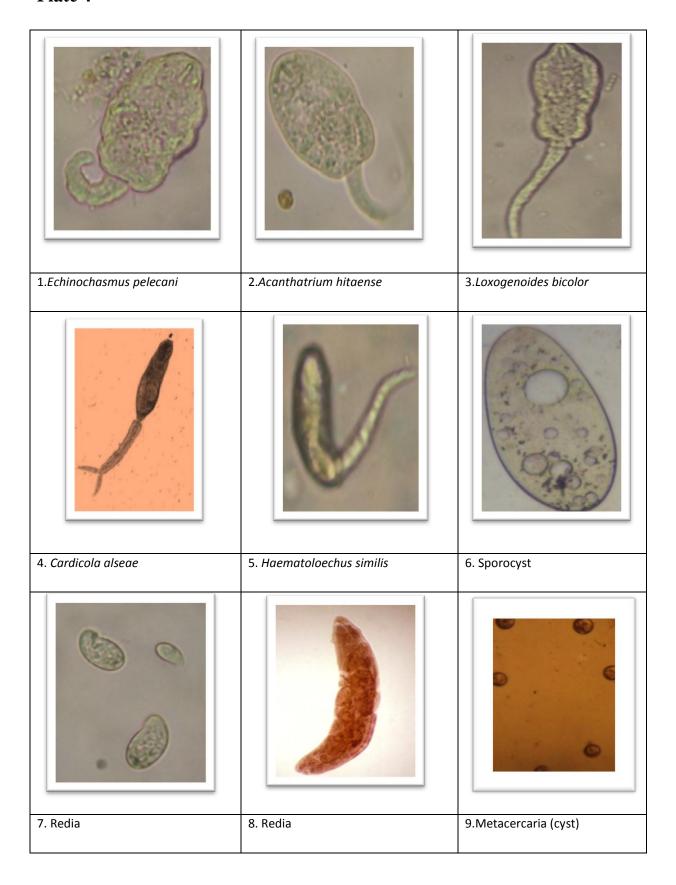


Plate 4 Different types of Cercaria and Radia found in the studied snail species.

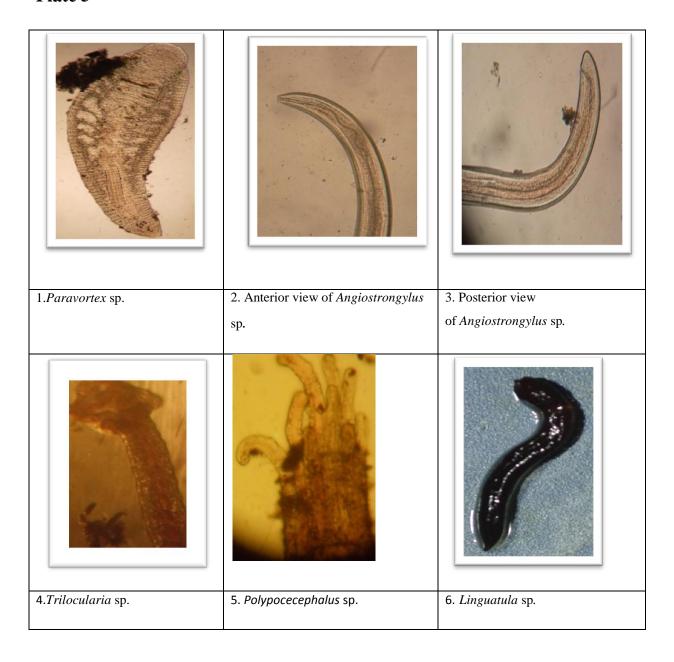


Plate 5 Other parasites found in the studied snail species.

RESULT AND OBSERVATION

Prevalence of larval trematodes

During 2011-13; 31,717 number of snails were examined and a total of 8 types of cercariae were identified which are as follows (the possible genera are mentioned within bracket).

The trematode cercariae types along with species are:

- 1) Parapleurophocercous (Haplorchis pumilio, Haplorchs taichui, Stictodora tridactyla);
- 2) Pleurophocercous(*Centrocestus fnusormosa*);
- 3) Xiphidiocercariae (Acanthatrium hitaense, Loxogenoides bicolor, Haematoloechus similis, Cloacitrema philippinum);
- 4) Furcocercous (Cardicola alseae, Alaria mustelae, Apatemon gracilis, Mesostephanus appendiculatus);
- 5) Echinostome (*Echinochasmus pelecani*);
- 6) Amphistome (Gastrothylax crumenife);
- 7) Renicolid (Cercaria caribbea);
- 8) Cotylomicrocercous (*Podocotyle lepomis*).

Beside these, two types of Nematodes, two types of Cestodes, one type of Turbellarian and one type of Penstomida were identified.

A total of 23 locations in Bangladesh were selected and the snails were collected within Dhaka and surrounding region (11 location), were selected and collected three species of snails were examined for larval trematodes and other parasites. Similarly from Khulna and surrounding region (6 locations) were selected for collected of snails and observed for larval trematodes and other parasites. From Kishoreganj and surrounding regions, 6 locations were selected for snails and examined them for parasites.

Three species of freshwater gastropods were selected which were belonging to two families (Viviparidae, Ampullariidae) and three genera (*Bellamya, Pila, Brotia*).

From each collection site, different sizes of specimen were obtained while the highest number of collection was from the shallow part of the ponds where the vegetation was more. The majority of collection sites were found mostly with *Pila* and that dominated more than the other molluscan species. Each of the study area has its own specificity of species and different parameters like pH, EC, dissolved oxygen, specificity of parasites and availability of aquatic plant species. All the collection sites are normal grounds with slight modifications as depth, shallow and medium in depth. Out of 23 collection sites, few are not deep enough while the other places are deep enough to contain water throughout the year. All the collection sites contain aquatic plants namely *Eichornia* sp (water hyacinth), *Azolla* sp (mosquito fern), *Potamogeton* sp (pond weeds), *Typha* sp (cattails) and algae of genera-*Cyanophycyeae*, *Anabaena* and *Gymnodinium* respectively without any differentiation.

Site 1: Dhaka and surrounding locations

• Ramna Park Pond

Increasing trend of trematode infection in *Bellamya* sp. was observed from July 2011 to September 2011 which was in decreasing trend up to February 2012 with slightly increased in January. Highest percentage of infection by trematodes in *Bellamya* sp. was found in September (24.24%) and lowest percentage of trematode infection was 6.25% in February 2012 (Fig 5.1).

Highest percentage of infection by trematodes in *Brotia* sp. was found in May (17.65%) and lowest infection was 6.25% in February. Highest percentage of infection by trematodes in *Pila* sp. was 21.21% in September 2011 and lowest infection was 7.14% in February 2012.

The trematode infection in *Bellamya* sp. started to increase from July 2012 to August 2012. Comparatively higher percentage of infection by trematodes in *Bellamya* sp. was found in August (23.33%) and lowest (8.33%) in February 2013 (Fig 5.2).

In *Brotia sp.*, highest infection was found in September (17.65%) and lowest (6.25%) in February 2013. Highest percentage of trematode infection in *Pila* sp. was 21.05% in September and lowest was (5.88%) in February 2013.

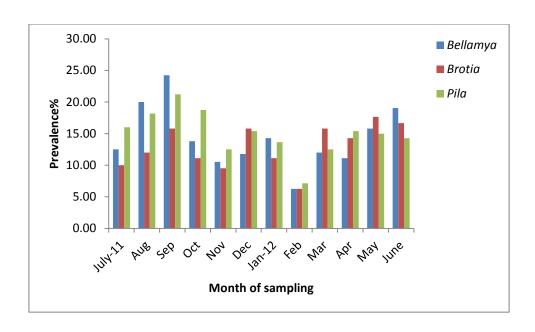


Figure 5.1 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Ramna Park Pond of Dhaka (2011-12).

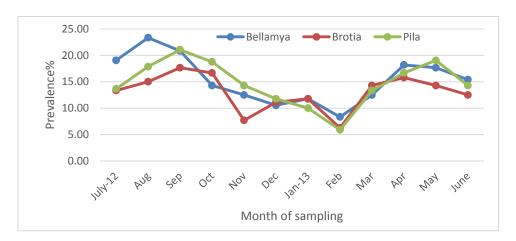


Figure 5.2 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Ramna Park Pond of Dhaka (2012-13).

• Curzon Hall Pond

The trematode infection in *Bellamya* sp. was observed to increase from July 2011 to September 2011 which was decreased up to February 2012 with slightly increased in December and January. Highest percentage of infection by trematodes in *Bellamya* sp was found in September (23.53%) and lowest infection (3.85%) in February 2012 (Fig 5.3).

Highest percentage of occurrence of infection by trematodes in *Brotia* sp. was found in October (13.79%) and lowest infection (3.70%) in February. In September 2011, in *Pila* sp.

highest percentage of infection by trematodes was 14.81% and lowest trematode infection was 4.76% in February 2012.

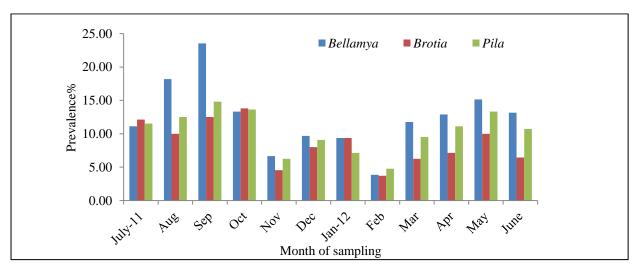


Figure 5.3Monthly prevalence of larval trematodes in three genera of freshwater snails sampled from Curzon Hall Pond of Dhaka (2011-12).

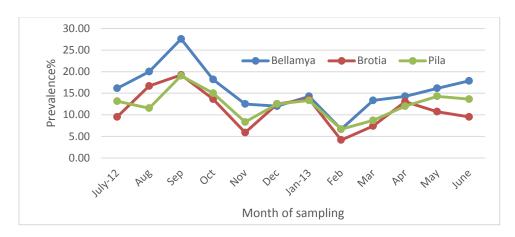


Figure 5.4Monthly prevalence of larval trematodes in three genera of freshwater snails sampled from Curzon Hall Pond of Dhaka (2012-13).

The trematode infection in *Bellamya* sp. was observed to increase from July 2012 to September 2012 in increasing trend which was decreased up to February 2013 with a little increase in December and January. Highest percentage of occurrence of infection by trematodes in *Bellamya* sp was found in September (27.59%) and lowest percentage of trematode infection was 6.67% in February 2013 (Fig 5.4).

Increasing trend of trematode infection in *Brotia* sp. was also observed from July 2012 to September 2012 which was decreased up to February 2013 with a little increase in December and January 2013. Highest percentage of occurrence of infection by trematodes in *Brotia* sp. was found in September which was 29.63%. Lowest percentage of parasite infection was found (4.17%) in February 2013.

Highest percentage of infection by trematodes in *Pila* sp. was (19.03%) found in September 2012 and lowest trematode infection was 6.67% in February 2013.

• Jagannath Hall Pond

Increasing trend of trematode infection in *Bellamya* sp. was getting higher from July 2011 to September 2011. However, highest percentage of infection by trematodes in *Bellamya* sp was found in September which was 30% while lowest percentage of trematode infection was 3.45% in December 2012 (Fig 5.5).

However, highest percentage of occurrence of infection by trematodes in *Brotia* sp. was found in September (18.18%) and lowest infection (3.33%) in February 2012.

Highest percentage of infection by trematodes in *Pila* sp. was nearly 21.21% in September 2011 and lowest trematode infection was 3.33% in February 2012.

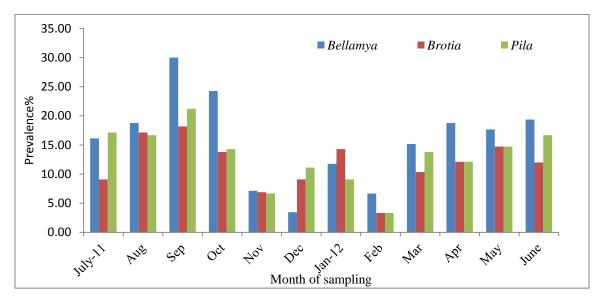


Figure 5.5 Monthly prevalence of larval trematodes in three genera of freshwater snails sampled from Jagannath hall Pond of Dhaka (2011-12).

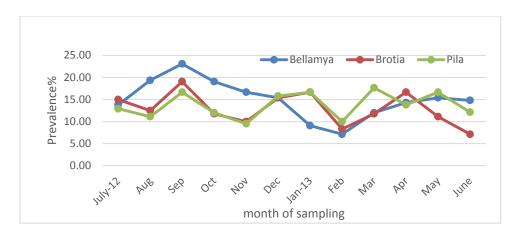


Figure 5.6Monthly prevalence of larval trematodes in three genera of freshwater snails sampled from Jagannath hall Pond of Dhaka (2012-13).

Comparatively highest percentage of infection by trematodes in *Bellamya* sp. was found in September which was 23.08% while, lowest percentage of trematode infection was 7.14% in February 2013 (Fig 5.6).

Increasing trend of trematode infection in *Brotia* sp. was also observed from July 2012 to September 2012 which was decreased up to February 2013. However, highest percentage of infection by trematodes in *Brotia* sp. was found in September which was nearly 19.05%. Lowest percentage of parasite infection (7.14%) in June 2013.

Highest percentage of infection by trematodes in *Pila* sp. was 17.65% found in March 2013 and lowest trematode infection was 9.52% in November 2012.

Keraniganj Pond

Trematode infection in *Bellamya* sp was found in September (18.92%) and lowest percentage was 6.06% in February2012 (Fig 5.7).

In *Brotia* sp. highest percentage of infection by trematodes was found in September which was 11.76% and lowest infection was 3.33% in November 2011.

Highest percentage of infection by trematodes in *Pila* sp. was 17.14% in September 2011 and lowest trematode infection was 3.70% in November 2011.

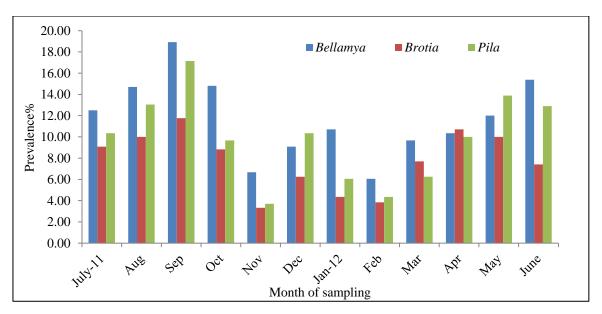


Figure 5.7 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Keraniganj Pond of Dhaka (2011-12).

The trematode infection in *Bellamya* sp. was observed to increase from July 2012 to September 2012 which was decreased up to February 2013. Highest percentage of infection by trematodes in *Bellamya* was found in September (24.24%) and lowest percentage of trematode infection (7.69%) in February 2013 (Fig 5.8).

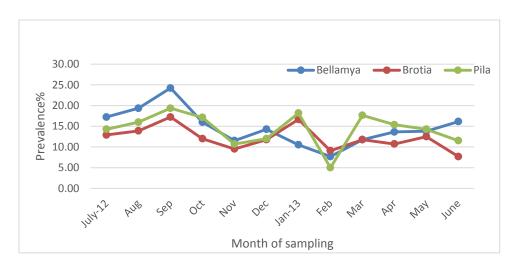


Figure 5.8 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Keraniganj Pond of Dhaka (2012-13).

Increasing trend of trematode infection in *Brotia* sp. was also observed from July 2012 to September 2012 which was decreased up to February 2013. Highest percentage of infection by trematodes in *Brotia* sp. was found in September which was 17.24%. Lowest percentage of parasite infection was found (9.09%) in February 2013.

Highest percentage of infection by trematodes in *Pila* sp. was 17.14% found in September 2012 and lowest trematode infection was 5% in February 2013.

Tongi Railway Station Pond

In the present study trematode infection in *Bellamya* sp. was observed to increase from July 2011 to September. Highest percentage of infection by trematodes in *Bellamya* sp. was found in September which was 25% while lowest percentage of trematode infection was 7.69% in February 2012 (Fig 5.9).

In *Brotia* sp. highest percentage of infection by trematodes was found in April (14.29%) and no infection was found in November 2011 and February 2012.

Highest percentage of infection by trematodes in *Pila* sp. was 19.44% in September 2011 and lowest trematode infection was 7.69% in February 2012.

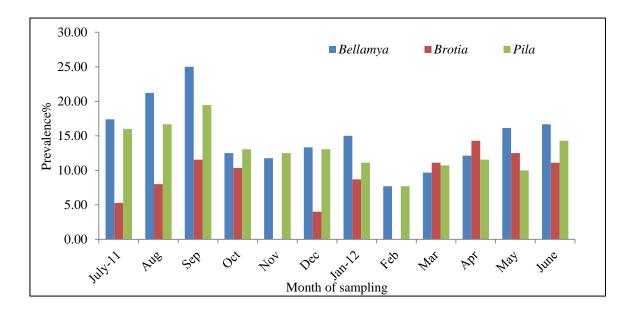


Figure 5.9 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Tongi Railway Station Pond of Gazipur (2011-12).

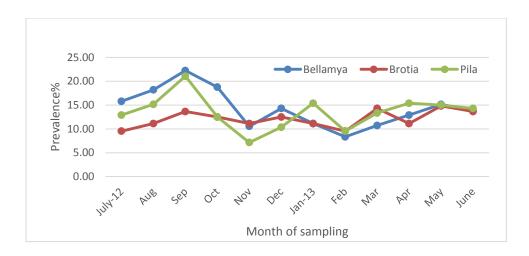


Figure 5.10 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Tongi Railway Station Pond of Gazipur (2012-13).

Increasing trend of trematode infection in *Bellamya* sp was observed from July 2012 to September 2012 which was in decreasing trend up to February 2013. Highest percentage of infection by trematodes in *Bellamya* sp. was found in September which was 22.22% while lowest percentage of trematode infection was 8.33% in February 2013 (Fig 5.10).

Highest percentage of infection by trematodes in *Brotia* sp. was found in March (14.29%) and lowest infection (9.52%) in July 2012 and February 2013.

Highest percentage of infection by trematodes in *Pila* sp. was 21.05% in September 2012 and lowest trematode infection was 7.14% in November 2012.

• Joydevpur Railway Station Pond

It was observed that, trematode infection in *Bellamya* sp. was highest in September which was 23.33% while lowest percentage of trematode infection was 5.56% in February 2012 (Fig 5.11).

Highest percentage of infection by trematodes in *Brotia* sp. was found in August which was 11.54% and lowest infection was found 7.14% in November and February 2012.

Highest percentage of infection by trematodes in *Pila* sp. was 19.05% in May 2012 and lowest trematode infection was 5.26% in February2012.

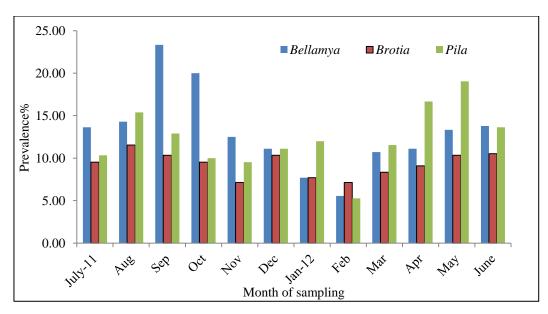


Figure 5.11 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Joydevpur Railway Station Pond of Gazipur (2011-12).

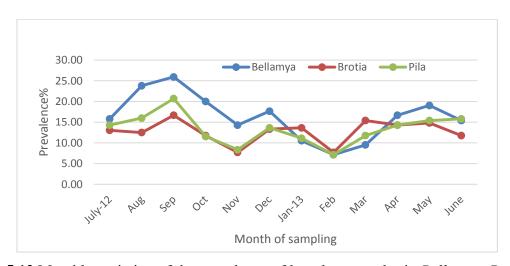


Figure 5.12 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Joydevpur Railway Station Pond of Gazipur (2012-13).

Trematode infection in *Bellamya* sp. was observed started ti increase from July 2012 to August 2012 which was decrease up to February 2013 with slightly increase in December and January 2013. Highest percentage of infection by trematodes in *Bellamya* sp was found in March which (25.93%) and lowest trematode infection (7.14%) in February 2013 (Fig 5.12).

In present observation highest percentage of infection by trematodes in *Brotia* sp. was found in April 2012 which was 16.67% and lowest infection was 7.69% in November 2012 and

February 2013.Highest percentage of infection by trematodes in *Pila* sp. was 20.69% in September 2012 and lowest trematode infection was 7.14% in February 2013.

• Bhawal Gazipur Railway Station Pond

In present investigation increasing trend of trematode infection in *Bellamya* sp. was observed from July 2011 to September. Highest percentage of infection by trematodes in *Bellamya* sp. was found in September which was 21.21% while lowest percentage of trematode infection was 6.25% in February 2012 (Fig 5.13).

Highest percentage of infection by trematodes in *Brotia* sp. was found in September which was 14.29% and lowest infection (6.25%) found in February2012.

Highest percentage of infection by trematodes in *Pila* sp. was 18.75% in September 2011 and lowest trematode infection was 8.33% in November and February 2012.

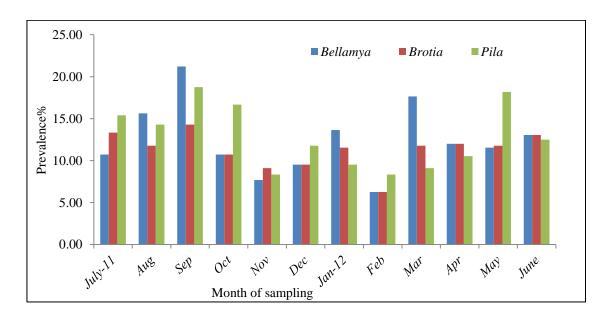


Figure 5.13 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Bhawal Gazipur Railway Station Pond of Gazipur (2011-12).

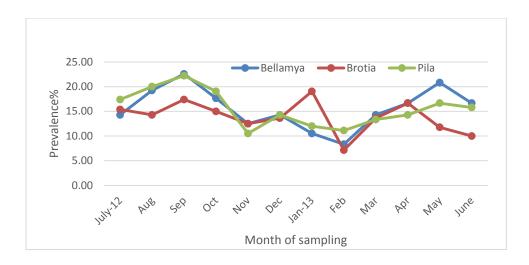


Figure 5.14 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Bhawal Gazipur Railway Station Pond of Gazipur (2012-13).

The highest percentage of infection by trematodes in *Bellamya* sp was found in March which was 22.58% and lowest percentage of trematode infection was 8.33% in February 2013 (Fig 5.14).

In *Brotia* sp highest percentage of infection by trematodes was found in January (19.05%) and lowest infection (7.14%) in February 2013.

Highest percentage of infection by trematodes in *Pila* sp. was 22.22% in September 2012 and lowest was 10.53% in November 2013.

• Salna BSMAU Pond

In the present study trematode infection in *Bellamya* sp. was observed to increase from July 2011 to September2011 which was then decreased up to February 2012 with slightly increase in December and January. Highest percentage of infection by trematodes in *Bellamya* sp was found in September which was 23.81% and lowest percentage of trematode infection was 3.45% in February 2012 (Fig 5.15).

Highest percentage of occurrence of infection by trematodes in *Brotia* sp. was found in September which was 19.05% and lowest infection was 7.69% in February.

Highest percentage of infection by trematodes in *Pila* sp. was 19.05% in April 2012 and lowest trematode infection was 6.67% in February 2012.

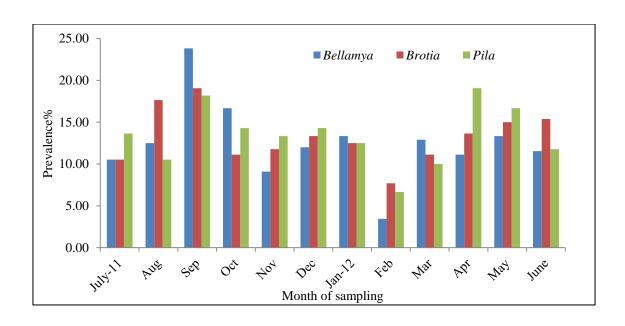


Figure 5.15 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Salna BSMAU Pond of Gazipur (2011-12).

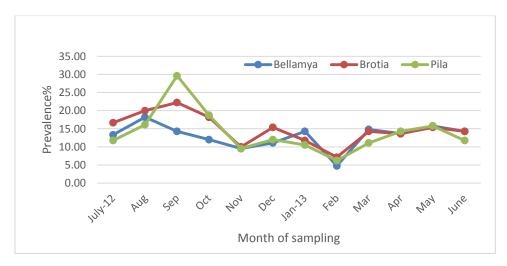


Figure 5.16 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Salna BSMAU Pond of Gazipur (2012-13).

Comparatively highest percentage of infection by trematodes in *Bellamya* sp was found in August which was 18.18% while lowest percentage of trematode infection was 4.76% in February 2013 (Fig 5.16).

Highest percentage of infection by trematodes in *Brotia* sp was found in September which was 22.22% and lowest infection was 7.14% in February.

Highest percentage of infection by trematodes in *Pila* sp. was 29.63% in September 2012 and lowest trematode infection was 6.25% in February 2013.

• Mawna Bazar Pond

In the present observation highest percentage of infection by trematodes in *Bellamya* sp. was found in September which was 28.57% and lowest percentage of trematode infection was 5.26% in February 2012 (Fig 5.17).

Highest percentage of occurrence of infection by trematodes in *Brotia* sp. was found in June which was 18.75% and lowest infection was 6.25% in February.

In *Pila* sp. highest percentage of infection by trematodes was 33.33% in September 2011 and lowest trematode infection was 7.69% in February 2012.

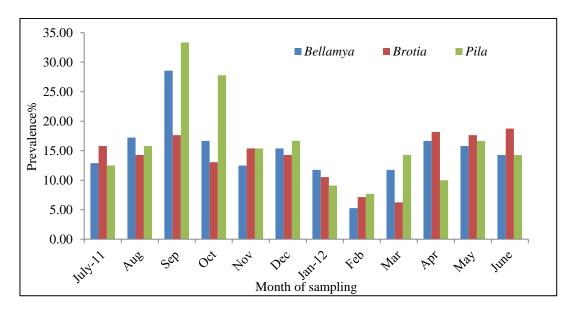


Figure 5.17 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Maona Bazar Pond of Gazipur (2011-12).

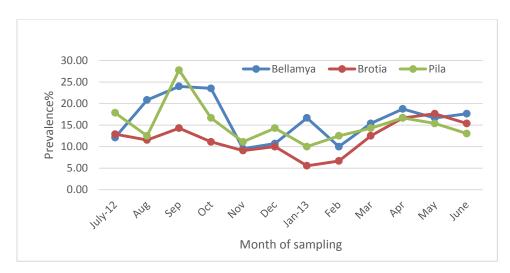


Figure 5.18 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Maona Bazar Pond of Gazipur (2012-13).

The highest percentage of *Bellamya* sp. was observed in September which was 24% and lowest was 9.52% in November 2012 (Fig 5.18).

In case of *Brotia* sp. the highest percentage was 17.65% May and the lowest was 5.56%. In January 2013. In case of *Pila* sp. the highest was 27.78% in September 2012 and lowest was 10% in January 2013.

• Tongi Bari Bazar Pond

Increasing trend of trematode infection in *Bellamya* sp. was observed from July 2011 to September 2011 which was then decreased up to February 2012. Highest percentage of infection by trematodes in *Bellamya* sp was found in September which was 25.81% and lowest was 5.88% in February 2012 (Fig 5.19).In *Brotia* sp. highest percentage of infection by trematodes was found in June (18.75%) and lowest infection (6.25%) in March. Highest percentage of infection by trematodes in *Pila* sp. was 20% in September 2011 and lowest trematode infection was 7.69% in February 2012.

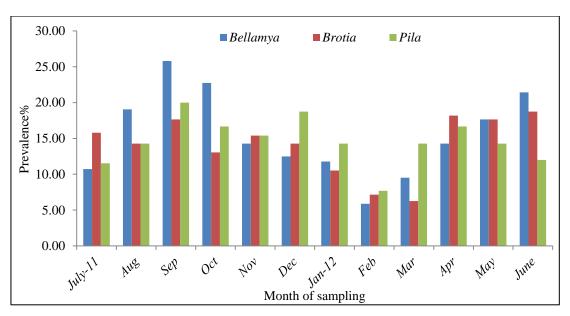


Figure 5.19 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Tong Bari Bazar Pond of Munshiganj (2011-12).

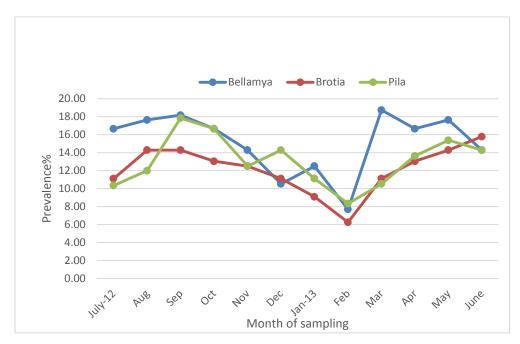


Figure 5.20 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Tong Bari Bazar Pond of Munshiganj (2012-13).

Trematode infection in *Bellamya* sp. was observed to increase from July 2012 to August 2012 in increasing trend which was decreased up to February 2013 with slightly increased in December and January. Highest percentage of infection by trematodes in *Bellamya* sp was found in March which was 18.75% and lowest percentage of trematode infection was 7.69% in February 2013 (Fig 5.20).

Highest percentage of infection by trematodes in *Brotia* sp. was found in June (15.79%) and lowest infection (6.25%) in February 2013.

Highest percentage of infection by trematodes in *Pila* sp. was 17.86% in September 2012 and lowest trematode infection was 8.33% in February 2013.

• Lohajang Bazar Pond, Munshiganj

Highest percentage of occurrence of infection by trematodes in *Bellamya* sp. was found in September which was 23.81% and lowest percentage of trematode infection was 6.25% in February 2012 (Fig 5.21).

In *Brotia* sp. highest percentage of infection by trematodes was found in May (17.65%) and lowest infection (7.41%) in October 2011.

Highest percentage of infection by trematodes in *Pila* sp. was 19.35% in September 2011 and lowest trematode infection was 7.69% in April 2012.

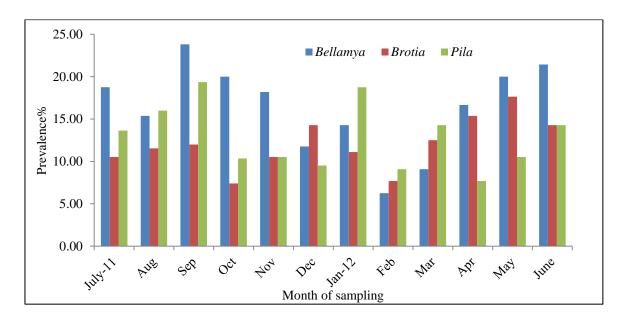


Figure 5.21 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Lohajang Bazar Pond, Munshiganj (2011-12).

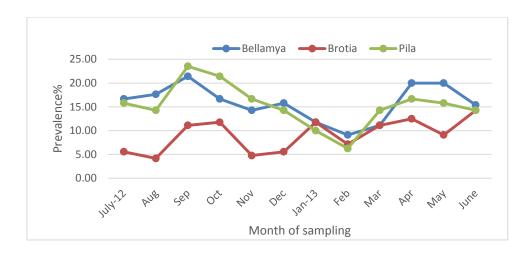


Figure 5.22 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Lohajang Bazar Pond, Munshiganj (2012-13).

Increasing trend of trematode infection in *Bellamya* sp. was observed from July 2012 to September 2012 which was then decreased up to February 2013 with slightly increase in December .However, highest percentage of infection by trematodes in *Bellamya* sp. was found in September which was 21.43% and lowest percentage of trematode infection was 9.09% in February 2013 (Fig 6.11).

Highest percentage of infection by trematodes in *Brotia* sp. was found in June which was 14.29% and lowest infection was 4.17% in August.

In *Pila* sp. highest percentage of infection by trematodes was 23.53% in September 2012 and lowest trematode infection was 6.25% in February 2013.

Site 2: Khulna and surrounding regions

• Khulna University Pond

In the present investigation increasing trend of trematode infection in *Bellamya* sp. was observed during July 2011 to September 2011 which was in decreasing trend up to March 2012 with slightly increased in February. Highest percentage of infection by trematodes in *Bellamya* sp. was found in September which was 36.3% and no infection was found in May 2012 (Fig 5.23).

Comparatively highest percentage of infection by trematodes in *Brotia* sp. was found in September (18.18%) and no infection was found in February. Highest percentage of infection

by trematodes in *Pila* sp. was 16.67% in September 2011 and lowest trematode infection (7.69%) in November and February 2012.

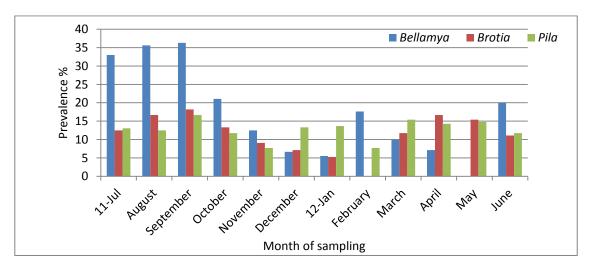


Figure 5.23 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Khulna University Pond of Dhaka (2011-12).

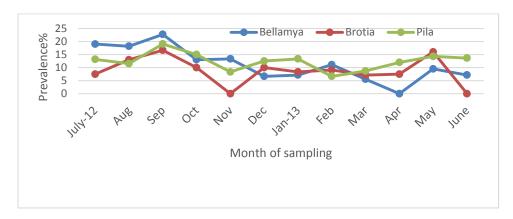


Figure 5.24 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Khulna University Pond of Dhaka (2012-13).

Increasing trend of trematode infection in *Bellamya* sp. was observed from July 2012 to September 2012 which was in decreasing trend up to March 2013 with slightly increased in February. In *Bellamya* sp. trematode infection was found in September which was 22.72% and no trematode infection was found in April 2013 (Fig 5.24).

Highest percentage of infection by trematodes in *Brotia* sp. was found in September 2012 which was 16.66% and no infection was found in November and June 2013. Highest percentage of infection by trematodes in *Pila* sp. was 19% in September 2012 and no trematode infection was in December, February and April 2013.

• Dacope Upazila Complex pond

The trematode infection in *Bellamya* sp. was observed to increase from July 2011 to September 2011. Highest percentage of infection by trematodes in *Bellamya* sp was found in September (19.04%) and lowest percentage of trematode infection (6.66%) in November 2011 and April 2012 (Fig 5.25).

In *Brotia* sp. highest percentage of infection by trematodes was found in September which was 12.5% and no infection was found in October, November and June 2012.

Highest percentage of infection by trematodes in *Pila* sp. was 15.78% in September 2011 and no trematode infection was found in February 2012.

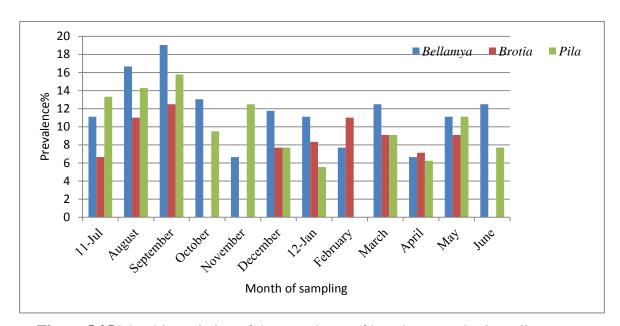


Figure 5.25 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Dacope Upazila Complex pond (2011-12).

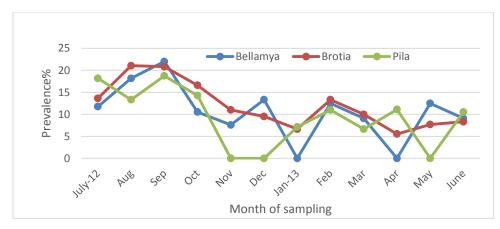


Figure 5.26 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Dacope Upazila Complex pond (2012-13).

Increasing trend of trematode infection in *Bellamya* sp. was observed from July 2012 to September 2012. Highest percentage of infection by trematodes in *Bellamya* sp. was found in September which was 22% while no trematode infection was found in January and April 2013 (Fig 5.26).

Comparatively highest infection in *Brotia* sp. was found in August 2012 which was 21.05% and lowest infection (5.55%) in April 2013. Highest percentage of infection by trematodes in *Pila* sp. was 18.75% in September 2012 and no trematode infection was in November, December, and May 2013.

