PREVALENCE OF FASCIOLIASIS IN CATTLE AT DINAJPUR DISTRICT OF BANGLADESH

A THESIS

BY

ABDIRISAK AHMED ALI

SEMESTER: JULY - DECEMBER, 2018

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A THESIS

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SEMESTER: JULY - DECEMBER, 2018

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DEDICATED

TO MY

BELOVED PARENTS

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ABSTRACT

Fascioliasis is a common hindrance in livestock development in Bangladesh and the present research experiment was carried out to investigate the prevalence of fascioliasis in cattle at Dinajpur district of Bangladesh, during July to December, 2018 using history, clinical signs, physical and coprological examinations. A total of 100 cattle (62 male and 38 female) were recorded as study population. Out of 100 cattle 17 cattle were found positive. Study population was divided into three age groups i.e. young (6 month-1year); adult (1-2 years); old (2- above years) on the basis of owner record and dental formula. The overall prevalence of fascioliasis was 17% and among which the highest prevalence was recorded in older animals (24.14 %) followed by adults (8.7 %) and young (5.26 %), respectively. As per sex-based sectary, higher prevalence of fascioliasis was found in female (28.95 %) followed by male (9.68 %). The higher prevalence of fascioliasis was recorded in poor healthy (22.39 %) followed by healthy ones (6.06 %). This study was preliminary one considering small population of cattle.

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LIST OF ABBREVIATIONS AND SYMBOLS

ALT	-	Alanine Amino Transferase
ASL	-	Aspartate Tgransaminase
BCS	-	Body Condition Score
Dot-Elisa	-	Dot Enzyme Linked Immunosorbent Assay
ELISA	-	Enzyme Linked Immunosorbent Assay
FABP	-	Fatty Acid Binding Proteins
GST	-	Glutathione Stransferase
HSTU	-	Hajee Mohammad Danesh Science and Technology University
iELISA	-	indirect Enzyme Linked Immunosorbent Assay
IEMVT	-	Institut d'élevage Et De Médecine Vétérinaire despay Tropicaux
Ig	-	Immunoglobulin
LNERV	-	Laboratoire National d'élevage Et De Recherches Vétérinaires
No.	-	Number
PHT	-	Passive Haemaglutination Test
Se	-	Sensitivity
Sp	-	Specificity
TIA	-	Thin Layer Immunoassay
WHO	-	World Health Organization

CHAPTER I

INTRODUCTION

CHAPTER I

INTRODUCTION

Bangladesh is located in South East Asia and it is one of the most densely populated countries in the world with an estimated 1,033 people/km^2 (United Nations, 2011). In Bangladesh, similar to human population density, livestock density is also highest (cattle, goats, sheep and buffaloes) in the world with an estimated 145 large ruminants/km^2 compared with 90 for India and 20 for Brazil (BARC Bangladesh, 2010). About 20% of the human population is directly and 50% is partly dependent on the livestock sector (Bangladesh Economic Review, 2009). Livestock is an important sub-sector considered to be the backbone of agriculture. Parasitism is one of the most vulnerable causes of livestock diseases which are the major obstacle in the growth and development of animal health (Mahfooz *et al.,* 2008).

Fascioliasis is recognized as one of the most important helminth diseases of the domesticated ruminants (Lessa *et al,*. 2000). The disease is usually characterized by a chronic, sometimes acute or subacute inflammation of the liver and bile ducts, accompanied by submandibular oedema, anaemia, anorexia, general intoxication, and death (Ogunrinade and Ogunrinade, 1980). Fasciolosis also known as Fascioliasis, Distomatosis and liver Rot is an important disease of cattle caused by trematodes i.e *Fasciola hepatica* and *Fasciola gigantica* (common liver flukes). This condition of internal parasitism is one of the major problems that lowers the livestock productivity throughout the world (Vercruysse and Claerebont, 2001).