• Shyamnagar Upazila Complex pond

Comparatively highest percentage of occurrence of infection by trematodes in *Bellamya* sp. was found in September (19.04%) and no infection was found in March and June (Fig 5.27). Highest percentage of infection by trematodes in *Brotia* sp. was found in September which

was 16.6% and no infection was found in March.

In *Pila* sp. highest percentage of infection by trematodes was 21.05% found in August 2011 and lowest trematode infection was 7.69% in March and June.

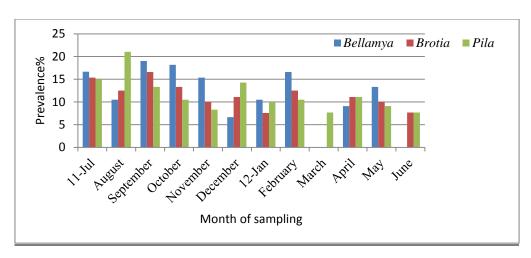


Figure 5.27 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Shyamnagar Upazila Complex pond (2011-12).

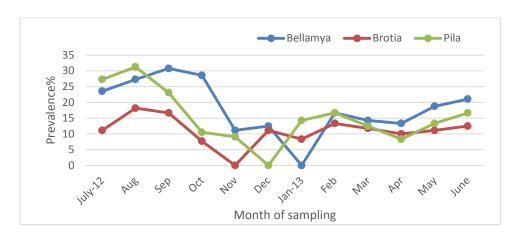


Figure 5.28 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Shyamnagar Upazila Complex pond (2012-13).

Trematode infection in *Bellamya* sp. was observed to increase from July 2012 to September 2012. Highest percentage of infection by trematodes in *Bellamya* sp was found in September which was 30.76% while no trematode infection was found in January 2013 (Fig 5.28).

Highest percentage of occurrence of infection by trematodes in *Brotia* sp. was found in August 2012 which was 18.18% and no infection was found in November 2012. Highest percentage of infection by trematodes in *Pila* sp. was 31.25% in August 2012 and no trematode infection was in December 2012.

Kaliganj Upazila Complex Pond

The present observation trematode infection in *Bellamya* sp. was found in September (22.27%) and lowest percentage of trematode infection (5.88%) in December and April 2012

(Fig 6.15). Highest percentage of infection by trematodes in *Brotia* sp. was found in July which was 15.38% and no infection was found in December, March and April. In *Pila* sp. highest percentage of infection by trematodes was 23.5% found in September 2011 and no infection was found in December and April.

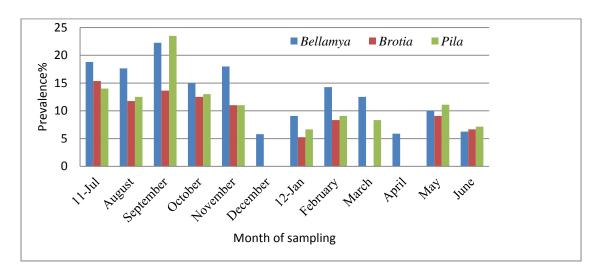


Figure 5.29 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Kaliganj Upazila Complex Pond (2011-12).

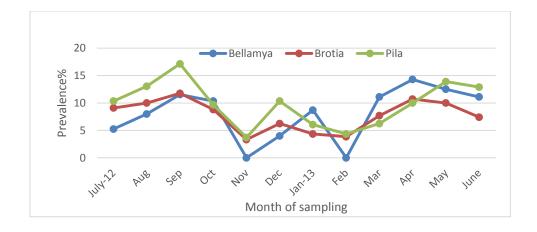


Figure 5.30 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Kaliganj Upazila Complex Pond (2012-13).

In *Bellamya* sp. highest percentage of infection by trematodes was found in April (14.29%) while no trematode infection was found in November and February 2013 (Fig5.30).

Highest percentage of infection by trematodes in *Brotia* sp. was found in September 2012 which was 11.76% and lowest infection was found in November (3.33%) 2012. The highest percentage of infection by trematodes in *Pila* sp. was 17.14% in September 2012 and lowest trematode infection was 3.7% in November 2012.

• Bagerhat PC college Pond

Trematode infection in *Bellamya* sp. was highest in August which was 21.05% and lowest was 5.55% in April 2012 (Fig 5.31).

In *Brotia* sp. highest percentage of infection by trematodes was found in September which was 18.75% and no infection was found in November, December and May 2012.

Highest percentage of infection by trematodes in *Pila* sp. was 22% in September 2011 and no infection was found in January and April 2012.

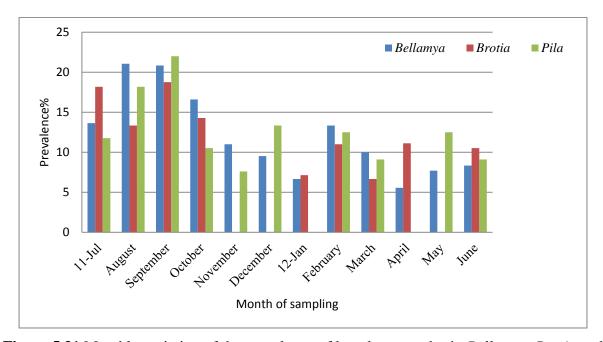


Figure 5.31 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Bagerhat PC College Pond (2011-12).

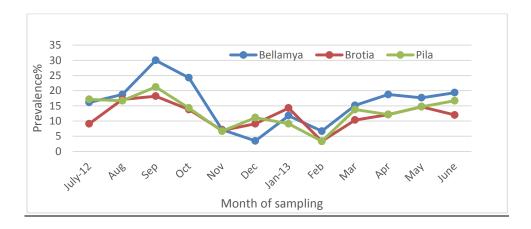


Figure 5.32 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Bagerhat PC College Pond (2012-13).

Increasing trend of trematode infection in *Bellamya* sp. was observed from July 2012 to September 2012 which was in decreasing trend up to February 2013 with slightly increased in January. Highest percentage of infection by trematodes in *Bellamya* sp was found in September (30%) and lowest trematode infection (3.45%) in December 2012 (Fig 5.32).

Highest percentage of infection by trematodes in *Brotia* sp. was found in September 2012 which was 18.18% and lowest infection (3.33%) in February 2013. Highest percentage of infection by trematodes in *Pila* sp. was 21.21% in September 2012 and lowest trematode infection was 3.33% in February 2013.

• Fakirhat Bazaar Pond

Trematode infection in *Bellamya* sp. was getting higher from July 2011 to September which was then decreased up to January. Highest percentage of infection by trematodes in *Bellamya* sp was found in September which was 36.3% and no infection was found in May 2012 (Fig 5.33).

Highest percentage of infection by trematodes in *Brotia* sp. was found in August which was 18.75% and no infection was found in December and January 2012.

Highest percentage of infection by trematodes in *Pila* sp. was 33% in September 2011and no infection was found in January and May 2012.

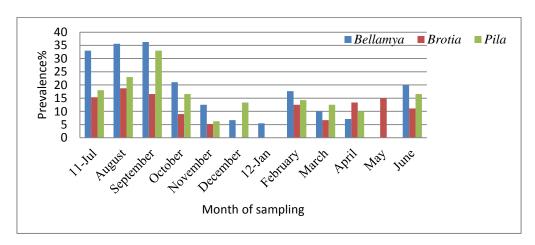


Figure 5.33 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Fakirhat Bazar Pond (2011-12).

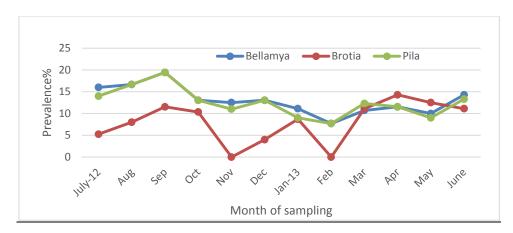


Figure 5.34 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Fakirhat Bazar Pond (2012-13).

In *Bellamya* sp. highest percentage of infection by trematodes was found in September (19.44%) and lowest trematode infection (7.69%) in February 2013 (Fig5.34).

Highest percentage of occurrence of infection by trematodes in *Brotia* sp. was found in April 2013 which was 14.29% and no infection was found in November and February 2013. Highest percentage of infection by trematodes in *Pila* sp. was 19.44% in September 2012 and lowest trematode infection was 7.69% in February 2013.

Site 3: Kishoreganj and surrounding regions

• Sholakia Bazar Pond, Kishoreganj

Increasing trend of trematode infection in *Bellamya* sp. was observed from July 2011 to September 2011 which was in decreasing trend up to January 2012 with slightly increased in December .However, highest percentage of infection by trematodes in *Bellamya* sp was found in September which was 30.76% while no trematode infection was found in January 2012 (Fig 5.35).Highest percentage of infection by trematodes in *Brotia* sp. was found in August which was 18.18% and no infection was found in November 2011. In *Pila* sp. highest percentage of infection by trematodes was 31.25% in August 2011 and no trematode infection was found in December 2011.

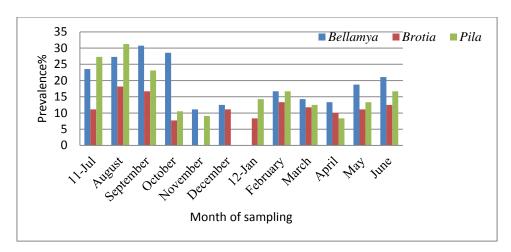


Figure 5.35 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Sholakia Bazar Pond, Kishoreganj (2011-12).

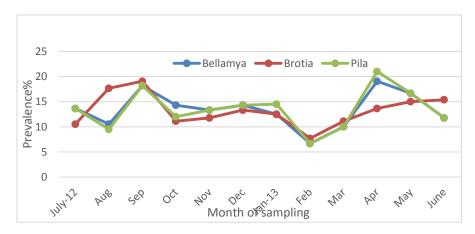


Figure 5.36 Monthly variation of the prevalence of larval trematodes in *Bellamya, Brotia* and *Pila* from Sholakia Bazar Pond, Kishoreganj (2012-13).

It was observed that highest percentage of infection by trematodes in *Bellamya* sp. was found in April (19.05%) and lowest trematode infection (6.67) in February 2013 (Fig5.36).

Highest percentage of infection by trematodes in *Brotia* sp. was found in September which was 19.05% and lowest infection was 7.69% in February 2013. In *Pila* sp. highest percentage of infection was 21% in April 2013 and lowest was 6.67 in February 2013.

• Astagram Upazila Complex pond

In the present investigation trematode infection in *Bellamya* sp. was observed from July 2011 to September 2011 which was in decreasing trend up to January 2012 with slightly increased in December. Highest percentage of infection by trematodes in *Bellamya* sp. was found in August (26.31%) and no trematode infection was found in February 2012 (Fig5.37).

Highest percentage of infection by trematodes in *Brotia* sp. was found in September which was 18.18% and no infection was found in February 2012. Highest percentage of infection by trematodes in *Pila* sp. was 20% in August 2011 and no trematode infection was found in December 2011.

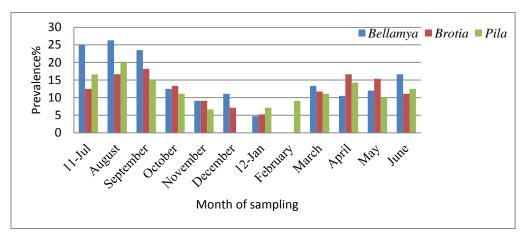


Figure 5.37 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Astagram Upazila Complex pond (2011-12).

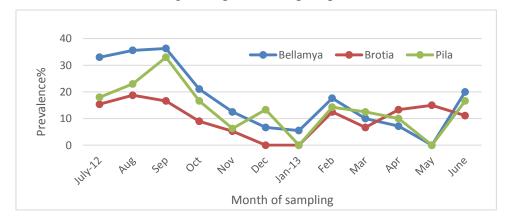


Figure 5.38 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Astagram Upazila Complex pond (2012-13).

In case of *Bellamya* sp. highest percentage of infection by trematodes was found in September which was 36.3% while no trematode infection was found in May 2013 (Fig 5.38).

Highest percentage of infection by trematodes in *Brotia* sp. was found in August which was 18.75% and no infection was found in December and January 2013. Highest percentage of infection by trematodes in *Pila* sp. was 33% in September 2013 and no trematode infection was found in January and May 2013.

Kuliarchar Bazarpond, Kishoreganj

The highest percentage of infection by trematodes in *Bellamya* sp. was found in August 2011 which was 27.27% and lowest trematode infection was 7.14% in November 2012 (Fig 5.39). Highest percentage of infection by trematodes in *Brotia* sp. was found in September which was 18.18% and no infection was found in November, December and March 2012. In *Pila* sp. highest percentage of infection by trematodes was 18.75% in July 2011 and no trematode infection was found in December 2011.

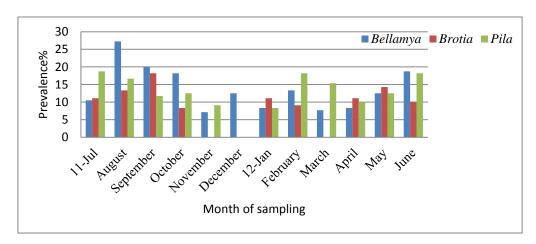


Figure 5.39 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Kuliarchar Bazar, Kishoreganj pond (2011-12).

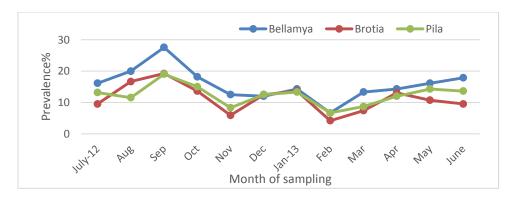


Figure 5.40 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Kuliarchar Bazar, Kishoreganj pond (2012-13).

In *Bellamya* sp. highest percentage of infection by trematodes was found in September 2012 (27.59%) and lowest trematode infection (6.67%) in February 2013 (Fig 5.40).

Highest percentage of infection by trematodes in *Brotia* sp. was found in September (19.23%) and lowest infection (4.17%) in February 2013. Highest percentage of infection by trematodes in *Pila* sp. was 19.05% in September 2012 and lowest trematode infection was 6.67% in February 2013.

• Mithamain bazar Pond

The trematode infection in *Bellamya* sp. was observed to increase during July 2011 to September 2011 which was in decreasing trend up to December 2011. Highest percentage of infection by trematodes in *Bellamya* sp. was found in September (37.5%) and lowest trematode infection (7.69%) in December 2011 (Fig 5.41).

In *Brotia* sp. highest percentage of infection by trematodes was found in August which was 18.18% and no infection was found in November, December and April 2012. Highest percentage of infection by trematodes in *Pila* sp. was 23.8% in September 2011 and lowest trematode infection was 5.55% in October 2011.

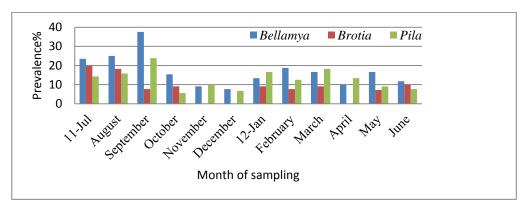


Figure 5.41Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Mithamain, Kishoreganj Bazar pond (2011-12).

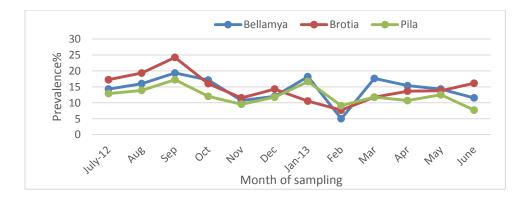


Figure 5.42Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Mithamain, Kishoreganj Bazar pond (2012-13).

Increasing trend of trematode infection in *Bellamya* sp. was observed from July 2012 to September 2012 which was in decreasing trend up to February 2013 with slightly increase in January. Highest percentage of infection by trematodes in *Bellamya* sp. was found in September which was 19.35% while lowest was 5% in February 2013 (Fig 5.42).

In case of *Brotia* sp. highest percentage of infection by trematodes was found in September (24.24%) and lowest trematode infection (7.69%) in February 2013.

Highest percentage of infection by trematodes in *Pila* sp. was 17.24% in September 2012 and lowest trematode infection was 7.69% in June 2013.

• BAU, Mymensingh Pond

It was observed that increasing trend of trematode infection in *Bellamya* sp. was found from July 2011 to September 2011 which was in decreasing trend up to December 2011. Highest percentage of infection by trematodes in *Bellamya* sp was found in September (25%) and no trematode infection was found in April 2012 (Fig5.43).

Highest percentage of infection by trematodes in *Brotia* sp. was found in March which was 13.33% and no infection was found in October, November, January and May 2012. In *Pila* sp. highest percentage of infection by trematodes was 23.07% in September 2011 and lowest was 6.66% in December 2011.

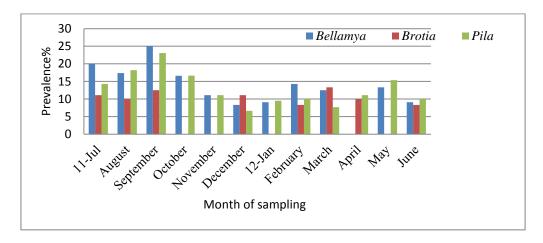


Figure 5.43 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from BAU, Mymensingh Pond (2011-12).

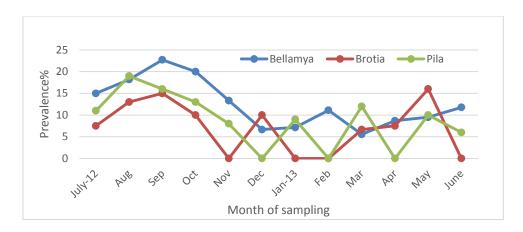


Figure 5.44 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from BAU, Mymensingh Pond (2012-13).

Increasing trend of trematode infection in *Bellamya* sp. was observed from July 2012 to September 2012. Highest percentage of infection by trematodes in *Bellamya* sp. was found in September (22.72%) and lowest trematode infection (5.55%) in March 2013 (Fig 5.44).

It was observed that highest percentage of infection by trematodes in *Brotia* sp. was found in May (16%) and no infection was found in November and January, February and June 2013. Highest percentage of infection by trematodes in *Pila* sp. was 19% in August 2012 and no trematode infection was found in December, February and April 2013.

• Trishal Kabi Nazrul University Pond

Highest percentage of occurrence of infection by trematodes in *Bellamya* sp. was found in August 2011 which was 17.64% while no trematode infection was found in April 2012 (Fig 5.45).

Similarly, highest percentage of infection by trematodes in *Brotia* sp. was found in September and May (16.6%) and lowest infection (6.25%) in August 2011. Highest percentage of infection by trematodes in *Pila* sp. was 23.52% in August 2011 and no trematode infection was found in December 2011.

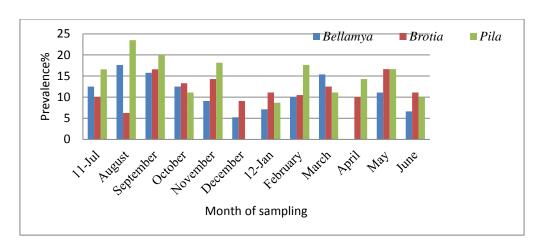


Figure 5.45 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Trishal Kabi Nazrul University Pond (2011-12).

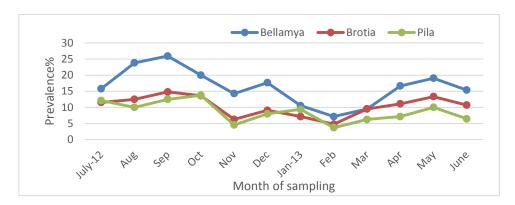


Figure 5.46 Monthly variation of the prevalence of larval trematodes in *Bellamya*, *Brotia* and *Pila* from Trishal Kabi Nazrul University Pond (2012-13).

Highest percentage of infection by trematodes in *Bellamya* sp. was found in September 2012 which was 25.93% and lowest trematode infection was 7.14% in February 2013 (Fig 5.46).

In present investigation highest percentage of infection by trematodes in *Brotia* sp. was found in September (14.81%) and lowest infection (4.76%) in February 2013. Highest percentage of infection by trematodes in *Pila* sp. was 13.79% in October 2012 and lowest trematode infection was 3.7% in February 2013.

Comparison of prevalence of trematode parasites

Ramna Park Pond

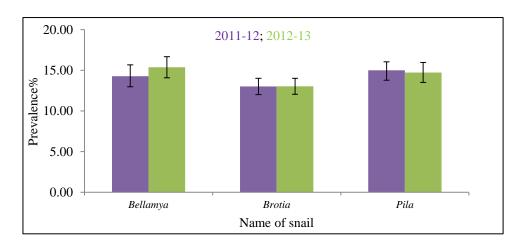


Figure 5.47Prevalenceof trematode parasites in different snails sampled from Ramna Park Pond in different two sampling years. Bar (Mean±SEM).

Similar prevalence of trematode parasites was found in all the snails in both years.

Curzon Hall Pond

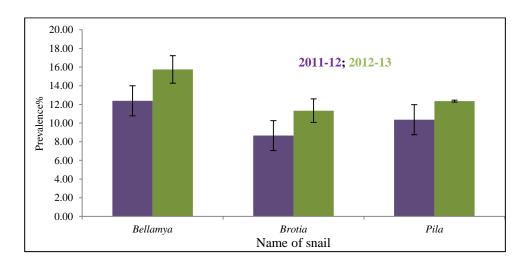


Figure 5.48Prevalence of trematode parasites in different snails sampled from Curzon Hall Pond in two different sampling years. Bar (Mean±SEM).

In 2011-12, occurrence of trematode parasite in *Bellamya* was significantly higher than that of *Brotia* and *Pila* (p<0.05). However, similar occurrence of this parasite in *Brotia* and *Pila* was detected. Moreover, in 2012-13 occurrence of trematode parasite in *Bellamya* was significantly higher than that of *Brotia* and *Pila*. However, similar occurrence of this parasite was found in *Brotia* and *Pila*. On the other hand, occurrence of this parasite in *Bellamya*

found in 2012-13 was significantly higher than that of 2011-12. No significant different was found in *Brotia* and *Pila* in this two years (p>0.05).

Jagannath Hall Pond

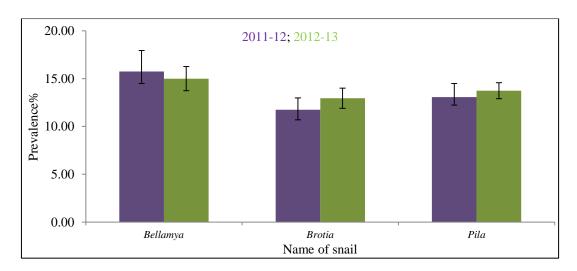


Figure 5.49 Occurrence of trematode parasites in different snails sampled from Jagannath Hall Pond in two different sampling years. Bar (Mean±SEM).

Similar prevalence of trematode parasites was found in all the snails in both years.

Keraniganj Pond

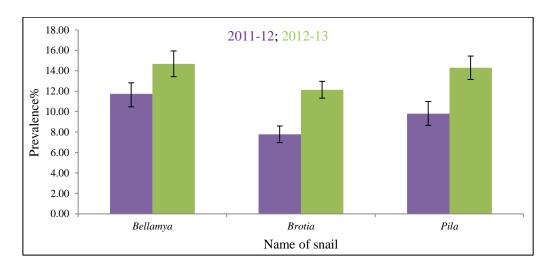


Figure 5.50 Occurrence of trematode parasites in different snails sampled from Keraniganj Pond in two different sampling years. Bar (Mean±SEM).

In 2011-12, occurrence of trematode parasite in *Bellamya* was significantly higher than that of *Brotia* and *Pila* (p<0.05). However, similar occurrence of this parasite in *Brotia* and *Pila* was detected. Moreover, in 2012-13 occurrence of trematode parasite in *Bellamya* and *Pila*

was similar but significantly higher than that of *Brotia*. Besides, occurrence of this parasites in all the snails in 2012-2013 was significantly higher than that of 2011-2012 (p<0.05).

Tongi Railway Station Pond

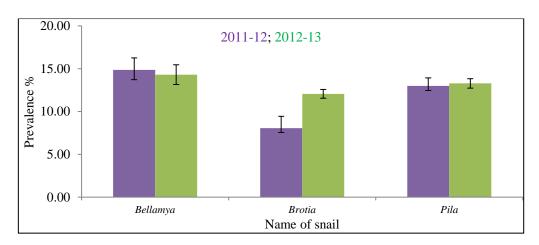


Figure 5.51 Occurrence of trematode parasites in different snails sampled from Tongi Railway Station Pond in two different sampling years. Bar (Mean±SEM).

In 2011-12, occurrence of trematode parasite in *Bellamya* and *Pila* was significantly higher than that of *Brotia* (p<0.05). Moreover, in 2012-13 occurrence of trematode parasite in *Bellamya* was significantly higher than that of *Brotia* and *Pila*. However, similar occurrence of this parasite was found in *Brotia* and *Pila*. On the other hand, occurrence of this parasite in *Brotia* found in 2012-13was significantly higher than that of 2011-12. No significant different was found in *Bellamya* and *Pila* in this two years (p>0.05).

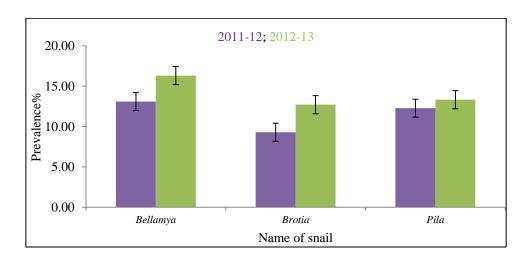


Figure 5.52 Occurrence of trematode parasites in different snails sampled from Jaydevpur Railway Station Pond in two different sampling years. Bar (Mean±SEM).

In 2011-12, occurrence of trematode parasite in *Bellamya* and Pila was significantly higher than that of *Brotia* (p<0.05). Moreover, in 2012-13 occurrence of trematode parasite in *Bellamya* was significantly higher than that of *Brotia* and *Pila*. However, similar occurrence of this parasite was found in *Brotia* and *Pila*. On the other hand, occurrence of this parasite *Bellamya* found in 2012-13 was significantly higher than that of 2011-12. No significant different was found in *Brotia* and *Pila* in this two years (p>0.05).

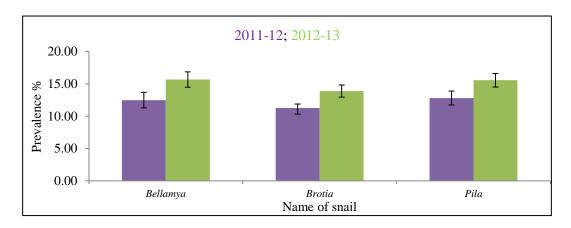


Figure 5.53 Occurrence of trematode parasites in different snails sampled from Bhawal Gazipur Railway Station Pond in two different sampling years. Bar (Mean±SEM).

In 2011-12, occurrence of trematode parasite in *Bellamya* and *Pila* was significantly higher than that of *Brotia* (p<0.05). Moreover, in 2012-13 occurrence of trematode parasite in *Bellamya* was significantly higher than that of *Brotia* and *Pila*. However, similar occurrence of this parasite was found in *Brotia* and *Pila*. On the other hand, occurrence of this parasite in *Bellamya* found in 2012-13 was significantly higher than that of 2011-12. No significant different was found in *Brotia* and *Pila* in this two years (p>0.05).

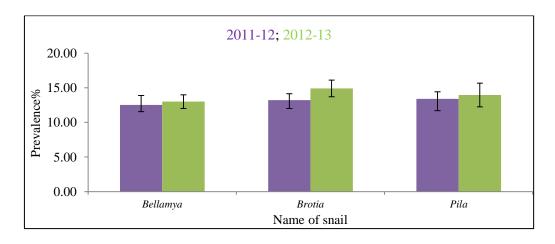


Figure 5.54 Occurrence of trematode parasites in different snails sampled from Salna BSMAU Pond in two different sampling years. Bar (Mean±SEM).

Similar prevalence of trematode parasites was found in all the snails in both years

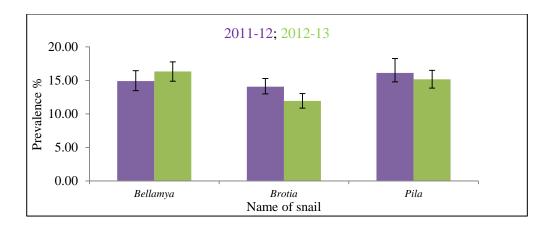


Figure 5.55 Occurrence of trematode parasites in different snails sampled from Maona Bazar Pond in two different sampling years. Bar (Mean±SEM).

In 2011-12, similar occurrence of trematode parasite in all the snails was found. In 2012-13 occurrence of trematode parasite in *Bellamya* and *Pila* was significantly higher than that of *Brotia* (p<0.05). However, similar occurrence of this parasite was found in *Bellamya* and *Pila*. No significant different was found in all the snails in this two years (p>0.05).

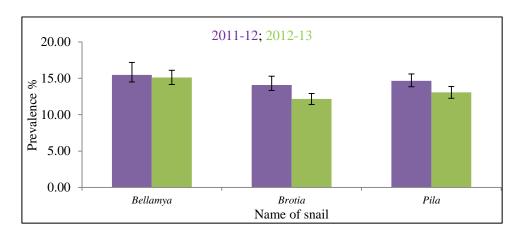


Figure 5.56 Occurrence of trematode parasites in different snails sampled from Tongi Bari Bazar Pond in two different sampling years. Bar (Mean±SEM).

Similar prevalence of trematode parasites was found in all the snails in both years.

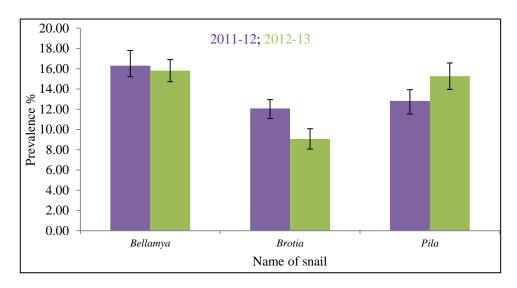


Figure 5.57 Occurrence of trematode parasites in different snails sampled from Lohajang Bazar Pond in two different sampling years. Bar (Mean±SEM).

In 2011-12, occurrence of trematode parasite in *Bellamya* was significantly higher than that of *Brotia* and *Pila* (p<0.05). However, similar occurrence of this parasite in *Brotia* and *Pila* was detected. Moreover, in 2012-13 occurrence of trematode parasite in *Bellamya* and *Pila* was significantly higher than that of *Brotia*. However, similar occurrence of this parasite was found in *Bellamya* and *Pila*. No significant different was found in all the snails in this two years (p>0.05).

Khulna University Pond

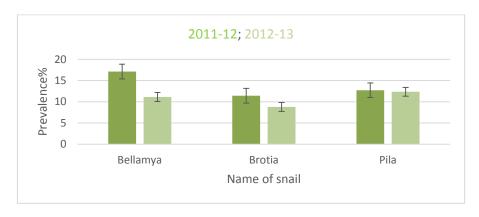


Figure 5.58 Occurrence of trematode parasites in different snails sampled from Khulna University Pond in two different sampling years.

In 2011-12, occurrence of trematode parasite in *Bellamya* was significantly higher than that of *Brotia* and *Pila* (p<0.05). However, similar occurrence of this parasite in *Brotia* and *Pila* was detected. Moreover, in 2012-13 occurrence of trematode parasite in *Pila* was

significantly higher than that of *Bellamya* and *Brotia*. No significant different was found in *Pila* in this two years (p>0.05).

Dacope upazila Complex Pond

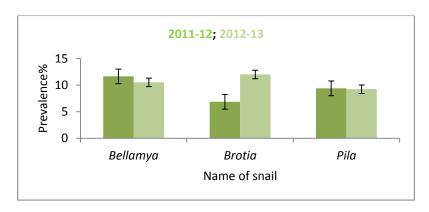


Figure 5.59 Occurrence of trematode parasites in different snails sampled from Dacope upazila Complex Pond in two different sampling years.

In 2011-12, occurrence of trematode parasite in *Bellamya* was significantly higher than that of *Brotia* and *Pila* (p<0.05). Moreover, in 2012-13 occurrence of trematode parasite in *Brotia* was significantly higher than that of *Bellamya* and *Pila*. No significant different was found in *Bellamya* and *Pila* in this two years (p<0.05).

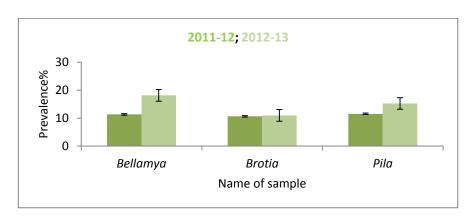


Figure 5.60 Occurrence of trematode parasites in different snails sampled from Syamnagar Upazila Complex pond in two different sampling years.

Similar prevalence of trematode parasites was found in all the snails in 2011-2012. Moreover, in 2012-13 occurrence of trematode parasite in *Bellamya* was significantly higher than that of *Brotia* and *Pila*. No significant different was found in *Brotia* in this two years (p<0.05).

Kaliganj Upazila Complex Pond

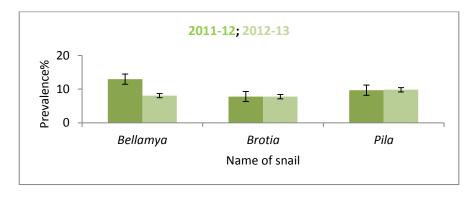


Figure 5.61 Occurrence of trematode parasites in different snails sampled from Kaliganj Upazila Complex Pond in two different sampling years.

In 2011-12, occurrence of trematode parasite in *Bellamya* was significantly higher than that of *Brotia* and *Pila* (p<0.05). Moreover, in 2012-13 occurrence of trematode parasite in Pila was significantly higher than that of *Bellamya* and *Brotia*. Similar occurrence of this parasite was found in *Pila* and *Brotia* in the two years (p<0.05).

Bagerhat DC Bunglow Pond

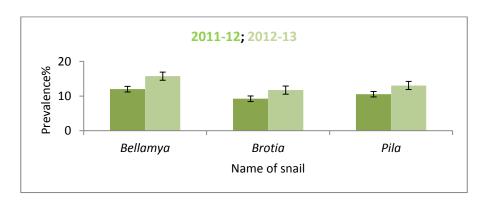


Figure 5.62 Occurrence of trematode parasites in different snails sampled from Bagerhat DC Bunglow Pond in two different sampling years.

In 2011-12, occurrence of trematode parasite in *Bellamya* and *Pila* was significantly higher than that of *Brotia* (p<0.05). Moreover, in 2012-13 occurrence of trematode parasite in *Bellamya* was significantly higher than that of *Brotia* and *Pila*. However, similar occurrence of this parasite was found in *Brotia* and *Pila*. On the other hand, occurrence of this parasite found in 2012-13 was significantly higher than that of 2011-12 in all snails (p>0.05).

Fakirhat bazar pond

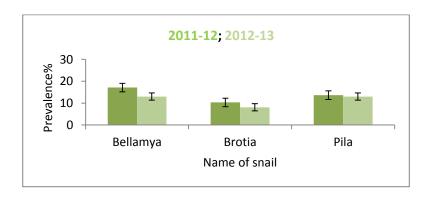


Figure 5.63 Occurrence of trematode parasites in different snails sampled from Fakirhat bazar pond in two different sampling years.

In 2011-12, the occurrence of trematode parasite in *Bellamya* was significantly higher than that of *Brotia* and *Pila* (p<0.05). Moreover, in 2012-13 occurrence of trematode parasite in *Bellamya* was significantly higher than that of *Brotia* and *Pila*. However, similar occurrence of this parasite was found in *Pila* in two years (p>0.05).

Solakia bazaar Pond, Kishoreganj

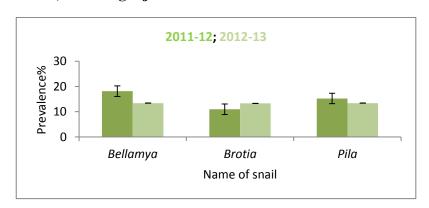


Figure 5.64 Occurrence of trematode parasites in different snails sampled from Solakia bazaar pond, Kishoreganj in two different sampling years.

In 2011-12, the occurrence of trematode parasite in *Bellamya* was significantly higher than that of *Brotia* and *Pila* (p<0.05). However, similar occurrence of this parasite was found in all snails in 2012-13.

Astagram Upazila Complex pond

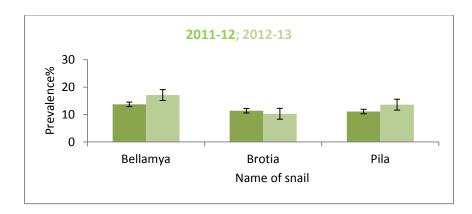


Figure 5.65 Occurrence of trematode parasites in different snails sampled from Astagram Upazila Complex pond in two different sampling years.

In 2012-13, the occurrence of trematode parasite in *Bellamya* was significantly higher than that of *Brotia* and *Pila* (p<0.05). However, similar occurrence of this parasite was found in *Brotia* in two years (p>0.05).

Kuliarchar bazar pond

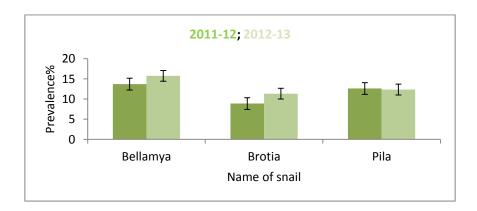


Figure 5.66 Occurrence of trematode parasites in different snails sampled from Kuliarchar bazar pond in two different sampling years.

In 2012-13, occurrence of trematode parasite in *Bellamya* was significantly higher than that of *Brotia* and *Pila* (p<0.05). However, similar occurrence of this parasite was found in *Pila* in two years (p>0.05).

Mitamain bazar pond

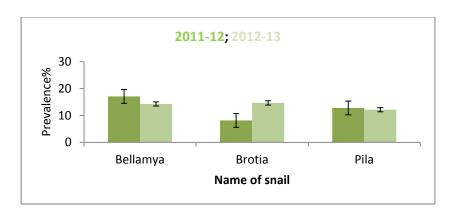


Figure 5.67 Occurrence of trematode parasites in different snails sampled from Mitamain bazar pond in two different sampling years.

In 2011-12, occurrence of trematode parasite in *Bellamya* and *Pila* was significantly higher than that of *Brotia* (p<0.05). However, similar occurrence of this parasite was found in *Bellamya* and *Pila*. On the other hand, occurrence of this parasite in *Brotia* found in 2012-13 was significantly higher than that of 2011-12. No significant different was found in *Bellamya* and *Pila* in this two years (p>0.05).

BAU pond, Mymensingh

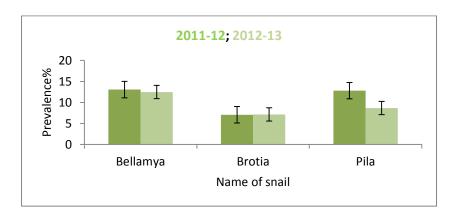


Figure 5.68 Occurrence of trematode parasites in different snails sampled from BAU pond, Mymensingh in two different sampling years.

In 2011-12, occurrence of trematode parasite in *Pila* was significantly higher than that of *Brotia* (p<0.05). However, similar occurrence of this parasite was found in *Bellamya* and *Brotia in two* years (p>0.05). Whereas, occurrence of this parasite in *Pila* found in 2011-12 was significantly higher than that of 2012-13(p<0.05).

Trishal Kabi Nazrul University Pond

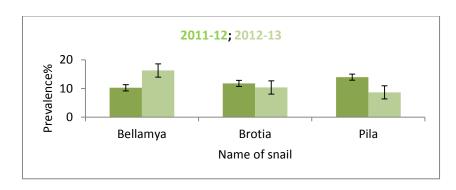


Figure 5.69 Occurrence of trematode parasites in different snails sampled from Trishal Kabi Nazrul University Pond in two different sampling years.

In 2012-13, occurrence of trematode parasite in *Bellamya* was significantly higher than that of *Brotia* and *Pila* (p<0.05). However, similar occurrence of this parasite was found in *Brotia in two* years (p>0.05). On the other hand, occurrence of this parasite in *Pila* found in 2011-12 was significantly higher than that of 2012-13(p<0.05).

Seasonal variation of Trematode and Environmental variables

The prevalence of trematode parasites in *Bellamya, Brotia* and *Pila* in 2011-2012 and 2012-2013 sampled from 23 different regions were determined. Each year was divided into three different seasons (Rainy, winter and summer) where the incidence of the parasites in rainy season in most of the cases was significantly higher than that of other seasons. However, the prevalence in summer (in most of the regions) was significantly higher than that of winter.

Table 5.1 Seasonal variation of occurrence (%) of parasite in *Bellamya* spp.(Site 1-Dhaka).

Region		Season 2011-1	2	F	Sig.		Season 2012-13	3	F	Sig.
	Rainy	Winter	Summer			Rainy	Winter	summer		
Ramna	11.63±2.74	10.71±1.68	14.49±1.8	2.63	0.126	19.38±1.91	10.78±0.91	15.93±1.29	9.127	.007
Curjon	16.54±2.76	13.24±0.70	7.4±1.36	6.46	0.018	20.46±2.5	11.37±1.64	15.4±1	6.283	.020
Jagannath	22.28±3.08	17.73±0.93	7.26±1.71	13.42	0.002	18.82±1.91	12.07±2.33	14.12±0.74	3.724	.066
Keranigonj	15.24±1.34	8.13±1.08	11.85±1.27	8.26	.009	19.21±1.81	11.01±1.36	13.83±0.9	8.739	.008
Tongi	19.03±2.67	11.95±1.56	13.65±1.67	3.31	.084	18.74±1.33	11.07±1.23	13.14±0.93	11.398	.003
Jaydebpur	17.82±2.33	9.22±1.58	12.24±0.78	6.7	.017	21.38±2.23	12.4±2.28	15.15±2.06	4.448	.008
Bhawal-	14.57±2.5	9.28±1.6	13.56±1.4	2.2	0.167	18.44±1.72	11.41±1.28	17.12±1.36	6.471	.018
Salna	15.88±2.94	9.47±2.19	12.22±0.53	2.26	.16	14.45±1.33	9.92±1.99	14.63±0.45	3.615	.070
Maona	18.85±3.38	11.23±2.14	14.63±1.07	2.55	.133	18.99±3.44	10.33±1.52	11.82±2.56	4.984	.035
Tongi Bari	19.58±3.26	11.11±1.82	15.72±2.53	2.65	.125	17.29±0.37	11.25±1.41	16.84±0.95	11.156	.004
Lahajang	19.49±1.74	12.62±2.5	16.8±2.76	2.13	.175	18.1±1.13	12.73±1.47	16.62±2.14	2.88	.108

Values are in mean \pm SEM. Multivariate comparison between seasons was performed using ANOVA at 5% level of significance (p<0.05).

Table 5.2 Seasonal variation of occurrence (%) of parasite in *Brotia* spp (Site 1-Dhaka).

Region		Season 2011-12	2	F	Sig.	\$	Season 2012-13	3	F	Sig.
	Rainy	Winter	Summer			Rainy	Winter	summer		
Ramna	12.23±1.26	1667±1.98	16.10±0.71	3.9	0.06		9.2±1.33	14.22±0.68	11.05	0.004
Curjon	12.10±0.79	6.4±1.36	7.46±0.87	8.6	0.008	14.77±2.09	9.05±2.36	10.17±1.78	2.43	0.143
Jagannath	14.55±2.05	8.4±2.29	12.29±0.90	2.83	0.112	14.58±1.64	12.6±2.02	11.67±1.95	.623	.558
Keranigonj	9.92±.66	4.41±.64	8.95±.82	16.77	.001	14.01±1.14	11.76±1.74	10.67±1.06	1.6	.255
Tongi	8.79±1.39	3.18±2.07	12.25±0.75	9.3	.006	11.69±0.89	11.06±0.61	13.46±0.82	2.54	0.134
Jaydebpur	10.23±0.48	8.08±.77	9.57±.52	3.36	.081	13.49±1.09	10.59±1.67	14.06±0.8	2.25	0.161
Bhawal	12.52±0.80	9.1±1.09	12.14±.31	5.5	.027	15.52±0.66	13.08±2.44	13.02±1.43	0.72	0.513
Salna	14.59±2.2	11.32±1.25	13.78±0.97	1.19	.35	19.27±1.2	11.07±1.72	14.4±0.36	11.25	.004
Maona	15.19±0.99	11.84±1.88	15.21±2.0	.84	.643	12.46±0.72	7.83±1.03	15.55±1.11	15.98	.001
Tongi Bari	15.19±.99	11.84±1.88	15.21±2.99	.84	.463	13.18±0.75	9.74±1.36	13.56±0.99	33.92	.059
Lahajang	10.37±1.03	10.91±1.35	14.96±1.08	4.6	.041	8.15±1.92	7.3±1.56	11.74±1.73	2.27	0.16

Values are in mean \pm SEM. Multivariate comparison between seasons was performed using ANOVA at 5% level of significance (p<0.05).

Table 5.3 Seasonal variation of occurrence (%) of parasite in *Pila* spp (Site 1-Dhaka).

Region		Season 2011-12	2	F	Sig.		Season 2012-13	3	F	Sig.
	Rainy	Winter	Summer			Rainy	Winter	summer		
Ramna	18.54±1.07	12.17±1.78	14.29±0.64	6.7	0.017	17.83±1.55	10.48±1.77	15.84±1.28	6.04	,022
Curjon	13.12±0.71	6.81±0.90	11.17±0.80	16.04	.001	14.69±1.61	10.21±1.61	12.16±1.25	2.24	.163
Jagannath	17.33±1.44	7.56±1.67	14.32±0.95	13.05	.002	13.17±1.22	12.99±1.88	15.06±1.28	.59	.575
Keranigonj	12.55±1.7	6.11±1.49	10.76±1.71	4.12	.054	16.7±1.06	11.47±2.7	14.72±1.27	2.08	.181
Tongi	16.29±1.31	11.09±1.20	11.64±0.94	6.04	.022	15.4±1.97	10.6±1.73	14.5±0.45	2.76	.116
Jaydebpur	12.16±1.25	9.47±1.49	15.23±1.65	3.8	.064	15.63±1.92	10.05±1.45	14.3±0.9	3.84	.062
Bhawal	16.27±.96	9.49±0.81	12.58±1.99	6.24	.020	19.66±1	11.98±0.83	15.02±0.75	19.89	.000
Salna	14.16±1.57	11.7±1.72	14.37±2.1	0.67	.534	19.07±3.8	9.57±1.72	13.24±1.09	4.01	.057
Maona	22.35±4.92	12.21±2.24	13.81±1.4	2.87	.109	18.7±3.24	11.97±0.93	14.84±0.77	2.9	0.11
Tongi Bari	15.63±1.8	14.28±2.32	14.31±.95	.229	.8	14.21±1.8	11.56±1.26	13.46±1.04	.950	.422
Lahajanj	14.83±1.9	11.97±2.28	11.7±1.6	.794	.481	18.76±2.21	11.8±2.3	15.26±0.59	3.44	.078

Values are in mean \pm SEM. Multivariate comparison between seasons was performed using ANOVA at 5% level of significance (p<0.05).

Table 5.4 Seasonal variation of parasite in *Bellamya* spp (Site 2 and 3-Khulna and Kishoreganj).

Region		Season 2011-12	2	F	Sig.		Season 2012-1	3	F	Sig.
	Rainy	Winter	Summer			Rainy	Winter	summer		
KU	31.49±3.54	10.58±2.81	9.29±4.14	12.37	0.003	18.24±1.99	9.56±1.60	5.55±2.02	11.87	0.003
Dacope	14.96±1.78	9.31±1.52	10.69±1.38	3.92	0.060	15.62±2.71	8.36±3.06	7.67±2.68	2.43	0.143
Symnagar	16.10±1.92	12.29±2.29	5.61±3.35	4.20	0.042	27.53±1.52	10.07±3.56	16.85±1.83	12.70	0.002
Kaligong	18.43±1.51	11.79±2.71	8.66±1.58	6.17	0.021	8.79±1.39	3.18±2.07	12.25±0.75	9.30	0.006
BagerhatDc	18.03±1.79	10.13±1.40	7.89±0.92	14.19	0.002	22.28±3.08	7.26±1.71	17.73±0.93	13.42	0.002
Fakirhat	31.49±3.55	10.58±2.81	9.29±4.41	12.37	0.003	16.29±1.31	11.09±1.20	11.64±0.94	6.05	0.022
Solakia	27.53±1.52	10.07±3.56	16.85±1.81	12.70	0.002	14.16±1.57	11.70±1.72	14.37±2.10	0.67	0.534
Astagram	21.83±3.16	6.24±2.47	13.13±1.31	10.29	0.005	31.49±3.55	10.58±2.81	9.29±4.14	12.37	0.003
Kuliarchar	18.99±3.44	10.33±1.52	11.82±2.56	3.12	0.093	20.48±2.50	11.37±1.64	15.40±1.00	6.28	0.02
Mithamain	25.35±4.57	12.22±2.49	13.74±1.69	5.18	0.032	16.70±1.06	11.47±2.70	14.72±1.27	2.08	0.181
BAU	19.75±1.90	10.70±1.33	8.73±3.05	7.06	0.014	18.98±1.62	9.56±1.60	8.88±1.28	13.95	0.002
Trisal	14.61±1.27	7.87±1.06	8.29±3.29	3.16	0.092	21.38±2.23	12.40±2.28	15.16±2.03	4.45	0.045

Values are in mean \pm SEM. Multivariate comparison between seasons was performed using ANOVA at 5% level of significance (p<0.05).

Table 5.5 Seasonal variation of trematode parasite in *Brotia* spp (Site 2 and 3-Khulna and Kishoreganj).

Region		Season 2011-12	2	F	Sig.		Season 2012-1	3	F	Sig.
	Rainy	Winter	Summer			Rainy	Winter	summer		
KU	15.17±1.35	5.37±1.95	13.73±1.36	11.23	0.004	11.79±1.97	6.86±2.31	7.66±3.27	1.05	0.388
Dacope	7.54±2.80	6.76±2.36	6.33±2.16	0.06	0.94	18.03±1.79	10.13±1.40	7.89±0.92	14.19	0.002
Symnagar	14.45±0.90	10.30±1.04	7.20±2.50	4.83	0.038	13.41±2.44	8.19±2.92	11.34±0.53	1.41	0.294
Kaligong	13.32±0.79	6.15±2.36	3.94±2.33	6.21	0.02	9.92±0.66	4.45±0.64	8.95±0.82	16.77	0.001
BagerhatDc	16.14±1.36	4.54±2.73	7.07±2.56	7.03	0.014	14.55±2.05	8.40±2.29	12.29±0.90	2.83	0.112
Fakirhat	14.93±2.10	4.44±2.96	11.53±1.81	5.24	0.031	8.79±1.39	3.18±2.07	12.25±0.75	9.30	0.006
Solakia	13.41±2.44	8.19±2.92	11.34±0.53	1.41	0.294	14.59±2.20	11.32±1.25	13.78±0.97	1.19	0.349

Astagram	15.17±1.35	5.37±1.95	13.73±1.36	11.23	0.004	14.93±2.10	4.44±2.96	11.53±1.81	5.24	0.031
Kuliarchar	12.74±2.08	5.05±2.94	8.85±3.09	1.97	0.195	14.77±2.09	9.05±2.36	10.17±1.18	2.43	0.143
Mithamain	13.74±3.12	4.20±2.44	6.56±2.27	3.56	0.073	19.21±1.81	11.01±1.36	13.83±0.90	8.74	0.008
BAU	8.40±2.85	4.86±2.86	7.92±2.84	0.45	0.649	11.38±1.65	2.50±2.50	7.54±3.28	3.01	0.100
Trisal	11.55±2.22	11.25±1.10	12.57±1.46	0.17	0.843	13.12±0.71	6.81±0.90	11.17±0.80	16.04	0.001

Values are in mean \pm SEM. Multivariate comparison between seasons was performed using ANOVA at 5% level of significance (p<0.05).

Table 5.6 Seasonal variation of prevalence (%) of parasite in *Pila* spp (Site 2 and 3-Khulna and Kishoreganj).

	Season 2011-12			518.	ig. Season 2012-13			F	Sig.
Rainy	Winter	Summer			Rainy	Winter	summer		
13.49±1.09	10.59±1.67	14.06±0.80	2.25	0.161	14.69±1.62	10.21±1.16	12.16±1.25	2.24	0.163
13.22±1.34	6.44±2.59	8.54±1.04	3.78	0.064	16.14±1.36	4.54±2.73	7.07±2.56	7.03	0.014
14.98±2.23	10.78±1.26	8.90±0.80	4.04	0.056	23.03±4.49	10.01±3.69	12.71±1.71	3.86	0.062
15.75±2.60	6.69±2.40	6.65±2.37	4.55	0.043	12.55±1.69	6.11±1.49	10.76±1.71	4.12	0.054
15.75±2.60	6.69±2.40	6.65±2.37	4.55	0.043	17.33±1.44	7.55±1.67	14.32±0.95	13.05	0.002
22.65±3.71	8.47±3.34	9.78±3.53	4.93	0.036	16.29±1.31	11.09±1.20	11.64±0.94	6.05	0.022
22.65±3.71	8.47±3.34	9.78±3.53	4.93	0.036	14.16±1.57	11.70±1.72	14.37±2.10	0.67	0.534
15.68±1.85	5.72±1.98	11.97±0.92	9.29	0.006	22.65±3.71	8.47±3.34	9.78±3.53	4.93	0.036
14.92±1.6	8.90±3.72	14.02±1.77	1.60	0.254	14.69±1.62	10.21±1.61	12.16±1.25	2.24	0.163
14.85±3.74	11.46±2.11	12.07±2.36	0.41	0.676	14.01±1.14	11.76±1.74	10.67±1.06	1.60	0.255
18.05±1.86	9.32±0.95	11.05±1.61	9.23	0.007	14.75±1.75	4.25±2.46	7.00±2.65	5.52	0.027
17.81±2.64	11.13±4.30	13.01±1.52	1.28	0.324	12.10±0.79	6.41±1.36	7.46±0.87	8.56	0.008
	13.22±1.34 14.98±2.23 15.75±2.60 15.75±2.60 22.65±3.71 22.65±3.71 15.68±1.85 14.92±1.6 14.85±3.74	13.22±1.34 6.44±2.59 14.98±2.23 10.78±1.26 15.75±2.60 6.69±2.40 15.75±2.60 6.69±2.40 22.65±3.71 8.47±3.34 22.65±3.71 8.47±3.34 15.68±1.85 5.72±1.98 14.92±1.6 8.90±3.72 14.85±3.74 11.46±2.11 18.05±1.86 9.32±0.95	13.22±1.34 6.44±2.59 8.54±1.04 14.98±2.23 10.78±1.26 8.90±0.80 15.75±2.60 6.69±2.40 6.65±2.37 15.75±2.60 6.69±2.40 6.65±2.37 22.65±3.71 8.47±3.34 9.78±3.53 22.65±3.71 8.47±3.34 9.78±3.53 15.68±1.85 5.72±1.98 11.97±0.92 14.92±1.6 8.90±3.72 14.02±1.77 14.85±3.74 11.46±2.11 12.07±2.36 18.05±1.86 9.32±0.95 11.05±1.61	13.22±1.34 6.44±2.59 8.54±1.04 3.78 14.98±2.23 10.78±1.26 8.90±0.80 4.04 15.75±2.60 6.69±2.40 6.65±2.37 4.55 15.75±2.60 6.69±2.40 6.65±2.37 4.55 22.65±3.71 8.47±3.34 9.78±3.53 4.93 22.65±3.71 8.47±3.34 9.78±3.53 4.93 15.68±1.85 5.72±1.98 11.97±0.92 9.29 14.92±1.6 8.90±3.72 14.02±1.77 1.60 14.85±3.74 11.46±2.11 12.07±2.36 0.41 18.05±1.86 9.32±0.95 11.05±1.61 9.23	13.22±1.34 6.44±2.59 8.54±1.04 3.78 0.064 14.98±2.23 10.78±1.26 8.90±0.80 4.04 0.056 15.75±2.60 6.69±2.40 6.65±2.37 4.55 0.043 15.75±2.60 6.69±2.40 6.65±2.37 4.55 0.043 22.65±3.71 8.47±3.34 9.78±3.53 4.93 0.036 22.65±3.71 8.47±3.34 9.78±3.53 4.93 0.036 15.68±1.85 5.72±1.98 11.97±0.92 9.29 0.006 14.92±1.6 8.90±3.72 14.02±1.77 1.60 0.254 14.85±3.74 11.46±2.11 12.07±2.36 0.41 0.676 18.05±1.86 9.32±0.95 11.05±1.61 9.23 0.007	13.22±1.34 6.44±2.59 8.54±1.04 3.78 0.064 16.14±1.36 14.98±2.23 10.78±1.26 8.90±0.80 4.04 0.056 23.03±4.49 15.75±2.60 6.69±2.40 6.65±2.37 4.55 0.043 12.55±1.69 15.75±2.60 6.69±2.40 6.65±2.37 4.55 0.043 17.33±1.44 22.65±3.71 8.47±3.34 9.78±3.53 4.93 0.036 16.29±1.31 22.65±3.71 8.47±3.34 9.78±3.53 4.93 0.036 14.16±1.57 15.68±1.85 5.72±1.98 11.97±0.92 9.29 0.006 22.65±3.71 14.92±1.6 8.90±3.72 14.02±1.77 1.60 0.254 14.69±1.62 14.85±3.74 11.46±2.11 12.07±2.36 0.41 0.676 14.01±1.14 18.05±1.86 9.32±0.95 11.05±1.61 9.23 0.007 14.75±1.75	13.22±1.34 6.44±2.59 8.54±1.04 3.78 0.064 16.14±1.36 4.54±2.73 14.98±2.23 10.78±1.26 8.90±0.80 4.04 0.056 23.03±4.49 10.01±3.69 15.75±2.60 6.69±2.40 6.65±2.37 4.55 0.043 12.55±1.69 6.11±1.49 15.75±2.60 6.69±2.40 6.65±2.37 4.55 0.043 17.33±1.44 7.55±1.67 22.65±3.71 8.47±3.34 9.78±3.53 4.93 0.036 16.29±1.31 11.09±1.20 22.65±3.71 8.47±3.34 9.78±3.53 4.93 0.036 14.16±1.57 11.70±1.72 15.68±1.85 5.72±1.98 11.97±0.92 9.29 0.006 22.65±3.71 8.47±3.34 14.92±1.6 8.90±3.72 14.02±1.77 1.60 0.254 14.69±1.62 10.21±1.61 14.85±3.74 11.46±2.11 12.07±2.36 0.41 0.676 14.01±1.14 11.76±1.74 18.05±1.86 9.32±0.95 11.05±1.61 9.23 0.007 14.75±1.75 4.25±2.46	13.22±1.34 6.44±2.59 8.54±1.04 3.78 0.064 16.14±1.36 4.54±2.73 7.07±2.56 14.98±2.23 10.78±1.26 8.90±0.80 4.04 0.056 23.03±4.49 10.01±3.69 12.71±1.71 15.75±2.60 6.69±2.40 6.65±2.37 4.55 0.043 12.55±1.69 6.11±1.49 10.76±1.71 15.75±2.60 6.69±2.40 6.65±2.37 4.55 0.043 17.33±1.44 7.55±1.67 14.32±0.95 22.65±3.71 8.47±3.34 9.78±3.53 4.93 0.036 16.29±1.31 11.09±1.20 11.64±0.94 22.65±3.71 8.47±3.34 9.78±3.53 4.93 0.036 14.16±1.57 11.70±1.72 14.37±2.10 15.68±1.85 5.72±1.98 11.97±0.92 9.29 0.006 22.65±3.71 8.47±3.34 9.78±3.53 14.92±1.6 8.90±3.72 14.02±1.77 1.60 0.254 14.69±1.62 10.21±1.61 12.16±1.25 14.85±3.74 11.46±2.11 12.07±2.36 0.41 0.676 14.01±1.14 11.76±1.74 10.67±1.06 18.05±1.86 9.32±0.95 11.05±1.61 9.23	13.22±1.34 6.44±2.59 8.54±1.04 3.78 0.064 16.14±1.36 4.54±2.73 7.07±2.56 7.03 14.98±2.23 10.78±1.26 8.90±0.80 4.04 0.056 23.03±4.49 10.01±3.69 12.71±1.71 3.86 15.75±2.60 6.69±2.40 6.65±2.37 4.55 0.043 12.55±1.69 6.11±1.49 10.76±1.71 4.12 15.75±2.60 6.69±2.40 6.65±2.37 4.55 0.043 17.33±1.44 7.55±1.67 14.32±0.95 13.05 22.65±3.71 8.47±3.34 9.78±3.53 4.93 0.036 16.29±1.31 11.09±1.20 11.64±0.94 6.05 22.65±3.71 8.47±3.34 9.78±3.53 4.93 0.036 14.16±1.57 11.70±1.72 14.37±2.10 0.67 15.68±1.85 5.72±1.98 11.97±0.92 9.29 0.006 22.65±3.71 8.47±3.34 9.78±3.53 4.93 14.92±1.6 8.90±3.72 14.02±1.77 1.60 0.254 14.69±1.62 10.21±1.61 12.16±1.25 2.24 14.85±3.74 11.46±2.11 12.07±2.36 0.41 0.676 14.01±1.14 1

Values are in mean \pm SEM. Multivariate comparison between seasons was performed using ANOVA at 5% level of significance (p<0.05).

Relation between hydro-climatic data and trematode prevalence

Various environmental factors may impact the emergence of larval trematodes (cercariae) from their molluscan first intermediate hosts. For example, the effects of temperature on

cercarial emergence are well documented. Many trematode species play increased emergence in response to increased temperature.

 Table 5.7 Limnological condition of the water bodies.

Serial	pН		EC		DO	
Dhaka region	2011-12	2012-13	2011-12	2012-13	2011-12	2012-13
1 Ramna	7.4	7.6	1330	1321	4.0	4.5
2 Curjon	7.5	7.8	1340	1325	6.5	6.25
3 Jagannath	7.3	7.6	1380	1245	6.25	7.0
4 Keraniganj	7.5	7.6	1340	1325	0.1	0.2
5 Tongi Railway	6.5	7.0	1200	1140	4.0	4.0
6 Jaydebpur Railway	7.25	7.5	1100	1265	4.5	4.9
7 Bhawal-Gazipur	6.5	7.8	1400	1349	5.5	5.0
8 Salna BSMAU	7.5	7.5	1100	945	7.5	8.0
9 Mawna	7.5	8.0	1280	1400	6.0	5.5
10 Tongi Bari	7.25	7.5	1100	980	7.5	7.25
11 Lahajang	7.5	7.5	1250	950	7.5	8.0

Khulna region						
	8.2	8.7	1400	1381	6.8	6.8
1.Khulna University						
	7.9	8.0	1540	1455	6.2	6.0
2.Dacope Upazila						
	9.8	8.9	1400	1605	6.8	7.0
3.Shyamnagar Upazila						
	9.1	8.25	1500	1305	7.0	7.5
4.Kaliganj Upazila						
	9.6	9.0	1405	1100	6.6	6.0
5.Bagerhat P.C College						
6. Fakirhat	7.25	7.5	1200	1205	6.6	6.5
BazarBagerhat						
Kishoreganj region						
	8.9	7.8	600	540	7.2	7.5
Solakia, Kishorganj						
Astagram Upazila	7.8	7.3	450	480	7.0	6.9

Kuliarchar, Kishorganj	7.4	8.0	510	500	6.5	6.5
Mithamain, Upazila	7.7	8.9	440	468	6.1	7.0
BAU Mymensingh	7.6	8.0	550	550	5.5	5.8
Kabi Nazrul University	7.5	7.8	600	630	5.2	5.1

 Table 5.8 Climatic condition of the study areas.

Rainfall pattern (in mm)

Location	Jan	Feb	Mar	Apr	May	June	July	Aug	Sep	Oct	Nov	Dec
Dhaka region												
2011	0	0	20	123	235	314	356	409	207	112	0	0
2012	10	1	37	269	137	175	226	282	81	38	68	5
2013	0	8	26	32	378	325	302	212	172	131	0	4
Khulna region												
2011	0	1	16	28	145	381	387	614	367	3	6	0
2012	66	18	1	52	63	255	391	254	374	89	80	2
2013	1	7	19	62	430	212	313	482	278	260	0	0
Kishoreganj re	gion											
2011	1	12	32	80	239	449	333	741	242	18	0	0
2012	18	0	1	202	85	241	409	238	221	45	19	0
2013	0	18	21	69	308	267	318	343	132	263	0	0

Temperature (in °C)

Location	Jan	Feb	Mar	Apr	May	June	July	Aug	Sep	Oct	Nov	Dec
Dhaka regi	ion			_					-		•	
2011	8.2-	13.0-	16.0-	20.2-	21.3-	23.2-	23.9-	24.5-	23.7-	22.0-	17.2-	11.0-
	27.8	31.0	34.5	35.8	35.3	36.0	35.4	35.0	36.2	34.5	32.4	30.0
2012	10.5-	12.2-	18.3-	19.0-	20.5-	23.2-	25.2-	24.4-	24.9-	20.3-	14.8-	9.6-
	28.5	33.0	37.3	37.1	36.2	36.7	34.3	34.5	36.5	34.4	32.4	28.5
2013	7.2-	14.0-	16.7-	19.8-	20.0-	22.0-	24.5-	24.5-	24.2-	20.1-	16.0-	11.8-
	28.1	32.4	36.0	37.0	37.1	34.4	34.6	35.0	35.7	35.2	32.1	30.5
Khulna reg	gion											
2011	8.0-	12.3-	12.5-	20.6-	21.3-	24.0-	24.2-	24.0-	24.5-	19.5-	15.5-	10.0-
	28.5	33.0	36.6	37.3	37.4	37.6	35.5	36.5	35.5	35.5	32.8	29.8
2012	10.2-	10.5-	17.5-	19.0-	20.4-	25.0-	25.2-	24.5-	19.5-	19.5-	13.4-	9.5-
	29.0	34.0	37.0	39.5	38.5	38.8	34.8	34.8	35.7	34.7	32.5	30.5
2013	7.0-	12.2-	15.0-	19.4-	21.4-	25.2-	24.8-	23.5-	21.0-	21.0-	15.5-	11.6-
	29.2	32.2	36.6	38.8	37.5	37.0	34.7	35.5	36.0	35.2	32.6	29.0
Kishoregai	nj regio	n										
2011	8.0-	12.2-	14.0-	20.0-	20.2-	21.7-	24.0-	24.6-	24.5-	20.0-	14.5-	9.4-
	25.6	29.5	33.0	34.4	34.2	35.2	35.1	35.5	36.5	35.0	31.7	29.8
2012	8.0-	9.4-	14.5-	18.2-	20.6-	20.6-	25.5-	25.0-	24.5-	17.8-	12.0-	9.5-
	27.2	31.3	34.8	35.6	36.2	35.2	34.6	34.5	37.0	34.5	31.8	28.0
2013	4.7-	10.4-	15.2	18.4-	18.8-	24.0-	25.0-	24.7-	23.8-	20.7-	11.8-	8.8-
	28.0	32.5	35.0	35.6	35.5	36.4	34.3	34.8	36.5	34.2	32.0	30.8

The overall natural environmental parameters varied within overlapping ranges among the ponds. Water temperature ranged between 4.7°C and 38°C depending on the day and time of collection. The hydrogen ion concentration was within the range providing protection for the aquatic life (7.2 to 9.8). The concentration of dissolved oxygen showed generally low in some locations due to pollution load (Dhaka region) and higher (> 7.0 mg/L) levels and exhibited a strong negative association with water conductivity (r_s = -0.665, p < 0.05). The following tables identified the correlation between temperature, pH, EC, DO, rainfall and prevalence of parasites in snails of three different regions in 2011-12 and 2012-13 academic years. Sometimes positive correlation with significant level (p>0.05) and without significant level were observed.

Table 5.9 Correlation between temperature and prevalence of parasites in snails of Dhaka regions in 2011-12.

		%prevalence in	Temperature	pH 2011-12	EC 2011-12	DO 2011-	Rain fall
		Dhaka region	2011-12			12	2011-12
	<u>, </u>	2011-12					
% prevalence in	Pearson Correlation	1	.437**	.027	.045	.034	.080
Dhaka region	Sig. (2-tailed)		.000	.586	.373	.506	.113
2011-12	N	396	396	396	396	396	396
	Pearson Correlation	.437**	1	.000	.000	.000	.029
Temperature	Sig. (2-tailed)	.000		1.000	1.000	1.000	.560
2011-12	N	396	396	396	396	396	396
	Pearson Correlation	.027	.000	1	094	.114*	.000
pH 2011-12	Sig. (2-tailed)	.586	1.000		.063	.023	1.000
	N	396	396	396	396	396	396
	Pearson Correlation	.045	.000	094	1	305**	.000
EC 2011-12	Sig. (2-tailed)	.373	1.000	.063		.000	1.000
	N	396	396	396	396	396	396
	Pearson Correlation	.034	.000	.114*	305**	1	.000
DO 2011-12	Sig. (2-tailed)	.506	1.000	.023	.000		1.000
	N	396	396	396	396	396	396
	Pearson Correlation	.080	.029	.000	.000	.000	1
Rain fall 2011-12	Sig. (2-tailed)	.113	.560	1.000	1.000	1.000	
	N	396	396	396	396	396	396

^{**.} Correlation is significant at the 0.01 level (2-tailed).

^{*.} Correlation is significant at the 0.05 level (2-tailed).

Significantly positive correlation was found between temperature and prevalence of parasites in snails of Dhaka regions in 2011-12 (p<0.01). Positive correlation was also found between % occurrence and pH, EC, DO and rainfall, but was not significant (p>0.05).

Table 5.10 Correlation between temperature and prevalence of parasites in snails of Dhaka regions in 2012-13.

		% prevalence in Dhaka region 2012-	Temperature 2012-13	pH 2012-13	EC 2012-13	DO 2012- 13	Rain fall 2012-13
		13					
	Pearson Correlation	1	.443**	.045	.025	004	.220**
% prevalence in Dhaka	Sig. (2-tailed)		.000	.377	.620	.934	.000
region 2012-13	N	396	396	396	396	396	396
	Pearson Correlation	.443**	1	.000	.000	.000	.578**
Temperature 2012-13	Sig. (2-tailed)	.000		1.000	1.000	1.000	.000
	N	396	396	396	396	396	396
	Pearson Correlation	.045	.000	1	.550**	.060	.000
pH 2012-13	Sig. (2-tailed)	.377	1.000		.000	.237	1.000
	N	396	396	396	396	396	396
	Pearson Correlation	.025	.000	.550**	1	597**	.000
EC 2012-13	Sig. (2-tailed)	.620	1.000	.000		.000	1.000
	N	396	396	396	396	396	396
	Pearson Correlation	004	.000	.060	597**	1	.000
DO 2012-13	Sig. (2-tailed)	.934	1.000	.237	.000		1.000
	N	396	396	396	396	396	396
	Pearson Correlation	.220**	.578**	.000	.000	.000	1
Rain fall 2012-13	Sig. (2-tailed)	.000	.000	1.000	1.000	1.000	
	N	396	396	396	396	396	396
**. Correlation is signific	ant at the 0.01 level (2-	-tailed).					

Significantly positive correlation was found between temperature and prevalence of parasites and rainfall and prevalence of parasites in snails of Dhaka regions in 2012-13 (p<0.01). Positive correlation was also found between % occurrence and pH, EC, and DO, but was not significant (p>0.05).

Table 5.11 Correlation between temperature and prevalence of parasites in snails of Khulna regions in 2011-12.

		% prevalence	Temperature	pH	EC	DO	Rain fall
		in Khulna	2011-12	2011-12	2011-12	2011-12	2011-12
		region 2011- 12					
		12					
% prevalence in Khulna region	Pearson Correlation	1	.336**	.070	.200**	086	.503**
2011-12	Sig. (2-tailed)		.000	.307	.003	.208	.000
	N	216	216	216	216	216	216
	Pearson Correlation	.336**	1	.239**	.100	.133	.581**
Temperature 2011-12	Sig. (2-tailed)	.000		.000	.144	.051	.000
	N	216	216	216	216	216	216
	Pearson Correlation	.070	.239**	1	.377**	.449**	233**
pH 2011-12	Sig. (2-tailed)	.307	.000		.000	.000	.001
	N	216	216	216	216	216	216
	Pearson Correlation	.200**	.100	.377**	1	118	.117
EC 2011-12	Sig. (2-tailed)	.003	.144	.000		.083	.085
	N	216	216	216	216	216	216
	Pearson Correlation	086	.133	.449**	118	1	323**
DO 2011-12	Sig. (2-tailed)	.208	.051	.000	.083		.000
	N	216	216	216	216	216	216
	Pearson Correlation	.503**	.581**	233**	.117	323**	1
Rain fall 2011-12	Sig. (2-tailed)	.000	.000	.001	.085	.000	
	N	216	216	216	216	216	216
Correlation is significant at the 0	0.01 level (2-tailed).**						

Correlation is significant at the 0.05 level (2-tailed).*

Significantly positive correlation was found between temperature, EC, rainfall and prevalence of parasites in snails of Khulna regions in 2011-12 (p<0.01). Positive correlation was also found between % occurrence and pH, but was not significant (p>0.05).