Fascioliasis, a serious infectious parasitic disease infecting domestic ruminants and humans, tops all the zoonotic helminthes worldwide (Haridy, *et al* 2002). The disease in predominantly caused by *F. hepatica* and / or *F. gigantica* (Soulsby, 1987). It causes acute and chronic infections (Sampaio *et al.*, 1996). Fascioliasis is endemic in 61 countries and has become a food-borne infection of public health importance in parts of the world such as the Andean Highlands of Bolivia, Ecuador, and Peru; the Nile Delta of Egypt; and Northern Iran (Soliman, 2008). It is estimated that more than 180 million people are at risk of infection, and infection rates are high enough to make Fascioliasis a serious public health concern (WHO, 1998). *Fasciola gigantica*, is endemic in domestic ruminants in Bangladesh (Mohanta *et al*, 2014).

The development of fascioliasis involves the presence of an intermediate host *(Lymnaea sp.)*, suitable habitats for mollusks and environmental factors such as high humidity, adequate temperature and rainfall. In livestock, fascioliasis is important for losses caused by either mortality in acute cases or weight loss, infertility and reduced production in chronic cases (Siddiki *et al.,* 2010). The significance of helminth infestation has been increased many folds in developing countries. The disease is of paramount importance due to its broad distribution and definite hosts (Rondelaud *et al.*, 2001). It is an emerging parasitic infection, having significant impacts on both veterinary and human health throughout the world (Lazara *et al.,* 2010). Fascioliasis is an economically important parasitic disease of herbivorous mammals caused by trematodes of the genus *Fasciola* that migrate in the hepatic parenchyma and establish in the bile ducts (Troncy, 1989). High prevalence of *F. hepatica* infection has been reported in dairy cattle in many countries (Mezo *et al.*, 2008).

Bangladesh has a tropical monsoon climate characterized by wide seasonal variations in rainfall, high temperatures, and humidity. The geo-climatic conditions of Bangladesh are highly favorable for the growth and multiplication of parasites. Due to the tropical climate, the causal agent *Fasciola gigantica* is prevalent in this part of the world (Amin & Samad, 1988). The prevalence of fascioliasis may differ in cattle. Earlier reports suggest around 19-53% prevalence of fascioliasis in cattle in various districts of Bangladesh (Rahman & Mondal, 1983); (Chowdhury *et al.,* 1994); (Affroze *et al.,* 2013). Fascioliasis is an economically important parasitic disease as it causes huge economic losses in terms of reduction of milk and meat and high morbidity in all ages of animals (Saleha, 1991). The disease causes considerable economic impact due to mortality, liver condemnation, reduced weight gain (up to 20%) and reduced quality and quantity (3–15% loss) of milk production (Piedrafita *et al.*, 2018). It could be zoonotic while constituting a major economic problem by lowering the productivity of cattle, in addition Hindawi Publishing Corporation to losses from condemnation of affected organs. Humans can accidentally ingest the eggs/larvae and become infected (Biu *et al.,* 2006).

However, there are major parasites of cattle in this area; therefore the present study was conduct with aim and fulfillment of the following objectives:

- To observe the overall prevalence of fascioliasis in cattle at Dinajpur district of Bangladesh.
- To determine the prevalence of fascioliasis in relation to age, sex and nutritional status.

CHAPTER II

REVIEW OF LITERATURE

CHAPTER II

REVIEW OF LITERATURE

2.1 Bovine Fascioliasis

Bovine fascioliasis (liver rot) is an economically important helminth disease caused by two trematodes viz. *Fasciola hepatica* (Linnaeus, 1758) (the common liver fluke) and *Fasciola gigantica* (Cobbold, 1855). This disease belongs to the plant-borne zoonosis. Fascioliasis is generally a disease of ruminants such as sheep, cattle and goats and is also recognized as occasional zoonotic disease of man. Fascioliasis has the widest geographic spread of any emerging vector-borne zoonotic disease and affects an estimated 17 million people in more than 51 countries, worldwide (Marcos *et al.*, 2008).

Fascioliasis causes thickening and dilation of the bile ducts and toxic degeneration of the adjacent liver tissues leading to liver condemnation at slaughter. Heavy infections cause serious disease and high mortality in cattle and sheep. Recently, fascioliasis has only been recognized as a significant human disease; studies to determine the global morbidity caused by the disease are ongoing. Chronic fascioliasis causes a chronic inflammation of the liver and bile ducts accompanied by loss of condition, digestive disturbances and a general reduction in productivity (Rana *et al.*, 2014). Depending on the disease prevalence in a herd, these reductions can be significant. The direct economic impact of fascioliasis infection is increased condemnation of liver meat, but the far more damaging effects are decreased animal productivity, lower calf birth weight, and reduced growth in effected animals (Hillyer, 2005).

2.2 History of Fascioliasis

Fasciola, amongst the helminthic parasites, does not find its description until before Ebers' Papyrus (1550 B.C.) described Guinea worm in Old Testament; Hippocrates (460-377 B.C.) described the beef tapeworm (*Taenia saginata*) and hydatid cyst (*Echinococcus granulosus)* infections and Anstali (384- 322 B.C.), the large round worm (Ascaris). The generic name Fasciola is from L. dim. of fascia, meaning a band of a fillet. Jehan de Brie recognized the Fasciola hepatica in 1379. The first report of human infection dates back to 17th century (Malpighi, 1697; Bidloo, 1697). Then specimen of Fasciola was detected in gall bladder of man during post-mortem (Fortassin, 1804; Duval, 1842; Partridge, 1846) and during operation in an eight-year-old girl in 1872. Leukart and Thomas in 1883 unravelled its life cycle and implicated the role of snails as its intermediate host. Adolph Lutz in 1892 described its mode of transmission to definitive host in Hawaii.

2.3 Taxonomy of Fascioliasis

The genus Fasciola Linnaeus, 1758 belongs to the family Fasciolidae established by Ralliet in 1895 followed by erection of subfamily Fasciolinae by (Stiles and Hassall, 1898); (Odhner, 1911) while describing the trematodes of North Africa divided the family Fasciolidae into three subfamilies, viz., 1) Fasciolinae Stiles et Hassall with the genuses Fasciola and a new genus for Dist. magnum Bassi later replaced as Fascioloides (Ward, 1917); 2) Fasciolopsinae (Odhner, 1911); and 3) Brachycladiinae (Odhner, 1905). Travassos, (1929) while analysing the family Fasciolidae conclusively divided it into four subfamilies with addition of subfamily Omphalometrinae to the above description by (Odhner, 1905).

Faust (1929) raised subfamily Brachycladiinae (Odhner, 1905) to a new family, Brachycladiidae (Faust, 1929) and along with family Fasciolidae (Ralliet, 1895) affiliated these with the superfamily Fascioloidea (Stiles, 1910). In 1930, Stunkard and Alvey briefly analysed the prevalent opinions on the taxonomy of Fasciolidae and from biological point of view reunited the families Fasciolopsidae and Campulidae as a single family Fasciolidae (Ralliet, 1895) with three subfamilies earlier suggested by (Odhner, 1911) but replacing Brachycladiinae (Odhner, 1905) by subfamily Campulinae (Stunkard and alvey, 1930).

In 1935, Skrjabin and Shul'ts, initially recognized the classification of (Stunkard and Alvey, 1930) but in the same year Ozaki, a Japanese researcher, recognized only the unified family Fasciolidae comprising four subfamilies, viz., Fasciolinae, Fasciolopsinae, Campulinae, and Nasitrematinae. Later they were convinced that these classifications could not withstand criticism at this particular juncture in the development of the taxonomy of fasciolid and reconciled with (Odhner, 1926) in elevating the subfamily Campulinae to the status of Family and suggested that these two families could be placed under single superfamily, Fascioloidea Stiles et Goldberger, 1910 affiliated with the suborder Fasciolata (Skrjabi, 1935). Further, they supported the concept of Fascioloidea formulated by (Faust, 1929) except proposal of a new family, Brachydadiidae (Faust, 1929), in place of considering the latter a synonym for the same Campulidae (Odhner, 1926).