Table 5.12 Correlation between temperature and prevalence of parasites in snails of Khulna regions in 2012-13.

		%	Temperature	pН	EC	DO	Rain fall
		prevalence	2012-13	2012-13	2012-13	2012-13	2012-13
		in Khulna					
		region					
	I	2012-13					
	Pearson Correlation	1	.358**	.083	.279**	.130	.441**
% prevalence in Khulna region 2012-13	Sig. (2-tailed)		.000	.223	.000	.057	.000
2012-13	N	216	216	216	216	216	216
	Pearson Correlation	.358**	1	.001	.009	.131	.705**
Temperature 2012-13	Sig. (2-tailed)	.000		.992	.899	.055	.000
	N	216	216	216	216	216	216
	Pearson Correlation	.083	.001	1	.156*	.056	.315**
pH 2012-13	Sig. (2-tailed)	.223	.992		.022	.409	.000
	N	216	216	216	216	216	216
	Pearson Correlation	.279**	.009	.156*	1	.322**	032
EC 2012-13	Sig. (2-tailed)	.000	.899	.022		.000	.643
	N	216	216	216	216	216	216
	Pearson Correlation	.130	.131	.056	.322**	1	174 [*]
DO 2012-13	Sig. (2-tailed)	.057	.055	.409	.000		.011
	N	216	216	216	216	216	216
	Pearson Correlation	.441**	.705**	.315**	032	174*	1
Rain fall 2012-13	Sig. (2-tailed)	.000	.000	.000	.643	.011	
	N	216	216	216	216	216	216

Significantly positive correlation was found between temperature, EC, rainfall and prevalence of parasites in snails of Khulna regions in 2012-13 (p<0.01). Positive correlation was also found between % occurrence and pH, but was not significant (p>0.05).

^{*.} Correlation is significant at the 0.05 level (2-tailed).

Table 5.13 Correlation between temperature and prevalence of parasites in snails of Kishoreganj regions in 2011-12.

		% prevalence	Temperature	pН	EC	DO	Rain fall
		in Kishoreganj	2011-12	2011-	2011-	2011-	2011-12
		region 2011-12		12	12	12	
% prevalence in	Pearson	1	.503**	.013	173*	.291**	.468**
Kishoreganj	Correlation						
region	Sig. (2-tailed)		.000	.849	.011	.000	.000
2011-12	N	216	216	216	216	216	216
Temperature	Pearson	.503**	1	209**	260**	008	.651**
2011-12	Correlation						
	Sig. (2-tailed)	.000		.002	.000	.912	.000
	N	216	216	216	216	216	216
рН	Pearson	.013	209**	1	.358**	.647**	.116
2011-12	Correlation						
	Sig. (2-tailed)	.849	.002		.000	.000	.090
	N	216	216	216	216	216	216
EC	Pearson	173*	260**	.358**	1	260**	193**
2011-12	Correlation						
	Sig. (2-tailed)	.011	.000	.000		.000	.004
	N	216	216	216	216	216	216
DO	Pearson	.291**	008	.647**	260**	1	.325**
2011-12	Correlation						
	Sig. (2-tailed)	.000	.912	.000	.000		.000
	N	216	216	216	216	216	216
Rain fall	Pearson	.468**	.651**	.116	193**	.325**	1
2011-12	Correlation						
	Sig. (2-tailed)	.000	.000	.090	.004	.000	_
	N	216	216	216	216	216	216

^{**.} Correlation is significant at the 0.01 level (2-tailed).

Significantly positive correlation was found between temperature, EC (p<0.05), rainfall and prevalence of parasites in snails of Kishoreganj regions in 2012-13 (p<0.01). Positive correlation was also found between % occurrence and pH, but was not significant (p>0.05).

^{*.} Correlation is significant at the 0.05 level (2-tailed).

Table 5.14 Correlation between temperature and prevalence of parasites in snails of Kishoreganj regions in 2012-13.

		% prevalence in	Temperature	pН	EC	DO	Rain fall
		Kishoreganj	2012-13	2012-13	2012-13	2012-	2012-13
		region 2012-13				13	
% prevalence in	Pearson	1	.436**	.076	154*	.101	.341**
Kishoreganj region	Correlation						
2012-13	Sig. (2-		.000	.266	.024	.139	.000
	tailed)						
	N	216	216	216	216	216	216
Temperature	Pearson	.436**	1	.117	194**	.017	.784**
2012-13	Correlation						
	Sig. (2-	.000		.087	.004	.805	.000
	tailed)						
	N	216	216	216	216	216	216
pН	Pearson	.076	.117	1	277**	.106	199**
2012-13	Correlation						
	Sig. (2-	.266	.087		.000	.121	.003
	tailed)						
	N	216	216	216	216	216	216
EC	Pearson	154 [*]	194**	277**	1	748**	.149*
2012-13	Correlation						
	Sig. (2-	.024	.004	.000		.000	.029
	tailed)						
	N	216	216	216	216	216	216
DO	Pearson	.101	.017	.106	748**	1	.015
2012-13	Correlation						
	Sig. (2-	.139	.805	.121	.000		.830
	tailed)						
	N	216	216	216	216	216	216
Rain fall	Pearson	.341**	.784**	199**	.149*	.015	1
2012-13	Correlation						
	Sig. (2-	.000	.000	.003	.029	.830	
	tailed)						
	N	216	216	216	216	216	216

^{**.} Correlation is significant at the 0.01 level (2-tailed).

st. Correlation is significant at the 0.05 level (2-tailed).

Significantly positive correlation was found between temperature, EC (p<0.05), rainfall and prevalence of parasites in snails of Kishoreganj regions in 2012-13 (p<0.01). Positive correlation was also found between % occurrence and pH, but was not significant (p>0.05).

The above mentioned physico-chemical and climatological parameters of different locations do play a vital role in the population dynamics of molluscan species. It is made clear from the results that more number of species related with low DO content. It can be inferred from the above points that DO content plays a vital role in affecting the population dynamics of a particular species than the other physico-chemical factors like pH, temperature and EC. Moreover the molluscan population is abundant in a place where the aquatic plants are available in diverse forms. Since there is not much variation in pH, temperature, EC and salinity in the collection sites, they do not have much influence in the availability of specimens in this present study. Through this analysis, it is inferred that the possibility of variation in pH, temperature, EC can play a vital role in the life cycle of any plant or animal group and when it is moderate or in low, it cannot have much influence on the population dynamics.

In the present study monthly fluctuations in the cercariae prevalence were observed due to the variations in the temperature, rainfall and dissolve oxygen of each month. The occurrence of cercariae are maximum in February to April (pre-monsoon) and September to November (post-monsoon) time which are dealt with elevated temperature, low rainfall and DO level in the Dhaka region. Whereas, In Khulna and Kishoreganj region only pre-monsoon time shows the maximum prevalence. Islam (2008) reported 14.5% infection and Alim (1997) and Rahman *et al.*, (1997) reported 3.7% infection during March-May.

The parasites collected from snails other than Trematode

Out of 23 locations nematode infection was very rare. Infection of Nematode was found only 3 locations i.e. Ramna Park pond, Mawna Bazar pond and Solakia, Kishoreganj pond during the study period.

Table 5.15 Prevalence of Nematoda in *Bellamya* sp. and *Pila* sp. in 3 different locations.

Location	Year		Bellamya			Pila			
		Tot	Inf	%	Tot	Inf	%		
Ramna park pond	2011-12	272	4	1.47%	224	10	4.46%		
	2012-13	221	4	1.81%	211	8	3.79%		
Mawna bazar pond	2011-12	232	5	2.16%	194	8	4.13%		
	2012-13	250	5	2.0%	224	7	3.13%		
Solakia bazar pond	2011-12	161	2	1.24%	160	7	4.38%		
	2012-13	220	7	3.18%	220	10	4.55%		

The overall prevalence on the two years period (2011-12 and 2012-13) showed that at Ramna, it was 1.47 to 4.46%, at Mawna 2.0 to 4.13% and at Solakia it was 1.24 to 4.55. No relation could not sort out with climatic or environmental factors of its existence due to its scarce nature. Prevalence was highest in March to May (2011-2013).

Table 5.16 Prevalence of parasites collected from snails other than Trematode

Location	Year	Hos	t	Parasites							
				Paravortex sp.		Trilocularia Polypod		ocephalus	cephalus Lingua		
						sp.		sp		Sp.	
		Total		Inf	%	Inf	%	Inf	%	Inf	%
Ramna park pond	2011-12	Pila globosa	272	6	2.21%	3	1.10%	3	1.10%	4	1.47%
Curjon hall pond			388	8	2.06%	4	1.03%	4	1.03%	3	1.29%
Jagannath hall pond			387	5	1.29%	5	1.29%	6	1.55%	4	1.03%
Ramna park pond	2012-13	Pila globosa	221	5	2.26%	6 4	1.80%	6 4	1.819	% 3	1.36%
Curjon hall pond			304	7	2.30%	ó 4	1.32%	6 4	1.329	% 4	1.32%
Jagannath hall pond			269	4	1.49%	6 3	1.12%	6 6	2.239	% 4	1.49%

In 2011-12 prevalence of *Paravortex* sp.. *Trilocularia* sp. *Polypocephalus* sp. and *Linguatula* sp. was found in 2.21%, 1.10%, 1.10% and 1.47% at Ramna Park Pond and 2.06%, 1.03% 1.03% and 1.29% at Curjon Hall Pond. Similarly at Jagannath Hall Pond (1.29%, 1.29%, 1.55% and 1.03% respectively. Highest prevalence was found in August to September.

In 2012-13 prevalence of *Paravortex* sp.. *Trilocularia* sp. *Polypocephalus* sp. and *Linguatula* sp. was found in 2.26%, 1.80% ,1.81% and 1.36% at Ramna Park Pond and 2.3%, 1.32% 1.32% and 1.32% at Curjon Hall Pond. Similarly at Jagannath Hall Pond (1.49%, 1.12%, 2.23% and 1.49%) respectively. Highest prevalence was found in August to September.

The parasites collected from snails other than Trematode

Phylum – Plathyhelminthes

Class - Turbellaria

Order-Rhabdocoela

Family-Graffillidae

Genus- Paravortex

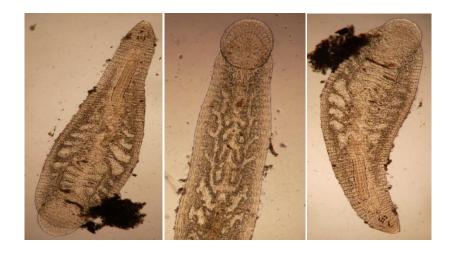


Figure 5.74: Anterior and posterior view of *Paravortex* sp.

Description: They are small, flattened organisms. They are generally colorless and somewhat transparent. They are all ectocommensals on fresh-water animals, primarily crayfishes, prawns, isopods and other crustaceans. They are less frequently found on turtles and snails, on which are attached to the external surface or in the branchial chamber. They are characterized by the absence of cilia or reduced ciliation, tentacles, a single genital pore and one or two adhesive discs at the posterior end of the body. They generally live on the gills or on the body surface of freshwater crustaceans of more rarely on gastropod mollusc or turtle. They usually have a ciliated epidermis, an undivided body and a simple life cycle. They are usually flattened dorso-ventrally without segmentation and are bilaterally symmetrical. They possess an incomplete digestive tract lack a body cavity and are without special skeleton, circulatory or respiratory structures. They have a flame- cell type of excretory system (after Cheng, 1964 and Noble, 1971).

Phylum - Plathyhelminthes

Class - Cestoda

Order- Tetraphyllidea

Family-Triloculariidae

Genus: Trilocularia





Figure 5.75: Anterior and posterior view of *Trilocularia* sp.

Description: The tetraphyllidean cestodes are recognizable by four prominent outgrowths on the Scolex. Scolex with four saccular or auricular bothridia and large myzorhynchus, without hooks or suckers. Strobila distinctly segmented proglotides acraspedote. Inner longitudinal musculature strongly developed. Scolex unarmed, with four sessile bothridia. Each bothridium triangular, with rounded corners, divided into three suckers like loculi placed in a triangle, one in front and two behind side byside. Neck long, anteriorly thick. Proglottides crowded anteriorly, then subquadrate to trapezoidal, longer than broad posteriorly. Common atrium is marginal. Vitellaria are in two lateral fields. Uterus appears as small uterine duct dorsal to vagina or as stemlike median uterine sac ventral to vagina. Gravid uterus is broad, occupying most of proglottid. Each proglottid contains single set male and female reproductive organs. Larvae have been found in cephalopod molluscs. The adults are all parasiticin the intestines of elasmobranch fish (after Cheng, 1964; Yamaguti, 1985 and Noble, 1971).

Phylum - Plathyhelminthes

Class - Cestoda

Order- Lecanicephalidea

Family- Lecanicephalidae

Genus-Polypocephalus



Figure 5.76 Anterior and posterior view of *Polypocephalus* sp.

Description: Scolex subglobular, flat and square In front view, anterior region represented by a crown of 14 to 16 retractile tentacles and posterior regions with four suckers symmetrically. Neck present or absent. Strobila more or less cylindrical. Proglottides craspedote. Cirrus pouch oblique may push inward the poral side of uterine sac. Genital pores irregularly alternating. Vitelline follicles large, extending in two lateral rows anterior to ovary. Uterus median, occupying most of proglottis length anterior as well as posterior to overy. Parasitic in elasmobranchus (after Cheng, 1964; Yamaguti, 1985 and Noble, 1971).

Phylum – Arthropoda (Pantastomida)

Class – Maxillopoda

Order-Porocephalida

Family- Linguatulidae

Genus- Linguatula



Figure 5.77 Ventral and Dorsal view of *Linguatula* sp.

Description: They are legless and worm like, but near the mouth are two pairs of hollow, curved, retractile hooklets that are rudimentary appendages. Sharp hooks on the anterior end are anchoring devices. Pentastomids possess elongate bodies that are cylindrical. Externally the bodies are annulated, but these rings are not true segments. Adults do not possess legs, but four or six rudimentary legs are present on larvae. Because of their vermiform bodies, these animals are referred as worms.

The larvae are found in the viscera of the intermediate host, which is usually a mammal or other vertebrates. Adult are parasitic in the respiratory tract and lungs of vertebrates. The mouth is located anteriorly and is sub terminal. The mouth leads into a buccal cavity, which is linked by a narrow prepharynx with the muscular pharynx. They have two pairs of hollow fanglike hooks, one pair on each side of the mouth which can be retracted into grooves. Their body surface is covered with a layer of chitin. The adult female is tongue-shaped, 100 to 130 mm long and up to 10mm wide, whereas the male is about 20 mm long and 3 to 4 mm wide. Egg containing embryos with rudimentary legs are deposited in the nasal passages and frontal sinuses of mammals and are discharged in nasal secretions. Upon reaching water or moist

vegetation, embryo nation is completed and if the eggs are ingested by the intermediate host hatching occurs in the digestive tract and the larvae migrate through the intestinal wall and lodged in the liver and other viscera. After number of molts the nymph stage is attained. Nymphs lie encapsulated with the tissue and they are the infective stage for carnivorous animals that feed on the herbivorous intermediate hosts. Although the general belief is that nymphs are digested out of their capsule and immediately migrate to the nasal passages of the definitive host (after Cheng, 1964 and Noble, 1971).

Phylum – Nematoda

Class - Secernentea

Order- Strongylida

Family- Metastrongylidae

Genus: Angiostrongylus

Description: Nematodes are usually round in cross section; hence they are called round worms. Compared with trematodes and cestodes, the life histories of nematodes are less varied and their anatomy less adapted to parasitism. In case of *Angiostrongylus* sp. definitive host rats and intermediate host snails and slugs. In infected rats, eggs containing uncleaved zygotes are deposited in the blood streams, where they become lodged as emboli in the smaller vessel in the lungs. Embryonic development occurs here.



Figure 5.78 Anterior and posterior view of *Angiostrongylus* sp.

However, these flatworms cannot be infected with first stage larvae and presumably serve as paratenic hosts, which acquire third stage larvae from feeding on naturally infected snails and slugs. The first stage larvae actively penetrate molluscs. Within the intermediate host, the larvae undergo two molts but do not exsheath. When an infected mollusk is ingested by a rat, the larvae exsheath in the rodents' stomach and burrow through the ileum and become bloodborne and finally they become mature in the rat (Cheng, 1964 and Noble, 1971).

DISCUSSION

General Discussion

Snails usually play a dominant role in the ecology of freshwaters by providing food for many other animals and by grazing on vast amounts of algae and detritus (debris). Freshwater snails occur across a variety of habitats, ranging from small temporary ponds and streams to large lakes and rivers, reflecting the wide-ranging biology of many different species. Many species spend their entire lives in a few square meters of habitat, making them extremely vulnerable to localized environmental habitat degradation. Although most species prefer clean, stable, and firm river bottoms, some prefer the soft substrates more common to ponds and lakes. Ponds are amongst the most diverse freshwater habitats and have been recently found to support more species, as well as more uncommon, rare, and threatened species compared to lakes, rivers, and streams (Wright, 1971).

Snails act as intermediate hosts of different trematode parasites, in which several developing larval stages such as sporocysts, rediae and cercariae are set up. The proportion of snails that release cercariae (prevalence of infection) and the number of cercariae released from each infected snail (intensity of infection) play important roles in the transmission of trematodes from the snail host. Age and size of snails, light conditions, temperature ranges, depth of water are some of the factors that appear to affect the prevalence and intensity of digenetic trematode infections in the snail intermediate hosts. The suitable conditions make cercariae able to develop in snails and fish which lead to perpetuation in their life cycle. Bojanus (1818) observed developing cercariae inside rediae and studied the emergence and swimming of cercariae. Cercariae are the important and interesting larval forms of trematodes.

Studies on developmental stages of helminthes in general, in particular, are of fundamental importance because establishment of a helminth parasite in the host depends primarily on the successful completion of the developmental stages. It may be noted here that these developmental stages pass a major part of their life in the external environment where they are subject to direct influence of the principle environmental factors. However, in Bangladesh,

works on this aspect of the life and biology of trematodes are practically not only little, but also virtually recent.

The present study was undertaken to record the prevalence of cercaria in the snail as intermediate host, in and around Dhaka, Khulna and Mymensingh. Results indicate a low overall infection rate of snails with larval trematodes. This is in line with findings from other studies (Anderson and May, 1979; Loker *et al.*, 1982; Mattison *et al.*, 1995; Kigadye, 1998; Toledo *et al.*, 1998). Wright (1966) and Sousa (1992) attributed such low infection rates in natural snail populations to a direct consequence of high rates of parasite-induced mortality. On the other hand, Begon *et al.* (1990) argued that as a result of host–parasite coevolution, hosts usually develop acquired resistance to infection and thus the observed low levels of prevalence. The low prevalences of infection could also be due to low parasite pressure, simply making contact between miracidia and snails a rare event.

Developmental stages of trematode

The present study represents the diversity of larval trematode parasites (cercariae) in the freshwater snails in different parts of Bangladesh. Eight morphologically distinguishable types of trematode cercariae were observed in three freshwater snail species with 12.92% prevalence rate in snail hosts. Such high infection rates in natural snail populations might be the direct consequence of high rates of parasite-induced emergence. Other influential factors may be the difference in snail species observed at different time periods of the year in different studies as well as limnological condition pond water that is stagnant in nature. Begum *et al.*, (2011) argued that because of host-parasite co-evolution, hosts usually acquire resistance to infection, resulting in low level of observed prevalence.

In the present study, the infection rate in different host snail species ranged from 10.95% to 14.7%. This finding can be compared with the Sewell's findings (1922) of 6.4–50 percent in *G. euphraticus*, 5.0–71.1% in *M. tuberculata*, and 0.5–44.2% in *I. exustus*in Calcutta. Trematode infection in *I. exustus*was recorded up to 50% in Wynaad, Madras, India (Sewell 1922). Cercarial infection was more prominent in *I. exustus*and *Lymnaeaovalis* than in other snail vectors of the Kaveri River in India (Amanullah and Hameed, 1996). One possible explanation of this observation could be the resistance of some snail species to trematode infection, a factor noted

by Bayne and Yoshino (1989). Caution must be applied when comparing levels of infection of intermediate host snails from different studies. The present study was based on "patent infection" (Ito, 1980). The infection levels of this study might have been significantly higher if the presence of "pre-patent infections" had been studied.

The prevalences of different types of trematode cercariae in different habitats are generally higher than those reported by Pandey *et al.*, (2001) for the rice fields (1.67%) and streams (0%). The highest prevalence rate in temporary ponds in this observation was likely due to the large number of birds perching on trees around the ponds. Many bird species act as final host for strigeoid and clinostomatoid digeneans. Hechinger and Lafferty (2005) demonstrated consistent, positive and significant associations between final host bird communities and trematode communities in intermediate host snail populations. The high prevalence rate in lakes was likely due to the presence of migratory and residential bird species and to several varieties of fishes and reptiles in and around the lakes. The fact that the lowest prevalence rate occurred in rivers may be due to the fast water currents that displace snails or at least keep them from congregating in large numbers and make snail-miracidia contact a rare occurrence. The degree of infection may vary in snails of the same species from one location to another, even though these areas might be apparently similar and geographically proximate to each other. Obtained results of the present study consistent with those of Sewell (1922).

The eight morphologically identified larval trematodes that are recorded from the present study showed similar trends in Zimbabwe, where Chingwena *et al.*, (2002) found eight types of the trematodecercariae. Likewise, the infection rate varies in studies carried out in other countries: six different trematode families in Northern Australia (Hurley *et al.*, 1980), eight trematode species afflicting *Batillaria attramentaria*in Japan (Miura and Chiba, 2007), and five morphologically distinct types of larval trematodes infecting freshwater snails in Zambia (Phiri *et al.*, 2007). Similarly, five morphologically distinguishable types of trematode cercariae were recorded in the Chitwan and Nawalparasi districts of Nepal (Devkota, 2008).

The present study revealed that some snail species are apparently capable of acting as primary host to large number of trematode species. By contrast, Sewell (1922) observed *M. tuberculata*

infected with seventeen forms of cercariae, *I. exustus* with fifteen forms and *Amnicola travencorica* with twelve forms. The present findings of lower trematode cercarial diversity in this case may be influenced by factors such as study seasons and the distribution and abundance of definitive hosts (Smith, 1999). Similarly, Sewell probably looked at far more snails from far more different areas.

Vector snails and the parthinate of trematodes inside them is a common problem in Bangladesh (Rahman *et al.*, 1997; Mondal *et al.*, 2003). Results of some studies showed the existence of vector snails and cercariae in Mymensingh district with overall 3.7% infection (Qadir. 1982). In the present study three different species of snails were detected such as, *Bellamya bengalensis*, *Pila globosa* and *Brotia costula*. Present findings confirm the findings of Mondal *et al.*, (2003), Alim (1997), Rahman *et al.*, (1999), Islam *et al.*, (2011) who reported the prevalence of snails in Bangladesh. In this study Gymnocephalus cercaria was recovered from *Pila globosa and Brotia costula* and Echinostome cercaria from *Pila globosa* which confirms the earlier findings of Alim (1997) and Sharif *et al.*, (2010).

The prevalence of gymnocephalus cercaria and echinostome cercaria in the study is lower than Islam (2008) and Chowdhury *et al.*, (1993) who reported 26.7% infection in India and higher than Alim (1997) who reported 0.8% gymnocephalus cercaria and 1.3% echinostome cercaria respectively. Findings of furcocercous cercariae from *Pila and Brotia* support the earlier findings of Alam (2010) and Mondal *et al.*, (2003).

In the present study amphistome cercariae were detected in *Bellamnya bengalensis*. The prevalence of amphistome cercariae in the study is lower than Islam (2008) who reported 10% infection and higher than Alim (1997) who reported 2.7%. Xiphidocercus cercariae were detected in all the three species.

The present study was undertaken to record the prevalence of snail's intermediate host infected with different helminth parasites mainly trematode in different regions of Bangladesh. An overall prevalence of snail's intermediate host infected with different trematode cercariae was recorded as 12.92%. Highest percentage of average infection by trematodes in *Bellamya* sp. was found in September which was 16.74% and lowest percentage of trematode infection was 7.25% in July,

in *Brotia sp* it was 13.53% in September and 9.38% in August whereas *Pila* sp. was having 19.12% in March and 9.43% in August.

The present study's findings supports where they reported that 5.3% snails were found infective with different trematode cercariae (Ismail *et al*, 2003). Whereas, Mendis and Fernando (2002) reported higher prevalence rate and their found that 16% snails were infected with different trematodes cercariae in Srilanka. Das Mahapatra *et al.*, (1982) also found similar findings about 17.27% in fresh water snails infected with different trematodes cercariae in West Bengal, India. Lockyer *et al.*, (2004) also reported a high prevalence rate of cercarial infection 71.56% in *Lymnaea auricularia* snails. Whereas, Sharif *et al.*, (2010) reported a low prevalence rate of cercarial infection in snails, they observed only 3.9% *L. gedrosiana* was infected by different trematode cercariae.

In the present study, monthly fluctuations in the cercarial prevalence were observed due to the variations in the temperature, rainfall and dissolve oxygen in each month. The occurrence of cercaria are maximum in February to April (pre-monsoon) and September to November (post-monsoon) time which are dealt with elevated temperature, low rainfall and DO level in the Dhaka region. Whereas In Khulna and Kishoreganj region only pre-monsoon time showed the maximum prevalence. Islam (2008) reported 14.5% infection and Alim (1997) and Rahman *et al.*, (1997) reported 3.7% infection during March-May. The cercarial shedding is related to temperature, humidity and rainfall; temperature between 25-30°C, relative humidity with a rainfall of more than 100mm is favorable for development and shedding of cercariae (Alim 1997; Rahman *et al.*, 1997). These conditions favor the growth and development and shedding of cercariae from snails by rapid development of miracidium to sporocyst, redia and finally cercariae. These developments reduced or ceased at a temperature less than 20°C, relative humidity 60% with a rainfall less than 30mm (Alim, 1997). This variation in cercarial prevalence might be due to hot weather, dry period or less rainfall followed by moderate rainfall, inadequate access of faeces to water bodies during study period etc.

In the present study, ponds were found to harbor cercariae of Furcocercus and Xiphidocercus which were higher than the earlier reports of Alim (1997) who reported 1.9% Furcocercus cercaria and lower than Alim (2010) and Islam (2008) who reported 2.5% Xiphidocercus cercaria. In the present study ponds were found to harbor cercariae of Furcocercus,

Xiphidocercus, Echinostome and Amphistome and marshy lands (beels) found to harbor cercariae of Gymnacephalus, Echinostome, Amphistome and Furcocercus. However there is no data available regarding these three location wise prevalence of cercariae. However, variations in the above findings might be due to differences in the climatic conditions, ecological factors, sample size, duration of study, access of livestock and duck to water bodies and also for using of raw cow dung as manure etc.

The habitat of the snails in present study is confirmed with the findings of Soulsby (1965), Alim (1997) and Islam (1991). The pH of water where the snails were found and weather conditions prevailing during study period (Table 3.2) confirms the reports of Soulsby (1965) and Islam (1991). It was assumed that, snails those like to live in water with vegetation perhaps provide their oxygen requirements and help to dilute and remove the products of metabolic activities and decay (Alim, 1997).

Similarly, a good number of works on this subject have been done in the South East Asian country of Thailand (e.g., Ngern- Klun *et al.*, 2006; Dechruksa *et al.*, 2007; Ukong *et al.* 2007, etc.) It is of particular importance to mention that trematode diseases like schistosomiasis, etc. are more prevalent in South East Asian countries. All these present a background for the cause of attempting this work which has been done on 3 snail host species (*Bellamya bengalensis*, *Brotia costula*, and *Pila globosa*) of 23 water bodies of three regions of Bangladesh. The developmental stages were cercariae, redia and sporocysts collected from the 3 snail hosts. The cercariae were of the richest form as far as diversity is concerned. They have been basically identified as of eight types (Parapleurophocercous, Xiphidiocercariae, Pleurophocercous, Echinostome, Amphistome, Furcocercous, Cotylomicrocercous and Renicolid cercaria having 17 species following Dechruksa *et al.*, (2007). Further down Category level identification was not possible because of in access to relevant literatures. Only Dechruksa *et al.*, (2007) and some down-loadings from Internet were the only sources for materials needed for identification; they were so meager that further identification was not possible.

Occurrence of the Developmental Stages of Trematoda (DST) in the 3 snail hosts:

In *B.bengalensis* the following types of DST were recorded-Parapleurophocercous (*Stictodora tridactyla*), Pleurophocercous (*Centrocestus fnusormosa*), Xiphidiocercariae (*Cloaci*

trema philippinum), Amphistome (*Gastrothylax crumenifer*), Cotylomicrocercous (*Podocotyle lepomis*) and Redia.

In the present work in *P. globosa*, 4 types of cercaria have been collected. They were Parapleurophocercous (*Haplorchis pumilio*, *Haplorchs taichui*), Xiphidiocercariae (*Acanthatrium hitaense*, *Loxogenoides bicolor*), Furcocercous (*Cardicola alseae*, *Apatemon gracilis*), Echinostome (*Echinochasmus pelecani*).

So, among the 8 cercarial types 7 were 2 hosts DST. They were Parapleurophocercous, Pleurophocercous, Xiphidiocercariae, Amphistome, Cotylomicrocercous, Furcocercous, Echinostome infesting B. *bengalensis* and *P. globosa*. It is thus evident that, of the 3 snail host species examined, *B. bengalensis* was the overwhelmingly susceptible host harbouring 7 cercarial types and 1 redial type. It It is also evident that of the present 3 host species, only one (*B. bengalensis*) harboured multiple infection, especially double infection.

The prevalence of parasitic infections (in the 3 hosts) was varied in the different water bodies and ecosystem. Of the 31717 hosts autopsied, 4099 were infected (12.92%). Infections were generally prevalent all over the year in *B. bengalensis* only and rare in *B. costula*. The highest infection rate was recorded in September (*B. bengalensis* 63%, *B. costula* 55% and *P. globosa* 57%). The rate of infection exhibits fluctuations, which may be due to variations in local habitats and in the susceptibility to infection which might be host-age related at the time of exposure to infective miracidia. This also may be due to the availability of the hosts and infective stages especially in sites 1 and 2, while no or limited infected snails were recorded in sites 3.

In the study areas, the values of prevalence, abundance and mean density depict the high rate of infection in snails during early winter and summer and less infection rate in the rainy season. This supports the idea of single peak in the infection rate in a year cited by Muraleedharan *et al.*, (1977).

In the present work Xiphidiocecaria, Cystocercous cercaria, Gymnocephalus cercaria, Amphistome, Leptocercous, Furcocercous cercaria are identified and obtained in the summer.

It has been also found that nematode infection in the 3 snail hosts have some seasonality. In *B. bengalensis* the highest prevalence was found in winter. In *P. globosa* that was found in winter season. In *B. costula* the highest prevalence was found in winter season. Whereas helminthes diseases of the vertebrate animals spread over during rainy season. So care should be taken to control the epidemic of helminth diseases in vertebrates before the rainy season through preventive measures.

Climatic and environmental factors

The present data indicated that the prevalence of infection in snails was higher in July to October. Yoder and Coggins (1998) also reported that the prevalence of the infection in snails was highest (4.18%) during the rainy season (July-October) whereas; it was lowest (0.76%) during the summer (March- June). Singh et al., 1998 also reported similar result; the number of snails shedding cercariae was highest during the rainy season (8.31%) followed by winter (6.8%) and summer (2.31%). No furcocercous cercariae were found in *L. exustus* during the summer. Loy and Hass also support our findings with the mean prevalence of most cercarial species increased from May- June to August- October. The results of the present study are (pre-monsoon and post-monsoon) in agreement with that of these works. However, prevalence of Fasciola infection in *L. auriularia* of Tarai region was highest in May (18.06%) and September (17.84%) and lowest in April (0.8%) and November (1.18%) reported by Garget al., (2009). Prevalence rate was high in the present observation and it may be due to the snails being infected by the parasitic stage i.e., cercarial stage during mid-rainy season when the eggs containing miracidium are being carried by water to low-lying areas where snails breed. The snails get infected by the parasites in about 1-1/2 months before and in the next 1-1/2 months the natural cercariae emerge out from the snails. The maximum emergence of cercariae from snails was noted in early winter months. The percentages of cercarial emergence during winter months have also been reported by Prasad, Sahai, Rajkhowa et al., and Kumar from different regions of the country.

As snail population is dependent on rainfall and temperature, it is no surprise to record change in snail population as per season which was highest during rainy season and lowest in summer. Accordingly, the percentage of positive snails is highest in monsoon or just after that, with the lowest number in summer. Almost all year-round the study recorded seasonal variation in snail population and positivity for schistosomes.

In a 2-year investigation, Soparkar (1921) reported negativeness of *I. exustus* for, *S. Spindale* between February and April, with the highest infection rate between September and October. In a survey near Bareilly, Uttar Pradesh, the population of L. Luteola var australis was high during March to May and October to February, low in June and September, and rare in July and August; "natural infection of S. incognitum could be detected in these snails mostly during March, April and May" (Sinha and Srivastava, 1960). Singh (1959) reported the highest positiveness of I. exustus for schistosome cercariae in the months of May and December, while Raut (1983) observed the maximum positive snails in the months of July and August. Year-round surveys have been made for S. nasale infection in the snails at four centers under All India project (Alwar, 1974) and for four mammalian schistosomes at Jabalpur (Kohli, 1991). In the two enzootic areas (Dhanayakanapura and Hunchipura) of Karnataka state, a 3-year survey of positive snails was made by collecting 5,286 I. exustus snails of which only 25 (0.47%) were found positive for S. nasale cercariae (Muraleedharan et al., 1976a). Interestingly, these positive snails could be observed only in the months of February, June, July, August, and October, in tandem with higher population during these months; they were negative in other months. A similar survey in Bhandara district of Maharashtra state between January 1995 and January 1996 revealed low snail population during late summer and early rainy season (April to August without finding any I. exustus in July and August), with its increase reaching a maximum in November. The highest positivity was recorded during November and January (maximum of 14.8% in January), whereas it was nil between April and August—when snail population was also low (Thakre and Bhilegaonkar, 1998). A 3-year (1987-1989) survey of I. Exustus at Jabalpur revealed 3.27% positivity for mammalian schistosome cercariae which was influenced by the season with recording of no positive snail in May and June but highest prevalence of 7.9% in January and 12.6% in April (Kohli, 1991). A similar study during the same years (1987-1989) on *I. exustus* made by another scientist (Mishra,1991) at Jabalpur, revealed 0.08–0.98% prevalence between April and July with the absence of any snails in the month of May; in other months, it varied between 0.37% and 11.35%, with the highest prevalence in October. Another survey on L. luteola at Jabalpur revealed overall prevalence of 3.35% of S. incognitum with observations of seasonal variation in the prevalence rate (Agrawal et al., 2000).

Nevertheless, the prevalence is not influenced by season alone, but other factors are also responsible as shown by us (Agrawal, 2000) when all the 74 *L. luteola* snails, collected from Marhai locality of Jabalpur, were found negative, but 100*L. luteola*, collected from the same pond just after 2 days, showed 13% being positive for *S. incognitum* infection. Likewise, variations in positiveness in other months have also been recorded suggesting that factors other than season are also influencing positiveness—perhaps a true assessment is difficult with present methodology.

Some of these influencing factors might be size and genetics of snail population, spot, time, weather, and number of snails collected. Simultaneously, these observations are indicative of how wrong it will be to declare a geographical area free from schistosomiasis merely on one-time survey of local snail population—that too during adverse season.

However, a general inference may be drawn by analyzing above data that positiveness of the snails is in tandem with their population in the environment—higher population, higher infection, and vice versa. This percentage of positive snails does not relate only with its population but also on miracidia population. During rainy season, there is increase in snail population, and a proportional increase should also be in miracidia number, thereby increasing not only percentage positivity but also absolute number of positive snails. As the absolute number of snails are decreasing post winter and summer, if the number of miracidia remains constant to the rainy season level, this would result in a higher percentage of positive snails due to the reduction in snail population (though not an absolute number), which is not happening. Indeed, a reverse is happening, decelerating positiveness of snails. This clearly hints that a lower number of snails are infected during winter and summer seasons, perhaps, because a lower number of miracidia are available in nature during these seasons. Or are some other factors responsible for lower positivity despite presence of constant number of miracidia.

Indeed, reduction in miracidia number as per season is supported by the observations of nasal scrapping and fecal examinations of domestic animals where schistosomiasis prevalence is shown to be influenced by the season—maximum during monsoon and minimum in summer (Muraleedharan *et al.*, 1976b; Kalwaghe *et al.*, 2003). As schistosomes are long lived (2–5 years), the only inference can be made is that egg extrusion is also influenced by the season,

though there is no season when animals completely stop excreting schistosome eggs. Therefore, this cannot be the sole reason for diminishing positivity in the snails as there are observations when in certain months no snail is found positive for schistosome cercariae. For instance, May and June were negative for *I. exustus* in the survey made by Kohli (1991).

As larval development in the snails takes about 30 days, it may be presumed that no *I. exustus* got infected during April/May. Thus, there is a situation in certain seasons of the year when snails are present but none became infected. It is important to work out the reasons for this negativity as it may greatly help in our control programs of schistosomiasis. What can be the reasons for this refractoriness of the snails, despite presence of miracidia in the environs?

To get a possible answer for the above refractoriness, it is important to note that with the change of the season, there are also changes in the snail's population dynamics, its habitats, and microclimates. The abiotic factors like dissolved oxygen, biochemical oxygen demand, pH, and mineral concentration of calcium, copper, iron, and zinc of soil and water of snail habitats differed as per season (Upadhyay, 2007). Change in type of aquatic weed, its quantity, pH, calcium, and total solid concentration as per season was also observed in the ponds of endemic foci of *S. nasale*in Karnataka (Muraleedharan and Kumar, 1974). The water sources start receding, and small ditches and pools may completely dry in summer season with changes in microclimate in all these snail habitats. There is a clear change in "biochemical ecology" as per season, and how the snail species are adjusting to these changes is not well understood.

Though the snails are considered being more flexible in their requirements, the same may not be true for schistosome miracidia or its activity. There is even a change in the proportion of snail population of gastropods with different percentage of *I. exustus* and *L. luteola* as per season (Upadhyay, 2007). Whether there is also a change in genetic makeup of these snail populations to withstand adverse environs (and so susceptibility to schistosomes) is not worked out, though it is well known that during pre-monsoons, snails have greater reproductive potentials. Obviously this phenomenon supports change in physiological and biochemical activities of the snails as per season; some might not be favorable for larval development resulting in encapsulation of miracidia that penetrate the snail during adverse seasons. There is also the possibility of changes

related to the miracidia. Change in hatchability of eggs as per season is the first possibility—the prime factor will be change in chloride ions in the water or salinity of water. There may be change in excretory products of snails which are responsible for miracidia attraction.

Additionally, there is evidence that all miracidia are not capable of infecting its compatible host. A study on *S. mansoni* and *B. glabrata* revealed that 30–60% miracidia do not penetrate the snail, and 15% miracidia which are penetrating the snails degenerate in the snail host in absence of hemocytic reaction—thus, this is beside encapsulation in the presence of an existing trematode infection (Jourdane and Theron, 1987). There is all the possibility that both infectivity and developing capability of miracidia are affected by the season also. This seasonal effect may vary as per sex of miracidia being more favorable to male miracidia during adverse season (e.g., summer), thereby resulting in more shedding of male cercariae. This assumption gets support from the recovery of only male *S. incognitum* flukes from rabbits and dogs which were infected subcutaneously with 390–1,200 *Schistosoma incognitummm* cercariae, pooled from six naturally infected *Lymnaea luteola* collected in March and April from in and around Jabalpur (Agrawal *et al.*, 1984).

Snail Habitats

The three species of snails, viz., *B.bengallensis, B.costula and Pila globosa* incriminated in schistosome infection in Indian continent, are lentic in nature, present in stagnant water sources such as lakes, ponds, ditches, and paddy fields. This situation is different from *Bulinus* or *Oncomelania* which have been associated with Nile or Yellow river. As Indian snails are not directly related with the rivers, itmay be argued that construction of dams and ramification of the canals from the rivers will not increase snail distribution and hence area of infectivity. This conjecture may partially be true as irrigation of any area increases the number of stagnant water bodies, thereby increasing snail population. The environment or ecology of a geographical area is mainly determined by rainfall or precipitation and temperature which indirectly are dependent on equatorial position and sea level of the geographical area. Bangladesh experiences extreme temperatures (7–42°C) and moderate temperatures (7–38°C) with two rainfalls, i.e., during monsoon and in winter. This is the reason for the abundance of snails (higher rainfall and moderate temperature). These snails are observed generally attached to floating leaves, stems,

and roots of vegetation, and their egg cocoons may easily be collected from these sources. There is a difference in biological requirements between *I. exustu s* and *L. luteola*, and hence their places of existence. Whereas *L. luteola* is observed generally in association with aquatic weeds, *I. exustus* is present even in transient water habitats devoid of any aquatic weed. Therefore, *I. exustus* could be collected from roadsides, low-lying land devoid of any aquatic weed but filled with rain water. Generally, workers have collected freshwater snails from lake, ponds, tanks, ditches, water pools, paddy fields, and marshy lands from an endemic area, separated and counted only *I. exustus* and *L. luteola* from their collection. A comparative study is made to estimate snail density in perennial ponds, marshy lands, water pools, ditches, and paddy fields at Jabalpur where the highest snail population was observed in perennial ponds (2,611 snail/m²), followed by marshy lands (1,146 snail/m²), ditches (304 snail/m²), and paddy fields (294 snail/m²), with the least snail population in the water pools (288 snail/m²); the difference proved statistically significant (P < 0.01) (Upadhyay, 2007).

Marshy lands also sustained snail population throughout the year which was not the case with water pools, ditches, and paddy fields. In the present study, the overall snail population in marshy lands ranged from 1,025 to 1,267 snails/m². The season influenced their percentage; it was 23.42%, 21%, and 17.67% for *Bellamya* in rainy, winter, and summer seasons, with 32%, 37.57%, and 30.7% being the corresponding figures for *Pila*. A lower snail population ranging between 235 and 303 snail/m² was seen in water pools, ditches, and paddy fields which sustained snail population during rainy season or winter season but dried up during summer. In all these places, *Brotia* has lower concentration (17–29%) with a comparatively higher concentration of *Pila* (31–39%).

Various workers have studied the impact of temperature, salinity, pH, dissolved oxygen, and water hardness on survival and breeding of *I. exustus* and *L. luteola* both under experimental conditions and analyzing them in natural habitats (Aziz and Raut, 1996; Mishra and Agrawal, 1996; Agrawal, 1999). In Karnataka, the water bodies with pH 6–9.2, calcium content of 17.5–91.6 ppm, and dissolved solids of 54.4–1,472 ppm with or without aquatic plants sustained *I. exustus* very well (Muraleedharan *et al.*, 1977). Devi *et al.*, (2009) provided evidence that a low dissolved carbon dioxide (2–6 ppm), high total alkalinity (70–85 ppm), and a pH range of 7.5–7.7 are suitable for diverse snail fauna.

These studies reflect difficulty in changing water environment detrimental to snail life (for snail control) but without damaging other aquatic fauna. Though season influences absolute snail population, the season of highest and lowest population differed as per geographical area of investigation. In Assam, there is abundant snail population in natural environment from February to July with differences as per snail species— *L. luteola* are prevalent with 5–10% prevalence throughout the year, whereas *I. exustus* starts falling in October with gradual disappearance from December onward (Devi *et al.*,2008). Even in the same season, the snail population differed among identical water sources—between two water ponds, or two marshy lands, etc. There may be different reasons for such variation, but important appears microenvironment of these similar habitats as differences in types of aquatic weeds, aquatic fauna (including *Daphnia, Cyclops*), soil texture, and mineral concentration have been demonstrated to exist between two perennial ponds or two marshy lands or water pools (Upadhyay, 2007). Moreover, it must be remembered that the snails are using discontinuous, temporary water habitats which create isolated local populations of snail fauna, giving rise to infra-specific variations that may also be responsible for these variations.

Human Activities Influencing the Habitats

Though water levels in these water sources are mainly depended on temperature and rainfall, human activities also play a crucial role in maintaining or otherwise the levels throughout the year. The prime reason is enhancing irrigation facilities which resulted in increase in inundated areas and prolongation of water in the water sources. Whether temporary water sources are used for agriculture or not also influences water levels during the year. For using ponds for fish culture, animal fecal material is added, which helps in the survival of snails; likewise, the ponds are used for cultivation of water chestnuts/lily or other aquatic crop, and the snails utilize the crop both as its food as well as for laying eggs. There are other human activities which affect snail population adversely by destroying their habitats. The first is adding pollutants in the form of detergents, chemicals, and industrial wastes which destroy ecological system of aquatic fauna, thus helping in destroying snail population. Other is urbanization or colonization in new areas resulting in filling of the ponds, tanks, and lakes for construction of the buildings.

SUMMARY AND CONCLUSION

Summary

The knowledge of the correlations between larval and adult forms, supported by environmental studies, gives us the possibility to decode the total life history of many Digenea species (Galaktionov and Skirnisson, 2000). There is full justification for conducting the research mentioned from the point of view of preventing schistosomiasis, fascioliasis, opisthorchiasis etc. (Esteban *et al.*, 2003; Karvoven *et al.*, 2006). However, bearing in mind the close relationship between trematodes and snails, results of research devoted to the relations of partners in the host–parasite system will be extraordinarily valuable. This requires, however, broad collaboration between parasitologist and malacologists. A comprehensive look at the causes and effects of fluctuations in trematodes in snail populations would make it possible to prepare a detailed analysis of such phenomena as the consequences of the accidental introduction of new species into the ecosystem (Niewiadomska and Pojmańska, 2004; Pointer *et al.*, 2005).

Efforts by malacologists and parasitologists conducting field studies and experimental research will make it possible to explain phenomena that are known incompletely today such as recognition in the snail–trematode system, the specificity of partners and defence responses, chaetotaxy, parasite manipulation, gigantism or snail shell hyper-calcification. These phenomena studied at the population and/or species level will also allow the evolution of both groups of animals to be better understood. It is sufficient to mention the fact that in recent years efforts have been undertaken with a view to constructing phylogenetic trees of Mollusca and Digenea based on data on interspecies relations in the host–parasite system (Bargues and Mas-Coma 1997; Bargues *et al.*, 2001). Developing this direction will be useful only after interspecies relationships of mollusc–trematode have first been studied in natural conditions.

Three species of freshwater gastropods were identified belonging to two families (Viviparidae, Ampullariidae) and three genera (*Bellamya, Pila, Brotia*). The available species of this particular study were found in abundance and dominated all the 23 lentic collection sites.

The identified trematodes belong to 8 types having 17 species. The trematode cercariae types along with species are: Parapleurophocercous (*Haplorchis pumilio*, *Haplorchs taichui*,

Stictodora tridactyla); Pleurophocercous(Centrocestus fnusormosa); Xiphidiocercariae (Acanth atrium hitaense, Loxogenoides bicolor, Haematoloechus similis,

Cloacitrema philippinum); Furcocercous (Cardicola alseae, Alaria mustelae, Apatemon gracilis, Mesostephanus appendiculatus); Echinostome (Echinochasmus pelecani); Amphistome (Gastrothylax crumenife); Renicolid (Cercaria caribbea); Cotylomicrocercous (Podocotyle lepomis).

Beside these, two types of Nematodes, two types of Cestodes, one type of Turbellarian and one type of Penstomida were identified.

The trend of trematode infection increased in *Bellamya* sp. from July 2011 to September 2011 and in decreasing trend up to February 2012 at Dhaka and surrounding region. Highest percentage of infection by trematodes in *Bellamya* sp. was found in September which was 17 to 30% and lowest infection was 3.85 to 6.25% in February 2012. Highest percentage of infection by trematodes in *Brotia* sp. was found in May which was 13.79 to 30% and lowest was 3.45 to 6.25% in February. Highest percentage of infection by trematodes in *Pila* sp. was 14.81 to 21.21% in September 2011 and lowest trematode infection was 4.76 to 7.14% in February 2012.

Whereas the trematode infection in *Bellamya* sp. started to increase from July 2012 to August 2012 which was in decreasing up to February 2013. Comparatively highest percentage of infection by trematodes in *Bellamya* sp. was found in August 23.33% and lowest 8.33% in February 2013. In *Brotia sp*. highest infection was found in September 17.65 to 29.63%. and lowest 4.17 to 6.25% in February 2013.

Highest percentage of trematode infection in *Pila* sp. was 19.03 to 21.05% in September and lowest was 5.88 to 7% in February 2013.

The similar nature was observed in Khulna and Kishoreganj region.

In 2011-12, occurrence of trematode parasite at Dhaka region in *Bellamya* was significantly higher than that of *Brotia* and *Pila* (p<0.05). However, similar occurrence of this parasite in *Brotia* and *Pila* was detected. Moreover, in 2012-13 occurrence of trematode parasite in *Bellamya* was significantly higher than that of *Brotia* and *Pila*. However, similar occurrence of this parasite was found in *Brotia* and *Pila*. On the other hand, occurrence of this parasite in *Bellamya* found in 2012-13 was significantly higher than that of 2011-12. No significant different was found in *Brotia* and *Pila* in this two years (p>0.05).

In 2011-12, occurrence of trematode parasite at Khulna region in *Bellamya* was significantly higher than that of *Brotia* and *Pila* (p<0.05). However, similar occurrence of this parasite in *Brotia* and *Pila* was detected. Moreover, in 2012-13 occurrence of trematode parasite in *Pila* was significantly higher than that of *Bellamya* and *Brotia*. No significant different was found in *Pila* in this two years (p>0.05).

In 2011-12, the occurrence of trematode parasite at Kishoreganj region in *Bellamya* was significantly higher than that of *Brotia* and *Pila* (p<0.05). However, similar occurrence of this parasite was found in all snails in 2012-13.

The prevalence of trematode parasites in *Bellamya*, *Brotia* and *Pila* in 2011-2012 and 2012-2013 sampled from 23 different regions were determined. Each year was divided into three different seasons (Rainy, winter and summer) where the incidence of the parasites in rainy season in most of the cases was significantly higher than that of other seasons. However, the prevalence in summer (in most of the regions) was significantly higher than that of winter.

Various environmental factors may impact the emergence of larval trematodes (cercariae) from their molluscan first intermediate hosts. For example, the effects of temperature on cercarial emergence are well documented. Many trematode species play increased emergence in response to increased temperature. The overall natural environmental parameters varied within overlapping ranges among the ponds. Water temperature ranged between 4.7°C and 38°C depending on the day and time of collection. The hydrogen ion concentration was within the range providing protection for the aquatic life (7.2 to 9.8). The concentration of dissolved oxygen showed generally low in some locations due to pollution load (Dhaka region) and higher

(> 7.0 mg/L) levels and exhibited a strong negative association with water conductivity ($r_s = -0.665$, p < 0.05).

Significantly positive correlation was found between temperature and prevalence of parasites in snails of Dhaka regions in 2011-12 (p<0.01). Positive correlation was also found between % occurrence and pH, EC, DO and rainfall, but was not significant (p>0.05). Also significant positive correlation was found between temperature and prevalence of parasites and rainfall and prevalence of parasites in snails of Dhaka regions in 2012-13 (p<0.01). Positive correlation was also found between % occurrence and pH, EC, and DO, but was not significant (p>0.05).

Significantly positive correlation was found between temperature, EC, rainfall and prevalence of parasites in snails of Khulna regions in 2011-12 (p<0.01). Positive correlation was also found between % occurrence and pH, but was not significant (p>0.05). Whereas significant positive correlation was found between temperature, EC, rainfall and prevalence of parasites in snails of Khulna regions in 2012-13 (p<0.01). Positive correlation was also found between % occurrence and pH, but was not significant (p>0.05).

Significantly positive correlation was found between temperature, EC (p<0.05), rainfall and prevalence of parasites in snails of Kishoreganj regions in 2012-13 (p<0.01). Positive correlation was also found between % occurrence and pH, but was not significant (p>0.05). Also significant positive correlation was found between temperature, EC (p<0.05), rainfall and prevalence of parasites in snails of Khulna regions in 2012-13 (p<0.01). Positive correlation was also found between % occurrence and pH, but was not significant (p>0.05).

The above mentioned physico-chemical and climatological parameters of different locations do play a vital role in the population dynamics of molluscan species. It is made clear from the results that more number of species related with low DO content. It can be inferred from the above points that DO content plays a vital role in affecting the population dynamics of a particular species than the other physico-chemical factors like pH, temperature and EC. Moreover the molluscan population is abundant in a place where the aquatic plants are available in diverse forms. Since there is not much variation in pH, temperature, EC and salinity in the collection sites, they do not have much influence in the availability of specimens in this present study. Through this analysis, it is inferred that the possibility of variation in pH, temperature, EC

can play a vital role in the life cycle of any plant or animal group and when it is moderate or in low, it can not have much influence on the population dynamics.

In the present study monthly fluctuations in the cercarial prevalence were observed due to the variations in the temperature, rainfall and dissolve oxygen of each month. The occurrence of cercaria are maximum in February to April (pre-monsoon) and September to November (post-monsoon) time which are dealt with elevated temperature, low rainfall and DO level in the Dhaka region. Whereas, In Khulna and Kishoreganj region only pre-monsoon time shows the maximum prevalence. Islam (2008) reported 14.5% infection and Alim (1997) and Rahman *et al.*, (1997) reported 3.7% infection during March-May.

Among the 23 locations nematode infection was very rare. The Nematodes are *Oionchus* (Order-Mononchida) and *Angiostrongylus* (Order-Strongylida). Rather than 2 Cestodes, 1Turballarian and 1 Pentastomida were found in *Pila globosa* at Ramna pond in September in 2011-12.

Prevalence of Nematodes was found only 3 locations i.e. Ramna park pond, Mawna bazaar pond and Solakia, Kishoreganj pond. The overall infection on the two year period (2011-12 and 2012-13) showed that at Ramna it was 1.47 to 1.81%, Mawna 3.13 to 4.64% and at Solakia it was 4.38 to 4.55%. No significant relation could not sort out with climatic or environmental factors of its existence due to its scarce nature.

Freshwater snails distributed in different habitats in different locations of Bangladesh were infected with eight types of trematode cercariae. *Bellamya bengalensis* was the most common host species for most of the trematode cercariae found in the study period. However the small ponds with livestock mobility were noticed as the richest habitats for snails infected by trematodes. These findings suggest that to control the snail-borne diseases, it is necessary to control the population of *Bellamya bengalensis* mainly around temporary ponds prone to cattle grazing. It is hopeful that the study will serve to stimulate further work on patent and pre-patent infections of snail hosts; larval behavior and morphology; geographical distribution of trematodes; infections in human beings, livestock and wildlife; and clinical manifestations of trematode infection in Bangladesh.

The snails are undergoing hibernation is confirmed by recovering alive snails from moist soil with dead snails when soil is completely dried, suggesting that temporary water sources are

replenished by a new batch of snails from perennial water sources. However, human activities are important for snails' ecology. Irrigation facilities increase inundation of new areas, prolongation of water levels, and cultivation of water chestnut/lily, which help in enhancing snail population, whereas addition of pollutants and industrial wastes in the water sources and construction of buildings by filling the water sources are helpful in diminishing their population.

Conclusion

- In the present study, *Bellamya bengalensis* is the appropriate host for the DST though *Pila globosa* is a common host.
- At the end of this research, the usefulness of continuing environmental research in presented field needs to be examined. This kind of environmental research is still necessary due to the incomplete list of trematode species whose larvae develop inside snails.
- Infection of snail is more common in September month due to runoff water inundated the ponds which carry eggs of the infectors from catchment areas.
- Among 23 locations nematode infection was very rare. The Nematodes are *Oionchus* (Order- Mononchida) and *Angiostrongylus* (Order- Strongylida). Rather than 2 Cestodes, 1Turballarian and 1 Pentastomida were identified in *Pila globosa*.
- Cercarial dermatitis should be considered a potential risk whenever warm-blooded and molluscan hosts share a water resource with man as it is a self-limited, severely itching rash that lasts about one week and may be easily mistaken for insect bites.
- The suggestion could be sort out that to control the snail-borne diseases, it is necessary to control the population of *Bellamya bengalensis* mainly around temporary ponds prone to cattle grazing.

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Appendix

Appendix A: Data sheet

Site 1(2011-12)

Table 1: Monthly variation of the abundance of snails and the prevalence of larval trematodes in Ramna Park Pond (2011-2012).

		Bellamye	а	Brotia			Pila		
Month	Tot	Inf	%		Inf	%	Tot	Inf	%
Jul-11	24	3	12.50	20	2	10.00	25	4	16.00
August	30	6	20.00	25	3	12.00	22	4	18.18
September	33	8	24.24	19	3	15.79	33	7	21.21
October	29	4	13.79	18	2	11.11	16	3	18.75
November	19	2	10.53	21	2	9.52	16	2	12.50
December	17	2	11.76	19	3	15.79	13	2	15.38
Jan-12	21	3	14.29	18	2	11.11	22	3	13.64
February	16	1	6.25	16	1	6.25	14	1	7.14
March	25	3	12.00	19	3	15.79	16	2	12.50
April	18	2	11.11	14	2	14.29	13	2	15.38
May	19	3	15.79	17	3	17.65	20	3	15.00
June	21	4	19.05	18	3	16.67	14	2	14.29
Total	272	41		224	29		224	35	

Table 2: Monthly variation of the abundance of snails and the prevalence of larval trematodes in Curzon Hall Pond (2011-2012).

		Bellamy	a		Brotia			Pila	
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
Jul-11	36	4	11.11	33	4	12.12	26	3	11.54
August	33	6	18.18	30	3	10.00	24	3	12.50
September	34	8	23.53	32	4	12.50	27	4	14.81
October	30	4	13.33	29	4	13.79	22	3	13.64
November	30	2	6.67	22	1	4.55	16	1	6.25
December	31	3	9.68	25	2	8.00	22	2	9.09
Jan-12	32	3	9.38	32	3	9.38	28	2	7.14
February	26	1	3.85	27	1	3.70	21	1	4.76
March	34	4	11.76	32	2	6.25	21	2	9.52
April	31	4	12.90	28	2	7.14	27	3	11.11
May	33	5	15.15	30	3	10.00	30	4	13.33
June	38	5	13.16	31	2	6.45	28	3	10.71
Total	388	49		351	21		292	31	

Table 3: Monthly variation of the prevalence of larval trematodes of snails in Jagannath Hall Pond (2011-12).

		Bellamya			Brotia			Pila	
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
Jul-11	31	5	16.13	33	3	9.09	35	6	17.14
August	32	6	18.75	35	6	17.14	30	5	16.67
September	30	9	30.00	33	6	18.18	33	7	21.21
October	33	8	24.24	29	4	13.79	28	4	14.29
November	28	2	7.14	29	2	6.90	30	2	6.67
December	29	1	3.45	33	3	9.09	27	3	11.11
Jan-12	34	4	11.76	28	4	14.29	33	3	9.09
February	30	2	6.67	30	1	3.33	30	1	3.33
March	33	5	15.15	29	3	10.34	29	4	13.79
April	32	6	18.75	33	4	12.12	33	4	12.12
May	34	6	17.65	34	5	14.71	34	5	14.71
June	31	6	19.35	25	3	12.00	30	5	16.67
Total	387	63		381	45		382	51	

Table 4: Monthly variation of the prevalence of larval trematodes of snails in Keraniganj Pond (2011-12).

		Bellamya			Brotia		Pila		
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
Jul-11	32	4	12.50	33	3	9.09	29	3	10.34
August	34	5	14.71	30	3	10.00	23	3	13.04
September	37	7	18.92	34	4	11.76	35	6	17.14
October	27	4	14.81	34	3	8.82	31	3	9.68
November	30	2	6.67	30	1	3.33	27	1	3.70
December	33	3	9.09	32	2	6.25	29	3	10.34
Jan-12	28	3	10.71	23	1	4.35	33	2	6.06
February	33	2	6.06	26	1	3.85	23	1	4.35
March	31	3	9.68	26	2	7.69	32	2	6.25
April	29	3	10.34	28	3	10.71	30	3	10.00
May	25	3	12.00	30	3	10.00	36	5	13.89
June	26	4	15.38	27	2	7.41	31	4	12.90
Total	365	39		353	28		359	36	

Table 5: Monthly variation of the abundance and the prevalence of larval trematodes in Tongi Railway Station Pond (2011-12).

		Bellamya			Brotia		Pila		
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
Jul-11	23	4	17.39	19	1	5.26	25	4	16.00
August	33	7	21.21	25	2	8.00	30	5	16.67
September	32	8	25.00	26	3	11.54	36	7	19.44
October	24	3	12.50	29	3	10.34	23	3	13.04
November	17	2	11.76	13	0	0.00	16	2	12.50
December	15	2	13.33	25	1	4.00	23	3	13.04
Jan-12	20	3	15.00	23	2	8.70	27	3	11.11
February	13	1	7.69	10	0	0.00	13	1	7.69
March	31	3	9.68	18	2	11.11	28	3	10.71
April	33	4	12.12	21	3	14.29	26	3	11.54
May	31	5	16.13	16	2	12.50	30	3	10.00
June	30	5	16.67	18	2	11.11	28	4	14.29
Total	302	47		243	21	8.64	305	41	13.44

Table 6: Monthly variation of the abundance and the prevalence of larval trematodes in Jaydevpur Railway Station Pond (2011-12).

		Bellamya			Brotia		Pila		
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
Jul-11	22	3	13.64	21	2	9.52	29	3	10.34
August	28	4	14.29	26	3	11.54	26	4	15.38
September	30	7	23.33	29	3	10.34	31	4	12.90
October	15	3	20.00	21	2	9.52	30	3	10.00
November	16	2	12.50	14	1	7.14	21	2	9.52
December	18	2	11.11	29	3	10.34	27	3	11.11
Jan-12	13	1	7.69	26	2	7.69	25	3	12.00
February	18	1	5.56	14	1	7.14	19	1	5.26
March	28	3	10.71	12	1	8.33	26	3	11.54
April	27	3	11.11	22	2	9.09	18	3	16.67
May	30	4	13.33	29	3	10.34	21	4	19.05
June	29	4	13.79	19	2	10.53	22	3	13.64
Total	274	34	12.41	262	25	9.54	295	36	12.20

Table 7: Monthly variation of the abundance of snails and the prevalence of larval trematodes in Bhawal- Gazipur Railway station Pond (2011-12).

		Bellamya			Brotia		Pila		
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
Jul-11	28	3	10.71	15	2.00	13.33	13	2	15.38462
August	32	5	15.63	17	2.00	11.76	21	3	14.28571
September	33	7	21.21	21	3.00	14.29	16	3	18.75
October	28	3	10.71	28	3.00	10.71	18	3	16.66667
November	13	1	7.69	22	2.00	9.09	12	1	8.333333
December	21	2	9.52	21	2.00	9.52	17	2	11.76471
Jan-12	22	3	13.64	26	3.00	11.54	21	2	9.52
February	16	1	6.25	16	1.00	6.25	12	1	8.33
March	17	3	17.65	17	2.00	11.76	11	1	9.09
April	25	3	12.00	25	3.00	12.00	19	2	10.53
May	26	3	11.54	17	2.00	11.76	11	2	18.18
June	23	3	13.04	23	3.00	13.04	16	2	12.50
Total	284	37		248	28.00		187	24	12.83

Table 8: Monthly variation of the abundance of snails and the prevalence of larval trematodes in Salna BSMAU Pond (2011-12).

		Bellamya		Brotia			Pila		
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
Jul-11	19.00	2.00	10.53	19	2	10.53	22	3	13.64
August	16.00	2.00	12.50	17	3	17.65	19	2	10.53
September	21.00	5.00	23.81	21	4	19.05	22	4	18.18
October	18.00	3.00	16.67	18	2	11.11	14	2	14.29
November	22.00	2.00	9.09	17	2	11.76	15	2	13.33
December	25.00	3.00	12.00	15	2	13.33	21	3	14.29
Jan-12	30.00	4.00	13.33	16	2	12.50	16	2	12.50
February	29.00	1.00	3.45	13	1	7.69	15	1	6.67
March	31.00	4.00	12.90	18	2	11.11	20	2	10.00
April	27.00	3.00	11.11	22	3	13.64	21	4	19.05
May	15.00	2.00	13.33	20	3	15.00	18	3	16.67
June	26.00	3.00	11.54	13	2	15.38	17	2	11.76
Total	279.00	34.00		209	28		220	30	13.64

Table 9: Monthly variation of the abundance of snails and the prevalence of larval trematodes in Mawna Bazar pond (2011-12).

		Bellamya		Brotia			Pila		
Month	Tot	Inf	%		Inf	%	Tot	Inf	%
Jul-11	31	4	12.90	19	3	15.79	24	3	12.50
August	29	5	17.24	14	2	14.29	19	3	15.79
September	21	6	28.57	17	3	17.65	21	7	33.33
October	18	3	16.67	23	3	13.04	18	5	27.78
November	16	2	12.50	13	2	15.38	13	2	15.38
December	13	2	15.38	21	3	14.29	12	2	16.67
Jan-12	17	2	11.76	19	2	10.53	11	1	9.09
February	19	1	5.26	14	1	7.14	13	1	7.69
March	17	2	11.76	16	1	6.25	14	2	14.29
April	18	3	16.67	11	2	18.18	10	1	10.00
May	19	3	15.79	17	3	17.65	18	3	16.67
June	14	2	14.29	16	3	18.75	21	3	14.29
Total	232	35		200	28		194	33	

Table 10: Monthly variation of the abundance of snails and the prevalence of larval trematodes in Tongi Bari Bazar Pond (2011-12).

		Bellamya		Brotia			Pila		
Month	Tot	Inf	%		Inf	%	Tot	Inf	%
Jul-11	28	3	10.71	19	3	15.79	26	3	11.54
August	21	4	19.05	14	2	14.29	21	3	14.29
September	31	8	25.81	17	3	17.65	25	5	20.00
October	22	5	22.73	23	3	13.04	18	3	16.67
November	14	2	14.29	13	2	15.38	13	2	15.38
December	16	2	12.50	21	3	14.29	16	3	18.75
Jan-12	17	2	11.76	19	2	10.53	14	2	14.29
February	17	1	5.88	14	1	7.14	13	1	7.69
March	21	2	9.52	16	1	6.25	14	2	14.29
April	14	2	14.29	11	2	18.18	18	3	16.67
May	17	3	17.65	17	3	17.65	21	3	14.29
June	14	3	21.43	16	3	18.75	25	3	12.00
Total	232	37		200	28		224	33	

Table 11: Monthly variation of the abundance of snails and the prevalence of larval trematodes in Lahajang Bazar Pond (2011-12).

		Bellamya		Brotia			Pila		
Month	Tot	Inf	%		Inf	%	Tot	Inf	%
Jul-11	16	3	18.75	19	2	10.53	22	3	13.64
August	13	2	15.38	26	3	11.54	25	4	16.00
September	21	5	23.81	25	3	12.00	31	6	19.35
October	20	4	20.00	27	2	7.41	29	3	10.34
November	11	2	18.18	19	2	10.53	19	2	10.53
December	17	2	11.76	21	3	14.29	21	2	9.52
Jan-12	21	3	14.29	18	2	11.11	16	3	18.75
February	16	1	6.25	13	1	7.69	11	1	9.09
March	22	2	9.09	16	2	12.50	14	2	14.29
April	18	3	16.67	13	2	15.38	13	1	7.69
May	15	3	20.00	17	3	17.65	19	2	10.53
June	14	3	21.43	21	3	14.29	14	2	14.29
Total	204	33		235	28		234	31	

Site 1 (2012-13)

Table 1: Monthly variation of the abundance of snails and the prevalence of larval trematodes in Ramna Park Pond (2012-2013).

		Bellamya		Brotia			Pila		
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
July/2012	21	4	19.05	15	2	13.33	22	3	13.64
August	30	7	23.33	20	3	15.00	28	5	17.86
September	24	5	20.83	17	3	17.65	19	4	21.05
October	14	2	14.29	12	2	16.67	16	3	18.75
November	16	2	12.50	13	1	7.69	14	2	14.29
December	19	2	10.53	18	2	11.11	17	2	11.76
January/2013	17	2	11.76	17	2	11.76	10	1	10.00
February	12	1	8.33	16	1	6.25	17	1	5.88
March	16	2	12.50	14	2	14.29	15	2	13.33
April	22	4	18.18	19	3	15.79	18	3	16.67
May	17	3	17.65	14	2	14.29	21	4	19.05
June	13	2	15.38	16	2	12.50	14	2	14.29
Total	221	36		191	25		211	32	

Table 2: Monthly variation of the abundance of snails and the prevalence of larval trematodes in Curzon Hall Pond (2012-2013).

		Bellamyo	ı		Brotia		Pila			
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%	
July/2012	31	5	16.13	21	2	9.52	38	5	13.16	
August	35	7	20.00	18	3	16.67	26	6	23.08	
September	29	8	27.59	26	5	19.23	27	8	29.63	
October	22	4	18.18	22	3	13.64	19	3	15.79	
November	16	2	12.50	17	1	5.88	12	1	8.33	
December	25	3	12.00	16	2	12.50	16	3	18.75	
January/2013	14	2	14.29	22	3	13.64	14	3	21.43	
February	15	1	6.67	24	1	4.17	15	1	6.67	
March	30	4	13.33	27	2	7.41	23	2	8.70	
April	28	4	14.29	23	3	13.04	25	4	16.00	
May	31	5	16.13	28	3	10.71	29	5	17.24	
June	28	5	17.86	21	2	9.52	22	3	13.64	
Total	304	50		265	21		266	44		

Table 3: Monthly variation of the prevalence of larval trematodes of snails in Jagannath Hall Pond (2012-13).

		Bellamya			Brotia		Pila			
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%	
July/2012	29	4	13.79	20	3	15.00	31	4	12.90	
August	31	6	19.35	16	2	12.50	27	3	11.11	
September	26	6	23.08	21	4	19.05	30	5	16.67	
October	21	4	19.05	17	2	11.76	25	3	12.00	
November	18	3	16.67	10	1	10.00	21	2	9.52	
December	13	2	15.38	13	2	15.38	19	3	15.79	
January/2013	11	1	9.09	18	3	16.67	18	3	16.67	
February	14	1	7.14	12	1	8.33	10	1	10.00	
March	25	3	12.00	17	2	11.76	17	3	17.65	
April	28	4	14.29	12	2	16.67	29	4	13.79	
May	26	4	15.38	18	2	11.11	30	5	16.67	
June	27	4	14.81	14	1	7.14	33	4	12.12	
Total	269	42		188	25		290	40		

Table 4: Monthly variation of the prevalence of larval trematodes of snails in Keraniganj Pond (2012-13).

	Bellamya			Brotia			Pila		
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
July/2012	29	5	17.24	31	4	12.90	21	3	14.29
August	31	6	19.35	36	5	13.89	25	4	16.00
September	33	8	24.24	29	5	17.24	31	6	19.35
October	25	4	16.00	25	3	12.00	35	6	17.14
November	26	3	11.54	21	2	9.52	28	3	10.71
December	21	3	14.29	17	2	11.76	25	3	12.00
January/2013	19	2	10.53	12	2	16.67	22	4	18.18
February	13	1	7.69	11	1	9.09	20	1	5.00
March	17	2	11.76	17	2	11.76	17	3	17.65
April	22	3	13.64	28	3	10.71	13	2	15.38
May	29	4	13.79	32	4	12.50	21	3	14.29
June	31	5	16.13	26	2	7.69	26	3	11.54
Total	296	41		285	35		284	41	

 Table 5: Monthly variation of the abundance of snails and the prevalence of larval trematodes in Tongi Railway Station Pond (2012-13).

		Bellamya	ı		Brotia		Pila		
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
July/2012	19	3	15.79	21	2	9.52	31	4	12.90
August	22	4	18.18	18	2	11.11	33	5	15.15
September	27	6	22.22	22	3	13.64	38	8	21.05
October	16	3	18.75	24	3	12.50	32	4	12.50
November	19	2	10.53	18	2	11.11	28	2	7.14
December	14	2	14.29	16	2	12.50	29	3	10.34
January/2013	18	2	11.11	18	2	11.11	26	4	15.38
February	12	1	8.33	21	2	9.52	21	2	9.52
March	28	3	10.71	21	3	14.29	30	4	13.33
April	31	4	12.90	18	2	11.11	26	4	15.38
May	33	5	15.15	27	4	14.81	20	3	15.00
June	29	4	13.79	22	3	13.64	28	4	14.29
Total	268	39		246	30		342	47	

Table 6: Monthly variation of the abundance of snails and the prevalence of larval trematodes in Jaydevpur Railway Station Pond (2012-13).