Taxonomic Classification

Kingdom *Animalia*

- **Phylum** *Platyhelminthes*
- **Class** *Trematoda*
- **Subclass** *Digenea*
- **Order** *Echinostomiformes*
- **Family** *Fasciolidae*
- **Genus** *Fasciola*
- **Species** *F. hepatica/ F. gigantica* (Soulsby, 1965)

Figure 1: Taxonomic Classification

2.4 Epidemiology of Fascioliasis

2.4.1 Etiology

Fasciolosis is caused by two digenetic trematodes *F. hepatica* and *F. gigantica*. Adult flukes of both species are localized in the bile ducts of the liver or gallbladder. Bovine fasciolosis is a parasitic disease of cattle caused by trematodes usually *Fasciola gigantica* and rarely *Fasciola hepatica* in the tropics. The life cycle of these trematodes involves snail as an intermediate host (Walker *et al.,* 2008). Fascioliasis is a common disease of cattle and other ruminants caused by *F. hepatica* and *F. gigantica* (Rana *et al.*, 2014).

2.4.2 Geographical Distribution

F. hepatica measures 2 to 3 cm and has a cosmopolitan distribution. *F. gigantica* measures 4 to 10 cm in length and the distribution of the species is limited to the tropics and has been recorded in Africa, the Middle East, Eastern Europe and south and eastern Asia (Torgerson, & Claxton, 1999). The disease is worldwide in distribution and is liable for causing extensive economic losses to the livestock industry encompassing reductions in weight gain, milk yield and fertility (Rana *et al.*, 2014).

Special conditions are needed for fascioliasis to be present in an area, and its geographic distribution is very patchy (focal). The eggs passed in the stool of infected mammals have to develop (mature) in a suitable aquatic snail host to be able to infect another mammalian host. Requirements include sufficient moisture and favorable temperatures (above 50°F) that allow the development of miracidia, reproduction of snails, and larval development within the snails. These factors also contribute to both the prevalence and level (intensity) of infection. Infective Fasciola larvae (metacercariae) are found in contaminated water, either stuck to (encysted on) water plants or floating in the water, often in marshy areas, ponds, or flooded pastures. People (and animals) typically become infected by eating raw watercress or other contaminated water plants. The plants may be eaten as a snack or in salads or sandwiches. People also can get infected by ingesting contaminated water, such as by drinking it or by eating vegetables that were washed or irrigated with contaminated water. Infection also can result from eating undercooked sheep or goat livers that contain immature forms of the parasite. In domestic livestock in Japan, diploid ($2n = 20$), triploid ($3n = 30$) and chimeric flukes ($2n/3n$) have been described, many of which reproduce parthenogenetically. As a result of this unclear classification, flukes in Japan are normally referred to as Fasciola spp (Sakaguchi, 1980). Recent reports based on mitochondrial genes analysis has shown that Japanese Fasciola spp. is more closely related to *F. gigantica* than to *F. hepatica* (Itagaki & Tsutsumi, 1998). In India, a species called F. jacksoni was described in elephants (Singh *et al.,*1994).

2.4.3 Host Range of Fascioliasis

Fascioliasis occurs in many areas of the world and usually is caused by *F. hepatica*, which is a common liver fluke of sheep and cattle. In general, fascioliasis is more common and widespread in animals than in people. Even so, the number of infected people in the world is thought to exceed 2 million. *Fasciola hepatica* is found in more than 50 countries, in all continents except Antarctica. It is found in parts of Latin America, the Caribbean, Europe, the Middle East, Africa, Asia, and Oceania. *Fasciola gigantica* is less widespread. Human cases have been reported in the tropics, in parts of Africa and Asia, and also in Hawaii. In some areas where fascioliasis is found, human cases are uncommon (sporadic). In other areas, human fascioliasis is very common (hyperendemic). For example, the areas with the highest known rates of human infection are in the Andean highlands of Bolivia and Peru (Valero & Mas-Coma, 2000).