		Bellamya			Brotia		Pila		
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
July/2012	19	3	15.79	23	3	13.04	21	3	14.29
August	21	5	23.81	16	2	12.50	25	4	16.00
September	27	7	25.93	24	4	16.67	29	6	20.69
October	20	4	20.00	17	2	11.76	26	3	11.54
November	14	2	14.29	13	1	7.69	24	2	8.33
December	17	3	17.65	15	2	13.33	22	3	13.64
January/2013	19	2	10.53	22	3	13.64	18	2	11.11
February	14	1	7.14	13	1	7.69	14	1	7.14
March	21	2	9.52	13	2	15.38	17	2	11.76
April	24	4	16.67	21	3	14.29	21	3	14.29
May	21	4	19.05	27	4	14.81	13	2	15.38
June	26	4	15.38	17	2	11.76	19	3	15.79
Total	243	38		221	29		249	34	

Table 7: Monthly variation of the abundance of snails and the prevalence of larval trematodes in Bhawal- Gazipur Railway station Pond (2012-13).

		Bellamya			Brotia		Pila		
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
July/2012	21	3	14.29	13	2.00	15.38	23	4	17.39
August	26	5	19.23	21	3.00	14.29	15	3	20
September	31	7	22.58	23	4.00	17.39	18	4	22.22
October	17	3	17.65	20	3.00	15.00	21	4	19.05
November	16	2	12.50	16	2.00	12.50	19	2	10.53
December	21	3	14.29	22	3.00	13.64	21	3	14.29
January/2013	19	2	10.53	21	4.00	19.05	25	3	12.00
February	12	1	8.33	14	1.00	7.14	18	2	11.11
March	21	3	14.29	22	3.00	13.64	15	2	13.33
April	18	3	16.67	18	3.00	16.67	14	2	14.29
May	24	5	20.83	17	2.00	11.76	18	3	16.67
June	18	3	16.67	10	1.00	10.00	19	3	15.79
Total	244	40		217	31.00		226	35	

Table 8: Monthly variation of the abundance of snails and the prevalence of larval trematodes in Salna BSMAU Pond (2012-13).

		Bellamya		Brotia			Pila		
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
July/2012	30	4	13.33	12	2	16.67	34	4	11.76
August	33	6	18.18	15	3	20.00	31	5	16.13
September	28	4	14.29	18	4	22.22	27	8	29.63
October	25	3	12.00	11	2	18.18	16	3	18.75
November	21	2	9.52	10	1	10.00	21	2	9.52
December	27	3	11.11	13	2	15.38	25	3	12.00
January/2013	28	4	14.29	17	2	11.76	19	2	10.53
February	21	1	4.76	14	1	7.14	16	1	6.25
March	27	4	14.81	21	3	14.29	18	2	11.11
April	22	3	13.64	22	3	13.64	21	3	14.29
May	19	3	15.79	26	4	15.38	19	3	15.79
June	21	3	14.29	21	3	14.29	17	2	11.76
Total	302	40		200	30		264	38	

Table 9: Monthly variation of the abundance of snails and the prevalence of larval trematodes in Mawna Bazar pond (2012-13).

		Bellamya		Brotia			Pila		
Month	Tot	Inf	%		Inf	%	Tot	Inf	%
July/2012	33	4	12.12	31	4	12.90	28	5	17.86
August	24	5	20.83	26	3	11.54	24	3	12.50
September	25	6	24.00	21	3	14.29	18	5	27.78
October	17	4	23.53	27	3	11.11	18	3	16.67
November	21	2	9.52	22	2	9.09	18	2	11.11
December	28	3	10.71	30	3	10.00	14	2	14.29
January/2013	24	4	16.67	18	1	5.56	20	2	10.00
February	20	2	10.00	15	1	6.67	16	2	12.50
March	13	2	15.38	16	2	12.50	14	2	14.29
April	16	3	18.75	18	3	16.67	18	3	16.67
May	12	2	16.67	17	3	17.65	13	2	15.38
June	17	3	17.65	13	2	15.38	23	3	13.04
Total	250	40		254	30		224	34	

Table 10: Monthly variation of the abundance of snails and the prevalence of larval trematodes in Tongi Bari Bazar Pond (2012-13).

		Bellamya		Brotia			Pila		
Month	Tot	Inf	%		Inf	%	Tot	Inf	%
July/2012	30	5	16.67	18	2	11.11	29	3	10.34
August	17	3	17.65	14	2	14.29	25	3	12.00
September	22	4	18.18	21	3	14.29	28	5	17.86
October	18	3	16.67	23	3	13.04	12	2	16.67
November	14	2	14.29	16	2	12.50	16	2	12.50
December	19	2	10.53	18	2	11.11	21	3	14.29
January/2013	16	2	12.50	22	2	9.09	18	2	11.11
February	13	1	7.69	16	1	6.25	12	1	8.33
March	16	3	18.75	18	2	11.11	19	2	10.53
April	18	3	16.67	23	3	13.04	22	3	13.64
May	17	3	17.65	21	3	14.29	26	4	15.38
June	21	3	14.29	19	3	15.79	21	3	14.29
Total	221	34		229	28		249	33	

Table 11: Monthly variation of the abundance of snails and the prevalence of larval trematodes in Lahajang Bazar Pond (2012-13).

		Bellamy	а	Brotia			Pila		
Month	Tot	Inf	%		Inf	%	Tot	Inf	%
July/2012	12	2	16.67	18	1	5.56	19	3	15.79
August	17	3	17.65	24	1	4.17	21	3	14.29
September	14	3	21.43	27	3	11.11	17	4	23.53
October	12	2	16.67	17	2	11.76	14	3	21.43
November	14	2	14.29	21	1	4.76	18	3	16.67
December	19	3	15.79	18	1	5.56	14	2	14.29
January/2013	17	2	11.76	17	2	11.76	10	1	10.00
February	11	1	9.09	14	1	7.14	16	1	6.25
March	18	2	11.11	18	2	11.11	14	2	14.29
April	15	3	20.00	16	2	12.50	18	3	16.67
May	15	3	20.00	11	1	9.09	19	3	15.79
June	13	2	15.38	21	3	14.29	14	2	14.29
Total	177	28		222	20		194	30	

Site 2 (2011-12)

1. Khulna university pond

J 1		Bellamya			Brotia			Pila	
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
Jul-11	09	3	33	13	1	12.5	23	3	13.04
August	14	5	35.6	15	2	16.66	16	2	12.50
September	11	4	36.3	14	2	18.18	24	4	16.67
October	19	4	21.05	10	1	13.33	17	2	11.76
November	16	2	12.50	05	0	9.09	13	1	7.69
December	15	1	6.66	10	1	7.14	15	2	13.33
Jan-12	18	1	5.5	12	0	5.26	22	3	13.64
February	17	3	17.64	07	0	0	13	1	7.69
March	20	2	10	15	1	11.76	13	2	15.38
April	14	1	7.14	13	1	16.66	21	3	14.29
May	08	0	0	12	2	15.38	27	4	14.81
June	15	3	20	12	0	11.11	17	2	11.76
Total	176	29		138	11	12.5	221	29	

2. Dacope upazila Complex pond

		Bellamyo	ı		Brotie	ı		Pila	
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
Jul-11	18	2	11.11	14	1	6.66	15	2	13.33
August	12	2	16.66	09	1	11	17	1	14.28
September	21	4	19.04	16	2	12.50	19	3	15.78
October	23	3	13.04	11	0	0	21	2	9.5
November	15	1	6.66	08	0	0	08	1	12.5
December	17	2	11.76	13	1	7.69	13	1	7.69
Jan-12	09	1	11.11	12	1	8.33	18	1	5.55
February	13	1	7.69	09	1	11	12	0	0
March	08	1	12.50	11	1	9.09	11	1	9.09
April	14	1	6.66	14	1	7.14	16	1	6.25
May	18	2	11.11	11	1	9.09	09	1	11.11
June	16	2	12.50	05	0	0	13	1	7.69
Total	184	22		133	10		172	15	

3. Syamnagar Upazila Complex pond

		Bellamyo	ı		Broti	а		Pila	
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
Jul-11	18	3	16.66	13	2	15.38	20	3	15
August	19	2	10.52	08	1	12.50	19	4	21.05
September	21	4	19.04	06	1	16.6	15	2	13.33
October	11	2	18.18	13	2	13.33	19	2	10.52
November	13	2	15.38	10	1	10	12	1	8.33
December	15	1	6.66	09	1	11.11	14	2	14.28
Jan-12	19	2	10.52	13	1	7.6	10	1	10
February	12	2	16.6	08	1	12.50	19	2	10.5
March	07	0	0	06	0	0	13	1	7.69
April	11	1	9.09	18	2	11.11	09	1	11.11
May	13	2	13.33	10	1	10	11	1	9.09
June	04	0	0	13	2	7.69	13	1	7.69
Total	163	21		127	15		174	22	

4. Kaliganj Upazila Complex Pond

		Bellamya			Broti	а		Pila	
	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
Jul-11	11	2	18.81	13	2	15.38	21	3	14
August	17	3	17.64	17	2	11.76	16	2	12.50
September	22	5	22.27	22	3	13.63	17	4	23.5
October	20	3	15	16	2	12.50	15	2	13
November	11	2	18	09	1	11	09	1	11
December	17	1	5.8	06	0	0	12	0	0
Jan-12	11	1	9.09	19	1	5.26	15	1	6.66
February	21	3	14.28	12	1	8.33	22	2	9.09
March	16	2	12.50	09	0	0	12	1	8.33
April	17	1	5.88	04	0	0	09	0	0
May	10	1	10	11	1	9.09	18	2	11.11
June	16	1	6.25	15	1	6.66	14	1	7.14
Total	189	26		153	14		180	19	

5. Bagerhat DC Bunglow Pond

		Bellamya	ı		Brotic	а		Pila	
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
Jul-11	22	3	13.63	22	4	18.18	17	2	11.76
August	19	4	21.05	15	2	13.33	22	4	18.18
September	24	5	20.83	16	3	18.75	18	4	22
October	12	2	16.6	14	2	14.28	19	2	10.52
November	09	1	11	06	0	0	13	1	7.6
December	21	2	9.52	04	0	0	15	2	13.33
Jan-12	15	1	6.66	14	1	7.14	12	0	0
February	15	2	13.33	09	1	11	16	2	12.5
March	10	1	10	15	1	6.66	11	1	9.09
April	18	1	5.55	09	1	11.11	12	0	0
May	13	1	7.69	12	0	0	16	2	12.5
June	12	1	8.33	19	2	10.52	11	1	9.09
Total	190	24		155	17		182	21	

6. Fakirhat Bazar pond

		Bellamyo	ı		Broti	а		Pila	
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
Jul-11	09	3	33	13	2	15.38	22	4	18
August	14	5	35.6	16	3	18.75	13	3	23
September	11	4	36.3	06	1	16.6	12	4	33
October	19	4	21.05	11	1	9	06	1	16.6
November	16	2	12.50	19	1	5.26	16	1	6.25
December	15	1	6.66	12	0	0	15	2	13.33
Jan-12	18	1	5.5	17	0	0	18	0	0
February	17	3	17.64	16	2	12.50	14	2	14.28
March	20	2	10	15	1	6.66	16	2	12.50
April	14	1	7.14	15	2	13.33	10	1	10
May	08	0	0	13	2	15	11	0	0
June	15	3	20	18	2	11.11	06	1	16.6
Total	176	29		171	17		159	21	

Site 2 (2012-13)

1. Khulna university pond

		Bellamyo	ı		Brotie	ı		Pila	
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
Jul-12	16	3	19	13	1	7.5	38	5	13.16
August	22	4	18.18	15	2	13	26	3	11.54
September	22	5	22.72	12	2	16.66	21	4	19.05
October	23	3	13.04	10	1	10	20	3	15.00
November	15	2	13.33	05	0	0	12	1	8.33
December	15	1	6.66	10	1	10	16	2	12.50
Jan-13	14	1	7.14	12	1	8.33	15	2	13.33
February	18	2	11.11	11	1	9.09	15	1	6.67
March	18	1	5.55	14	1	7.14	23	2	8.70
April	11	0	0	13	1	7.5	25	3	12.00
May	21	2	9.5	12	2	16	28	4	14.29
June	14	1	7.14	12	0	0	22	3	13.64
Total	209	25		139	13		261	33	

2. Dacope upazila Complex pond

		Bellamya	!		Brotia			Pila	
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
Jul-12	17	2	11.76	22	3	13.63	22	4	18.18
August	22	4	18.18	19	4	21.05	15	2	13.33
September	18	4	22	24	5	20.83	16	3	18.75
October	19	2	10.52	12	2	16.6	14	2	14.28
November	13	1	7.6	09	1	11	06	0	0
December	15	2	13.33	21	2	9.52	04	0	0
Jan-13	12	0	0	15	1	6.66	14	1	7.14
February	16	2	12.5	15	2	13.33	09	1	11
March	11	1	9.09	10	1	10	15	1	6.66
April	12	0	0	18	1	5.55	09	1	11.11
May	16	2	12.5	13	1	7.69	12	0	0
June	11	1	9.09	12	1	8.33	19	2	10.52
Total	182	21		190	24		155	17	

3. Syamnagar Upazila Complex pond

		Bellamyo	ı		Brotic	а		Pila	
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
Jul-12	17	4	23.52	09	1	11.11	11	3	27.27
August	11	3	27.27	22	4	18.18	16	5	31.25
September	13	4	30.76	18	3	16.66	13	3	23.07
October	14	4	28.57	13	1	7.69	19	2	10.52
November	09	1	11.11	11	0	0	11	1	9.09
December	16	2	12.5	09	1	11.11	09	0	0
Jan-13	05	0	0	12	1	8.33	14	2	14.28
February	12	2	16.66	15	2	13.33	18	3	16.66
March	14	2	14.28	17	2	11.76	16	2	12.5
April	15	2	13.33	10	1	10	12	1	8.33
May	16	3	18.75	09	1	11.11	15	2	13.33
June	19	4	21.05	08	1	12.5	06	1	16.66
Total	161	31		153	18		160	25	

221

4. Kaliganj Upazila Complex Pond

		Bellamya			Brotie	a		Pila	
	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
Jul-12	19	1	5.26	33	3	9.09	29	3	10.34
August	25	2	8.00	30	3	10.00	23	3	13.04
September	26	3	11.54	34	4	11.76	35	6	17.14
October	29	3	10.34	34	3	8.82	31	3	9.68
November	13	0	0.00	30	1	3.33	27	1	3.70
December	25	1	4.00	32	2	6.25	29	3	10.34
Jan-13	23	2	8.70	23	1	4.35	33	2	6.06
February	10	0	0.00	26	1	3.85	23	1	4.35
March	18	2	11.11	26	2	7.69	32	2	6.25
April	21	3	14.29	28	3	10.71	30	3	10.00
May	16	2	12.50	30	3	10.00	36	5	13.89
June	18	2	11.11	27	2	7.41	31	4	12.90
Total	243	21	8.64	353	28		359	36	

5. Bagerhat DC Bunglow Pond

		Bellamyo	ı		Brotio	ı		Pila	
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
Jul-12	31	5	16.13	33	3	9.09	35	6	17.14
August	32	6	18.75	35	6	17.14	30	5	16.67
September	30	9	30.00	33	6	18.18	33	7	21.21
October	33	8	24.24	29	4	13.79	28	4	14.29
November	28	2	7.14	29	2	6.90	30	2	6.67
December	29	1	3.45	33	3	9.09	27	3	11.11
Jan-13	34	4	11.76	28	4	14.29	33	3	9.09
February	30	2	6.67	30	1	3.33	30	1	3.33
March	33	5	15.15	29	3	10.34	29	4	13.79
April	32	6	18.75	33	4	12.12	33	4	12.12
May	34	6	17.65	34	5	14.71	34	5	14.71
June	31	6	19.35	25	3	12.00	30	5	16.67
Total	387	60		381	44		382	49	

6. Fakirhat bazar pond

		Bellamyo	ı		Brotie	a		Pila	
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
Jul-12	25	4	16.00	19	1	5.26	25	4	16.00
August	30	5	16.67	25	2	8.00	30	5	16.67
September	36	7	19.44	26	3	11.54	36	7	19.44
October	23	3	13.04	29	3	10.34	23	3	13.04
November	16	2	12.50	13	0	0.00	16	2	12.50
December	23	3	13.04	25	1	4.00	23	3	13.04
Jan-13	27	3	11.11	23	2	8.70	27	3	11.11
February	13	1	7.69	10	0	0.00	13	1	7.69
March	28	3	10.71	18	2	11.11	28	3	10.71
April	26	3	11.54	21	3	14.29	26	3	11.54
May	30	3	10.00	16	2	12.50	30	3	10.00
June	28	4	14.29	18	2	11.11	28	4	14.29
Total	305	41	13.44	243	21	8.64	305	41	13.44

Site 3 (2011-12)

1. Solakia, Kishoreganj Bazar Pond

		Bellamyo	ı		Brotic	а		Pila	
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
Jul-11	17	4	23.52	09	1	11.11	11	3	27.27
August	11	3	27.27	22	4	18.18	16	5	31.25
September	13	4	30.76	18	3	16.66	13	3	23.07
October	14	4	28.57	13	1	7.69	19	2	10.52
November	09	1	11.11	11	0	0	11	1	9.09
December	16	2	12.5	09	1	11.11	09	0	0
Jan-12	05	0	0	12	1	8.33	14	2	14.28
February	12	2	16.66	15	2	13.33	18	3	16.66
March	14	2	14.28	17	2	11.76	16	2	12.5
April	15	2	13.33	10	1	10	12	1	8.33
May	16	3	18.75	09	1	11.11	15	2	13.33
June	19	4	21.05	08	1	12.5	06	1	16.66
Total	161	31		153	18		160	25	

2. Astagram Upazila Complex pond

		Bellamyo	ı		Brotio	a		Pila	
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
Jul-11	24	6	25	08	1	12.5	18	3	16.6
August	19	5	26.31	06	1	16.66	15	3	20
September	17	4	23.52	11	2	18.18	20	3	15
October	16	2	12.5	15	2	13.33	09	1	11.11
November	11	1	9.09	11	1	9.09	15	1	6.66
December	18	2	11.11	14	1	7.14	16	0	0
Jan-12	21	1	4.76	19	1	5.26	14	1	7.14
February	12	0	0	09	0	0	12	1	9.09
March	15	2	13.33	17	2	11.76	18	2	11.11
April	19	2	10.52	12	2	16.66	07	1	14.28
May	25	3	12	13	2	15.38	10	1	10
June	06	1	16.66	09	1	11.11	16	2	12.5
Total	203	29		144	16		170	19	

3. Kuliarchar Bazar, Kishoreganj pond

		Bellamyo	ı		Broti	а		Pila	
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
Jul-11	19	2	10.52	18	2	11.11	16	3	18.75
August	22	6	27.27	15	2	13.33	18	3	16.66
September	20	4	20	11	2	18.18	17	2	11.76
October	11	2	18.18	12	1	8.33	08	1	12.5
November	14	1	7.14	08	0	0	11	1	9.09
December	08	1	12.5	12	0	0	13	0	0
Jan-12	12	1	8.33	09	1	11.11	12	1	8.33
February	15	2	13.33	11	1	9.09	11	2	18.18
March	13	1	7.69	11	0	0	13	2	15.38
April	12	1	8.33	09	1	11.11	10	1	10
May	16	2	12.5	07	1	14.28	08	1	12.5
June	16	3	18.75	10	1	10	11	2	18.18
Total	178	26		133	12		148	19	

4. Mithamon, Kishoreganj Bazar pond

		Bellamy	а		Brot	ia		Pila	
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
Jul-11	17	4	23.52	10	2	20	14	2	14.28
August	12	3	25	11	2	18.18	19	3	15.78
September	16	6	37.5	13	1	7.69	21	5	23.8
October	13	2	15.38	11	1	9.09	18	1	5.55
November	11	1	9.09	14	0	0	10	1	10
December	13	1	7.69	07	0	0	15	1	6.66
Jan-12	15	2	13.33	11	1	9.09	12	2	16.66
February	16	3	18.75	13	1	7.69	16	2	12.5
March	12	2	16.6	11	1	9.09	11	2	18.18
April	10	1	10	06	0	0	15	2	13.33
May	12	2	16.6	14	1	7.14	11	1	9.09
June	17	2	11.76	10	1	10	13	1	7.69
Total	164	29		131	11		175	23	

5. BAU, Mymensingh Pond

		Bellamy	ı		Broti	а		Pila	
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
Jul-11	25	5	20	09	1	11.11	14	4	14.28
August	23	4	17.39	10	1	10	11	2	18.18
September	16	4	25	08	1	12.5	13	3	23.07
October	12	2	16.6	10	0	0	12	2	16.66
November	09	1	11.11	12		0	09	1	11.11
December	12	1	8.33	09	1	11.11	15	1	6.66
Jan-12	11	1	9.09	08	0	0	21	2	9.52
February	07	1	14.28	12	1	8.33	10	1	10
March	08	1	12.5	15	2	13.33	13	1	7.69
April	11	0	0	10	1	10	09	1	11.11
May	153	2	13.33	07	0	0	13	2	15.38
June	11	1	9.09	12	1	8.33	10	1	10
Total	298	23		122	09		150	21	

6. Trishal Kabi Nazrul University Pond

		Bellamy	a		Broti	a		Pila	
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
Jul-11	24	3	12.5	10	1	10	18	3	16.6
August	17	3	17.64	16	1	6.25	17	4	23.52
September	19	3	15.78	12	2	16.6	15	3	20
October	16	2	12.5	15	2	13.33	09	1	11.11
November	11	1	9.09	07	1	14.28	11	2	18.18
December	19	1	5.26	11	1	9.09	16	0	0
Jan-12	14	1	7.14	18	2	11.11	23	2	8.69
February	10	1	10	19	2	10.52	17	3	17.64
March	13	2	15.38	08	1	12.5	18	2	11.11
April	09	0	0	10	1	10	07	1	14.28
May	18	2	11.11	12	2	16.66	18	3	16.66
June	15	1	6.66	09	1	11.11	10	1	10
Total	185	20		147	17		179	25	

Site 3 (2012-13)

1. Solakia Bazar Pond, Kishoreganj

		Bellamyo	ı		Brotie	а		Pila	
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
Jul-12	22	3	13.64	19	2	10.53	22	3	13.64
August	19	2	10.53	17	3	17.65	19	2	10.53
September	22	4	18.18	21	4	19.05	22	4	18.18
October	14	2	14.29	18	2	11.11	14	2	14.29
November	15	2	13.33	17	2	11.76	15	2	13.33
December	21	3	14.29	15	2	13.33	21	3	14.29
Jan-13	16	2	12.50	16	2	12.50	16	2	12.50
February	15	1	6.67	13	1	7.69	15	1	6.67
March	20	2	10.00	18	2	11.11	20	2	10.00
April	21	4	19.05	22	3	13.64	21	4	19.05
May	18	3	16.67	20	3	15.00	18	3	16.67
June	17	2	11.76	13	2	15.38	17	2	11.76
Total	220	30	13.64	209	28		220	30	13.64

2. Astagram Upazila Complex pond

		Bellamyo	ı		Brotie	a		Pila	
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
Jul-12	09	3	33	13	2	15.38	22	4	18
August	14	5	35.6	16	3	18.75	13	3	23
September	11	4	36.3	06	1	16.6	12	4	33
October	19	4	21.05	11	1	9	06	1	16.6
November	16	2	12.50	19	1	5.26	16	1	6.25
December	15	1	6.66	12	0	0	15	2	13.33
Jan-13	18	1	5.5	17	0	0	18	0	0
February	17	3	17.64	16	2	12.50	14	2	14.28
March	20	2	10	15	1	6.66	16	2	12.50
April	14	1	7.14	15	2	13.33	10	1	10
May	08	0	0	13	2	15	11	0	0
June	15	3	20	18	2	11.11	06	1	16.6

3. Kuliarchar Bazar pond

		Bellamyo	ı		Brotic	а		Pila	
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
Jul-12	31	5	16.13	21	2	9.52	38	5	13.16
August	35	7	20.00	18	3	16.67	26	3	11.54
September	29	8	27.59	26	5	19.23	21	4	19.05
October	22	4	18.18	22	3	13.64	20	3	15.00
November	16	2	12.50	17	1	5.88	12	1	8.33
December	25	3	12.00	16	2	12.50	16	2	12.50
Jan-13	14	2	14.29	22	3	13.64	15	2	13.33
February	15	1	6.67	24	1	4.17	15	1	6.67
March	30	4	13.33	27	2	7.41	23	2	8.70
April	28	4	14.29	23	3	13.04	25	3	12.00
May	31	5	16.13	28	3	10.71	28	4	14.29
June	28	5	17.86	21	2	9.52	22	3	13.64
Total	304	50		265	30		261	33	

4. Mithamain Bazar pond

		Bellamya	ı		Brotic	a		Pila	
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
Jul-12	21	3	14.29	29	5	17.24	31	4	12.90
August	25	4	16.00	31	6	19.35	36	5	13.89
September	31	6	19.35	33	8	24.24	29	5	17.24
October	35	6	17.14	25	4	16.00	25	3	12.00
November	28	3	10.71	26	3	11.54	21	2	9.52
December	25	3	12.00	21	3	14.29	17	2	11.76
Jan-13	22	4	18.18	19	2	10.53	12	2	16.67
February	20	1	5.00	13	1	7.69	11	1	9.09
March	17	3	17.65	17	2	11.76	17	2	11.76
April	13	2	15.38	22	3	13.64	28	3	10.71
May	21	3	14.29	29	4	13.79	32	4	12.50
June	26	3	11.54	31	5	16.13	26	2	7.69
Total	284	41		296	46		285	35	

5. BAU, Mymensingh Pond

		Bellamyo	ı		Brotic	а		Pila	
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
Jul-12	20	3	15	13	1	7.5	18	2	11
August	22	4	18.18	15	2	13	16	3	19
September	22	5	22.72	14	2	15	19	3	16
October	15	3	20	10	1	10	15	2	13
November	15	2	13.33	05	0	0	12	1	8
December	15	1	6.66	10	1	10	09	0	0
Jan-13	14	1	7.14	12	0	0	11	1	9
February	18	2	11.11	07	0	0	13	0	0
March	18	1	5.55	15	1	6.66	17	2	12
April	23	2	8.69	13	1	7.5	08	0	0
May	21	2	9.5	12	2	16	10	1	10
June	17	2	11.76	12	0	0	15	1	6
Total	220	28		138	11		163	16	

6. Trishal Kabi Nazrul University Pond

		Bellamyo	ı		Brotie	ı		Pila	
Month	Tot	Inf	%	Tot	Inf	%	Tot	Inf	%
Jul-12	19	3	15.79	26	3	11.54	33	4	12.12
August	21	5	23.81	24	3	12.50	30	3	10.00
September	27	7	25.93	27	4	14.81	32	4	12.50
October	20	4	20.00	22	3	13.64	29	4	13.79
November	14	2	14.29	16	1	6.25	22	1	4.55
December	17	3	17.65	22	2	9.09	25	2	8.00
Jan-13	19	2	10.53	28	2	7.14	32	3	9.38
February	14	1	7.14	21	1	4.76	27	1	3.70
March	21	2	9.52	21	2	9.52	32	2	6.25
April	24	4	16.67	27	3	11.11	28	2	7.14
May	21	4	19.05	30	4	13.33	30	3	10.00
June	26	4	15.38	28	3	10.71	31	2	6.45
Total	243	41		292	31		351	31	

Appendix B: Yearly percentage of occurrence

Site 1: Dhaka region

Ramna	%occurrance 11-12	%occurrance 12-13
Bellamva	14.27	15.36
Brotia	13.00	13.03
Pila	15.00	14.72
Curjon	%occurance 11-12	%occurance 12-13
Bellamya	12.39	15.75
Brotia	8.66	11.33
Pila	10.37	12.35
JH	%occurance 11-12	%occurance 12-13
Bellamya	15.75	15,0000
Brotia	11.75	12.9500
Pila	13.07	13.7400
Keraniganj	%occurance 11-12	%occurance 12-13
Bellamya	11.74	14.6800
Brotia	7.78	12.1400
Pila	9.81	14.3000
Tongi railway	%occurance 11-12	%occurance 12-13
Bellamya	14.87	14.31
Brotia	8.07	12.07
Pila	13.00	13.29
Joydevpur	%occurance 11-12	%occurance 12-13
Bellamya	13.08	16.31
Brotia	9.29	12.71
Pila	12.28	13.33
Bhawal	%occurance 11-12	%occurance 12-13
Bellamya	12.46	15.65
Brotia	11.25	13.87
Pila	12.78	15.55
Salna BKB	%occurance 11-12	%occurance 12-13
Bellamya	12.52	13.00
Brotia	13.22	14.91
Pila	13.41	13.96
Mawna	%occurance 11-12	%occurance 12-13
Bellamya	13.09	16.31
Brotia	9.29	12.71
Pila	12.28	13.33
Tongi Bari	%occurance 11-12	%occurance 12-13
Bellamya	14.87	14.31
Brotia	8.07	12.07
Pila	13.00	13.29
Lohajang	%occurance 11-12	%occurance 12-13
Bellamya	16.30	15.82
Brotia	12.08	9.07
Pila	12.83	15.27

Site 2: Khulna region

Khulna	%occurrence 11-12	%occurrence 12-13
Bellamya	17.115	11.114
Brotia	11.422	8.768
Pila	12.713	12.350
Dacope	%occurrence 11-12	%occurrence 12-13
Bellamya	11.653	10.547
Brotia	6.875	12.015
Pila	9.397	9.247
Syamnagar	%occurrence 11-12	%occurrence 12-13
Bellamya	11.331	18.15
Brotia	10.651	10.981
Pila	11.549	15.246
Kaliganj	%occurrence 11-12	%occurrence 12-13
Bellamya	12.964	8.07
Brotia	7.80	7.771
Pila	9.69	9.807
Bagerhat	%occurrence 11-12	%occurrence 12-13
Bellamya	12.015	15.753
Brotia	9.247	11.748
Pila	10.547	13.066
Fakirhat	%occurrence 11-12	%occurrence 12-13
Bellamya	17.115	13.002
Brotia	10.299	8.07
Pila	13.63	13.002

Site 3: Kishoreganj region

Solakia	%occurrence 11-12	%occurrence 12-13
Bellamya	18.15	13.409
Brotia	10.981	13.299
Pila	15.246	13.409
Austagram	%occurrence 11-12	%occurrence 12-13
Bellamya	13.733	17.115
Brotia	11.422	10.299
Pila	11.124	13.63
Kuliarchar	%occurrence 11-12	%occurrence 12-13
Bellamya	13.711	15.745
Brotia	8.878	11.327
Pila	12.61	12.35
Mitamon	%occurrence 11-12	%occurrence 12-13
Bellamya	17.101	14.294
Brotia	8.164	14.683
Pila	12.793	12.144
BAU	%occurrence 11-12	%occurrence 12-13
Bellamya	13.06	12.47
Brotia	7.059	7.138
Pila	12.805	8.666
Trisal	%occurrence 11-12	%occurrence 12-13
Bellamya	10.255	16.313
Brotia	11.787	10.366
Pila	13.982	8.656

Appendix C: List of prevalence of cercaria on different host.

P- Parapleurophocercous, X- Xiphidiocercariae, F- Furcocercous , E- Echinostome

Sampling area	Pila globosa(2011-12)											
	Sun	nmer			Rainy				Winte	er		
	P	X	F	Е	P	X	F	Е	P	X	F	E
Ramna Park Pond	4	-	-	2	9	5	4	4	3	1	1	2
Curjon Hall Pond	-	2	2	3	4	6	4	4	1	-	2	3
Jagannath Hall Pond	4	3	1	3	11	9	5	7	-	4	1	3
Keranigonj Pond	2	-	-	3	8	5	3	5	2	2	3	3
Tongi Railway Station Pond	3	-	2	2	6	9	-	5	-	3	6	5
Jaydebpur Railway Station Pond	4	1	-	2	8	5	3	6	-	4	1	3
Bhawal-Gazipur Railway Station Pond	-	3	2	-	5	3	3	4	2	-	2	-
Salna BSMAU Pond	-	-	3	3	6	3	2	5	2	3	-	3
Maona Bazar Pond	3	-	2	5	4	-	6	3	5	1	4	-
Tongi Bari Bazar Pond	-	3	2	3	1	9	3	5	-	4	3	-
Lahajanj Bazar Pond	2	-	3	2	4	-	6	3	4	3	2	2
Khulna university pond	2	-	1	1	3	4	-	1	2	-	-	2
Dacope Upazila Complex pond	1	-	2	-	3	4	2	1	1	-	-	1
Shyamnagar Upazila complex pond	2	1	-	1	1	5	3	3	2	-	2	2
Kaliganj Upazila Complex pond	1	2	1	-	4	3	3	1	1	2	-	1
Bagerhat P.C College Pond	1	-	2	1	3	4	3	1	2	1	2	1
Fakirhat Bazar Pond Bagerhat	-	2	2	1	4	4	3	2	-	1	-	2
Solakia Kishorganj Pond	2	2	1	1	5	7	-	1	2	-	2	2
Astagram Upazila Complex pond	1	-	2	-	4	5	2	2	1	-	-	2
Kuliarchar Kishorganj	-	1	2	1	3	4	1	1	2	-	2	2
Mithamain Upazila Complex Pond	3	2	1	-	4	5	3	2	1	2	-	-
BAU Mymensingh Pond	1	2	2	3	3	4	-	2	1	-	2	1
Kazi Nazrul Islam Univeristy Pond	-	3	2	-	5	6	2	2	1	1	2	1

P-Parapleurophocercous , E-Echinostome, F-Furcocercous, X- Xiphidiocercous

Sampling area	Pila globosa(2012-13)											
	Sum	mer			Rainy	,			Winte	er		
	P	X	F	Е	P	X	F	E	P	X	F	Е
Ramna Park Pond	4	-	2	2	4	6	4	5	-	2	-	3
Curjon Hall Pond	5	3	-	3	9	6	4	6	3	-	4	1
Jagannath Hall Pond	4	3	-	-	7	-	9	5	4	-	5	3
Keranigonj Pond	6	3	2	-	9	4	-	6	1	4	5	1
Tongi Railway Station Pond	5	5	-	7	8	6	4	3	4	2	-	3
Jaydebpur Railway Station Pond	3	4	2	3	-	5	3	-	5	6	3	-
Bhawal-Gazipur Railway Station Pond	4	-	3	5	6	4	2	7	-	-	3	1
Salna BSMAU Pond	-	5	4	5	7	-	4	6	2	-	1	4
Maona Bazar Pond	3	3	5	-	4	3	6	6	3	1	-	-
Tongi Bari Bazar Pond	1	5	2	1	5	3	8	3	-	2	3	-
Lahajanj Bazar Pond	-	1	1	4	3	-	5	7	2	-	5	2
Khulna university pond	1	-	-	2	2	5	2	1	2	-	1	-
Dacope Upazila Complex pond	1	1	2	-	3	3	2	1	2	-	1	1
Shyamnagar Upazila complex pond	2	3	1	2	2	4	2	3	3	-	2	1
Kaliganj Upazila Complex pond	3	4	2	2	7	4	4	2	2	1	3	2
Bagerhat P.C College Pond	3	3	4	5	7	9	4	3	3	3	4	1
Fakirhat Bazar Pond Bagerhat	2	3	4	1	6	7	3	4	2	4	3	2
Solakia Kishorganj Pond	2	2	3	2	4	6	3	2	2	2	-	2
-Astagram Upazila Complex pond	1	2	-	2	3	3	2	3	2	1	1	1
Kuliarchar Kishorganj	3	4	-	3	6	8	3	3	-	2	-	1
Mithamain Upazila Complex Pond	3	5	-	2	6	5	2	3	3	3	1	2
BAU Mymensingh Pond	3	5	-	2	3	4	2	1	5	2	2	-
Kazi Nazrul Islam Univeristy Pond	3	2	2	2	7	4	4	3	2	-	2	-

 $Brotia = \ P-Parapleurophocercous, \ X-\ Xiphidiocercariae, F-Furcocercous, \ R-Renicolid$

Sampling area					Bro	tia cos	tula(20	012-13)			
	Sur	nmer			Rain	y			Wint	er		
	P	X	F	R	P	X	F	R	P	X	F	R
Ramna Park Pond	2	-	4	1	3	2	4	3	2	-	2	2
Curjon Hall Pond	-	-	1	1	-	4	6	4	-	3	2	T -
Jagannath Hall Pond	-	3	2	2	4	6	2	3	2	1	-	-
Keranigonj Pond	2	1	1	-	1	5	7	9	-	4	2	3
Tongi Railway Station Pond	3	-	2	3	2	3	6	4	2	-	3	2
Jaydebpur Railway Station Pond	2	-	5	2	5	4	4	-	1	3	2	1
Bhawal-Gazipur Railway Station Pond	2	4	-	3	5	3	6	3	4	-	-	1
Salna BSMAU Pond	4	-	3	2	-	4	5	4	2	1	3	2
Maona Bazar Pond	2	-	5	3	4	3	5	3	-	-	3	2
Tongi Bari Bazar Pond	2	3	3	2	5	3	2	2	-	3	1	2
Lahajanj Bazar Pond	-	2	-	-	1	2	4	3	2	-	3	3
Khulna university pond	-	-	2	2	3	2	1	1	-	1	1	-
Dacope Upazila Complex pond	3	3	2	1	3	5	2	1	2	1	-	1
Shyamnagar Upazila complex pond	2	-	1	2	3	6	2	1	-	1	-	-
Kaliganj Upazila Complex pond	2	3	-	3	6	7	2	2	1	-	2	-
Bagerhat P.C College Pond	3	4	3	3	6	8	5	4	3	2	3	-
Fakirhat Bazar Pond Bagerhat	2	1	-	3	4	3	2	3	2	-	1	-
Solakia Kishorganj Pond	2	3	-	3	5	4	3	3	2	2	-	1
Astagram Upazila Complex pond	1	2	-	1	3	4	2	1	1	2	-	-
-Kuliarchar Kishorganj	3	2	3	1	5	2	4	3	2	1	2	2
Mithamain Upazila Complex Pond	4	5	2	3	8	9	4	3	3	3	-	2
BAU Mymensingh Pond	2	1	1	2	4	5	3	2	1	-	-	-
Kazi Nazrul Islam Univeristy Pond	3	1	-	5	6	3	3	4	2	3	-	1

Seasonal prevalence of trematode cercaria in Brotia costula collected from different areas during the study period.

Sampling area	Brotia costula (2011-12)											
	Sun	nmer			Rain	ıy			Wint	er		
	P	X	F	R	P	X	F	R	P	X	F	R
Ramna Park Pond	2	3	2	1	4	3	4	4	2	-	3	1
Curjon Hall Pond	3	-	2	5	-	4	3	-	2	-	-	2
Jagannath Hall Pond	-	5	4	3	3	4	5	6	-	6	6	3
Keranigonj Pond	2	3	-	5	3	5	-	6	-	2	1	1
Tongi Railway Station Pond	1	3	-	3	2	-	4	3	2	2	1	-
Jaydebpur Railway Station Pond	2	2	1	4	5	3	-	2	4	1	-	1
Bhawal-Gazipur Railway Station Pond	3	4	-	2	4	2	3	3	1	3	-	3
Salna BSMAU Pond	2	2	1	1	5	3	3	4	3	2	1	1
Maona Bazar Pond	1	-	3	2	4	2	4	4	-	1	4	3
Tongi Bari Bazar Pond	1	3	-	4	3	-	5	4	3	3	-	2
Lahajanj Bazar Pond	-	2	4	2	6	2	1	4	-	2	1	4
Khulna university pond	1	-	-	2	3	2	1	-	-	1	-	1
Dacope Upazila Complex pond	1	-	-	1	2	-	2	1	2	-	-	1
Shyamnagar Upazila complex pond	2	2	1	-	2	4	2	1	1	-	-	-
Kaliganj Upazila Complex pond	-	2	-	-	2	4	3	1	1	-	1	-
Bagerhat P.C College Pond	2	2	-	1	4	4	2	1	-	-	1	-
Fakirhat Bazar Pond Bagerhat	3	2	1	-	3	1	1	1	2	2	-	1
Solakia Kishorganj Pond	2	1	2	-	4	3	2	2	1	-	1	-
Astagram Upazila Complex pond	1	1	2	1	3	4	-	1	-	1	1	1
Kuliarchar Kishorganj	1	2	-	-	3	2	2	1	-	-	1	-
Mithamain Upazila Complex Pond	1	2	-	1	2	1	-	1	1		1	-
BAU Mymensingh Pond	-	-	1	2	3	2	1	-	-	-	-	-
Kazi Nazrul Islam Univeristy Pond	2	2	1	-	3	4	1	1	2	-	1	-

 $Seasonal\ prevalence\ of\ trematode\ cercaria\ in\ \textit{Bellamya}\ bengalens is\ collected\ from\ different\ areas\ during\ the\ study\ period.$

Bellamya: P-Pleurophocercous, X-Xiphidiocercariae, A-Amphistome, C- Cotylomicrocercous

Sampling area				1	Bellam	ya beng	alensi	s(2011	'-12)			
	Sur	nmer			Rai	ny			Wint	er		
	P	X	A	С	P	X	Α	С	P	X	Α	C
Ramna Park Pond	3	5	4	4	3	6	5	2	1	4	3	1
Curjon Hall Pond	2	4	3	2	6	8	5	5	2	6	4	2
Jagannath Hall Pond	6	7	-	5	8	11	6	4	5	6	5	-
Keranigonj Pond	-	5	3	4	3	6	5	3	3	6	1	-
Tongi Railway Station Pond	4	6	4	-	5	9	3	5	2	6	-	3
Jaydebpur Railway Station Pond	2	4	2	-	3	6	2	-	3	5	3	4
Bhawal-Gazipur Railway Station Pond	-	4	3	3	6	7	3	2	4	4	1	-
Salna BSMAU Pond	1	4	-	6	3	7	3	2	2	4	2	-
Maona Bazar Pond	2	6	3	2	3	7	2	1	2	5	-	2
Tongi Bari Bazar Pond	-	3	4	5	3	9	3	-	4	2	4	-
Lahajanj Bazar Pond	2	-	5	4	6	4	2	3	-	3	-	4
Khulna university pond	3	2	4	-	5	4	2	3	2	1	2	-
Dacope Upazila Complex pond	3	2	2	1	4	5	2	1	-	-	1	1
Shyamnagar Upazila complex pond	2	2	1	2	4	3	4	2	-	-	-	1
Kaliganj Upazila Complex pond	2	3	3	1	4	3	5	2	-	2	-	1
Bagerhat P.C College Pond	-	3	2	3	4	3	2	1	3	-	1	2
Fakirhat Bazar Pond Bagerhat	2	-	2	2	6	5	4	3	2	-	2	1
Solakia Kishorganj Pond	3	2	2	-	5	7	3	4	2	1	-	2
Astagram Upazila Complex pond	1	2	-	3	4	6	5	3	2	1	1	1
Kuliarchar Kishorganj	2	3	2		5	4	5	2	1	-	2	-
Mithamain Upazila Complex Pond	3	-	2	1	5	8	4	3	-	2	1	-
BAU Mymensingh Pond	1	2	-	1	3	5	3	2	3	-	2	1
Kazi Nazrul Islam Univeristy Pond	2	1	2	-	-	5	4	3	2	-	1	-

Seasonal prevalence of trematode cercaria in Bellamya bengalensis collected from different areas during the study period.

Sampling area	Bellamya bengalensis(2012-13)											
	Sum	mer			Rain	y			Winte	er		
	P	X	Α	C	P	X	Α	С	P	X	Α	C
Ramna Park Pond	2	4	3	3	2	6	3	2	2	3	4	2
Curjon Hall Pond	4	6	2	4	3	7	4	6	3	5	3	3-
Jagannath Hall Pond	-	3	5	2	5	6	5	-	4	6	3	3
Keranigonj Pond	4	-	4	3	2	7	3	-	4	4	5	5
Tongi Railway Station Pond	2	6	3	5	5	3	4	2	-	6	2	1
Jaydebpur Railway Station Pond	1	4	3	5	-	6	3	3	6	4	-	3
Bhawal-Gazipur Railway Station Pond	4	3	3	6	4	3	5	5	4	-	2	1
Salna BSMAU Pond	-	3	-	5	6	8	4	3	5	2	-	4
Maona Bazar Pond	3	-	6	4	3	9	-	1	4	6	3	1
Tongi Bari Bazar Pond	-	4	-	3	5	7	4	2	5	-	2	2
Lahajanj Bazar Pond	2	4	7	2	-	3	4	1	-	3	1	1
Khulna university pond	3	2	1	-	4	4	3	3	2	1	-	2
Dacope Upazila Complex pond	1	2	1	-	4	3	1	2	3	2	1	1
Shyamnagar Upazila complex pond	3	4	2	2	5	4	4	2	1	-	3	1
Kaliganj Upazila Complex pond	2	-	2	3	5	3	3	2	-	-	1	-
Bagerhat P.C College Pond	3	4	5	4	7	9	6	9	4	4	3	2
Fakirhat Bazar Pond Bagerhat	4	3	2	3	6	8	4	5	2	2	-	2
Solakia Kishorganj Pond	2	-	3	2	5	4	4	-	5	3	-	2
Astagram Upazila Complex pond	3	2	-	2	6	5	5	3	2	-	-	1
Kuliarchar Kishorganj	5	1	3	5	8	7	6	4	5	2	1	3
Mithamain Upazila Complex Pond	4	3	1	2	6	7	7	3	4	1	1	2
BAU Mymensingh Pond	2	-	3	2	6	5	3	3	5	3	2	-
Kazi Nazrul Islam Univeristy Pond	4	4	2	3	4	6	6	4	3	3	1	1

Appendix D

 $Appendix\ 1: Mean\ \pm SEM\ of\ seasonal\ variation\ of\ prevalence\ of\ trematode\ parasites\ in\ \textit{Bellamya}\ sampled\ from\ Khulna\ regions\ in\ 2011\ -2012$

				KU	Dacope	Symnagar	Kaligong	BagerhatDc	Fakirhat
Season	Rainy	1		33.00	11.11	16.66	18.81	13.63	33.00
		2		35.60	16.66	10.52	17.64	21.05	35.60
		3		36.30	19.04	19.04	22.27	20.83	36.30
		4		21.05	13.04	18.18	15.00	16.60	21.05
		Total	N	4	4	4	4	4	4
			Mean	31.4875	14.9625	16.1000	18.4300	18.0275	31.4875
			SEM	3.55084	1.78057	1.92397	1.50773	1.78805	3.55084
	Winter	1		12.50	6.66	15.38	18.00	11.00	12.50
		2		6.66	11.76	6.66	5.80	9.52	6.66
		3		5.50	11.11	10.52	9.09	6.66	5.50
		4		17.64	7.69	16.60	14.28	13.33	17.64
		Total	N	4	4	4	4	4	4
			Mean	10.5750	9.3050	12.2900	11.7925	10.1275	10.5750
			SEM	2.80925	1.25463	2.29045	2.70700	1.39672	2.80925
	Summer	1		10.00	12.50	.00	12.50	10.00	10.00
		2		7.14	6.66	9.09	5.88	5.55	7.14
		3		.00	11.11	13.33	10.00	7.69	.00
		4		20.00	12.50	.00	6.25	8.33	20.00
		Total	N	4	4	4	4	4	4
			Mean	9.2850	10.6925	5.6050	8.6575	7.8925	9.2850
		SEM		4.14462	1.38352	3.34979	1.58318	.92020	4.14462
	Total	N		12	12	12	12	12	12
		Mean		17.1158	11.6533	11.3317	12.9600	12.0158	17.1158
		SEM		3.58314	1.06408	1.88259	1.61544	1.50446	3.58314

Appendix 2: Mean ±SEM of seasonal variation of prevalence of trematode parasites in *Bellamya* sampled from Kishoreganj regions in 2011 - 2012

				Solakia	Astagram	Mithamain	BAU	Trisal
		1		23.52	25.00	23.52	20.00	12.50
		2		27.27	26.31	25.00	17.39	17.64
		3		30.76	23.52	37.50	25.00	15.78
	Rainy	4		28.57	12.50	15.38	16.60	12.50
			N	4	4	4	4	4
		Total	Mean	27.5300	21.8325	25.3500	19.7475	14.6050
			SEM	1.51828	3.16260	4.56887	1.89554	1.27325
		1		11.11	9.09	9.09	11.11	9.09
		2		12.50	11.11	7.69	8.33	5.26
		3		.00	4.76	13.33	9.09	7.14
	Winter	4		16.66	.00	18.75	14.28	10.00
G			N	4	4	4	4	4
Season		Total	Mean	10.0675	6.2400	12.2150	10.7025	7.8725
			SEM	3.55691	2.46589	2.48648	1.32896	1.05555
		1		14.28	13.33	16.60	12.50	15.38
		2		13.33	10.52	10.00	.00	.00
		3		18.75	12.00	16.60	13.33	11.11
	Summer	4		21.05	16.66	11.76	9.09	6.66
			N	4	4	4	4	4
		Total	Mean	16.8525	13.1275	13.7400	8.7300	8.2875
			SEM	1.83133	1.30989	1.68985	3.05117	3.28635
		N		12	12	12	12	12
	Total	Mean		18.1500	13.7333	17.1017	13.0600	10.2550
		SEM		2.52218	2.30618	2.41807	1.85076	1.44680

Appendix 3: Mean ±SEM of seasonal variation of prevalence of trematode parasites in *Bellamya* sampled from Khulna regions in 2012 -2013

		_		KU	Dacope	Symnagar	Kaligong	BagerhatDc	Fakirhat
Season	Rainy	1		19.00	11.76	23.52	5.26	16.13	16.00
		2		18.18	18.18	27.27	8.00	18.75	16.67
		3		22.72	22.00	30.76	11.54	30.00	19.44
		4		13.04	10.52	28.57	10.34	24.24	13.04
		Total	N	4	4	4	4	4	4
			Mean	18.2350	15.6150	27.5300	8.7850	22.2800	16.2875
			SEM	1.99356	2.71059	1.51828	1.38594	3.07847	1.31382
	Winter	1		13.33	7.60	11.11	.00	7.14	12.50
		2		6.66	13.33	12.50	4.00	3.45	13.04
		3		7.14	.00	.00	8.70	11.76	11.11
		4		11.11	12.50	16.66	.00	6.67	7.69
		Total	N	4	4	4	4	4	4
			Mean	9.5600	8.3575	10.0675	3.1750	7.2550	11.0850
			SEM	1.60421	3.05924	3.55691	2.06897	1.71096	1.20246
	Summer	1		5.55	9.09	14.28	11.11	15.15	10.71
		2		.00	.00	13.33	14.29	18.75	11.54
		3		9.50	12.50	18.75	12.50	17.65	10.00
		4		7.14	9.09	21.05	11.11	19.35	14.29
		Total	N	4	4	4	4	4	4
			Mean	5.5475	7.6700	16.8525	12.2525	17.7250	11.6350
			SEM	2.01935	2.68003	1.83133	.75406	.92770	.93928
	Total	N		12	12	12	12	12	12
		Mean		11.1142	10.5475	18.1500	8.0708	15.7533	13.0025
		SEM		1.87476	1.82924	2.52218	1.37373	2.19159	.92934

Appendix 4: Mean ±SEM of seasonal variation of prevalence of trematode parasites in *Bellamya* sampled from Kishoreganj regions in 2012 - 2013

				Solakia	Astagram	Kuliarchar	Mithamain	BAU	Trisal
Season	Rainy	1		13.64	33.00	16.13	14.29	15.00	15.79
		2		10.53	35.60	20.00	16.00	18.18	23.81
		3		18.18	36.30	27.59	19.35	22.72	25.93
		4		14.29	21.05	18.18	17.14	20.00	20.00
		Total	N	4	4	4	4	4	4
			Mean	14.1600	31.4875	20.4750	16.6950	18.9750	21.3825
			SEM	1.57122	3.55084	2.49991	1.06121	1.62040	2.23159
	Winter	1		13.33	12.50	12.50	10.71	13.33	14.29
		2		14.29	6.66	12.00	12.00	6.66	17.65
		3		12.50	5.50	14.29	18.18	7.14	10.53
		4		6.67	17.64	6.67	5.00	11.11	7.14
		Total	N	4	4	4	4	4	4
			Mean	11.6975	10.5750	11.3650	11.4725	9.5600	12.4025
			SEM	1.71527	2.80925	1.64038	2.70406	1.60421	2.27851
	Summer	1		10.00	10.00	13.33	17.65	5.55	9.52
		2		19.05	7.14	14.29	15.38	8.69	16.67
		3		16.67	.00	16.13	14.29	9.50	19.05
		4		11.76	20.00	17.86	11.54	11.76	15.38
		Total	N	4	4	4	4	4	4
			Mean	14.3700	9.2850	15.4025	14.7150	8.8750	15.1550
			SEM	2.10360	4.14462	1.00422	1.26878	1.28464	2.02629
	Total	N		12	12	12	12	12	12
		Mean		13.4092	17.1158	15.7475	14.2942	12.4700	16.3133
		SEM		1.01391	3.58314	1.47218	1.15531	1.59787	1.60647

Appendix 5: Mean ±SEM of seasonal variation of prevalence of trematode parasites in *Brotia* sampled from Khulna regions in 2011 -2012

		_		KU	Dacope	Symnagar	Kaligong	BagerhatDc	Fakirhat
		1		12.50	6.66	15.38	15.38	18.18	15.38
		2		16.66	11.00	12.50	11.76	13.33	18.75
		3		18.18	12.50	16.60	13.63	18.75	16.60
	Rainy	4		13.33	.00	13.33	12.50	14.28	9.00
			N	4	4	4	4	4	4
		Total	Mean	15.1675	7.5400	14.4525	13.3175	16.1350	14.9325
			SEM	1.34767	2.80177	.93738	.78770	1.36410	2.09660
		1		9.09	.00	10.00	11.00	.00	5.26
		2		7.14	7.69	11.11	.00	.00	.00
		3		5.26	8.33	7.60	5.26	7.14	.00
	Winter	4		.00	11.00	12.50	8.33	11.00	12.50
			N	4	4	4	4	4	4
Season		Total	Mean	5.3725	6.7550	10.3025	6.1475	4.5350	4.4400
			SEM	1.95406	2.36299	1.03586	2.36096	2.73427	2.95893
		1		11.76	9.09	.00	.00	6.66	6.66
		2		16.66	7.14	11.11	.00	11.11	13.33
		3		15.38	9.09	10.00	9.09	.00	15.00
	Summer	4		11.11	.00	7.69	6.66	10.52	11.11
			N	4	4	4	4	4	4
		Total	Mean	13.7275	6.3300	7.2000	3.9375	7.0725	11.5250
			SEM	1.35563	2.15948	2.50347	2.32680	2.55566	1.80680
		N		12	12	12	12	12	12
	Total	Mean		11.4225	6.8750	10.6517	7.8008	9.2475	10.2992
		SEM		1.54093	1.29152	1.24486	1.58514	1.92260	1.79686

 $Appendix\ 6:\ Mean\ \pm SEM\ of\ seasonal\ variation\ of\ prevalence\ of\ trematode\ parasites\ in\ \textit{Brotia}\ sampled\ from\ Kishoreganj\ regions\ in\ 2011\ -2012$

				Solakia	Astagram	Kuliarchar	Mithamon	BAU	Trisal
		1		11.11	12.50	11.11	20.00	11.11	10.00
		2		18.18	16.66	13.33	18.18	10.00	6.25
		3		16.66	18.18	18.18	7.69	12.50	16.60
	Rainy	4		7.69	13.33	8.33	9.09	.00	13.33
			N	4	4	4	4	4	4
		Total	Mean	13.4100	15.1675	12.7375	13.7400	8.4025	11.5450
			SEM	2.43796	1.34767	2.08260	3.12418	2.84713	2.22042
		1		.00	9.09	.00	.00	.00	14.28
		2		11.11	7.14	.00	.00	11.11	9.09
		3		8.33	5.26	11.11	9.09	.00	11.11
	Winter	4		13.33	.00	9.09	7.69	8.33	10.52
		Total	N	4	4	4	4	4	4
Season			Mean	8.1925	5.3725	5.0500	4.1950	4.8600	11.2500
			SEM	2.91607	1.95406	2.94463	2.43879	2.86273	1.09541
		1		11.76	11.76	.00	9.09	13.33	12.50
		2		10.00	16.66	11.11	.00	10.00	10.00
		3		11.11	15.38	14.28	7.14	.00	16.66
	Summer	4	•	12.50	11.11	10.00	10.00	8.33	11.11
			N	4	4	4	4	4	4
		Total	Mean	11.3425	13.7275	8.8475	6.5575	7.9150	12.5675
			SEM	.52997	1.35563	3.08542	2.26577	2.83562	1.45687
		N		12	12	12	12	12	12
	Total	Mean		10.9817	11.4225	8.8783	8.1642	7.0592	11.7875
		SEM		1.32563	1.54093	1.71565	1.84186	1.56088	.88273

Appendix 7: Mean ±SEM of seasonal variation of prevalence of trematode parasites in *Brotia* sampled from Khulna regions in 2012 -2013

				KU	Dacope	Symnagar	Kaligong	BagerhatDc	Fakirhat
		1		7.50	13.63	11.11	9.09	9.09	5.26
		2	2		21.05	18.18	10.00	17.14	8.00
		3		16.66	20.83	16.66	11.76	18.18	11.54
	Rainy	4		10.00	16.60	7.69	8.82	13.79	10.34
			N	4	4	4	4	4	4
		Total	Mean	11.7900	18.0275	13.4100	9.9175	14.5500	8.7850
			SEM	1.97461	1.78805	2.43796	.66401	2.04683	1.38594
		1		.00	11.00	.00	3.33	6.90	.00
		2		10.00	9.52	11.11	6.25	9.09	4.00
		3		8.33	6.66	8.33	4.35	14.29	8.70
	Winter	4		9.09	13.33	13.33	3.85	3.33	.00
		Total	N	4	4	4	4	4	4
Season			Mean	6.8550	10.1275	8.1925	4.4450	8.4025	3.1750
			SEM	2.31036	1.39672	2.91607	.63668	2.29352	2.06897
		1		7.14	10.00	11.76	7.69	10.34	11.11
		2		7.50	5.55	10.00	10.71	12.12	14.29
		3		16.00	7.69	11.11	10.00	14.71	12.50
	Summer	4		.00	8.33	12.50	7.41	12.00	11.11
			N	4	4	4	4	4	4
		Total	Mean	7.6600	7.8925	11.3425	8.9525	12.2925	12.2525
			SEM	3.27271	.92020	.52997	.82458	.90240	.75406
		N		12	12	12	12	12	12
	Total	Mean		8.7683	12.0158	10.9817	7.7717	11.7483	8.0708
		Std. Error	of Mean	1.49608	1.50446	1.32563	.80987	1.23254	1.37373

Appendix 8: Mean ±SEM of seasonal variation of prevalence of trematode parasites in *Brotia* sampled from Kishoreganj regions in 2012 -2013

				Solakia	Astagram	Kuliarchar	Mithamon	BAU	Trisal
		1		10.53	15.38	9.52	17.24	7.50	11.54
		2	2		18.75	16.67	19.35	13.00	12.50
		3	3		16.60	19.23	24.24	15.00	14.81
	Rainy	4	4		9.00	13.64	16.00	10.00	13.64
			N	4	4	4	4	4	4
		Total	Mean	14.5850	14.9325	14.7650	19.2075	11.3750	13.1225
			SEM	2.19562	2.09660	2.08848	1.81442	1.65044	.70754
		1		11.76	5.26	5.88	11.54	.00	6.25
		2		13.33	.00	12.50	14.29	10.00	9.09
		3		12.50	.00	13.64	10.53	.00	7.14
	Winter	4		7.69	12.50	4.17	7.69	.00	4.76
		Total	N	4	4	4	4	4	4
Season			Mean	11.3200	4.4400	9.0475	11.0125	2.5000	6.8100
			SEM	1.25177	2.95893	2.35998	1.36296	2.50000	.90477
		1		11.11	6.66	7.41	11.76	6.66	9.52
		2		13.64	13.33	13.04	13.64	7.50	11.11
		3		15.00	15.00	10.71	13.79	16.00	13.33
	Summer	4	4		11.11	9.52	16.13	.00	10.71
			N	4	4	4	4	4	4
		Total	Mean	13.7825	11.5250	10.1700	13.8300	7.5400	11.1675
			SEM	.96596	1.80680	1.17504	.89501	3.28125	.79599
		N	N		12	12	12	12	12
	Total	Mean		13.2292	10.2992	11.3275	14.6833	7.1383	10.3667
		Std. Error	r of Mean	.91704	1.79686	1.25879	1.26160	1.73071	.90024

Appendix 9: Mean ±SEM of seasonal variation of prevalence of trematode parasites in *Pila* sampled from Khulna regions in 2011 -2012

			KU	Dacope	Symnagar	Kaligong	BagerhatDc	Fakirhat	
		1		13.16	18.18	27.27	10.34	17.14	16.00
		2		11.54	13.33	31.25	13.04	16.67	16.67
		3		19.05	18.75	23.07	17.14	21.21	19.44
	Rainy	4		15.00	14.28	10.52	9.68	14.29	13.04
			N	4	4	4	4	4	4
		Total	Mean	14.6875	16.1350	23.0275	12.5500	17.3275	16.2875
			SEM	1.61681	1.36410	4.49117	1.69384	1.43665	1.31382
		1		8.33	.00	9.09	3.70	6.67	12.50
		2		12.50	.00	.00	10.34	11.11	13.04
		3		13.33	7.14	14.28	6.06	9.09	11.11
	Winter	4		6.67	11.00	16.66	4.35	3.33	7.69
		Total	N	4	4	4	4	4	4
Season			Mean	10.2075	4.5350	10.0075	6.1125	7.5500	11.0850
			SEM	1.60843	2.73427	3.69123	1.49446	1.67402	1.20246
		1		8.70	6.66	12.50	6.25	13.79	10.71
		2		12.00	11.11	8.33	10.00	12.12	11.54
		3		14.29	.00	13.33	13.89	14.71	10.00
	Summer	4		13.64	10.52	16.66	12.90	16.67	14.29
			N	4	4	4	4	4	4
		Total	Mean	12.1575	7.0725	12.7050	10.7600	14.3225	11.6350
			SEM	1.24915	2.55566	1.71307	1.71499	.94848	.93928
		N		12	12	12	12	12	12
	Total	Mean		12.3508	9.2475	15.2467	9.8075	13.0667	13.0025
		SEM		.95942	1.92260	2.49023	1.18338	1.42980	.92934

Appendix 10: Mean ±SEM of seasonal variation of prevalence of trematode parasites in *Pila* sampled from Kishoreganj regions in 2011 -2012

				Solakia	Astagram	Kuliarchar	Mithamon	BAU	Trisal
		1		13.64	18.00	13.16	12.90	11.00	12.12
		2		10.53	23.00	11.54	13.89	19.00	10.00
		3		18.18	33.00	19.05	17.24	16.00	12.50
	Rainy	4		14.29	16.60	15.00	12.00	13.00	13.79
			N	4	4	4	4	4	4
		Total	Mean	14.1600	22.6500	14.6875	14.0075	14.7500	12.1025
			SEM	1.57122	3.71338	1.61681	1.14453	1.75000	.78668
		1		13.33	6.25	8.33	9.52	8.00	4.55
		2		14.29	13.33	12.50	11.76	.00	8.00
		3		12.50	.00	13.33	16.67	9.00	9.38
	Winter	4		6.67	14.28	6.67	9.09	.00	3.70
		Total	N	4	4	4	4	4	4
Season			Mean	11.6975	8.4650	10.2075	11.7600	4.2500	6.4075
			SEM	1.71527	3.34222	1.60843	1.73816	2.46221	1.35870
		1		10.00	12.50	8.70	11.76	12.00	6.25
		2		19.05	10.00	12.00	10.71	.00	7.14
		3		16.67	.00	14.29	12.50	10.00	10.00
	Summer	4		11.76	16.60	13.64	7.69	6.00	6.45
		Total	N	4	4	4	4	4	4
			Mean	14.3700	9.7750	12.1575	10.6650	7.0000	7.4600
			SEM	2.10360	3.53091	1.24915	1.05747	2.64575	.86786
		N		12	12	12	12	12	12
	Total	Mean		13.4092	13.6300	12.3508	12.1442	8.6667	8.6567
		SEM		1.01391	2.66958	.95942	.81938	1.80627	.92144

Appendix 11: Mean ±SEM of seasonal variation of prevalence of trematode parasites in *Pila* sampled from Khulna regions in 2012 -2013

				KU	Dacope	Symnagar	Kaligong	BagerhatDc	Fakirhat
		1		13.16	18.18	27.27	10.34	17.14	16.00
		2		11.54	13.33	31.25	13.04	16.67	16.67
		3		19.05	18.75	23.07	17.14	21.21	19.44
	Rainy	4		15.00	14.28	10.52	9.68	14.29	13.04
			N	4	4	4	4	4	4
		Total	Mean	14.6875	16.1350	23.0275	12.5500	17.3275	16.2875
			SEM	1.61681	1.36410	4.49117	1.69384	1.43665	1.31382
		1		8.33	.00	9.09	3.70	6.67	12.50
		2		12.50	.00	.00	10.34	11.11	13.04
		3		13.33	7.14	14.28	6.06	9.09	11.11
	Winter	4		6.67	11.00	16.66	4.35	3.33	7.69
		Total	N	4	4	4	4	4	4
Season			Mean	10.2075	4.5350	10.0075	6.1125	7.5500	11.0850
			SEM	1.60843	2.73427	3.69123	1.49446	1.67402	1.20246
		1		8.70	6.66	12.50	6.25	13.79	10.71
		2		12.00	11.11	8.33	10.00	12.12	11.54
		3		14.29	.00	13.33	13.89	14.71	10.00
	Summer	4		13.64	10.52	16.66	12.90	16.67	14.29
		Total	N	4	4	4	4	4	4
			Mean	12.1575	7.0725	12.7050	10.7600	14.3225	11.6350
			SEM	1.24915	2.55566	1.71307	1.71499	.94848	.93928
		N		12	12	12	12	12	12
	Total	Mean		12.3508	9.2475	15.2467	9.8075	13.0667	13.0025
		SEM		.95942	1.92260	2.49023	1.18338	1.42980	.92934

Appendix 12: Mean ±SEM of seasonal variation of prevalence of trematode parasites in *Pila* sampled from Kishoreganj regions in 2012 -2013

				Solakia	Astagram	Kuliarchar	Mithamain	BAU	Trisal
		1		13.64	18.00	13.16	12.90	11.00	12.12
		2		10.53	23.00	11.54	13.89	19.00	10.00
		3		18.18	33.00	19.05	17.24	16.00	12.50
	Rainy	4		14.29	16.60	15.00	12.00	13.00	13.79
			N	4	4	4	4	4	4
	Total	Mean	14.1600	22.6500	14.6875	14.0075	14.7500	12.1025	
			SEM	1.57122	3.71338	1.61681	1.14453	1.75000	.78668
		1		13.33	6.25	8.33	9.52	8.00	4.55
		2		14.29	13.33	12.50	11.76	.00	8.00
		3		12.50	.00	13.33	16.67	9.00	9.38
	Winter	4		6.67	14.28	6.67	9.09	.00	3.70
			N	4	4	4	4	4	4
Season		Total	Mean	11.6975	8.4650	10.2075	11.7600	4.2500	6.4075
			SEM	1.71527	3.34222	1.60843	1.73816	2.46221	1.35870
		1		10.00	12.50	8.70	11.76	12.00	6.25
		2		19.05	10.00	12.00	10.71	.00	7.14
		3		16.67	.00	14.29	12.50	10.00	10.00
	Summer	4		11.76	16.60	13.64	7.69	6.00	6.45
			N	4	4	4	4	4	4
		Total	Mean	14.3700	9.7750	12.1575	10.6650	7.0000	7.4600
			SEM	2.10360	3.53091	1.24915	1.05747	2.64575	.86786
		N		12	12	12	12	12	12
	Total	Mean		13.4092	13.6300	12.3508	12.1442	8.6667	8.6567
		SEM		1.01391	2.66958	.95942	.81938	1.80627	.92144

Table 13: Multiple comparison of percentage occurrence of trematode parasites in *Bellamya* between seasons in Khulna and Kishoreganj regions in 2011-12

	LSD					Multiple Comparis	sons
Dependent	(I) Season	(J) Season	Mean Difference (I-	Std. Error	Sig.	95% Confide	ence Interval
Variable			J)			Lower Bound	Upper Bound
		Winter	20.91250*	5.01187	.002	9.5749	32.2501
	Rainy	Summer	22.20250*	5.01187	.002	10.8649	33.5401
		Rainy	-20.91250*	5.01187	.002	-32.2501	-9.5749
KU	Winter	Summer	1.29000	5.01187	.803	-10.0476	12.6276
	Cummon	Rainy	-22.20250*	5.01187	.002	-33.5401	-10.8649
	Summer	Winter	-1.29000	5.01187	.803	-12.6276	10.0476
		Winter	5.65750*	2.10692	.025	.8913	10.4237
	Rainy	Summer	4.27000	2.10692	.073	4962	9.0362
-		Rainy	-5.65750*	2.10692	.025	-10.4237	8913
Dacope	Winter	Summer	-1.38750	2.10692	.527	-6.1537	3.3787
		Rainy	-4.27000	2.10692	.073	-9.0362	.4962
	Summer	Winter	1.38750	2.10692	.527	-3.3787	6.1537
		Winter	3.81000	3.66687	.326	-4.4850	12.1050
	Rainy	Summer	10.49500*	3.66687	.019	2.2000	18.7900
_	Winter	Rainy	-3.81000	3.66687	.326	-12.1050	4.4850
Symnagar		Summer	6.68500	3.66687	.102	-1.6100	14.9800
	Summer	Rainy	-10.49500*	3.66687	.019	-18.7900	-2.2000
		Winter	-6.68500	3.66687	.102	-14.9800	1.6100
		Winter	6.63750*	2.84108	.044	.2105	13.0645
	Rainy	Summer	9.77250*	2.84108	.007	3.3455	16.1995
** **		Rainy	-6.63750*	2.84108	.044	-13.0645	2105
Kaligong	Winter	Summer	3.13500	2.84108	.298	-3.2920	9.5620
		Rainy	-9.77250*	2.84108	.007	-16.1995	-3.3455
	Summer	Winter	-3.13500	2.84108	.298	-9.5620	3.2920
		Winter	7.90000 [*]	1.99912	.003	3.3777	12.4223
	Rainy	Summer	10.13500*	1.99912	.001	5.6127	14.6573
		Rainy	-7.90000 [*]	1.99912	.003	-12.4223	-3.3777
BagerhatDc	Winter	Summer	2.23500	1.99912	.293	-2.2873	6.7573
		Rainy	-10.13500 [*]	1.99912	.001	-14.6573	-5.6127
	Summer	Winter	-2.23500	1.99912	.293	-6.7573	2.2873
		Winter	20.91250*	5.01187	.002	9.5749	32.2501
	Rainy	Summer	22.20250*	5.01187	.002	10.8649	33.5401
Fakirhat		Rainy	-20.91250*	5.01187	.002	-32.2501	-9.5749
	Winter	Summer	1.29000	5.01187	.803	-10.0476	12.6276

	1		,				
	Summer	Rainy	-22.20250*	5.01187	.002	-33.5401	-10.8649
	Summer	Winter	-1.29000	5.01187	.803	-12.6276	10.0476
	D-:	Winter	17.46250*	3.49386	.001	9.5588	25.3662
	Rainy	Summer	10.67750*	3.49386	.014	2.7738	18.5812
0.111	777	Rainy	-17.46250*	3.49386	.001	-25.3662	-9.5588
Solakia	Winter	Summer	-6.78500	3.49386	.084	-14.6887	1.1187
		Rainy	-10.67750*	3.49386	.014	-18.5812	-2.7738
	Summer	Winter	6.78500	3.49386	.084	-1.1187	14.6887
	D .	Winter	15.59250*	3.44465	.001	7.8002	23.3848
	Rainy	Summer	8.70500*	3.44465	.032	.9127	16.4973
		Rainy	-15.59250*	3.44465	.001	-23.3848	-7.8002
Astagram	Winter	Summer	-6.88750	3.44465	.077	-14.6798	.9048
		Rainy	-8.70500*	3.44465	.032	-16.4973	9127
	Summer	Winter	6.88750	3.44465	.077	9048	14.6798
		Winter	8.66750*	3.70873	.044	.2778	17.0572
	Rainy	Summer	7.17500	3.70873	.085	-1.2147	15.5647
		Rainy	-8.66750*	3.70873	.044	-17.0572	2778
Kuliarchar	Winter	Summer	-1.49250	3.70873	.697	-9.8822	6.8972
	Summer	Rainy	-7.17500	3.70873	.085	-15.5647	1.2147
		Winter	1.49250	3.70873	.697	-6.8972	9.8822
		Winter	13.13500*	4.46563	.016	3.0331	23.2369
	Rainy	Summer	11.61000*	4.46563	.029	1.5081	21.7119
		Rainy	-13.13500*	4.46563	.016	-23.2369	-3.0331
Mithamon	Winter	Summer	-1.52500	4.46563	.741	-11.6269	8.5769
		Rainy	-11.61000*	4.46563	.029	-21.7119	-1.5081
	Summer	Winter	1.52500	4.46563	.741	-8.5769	11.6269
		Winter	9.04500*	3.12717	.018	1.9708	16.1192
	Rainy	Summer	11.01750*	3.12717	.006	3.9433	18.0917
		Rainy	-9.04500*	3.12717	.018	-16.1192	-1.9708
BAU	Winter	Summer	1.97250	3.12717	.544	-5.1017	9.0467
		Rainy	-11.01750*	3.12717	.006	-18.0917	-3.9433
	Summer	Winter	-1.97250	3.12717	.544	-9.0467	5.1017
		Winter	6.73250	3.00394	.052	0629	13.5279
	Rainy	Summer	6.31750	3.00394	.065	4779	13.1129
		Rainy	-6.73250	3.00394	.052	-13.5279	.0629
Trisal	Winter	Summer	41500	3.00394	.893	-7.2104	6.3804
		Rainy	-6.31750	3.00394	.065	-13.1129	.4779
<u> </u>	Summer	Winter	.41500	3.00394	.893	-6.3804	7.2104
*. The mean	difference is sig	gnificant at the 0.05	level.				
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Table 14: Multiple comparison of percentage occurrence of trematode parasites in *Bellamya* in Khulna and Kishoreganj regions in 2012-13

	LSD			Multiple Comparisons						
Dependent	(I) Season	(J) Season	Mean Difference (I-J)	Std. Error	Sig.	95% Confide	ence Interval			
Variable						Lower Bound	Upper Bound			
		Winter	8.67500*	2.66152	.010	2.6542	14.6958			
	Rainy	Summer	12.68750*	2.66152	.001	6.6667	18.7083			
		Rainy	-8.67500*	2.66152	.010	-14.6958	-2.6542			
KU	Winter	Summer	4.01250	2.66152	.166	-2.0083	10.0333			
		Rainy	-12.68750*	2.66152	.001	-18.7083	-6.6667			
	Summer	Winter	-4.01250	2.66152	.166	-10.0333	2.0083			
		Winter	7.25750	3.99072	.102	-1.7701	16.2851			
	Rainy	Summer	7.94500	3.99072	.078	-1.0826	16.9726			
	**************************************	Rainy	-7.25750	3.99072	.102	-16.2851	1.7701			
Dacope	Winter	Summer	.68750	3.99072	.867	-8.3401	9.7151			
		Rainy	-7.94500	3.99072	.078	-16.9726	1.0826			
	Summer	Winter	68750	3.99072	.867	-9.7151	8.3401			
	D :	Winter	17.46250*	3.49386	.001	9.5588	25.3662			
	Rainy	Summer	10.67750*	3.49386	.014	2.7738	18.5812			
G	Winter	Rainy	-17.46250*	3.49386	.001	-25.3662	-9.5588			
Symnagar		Summer	-6.78500	3.49386	.084	-14.6887	1.1187			
		Rainy	-10.67750*	3.49386	.014	-18.5812	-2.7738			
		Winter	6.78500	3.49386	.084	-1.1187	14.6887			
	D - i	Winter	5.61000*	2.12447	.027	.8041	10.4159			
	Rainy	Summer	-3.46750	2.12447	.137	-8.2734	1.3384			
W-B	W7:4	Rainy	-5.61000 [*]	2.12447	.027	-10.4159	8041			
Kaligong	Winter	Summer	-9.07750*	2.12447	.002	-13.8834	-4.2716			
	G	Rainy	3.46750	2.12447	.137	-1.3384	8.2734			
	Summer	Winter	9.07750*	2.12447	.002	4.2716	13.8834			
	D - i	Winter	15.02500*	2.97377	.001	8.2979	21.7521			
	Rainy	Summer	4.55500	2.97377	.160	-2.1721	11.2821			
BagerhatDc	Winter	Rainy	-15.02500*	2.97377	.001	-21.7521	-8.2979			
Бадетапъс	willer	Summer	-10.47000*	2.97377	.007	-17.1971	-3.7429			
	Summan.	Rainy	-4.55500	2.97377	.160	-11.2821	2.1721			
	Summer	Winter	10.47000*	2.97377	.007	3.7429	17.1971			
	Rainy	Winter	5.20250*	1.64403	.011	1.4834	8.9216			
Folsiub ct	Kainy	Summer	4.65250*	1.64403	.020	.9334	8.3716			
Fakirhat	Winter	Rainy	-5.20250*	1.64403	.011	-8.9216	-1.4834			
	Winter	Summer	55000	1.64403	.746	-4.2691	3.1691			

	C	Rainy	-4.65250*	1.64403	.020	-8.3716	9334
	Summer	Winter	.55000	1.64403	.746	-3.1691	4.2691
	D .	Winter	2.46250	2.56073	.361	-3.3303	8.2553
	Rainy	Summer	21000	2.56073	.936	-6.0028	5.5828
~		Rainy	-2.46250	2.56073	.361	-8.2553	3.3303
Solakia	Winter	Summer	-2.67250	2.56073	.324	-8.4653	3.1203
		Rainy	.21000	2.56073	.936	-5.5828	6.0028
	Summer	Winter	2.67250	2.56073	.324	-3.1203	8.4653
		Winter	20.91250 [*]	5.01187	.002	9.5749	32.2501
	Rainy	Summer	22.20250*	5.01187	.002	10.8649	33.5401
		Rainy	-20.91250*	5.01187	.002	-32.2501	-9.5749
Astagram	Winter	Summer	1.29000	5.01187	.803	-10.0476	12.6276
		Rainy	-22.20250*	5.01187	.002	-33.5401	-10.8649
	Summer	Winter	-1.29000	5.01187	.803	-12.6276	10.0476
		Winter	9.11000*	2.57538	.006	3.2841	14.9359
	Rainy	Summer	5.07250	2.57538	.080	7534	10.8984
		Rainy	-9.11000*	2.57538	.006	-14.9359	-3.2841
Kuliarchar	Winter	Summer	-4.03750	2.57538	.151	-9.8634	1.7884
	Summer	Rainy	-5.07250	2.57538	.080	-10.8984	.7534
		Winter	4.03750	2.57538	.151	-1.7884	9.8634
		Winter	5.22250	2.58817	.074	6323	11.0773
	Rainy	Summer	1.98000	2.58817	.464	-3.8748	7.8348
		Rainy	-5.22250	2.58817	.074	-11.0773	.6323
Mithamon	Winter	Summer	-3.24250	2.58817	.242	-9.0973	2.6123
		Rainy	-1.98000	2.58817	.464	-7.8348	3.8748
	Summer	Winter	3.24250	2.58817	.242	-2.6123	9.0973
		Winter	9.41500*	2.13690	.002	4.5810	14.2490
	Rainy	Summer	10.10000*	2.13690	.001	5.2660	14.9340
		Rainy	-9.41500*	2.13690	.002	-14.2490	-4.5810
BAU	Winter	Summer	.68500	2.13690	.756	-4.1490	5.5190
		Rainy	-10.10000*	2.13690	.001	-14.9340	-5.2660
	Summer	Winter	68500	2.13690	.756	-5.5190	4.1490
		Winter	8.98000*	3.08517	.017	2.0009	15.9591
	Rainy	Summer	6.22750	3.08517	.074	7516	13.2066
		Rainy	-8.98000*	3.08517	.017	-15.9591	-2.0009
Trisal	Winter	Summer	-2.75250	3.08517	.396	-9.7316	4.2266
		Rainy	-6.22750	3.08517	.074	-13.2066	.7516
	Summer	Winter	2.75250	3.08517	.396	-4.2266	9.7316

Table 15: Multiple comparison of percentage occurrence of trematode parasites in *Brotia* in Khulna and Kishoreganj regions in 2011-12

	L	SD			Multiple Com	parisons	
Dependent	(I) Season	(J) Season	Mean Difference	Std. Error	Sig.	95% Confid	ence Interval
Variable			(I-J)			Lower Bound	Upper Bound
		Winter	9.79500*	2.23194	.002	4.7460	14.8440
	Rainy	Summer	1.44000	2.23194	.535	-3.6090	6.4890
		Rainy	-9.79500*	2.23194	.002	-14.8440	-4.7460
KU	Winter	Summer	-8.35500*	2.23194	.005	-13.4040	-3.3060
		Rainy	-1.44000	2.23194	.535	-6.4890	3.6090
	Summer	Winter	8.35500*	2.23194	.005	3.3060	13.4040
	ъ.	Winter	.78500	3.47343	.826	-7.0724	8.6424
	Rainy	Summer	1.21000	3.47343	.736	-6.6474	9.0674
D	337° 4	Rainy	78500	3.47343	.826	-8.6424	7.0724
Dacope	Winter	Summer	.42500	3.47343	.905	-7.4324	8.2824
		Rainy	-1.21000	3.47343	.736	-9.0674	6.6474
	Summer	Winter	42500	3.47343	.905	-8.2824	7.4324
	ъ.	Winter	4.15000	2.34080	.110	-1.1453	9.4453
	Rainy	Summer	7.25250*	2.34080	.013	1.9572	12.5478
a a	Winter	Rainy	-4.15000	2.34080	.110	-9.4453	1.1453
Symnagar		Summer	3.10250	2.34080	.218	-2.1928	8.3978
	Summer	Rainy	-7.25250*	2.34080	.013	-12.5478	-1.9572
		Winter	-3.10250	2.34080	.218	-8.3978	2.1928
	D .	Winter	7.17000*	2.78192	.030	.8769	13.4631
	Rainy	Summer	9.38000*	2.78192	.008	3.0869	15.6731
IZ 1'	337° 4	Rainy	-7.17000*	2.78192	.030	-13.4631	8769
Kaligong	Winter	Summer	2.21000	2.78192	.447	-4.0831	8.5031
	C	Rainy	-9.38000*	2.78192	.008	-15.6731	-3.0869
	Summer	Winter	-2.21000	2.78192	.447	-8.5031	4.0831
	D .	Winter	11.60000*	3.25253	.006	4.2423	18.9577
	Rainy	Summer	9.06250*	3.25253	.021	1.7048	16.4202
D d 4D -	W/:4	Rainy	-11.60000*	3.25253	.006	-18.9577	-4.2423
BagerhatDc	Winter	Summer	-2.53750	3.25253	.455	-9.8952	4.8202
	C	Rainy	-9.06250*	3.25253	.021	-16.4202	-1.7048
	Summer	Winter	2.53750	3.25253	.455	-4.8202	9.8952
	Daim	Winter	10.49250*	3.30812	.011	3.0090	17.9760
Table of	Rainy	Summer	3.40750	3.30812	.330	-4.0760	10.8910
Fakirhat	337:	Rainy	-10.49250*	3.30812	.011	-17.9760	-3.0090
	Winter	Summer	-7.08500	3.30812	.061	-14.5685	.3985

	Summer	Rainy	-3.40750	3.30812	.330	-10.8910	4.0760
	Summer	Winter	7.08500	3.30812	.061	3985	14.5685
	Daine	Winter	5.21750	3.13347	.130	-1.8709	12.3059
	Rainy	Summer	2.06750	3.13347	.526	-5.0209	9.1559
0.11:	***	Rainy	-5.21750	3.13347	.130	-12.3059	1.8709
Solakia	Winter	Summer	-3.15000	3.13347	.341	-10.2384	3.9384
		Rainy	-2.06750	3.13347	.526	-9.1559	5.0209
	Summer	Winter	3.15000	3.13347	.341	-3.9384	10.2384
		Winter	9.79500*	2.23194	.002	4.7460	14.8440
	Rainy	Summer	1.44000	2.23194	.535	-3.6090	6.4890
		Rainy	-9.79500*	2.23194	.002	-14.8440	-4.7460
Astagram	Winter	Summer	-8.35500*	2.23194	.005	-13.4040	-3.3060
		Rainy	-1.44000	2.23194	.535	-6.4890	3.6090
	Summer	Winter	8.35500*	2.23194	.005	3.3060	13.4040
		Winter	7.68750	3.87538	.079	-1.0792	16.4542
	Rainy	Summer	3.89000	3.87538	.342	-4.8767	12.6567
		Rainy	-7.68750	3.87538	.079	-16.4542	1.0792
Kuliarchar	Winter	Summer	-3.79750	3.87538	.353	-12.5642	4.9692
		Rainy	-3.89000	3.87538	.342	-12.6567	4.8767
	Summer	Winter	3.79750	3.87538	.353	-4.9692	12.5642
		Winter	9.54500*	3.72755	.031	1.1127	17.9773
	Rainy	Summer	7.18250	3.72755	.086	-1.2498	15.6148
		Rainy	-9.54500*	3.72755	.031	-17.9773	-1.1127
Mithamon	Winter	Summer	-2.36250	3.72755	.542	-10.7948	6.0698
		Rainy	-7.18250	3.72755	.086	-15.6148	1.2498
	Summer	Winter	2.36250	3.72755	.542	-6.0698	10.7948
		Winter	3.54250	4.02841	.402	-5.5704	12.6554
	Rainy	Summer	.48750	4.02841	.906	-8.6254	9.6004
		Rainy	-3.54250	4.02841	.402	-12.6554	5.5704
BAU	Winter	Summer	-3.05500	4.02841	.468	-12.1679	6.0579
		Rainy	48750	4.02841	.906	-9.6004	8.6254
	Summer	Winter	3.05500	4.02841	.468	-6.0579	12.1679
		Winter	.29500	2.34558	.903	-5.0111	5.6011
	Rainy	Summer	-1.02250	2.34558	.673	-6.3286	4.2836
		Rainy	29500	2.34558	.903	-5.6011	5.0111
Trisal	Winter	Summer	-1.31750	2.34558	.588	-6.6236	3.9886
		Rainy	1.02250	2.34558	.673	-4.2836	6.3286
	Summer	Winter	1.31750	2.34558	.588	-3.9886	6.6236
*. The mean di	fference is signif	ficant at the 0.05 leve	el.				

Table 16: Multiple comparison of percentage occurrence of trematode parasites in *Brotia* between seasons in Khulna and Kishoreganj regions in 2012-13

	LS	SD	Multi	iple Compa	risons		
Dependent Variable	(I) Season	(J) Season	Mean Difference (I-J)	Std. Error	Sig.	95% Confide	ence Interval
						Lower Bound	Upper Bound
		Winter	4.93500	3.64668	.209	-3.3144	13.1844
	Rainy	Summer	4.13000	3.64668	.287	-4.1194	12.3794
KU		Rainy	-4.93500	3.64668	.209	-13.1844	3.3144
	Winter	Summer	80500	3.64668	.830	-9.0544	7.4444
	_	Rainy	-4.13000	3.64668	.287	-12.3794	4.1194
	Summer	Winter	.80500	3.64668	.830	-7.4444	9.0544
		Winter	7.90000*	1.99912	.003	3.3777	12.4223
	Rainy	Summer	10.13500*	1.99912	.001	5.6127	14.6573
_		Rainy	-7.90000 [*]	1.99912	.003	-12.4223	-3.3777
Dacope	Winter	Summer	2.23500	1.99912	.293	-2.2873	6.7573
	_	Rainy	-10.13500*	1.99912	.001	-14.6573	-5.6127
	Summer	Winter	-2.23500	1.99912	.293	-6.7573	2.2873
		Winter	5.21750	3.13347	.130	-1.8709	12.3059
	Rainy	Summer	2.06750	3.13347	.526	-5.0209	9.1559
_	Winter	Rainy	-5.21750	3.13347	.130	-12.3059	1.8709
Symnagar		Summer	-3.15000	3.13347	.341	-10.2384	3.9384
	<u> </u>	Rainy	-2.06750	3.13347	.526	-9.1559	5.0209
	Summer	Winter	3.15000	3.13347	.341	-3.9384	10.2384
		Winter	5.47250*	1.00870	.000	3.1907	7.7543
	Rainy	Summer	.96500	1.00870	.364	-1.3168	3.2468
		Rainy	-5.47250*	1.00870	.000	-7.7543	-3.1907
Kaligong	Winter	Summer	-4.50750*	1.00870	.002	-6.7893	-2.2257
		Rainy	96500	1.00870	.364	-3.2468	1.3168
	Summer	Winter	4.50750*	1.00870	.002	2.2257	6.7893
		Winter	6.14750 [*]	2.61586	.043	.2300	12.0650
	Rainy	Summer	2.25750	2.61586	.411	-3.6600	8.1750
		Rainy	-6.14750 [*]	2.61586	.043	-12.0650	2300
BagerhatDc	Winter	Summer	-3.89000	2.61586	.171	-9.8075	2.0275
	G	Rainy	-2.25750	2.61586	.411	-8.1750	3.6600
	Summer	Winter	3.89000	2.61586	.171	-2.0275	9.8075
	D .	Winter	5.61000 [*]	2.12447	.027	.8041	10.4159
E 11 1	Rainy	Summer	-3.46750	2.12447	.137	-8.2734	1.3384
Fakirhat	***	Rainy	-5.61000 [*]	2.12447	.027	-10.4159	8041
	Winter	Summer	-9.07750*	2.12447	.002	-13.8834	-4.2716

	1		1				
	G	Rainy	3.46750	2.12447	.137	-1.3384	8.2734
	Summer	Winter	9.07750*	2.12447	.002	4.2716	13.8834
	ъ.	Winter	3.26500	2.20919	.174	-1.7325	8.2625
	Rainy	Summer	.80250	2.20919	.725	-4.1950	5.8000
		Rainy	-3.26500	2.20919	.174	-8.2625	1.7325
Solakia	Winter	Summer	-2.46250	2.20919	.294	-7.4600	2.5350
		Rainy	80250	2.20919	.725	-5.8000	4.1950
	Summer	Winter	2.46250	2.20919	.294	-2.5350	7.4600
		Winter	10.49250*	3.30812	.011	3.0090	17.9760
	Rainy	Summer	3.40750	3.30812	.330	-4.0760	10.8910
		Rainy	-10.49250*	3.30812	.011	-17.9760	-3.0090
Astagram	Winter	Summer	-7.08500	3.30812	.061	-14.5685	.3985
		Rainy	-3.40750	3.30812	.330	-10.8910	4.0760
	Summer	Winter	7.08500	3.30812	.061	3985	14.5685
		Winter	5.71750	2.74614	.067	4947	11.9297
	Rainy	Summer	4.59500	2.74614	.129	-1.6172	10.8072
		Rainy	-5.71750	2.74614	.067	-11.9297	.4947
Kuliarchar	Winter	Summer	-1.12250	2.74614	.692	-7.3347	5.0897
		Rainy	-4.59500	2.74614	.129	-10.8072	1.6172
	Summer	Winter	1.12250	2.74614	.692	-5.0897	7.3347
		Winter	8.19500*	1.99179	.003	3.6893	12.7007
	Rainy	Summer	5.37750*	1.99179	.024	.8718	9.8832
		Rainy	-8.19500*	1.99179	.003	-12.7007	-3.6893
Mithamon	Winter	Summer	-2.81750	1.99179	.191	-7.3232	1.6882
		Rainy	-5.37750*	1.99179	.024	-9.8832	8718
	Summer	Winter	2.81750	1.99179	.191	-1.6882	7.3232
		Winter	8.87500*	3.62772	.037	.6685	17.0815
	Rainy	Summer	3.83500	3.62772	.318	-4.3715	12.0415
		Rainy	-8.87500*	3.62772	.037	-17.0815	6685
BAU	Winter	Summer	-5.04000	3.62772	.198	-13.2465	3.1665
		Rainy	-3.83500	3.62772	.318	-12.0415	4.3715
	Summer	Winter	5.04000	3.62772	.198	-3.1665	13.2465
		Winter	6.31250*	1.14100	.000	3.7314	8.8936
	Rainy	Summer	1.95500	1.14100	.121	6261	4.5361
		Rainy	-6.31250*	1.14100	.000	-8.8936	-3.7314
Trisal	Winter	Summer	-4.35750*	1.14100	.004	-6.9386	-1.7764
		Rainy	-1.95500	1.14100	.121	-4.5361	.6261
	Summer	Winter	4.35750*	1.14100	.004	1.7764	6.9386
	ference is signifi	•					

Table 17: Multiple comparison of percentage occurrence of trematode parasites in *Pila* between seasons in Khulna and Kishoreganj regions in 2011-12

	LSD			Multiple Comparisons					
Dependent	(I) Season	(J) Season	Mean Difference	Std. Error	Sig.	95% Confide	ence Interval		
Variable			(I-J)			Lower Bound	Upper Bound		
		Winter	2.90500	1.75698	.133	-1.0696	6.8796		
	Rainy	Summer	56750	1.75698	.754	-4.5421	3.4071		
		Rainy	-2.90500	1.75698	.133	-6.8796	1.0696		
KU	Winter	Summer	-3.47250	1.75698	.080	-7.4471	.5021		
		Rainy	.56750	1.75698	.754	-3.4071	4.5421		
	Summer	Winter	3.47250	1.75698	.080	5021	7.4471		
	ъ.	Winter	6.78750*	2.52711	.025	1.0708	12.5042		
	Rainy	Summer	4.68750	2.52711	.097	-1.0292	10.4042		
D	337	Rainy	-6.78750*	2.52711	.025	-12.5042	-1.0708		
Dacope	Winter	Summer	-2.10000	2.52711	.427	-7.8167	3.6167		
		Rainy	-4.68750	2.52711	.097	-10.4042	1.0292		
	Summer	Winter	2.10000	2.52711	.427	-3.6167	7.8167		
	ъ.	Winter	4.19750	2.18895	.087	7542	9.1492		
	Rainy	Summer	6.08000*	2.18895	.021	1.1283	11.0317		
_	Winter	Rainy	-4.19750	2.18895	.087	-9.1492	.7542		
Symnagar		Summer	1.88250	2.18895	.412	-3.0692	6.8342		
	Summer	Rainy	-6.08000*	2.18895	.021	-11.0317	-1.1283		
		Winter	-1.88250	2.18895	.412	-6.8342	3.0692		
		Winter	9.06250*	3.47627	.028	1.1986	16.9264		
	Rainy	Summer	9.10500 [*]	3.47627	.028	1.2411	16.9689		
		Rainy	-9.06250*	3.47627	.028	-16.9264	-1.1986		
Kaligong	Winter	Summer	.04250	3.47627	.991	-7.8214	7.9064		
		Rainy	-9.10500*	3.47627	.028	-16.9689	-1.2411		
	Summer	Winter	04250	3.47627	.991	-7.9064	7.8214		
		Winter	9.06250*	3.47627	.028	1.1986	16.9264		
	Rainy	Summer	9.10500*	3.47627	.028	1.2411	16.9689		
D 1 D		Rainy	-9.06250*	3.47627	.028	-16.9264	-1.1986		
BagerhatDc	Winter	Summer	.04250	3.47627	.991	-7.8214	7.9064		
	g	Rainy	-9.10500*	3.47627	.028	-16.9689	-1.2411		
	Summer	Winter	04250	3.47627	.991	-7.9064	7.8214		
	D .	Winter	14.18500*	4.99512	.019	2.8852	25.4848		
T 111 -	Rainy	Summer	12.87500*	4.99512	.030	1.5752	24.1748		
Fakirhat	W.	Rainy	-14.18500*	4.99512	.019	-25.4848	-2.8852		
	Winter	Summer	-1.31000	4.99512	.799	-12.6098	9.9898		

Summer W W Su Summer W W Su Summer W W Su Summer W W Su Summer Su Summer Su Su Summer Su Su Su Su Su Su Su S						
Solakia Rainy W Su	Rainy	-12.87500*	4.99512	.030	-24.1748	-1.5752
Solakia Winter Su	Winter	1.31000	4.99512	.799	-9.9898	12.6098
Solakia Winter Su	Winter	14.18500*	4.99512	.019	2.8852	25.4848
Solakia Winter Summer Rainy Winter Winter Rainy Rainy Rainy Summer Winter Rainy Kuliarchar Winter Rainy Summer Winter Rainy Mithamon Winter Rainy Summer Winter Rainy BAU Winter Rainy Summer Winter Rainy Summer Winter Rainy Trisal Winter Rainy Summer Winter Rainy Summer Winter Rainy	Summer	12.87500*	4.99512	.030	1.5752	24.1748
Summer Ra Ra W Summer Ra Summer Summer Ra Summer Summer Summer Summer Summer Ra Summer	Rainy	-14.18500*	4.99512	.019	-25.4848	-2.8852
Summer W	Summer	-1.31000	4.99512	.799	-12.6098	9.9898
Rainy W	Rainy	-12.87500*	4.99512	.030	-24.1748	-1.5752
Astagram Rainy Su	Winter	1.31000	4.99512	.799	-9.9898	12.6098
Astagram Winter Su Summer W Rainy Su Ra Summer Ra Summer W Rainy Mithamon	Winter	9.95500 [*]	2.33415	.002	4.6748	15.2352
Astagram Winter Su	Summer	3.70500	2.33415	.147	-1.5752	8.9852
Summer Ra Ra W Rainy Summer W Rainy Summer Ra Summer Ra Ra W Rainy Summer Ra Ra Rainy Summer Rainy Summer Rainy Summer Rainy Summer Rainy Summer Rainy Rainy Rainy Rainy Rainy Rainy Summer Rainy Rainy Summer Summer Rainy Summer Summer Summer Rainy Summer Summe	Rainy	-9.95500*	2.33415	.002	-15.2352	-4.6748
Kuliarchar Rainy Rainy Winter Ra Summer W Rainy Mithamon Mi	Summer	-6.25000*	2.33415	.025	-11.5302	9698
Rainy W	Rainy	-3.70500	2.33415	.147	-8.9852	1.5752
Kuliarchar Winter Rainy Su Summer W Rainy Su Summer Rainy Su Summer W Rainy Su Summer W Rainy Su Summer W Rainy Su Rainy Rainy Su Summer Rainy Rainy Su Summer W Rainy Su Summer Rainy Rainy Su Su Su Trisal	Winter	6.25000*	2.33415	.025	.9698	11.5302
Summer Ra Summer Ra Summer Ra Summer Ra Summer Ra Summer Ra Summer Summer Summer Summer Summer Summer Ra Summer Summer Summer Summer Ra Summer	Winter	6.01750	3.62747	.132	-2.1884	14.2234
Kuliarchar Winter Su Summer W Rainy Mithamon Winter Su Summer W Rainy Ra Summer W Rainy Su Summer W Rainy Su Summer W Rainy Su Summer Ra Summer W Rainy Su Summer Ra Summer W Rainy Su Summer Ra Summer W Rainy Su Summer Su Summer Su Summer Su Summer Su Su Su Su Su Su Su Su Su S	Summer	.90250	3.62747	.809	-7.3034	9.1084
Summer Ra	Rainy	-6.01750	3.62747	.132	-14.2234	2.1884
Summer W Rainy Rainy Rainy Rainy Rainy Rainy Rainy Rainy Summer W Rainy Summer Rainy Rainy Rainy Rainy Rainy Rainy Trisal Winter Rainy Summer Summer Rainy Summer Summer Rainy Summer Summer Rainy Summer Summer Summer Rainy Summer Summer Summer Summer Summer	Summer	-5.11500	3.62747	.192	-13.3209	3.0909
Rainy Rainy Rainy Mithamon Winter Su Summer W Rainy Rainy Su Summer W Rainy Rainy Rainy Rainy Rainy Trisal Winter Su Summer Rainy Rainy Su Su Summer	Rainy	90250	3.62747	.809	-9.1084	7.3034
Mithamon Winter Su Summer W Rainy Ra W Rainy Su Ra W Su Summer Ra W Rainy Ra Summer Ra Summer Ra Summer Ra Summer W Rainy Su Ra Summer Ra Summer W Rainy Su Summer Su Su Su Su Su Su Su Su Su S	Winter	5.11500	3.62747	.192	-3.0909	13.3209
Mithamon Winter Su Summer W Rainy Su Rainy Su Rainy Rainy Rainy Rainy Trisal Winter Su Rainy Rainy Su Rainy Rainy Su Su Summer	Winter	3.39750	4.00042	.418	-5.6521	12.4471
Mithamon Winter Su Summer W Rainy Su Ra Summer Su	Summer	2.78000	4.00042	.505	-6.2696	11.8296
Summer Ra	Rainy	-3.39750	4.00042	.418	-12.4471	5.6521
BAU Rainy Rainy Su What is a simple of the series of t	Summer	61750	4.00042	.881	-9.6671	8.4321
$\begin{array}{c c} & & & & & & & & \\ & & & & & & & \\ & & & & & & \\ & & & & & & \\ & & & & & \\ & & & & & \\ & & & & & \\ & & & & & \\ & & & & & \\ & & & & & \\ & & & & \\ & & & & \\ & & & & \\ & & & & \\ & & & \\ & & & & \\ & & & \\ & & & \\ & & & \\ & & & \\ & & & \\ & & & \\ & & & \\ & & & \\ & & & \\ & & & \\ & & & \\ & & & \\ & & & \\ & & & \\ & & \\ & & & \\ & & \\ & & & \\ & \\ & & \\ & & \\ & & \\ & & \\ & & \\ & & \\ & & \\ & & \\ & & \\ & & \\ & & \\ & & \\ & & \\ & &$	Rainy	-2.78000	4.00042	.505	-11.8296	6.2696
BAU Winter Su Summer W Rainy Ra W Rainy Ra W Rainy Su Summer W Rainy Su Su Su	Winter	.61750	4.00042	.881	-8.4321	9.6671
BAU Winter Su Summer W Rainy Ra W Rainy Ra W Rainy Su Summer W Rainy Su Su Su	Winter	8.72500*	2.15108	.003	3.8589	13.5911
BAU Winter Su Summer W Rainy Su Trisal Winter Su	Summer	7.00250*	2.15108	.010	2.1364	11.8686
Summer Rainy Rainy Summer W Rainy Su Rainy Su Rainy Su Rainy Rainy Rainy Rainy Rainy	Rainy	-8.72500*	2.15108	.003	-13.5911	-3.8589
Summer Rainy Su Rainy	Summer	-1.72250	2.15108	.444	-6.5886	3.1436
Rainy Rainy Trisal Winter Su	Rainy	-7.00250*	2.15108	.010	-11.8686	-2.1364
Rainy W Su Trisal Winter Su	Winter	1.72250	2.15108	.444	-3.1436	6.5886
Rainy Su Ra Trisal Winter Su	Winter	6.68000	4.30292	.155	-3.0539	16.4139
Trisal Winter Su	Summer	4.79500	4.30292	.294	-4.9389	14.5289
Trisal Winter Su	Rainy	-6.68000	4.30292	.155	-16.4139	3.0539
	Summer	-1.88500	4.30292	.672	-11.6189	7.8489
	Rainy	-4.79500	4.30292	.294	-14.5289	4.9389
Summer	Winter	1.88500	4.30292	.672	-7.8489	11.6189
*. The mean difference is significant at			502,2	.072	7.0107	11.010)

Table 18: Multiple comparison of percentage occurrence of trematode parasites in *Pila* in Khulna and Kishoreganjregions in 2012-13

	1	LSD		Multiple C	Comparisons		
Dependent	(I) Season	(J) Season	Mean Difference	Std. Error	Sig.	95% Confide	ence Interval
Variable			(I-J)			Lower Bound	Upper Bound
		Winter	11.60000°	3.25253	.006	4.2423	18.9577
	Rainy	Summer	9.06250*	3.25253	.021	1.7048	16.4202
_		Rainy	-11.60000*	3.25253	.006	-18.9577	-4.2423
Dacope	Winter	Summer	-2.53750	3.25253	.455	-9.8952	4.8202
	C	Rainy	-9.06250*	3.25253	.021	-16.4202	-1.7048
	Summer	Winter	2.53750	3.25253	.455	-4.8202	9.8952
		Winter	13.02000*	4.94843	.027	1.8259	24.2141
	Rainy	Summer	10.32250	4.94843	.067	8716	21.5166
		Rainy	-13.02000*	4.94843	.027	-24.2141	-1.8259
Symnagar	Winter	Summer	-2.69750	4.94843	.599	-13.8916	8.4966
		Rainy	-10.32250	4.94843	.067	-21.5166	.8716
	Summer	Winter	2.69750	4.94843	.599	-8.4966	13.8916
		Winter	6.43750*	2.31570	.021	1.1990	11.6760
	Rainy	Summer	1.79000	2.31570	.459	-3.4485	7.0285
	Winter	Rainy	-6.43750*	2.31570	.021	-11.6760	-1.1990
Kaligong		Summer	-4.64750	2.31570	.076	-9.8860	.5910
	Summer	Rainy	-1.79000	2.31570	.459	-7.0285	3.4485
		Winter	4.64750	2.31570	.076	5910	9.8860
		Winter	9.77750*	1.96060	.001	5.3423	14.2127
	Rainy	Summer	3.00500	1.96060	.160	-1.4302	7.4402
		Rainy	-9.77750*	1.96060	.001	-14.2127	-5.3423
BagerhatDc	Winter	Summer	-6.77250*	1.96060	.007	-11.2077	-2.3373
		Rainy	-3.00500	1.96060	.160	-7.4402	1.4302
	Summer	Winter	6.77250*	1.96060	.007	2.3373	11.2077
		Winter	5.20250*	1.64403	.011	1.4834	8.9216
	Rainy	Summer	4.65250*	1.64403	.020	.9334	8.3716
		Rainy	-5.20250*	1.64403	.011	-8.9216	-1.4834
Fakirhat	Winter	Summer	55000	1.64403	.746	-4.2691	3.1691
		Rainy	-4.65250*	1.64403	.020	-8.3716	9334
	Summer	Winter	.55000	1.64403	.746	-3.1691	4.2691
		Winter	2.46250	2.56073	.361	-3.3303	8.2553
	Rainy	Summer	21000	2.56073	.936	-6.0028	5.5828
Solakia		Rainy	-2.46250	2.56073	.361	-8.2553	3.3303
	Winter	Summer	-2.67250	2.56073	.324	-8.4653	3.1203

		1				1	
	Summer	Rainy	.21000	2.56073	.936	-5.5828	6.0028
	Summer	Winter	2.67250	2.56073	.324	-3.1203	8.4653
Astagram	Daine	Winter	14.18500*	4.99512	.019	2.8852	25.4848
	Rainy	Summer	12.87500*	4.99512	.030	1.5752	24.1748
	Winter	Rainy	-14.18500*	4.99512	.019	-25.4848	-2.8852
		Summer	-1.31000	4.99512	.799	-12.6098	9.9898
	Summer	Rainy	-12.87500*	4.99512	.030	-24.1748	-1.5752
		Winter	1.31000	4.99512	.799	-9.9898	12.6098
Kuliarchar	Rainy	Winter	4.48000	2.12313	.064	3228	9.2828
		Summer	2.53000	2.12313	.264	-2.2728	7.3328
	Winter	Rainy	-4.48000	2.12313	.064	-9.2828	.3228
		Summer	-1.95000	2.12313	.382	-6.7528	2.8528
	Summer	Rainy	-2.53000	2.12313	.264	-7.3328	2.2728
		Winter	1.95000	2.12313	.382	-2.8528	6.7528
Mithamon	Rainy	Winter	2.24750	1.90603	.269	-2.0642	6.5592
		Summer	3.34250	1.90603	.113	9692	7.6542
	Winter	Rainy	-2.24750	1.90603	.269	-6.5592	2.0642
		Summer	1.09500	1.90603	.580	-3.2167	5.4067
	Summer	Rainy	-3.34250	1.90603	.113	-7.6542	.9692
		Winter	-1.09500	1.90603	.580	-5.4067	3.2167
BAU	Rainy	Winter	10.50000*	3.27872	.011	3.0830	17.9170
		Summer	7.75000*	3.27872	.042	.3330	15.1670
	Winter	Rainy	-10.50000*	3.27872	.011	-17.9170	-3.0830
		Summer	-2.75000	3.27872	.423	-10.1670	4.6670
	Summer	Rainy	-7.75000 [*]	3.27872	.042	-15.1670	3330
		Winter	2.75000	3.27872	.423	-4.6670	10.1670
Trisal	Rainy	Winter	5.69500*	1.46472	.004	2.3816	9.0084
		Summer	4.64250*	1.46472	.011	1.3291	7.9559
	***	Rainy	-5.69500*	1.46472	.004	-9.0084	-2.3816
	Winter	Summer	-1.05250	1.46472	.491	-4.3659	2.2609
		Rainy	-4.64250*	1.46472	.011	-7.9559	-1.3291
	Summer	Winter	1.05250	1.46472	.491	-2.2609	4.3659
KU	Rainy	Winter	4.48000	2.12313	.064	3228	9.2828
		Summer	2.53000	2.12313	.264	-2.2728	7.3328
	Winter	Rainy	-4.48000	2.12313	.064	-9.2828	.3228
		Summer	-1.95000	2.12313	.382	-6.7528	2.8528
	Summer	Rainy	-2.53000	2.12313	.264	-7.3328	2.2728
		Winter	1.95000	2.12313	.382	-2.8528	6.7528
*. The mean di	fference is signif	icant at the 0.05 leve	el.				