2.4.4 Source and Mode of Transmission

Favourable ecological conditions consisting of ideal temperature and presence of natural water sources such as ponds, lakes, streams, canals, rivers, etc., necessary for the survival of the snail intermediate hosts and presence of definitive host allows the parasite complete its life cycle. The infected definitive hosts contaminate the environment by spreading parasite eggs with their faeces. By an estimate, a sheep with mild clinical infection can contaminate a pasture with more than 500,000 eggs a day, and one with moderate infection can shed nearly 3 million eggs a day. Similarly, a moderately infected buffalo passes 7 to 10 million eggs per day. Temperatures of above 10^0 C and up to 28^0 C promote the development and hatching of miracidium in 2 weeks, which infect the appropriate snail intermediate host. The infected snail starts shedding cercariae in 4 to 8 weeks depending on temperature range. The cercariae encyst on the pasture or the aquatic vegetation or even water surface. Animals acquire infection by ingesting the metacercariae while grazing on the infested pasture or with contaminated water and aquatic vegetation (Gupta & Singh, 2002).

2.4.5 Risk Factors

Many factors enhanced the persistence of fascioliasis: The suitability of the climate and canals for the intermediate host; the resistance of metacercariae for dissociation, especially with the presence of shallow water, enough vegetation, and/or humidity; and continued exposure of the animals to encysted metacercariae, grazing habits, and movement between the infected and treated localities (El-Bahy, 1998).

Climatic factors are of supreme importance influencing epidemiology of Fascioliasis. (Claxton *et al.*, 1997; Rangel-Ruiz *et al.*,1999; Phiri *et al.*, 2005; Ansari-Lari and Moazzeni, 2006).

2.4.6 Incidence of Fascioliasis

Fascioliasis is a parasitic disease in ruminants that can cause major economic losses. In Africa, fascioliasis is a serious problem in the humid and subhumid zones (Daynes, 1969; Ogunrinade and Ogunrinade, 1980). In the arid and semi-arid zones, where climatic conditions are less favourable for liver flukes, fascioliasis incidence has until recently been comparatively low. However, the increasing number of dams and irrigation canals built to boost energy and food production has also increased the number of potential snail habitats, and with them the danger of liver fluke infestation. Hammond (1973) and Graber (1976) reported that sheep are very susceptible to acute fascioliasis and that its periodic outbreaks cause high economic losses. Once infected with flukes, goats are also very susceptible to the disease. In cattle, acute fascioliasis often remains undetected and develops into chronic fascioliasis. (Ollerenshaw and Rowlands, 1959)

The probable reasons of increased infection rate of Fascioliasis may include, (i) development of resistance due to improper use of *Fasciolicides* including frequent use of same drug for a longer time with inappropriate doses (Boray, 1990; Fairweather and Boray, 1999) (ii) Lack of regular evaluation of local available drugs against any parasitic disease or no use of specific drug against any parasitic disease (Jabbar *et al*., 2006). (iii) Socio-economic status of the farmers to treat the nuisance (Jabbar *et al*., 2006). The probable reason for highest prevalence in winter might be the availability of optimal conditions of environment for the transmission, growth and development of parasitic life cycle stages (Rowcliffe and Ollerenshaw, 1960). This includes temperature ranging from 23-26 C^0 for development of eggs (Rowcliffe and Ollerenshaw, 1960; Thomas, 1883) and maximal growth of snails (Kendall, 1953) and humidity level up to 90 % caused by plenty of water available facilitates embryonation (Andrew, 1999), emergence of miracidium from eggs due to increased activity of cilia (Thomas, 1883 a,b) and liberation of cercariae from snails (Alicata, 1938; Dixon, 1966). However, *Fasciolosis* was recorded throughout the year (Maqbool *et al*., 2002).

2.4.7 Prevalence of Fascioliasis

A study was conducted during the period from October, 2016 to January, 2017 to determine the prevalence of bovine fascioliasis at Mirzaganj upazilla under the Patuakhali district in Bangladesh. A total of 92 bovine cases was recorded as study population for the present study. Tentative diagnosis of fascioliasis was made on the basis of history, clinical signs, physical examination findings; whereas confirmatory diagnosis was made on the basis of coprological examination. The overall prevalence of fascioliasis was 44.57%. The age of the study population (cattle) were divided into three groups i.e. □1-2 years; 2-4 years; 4-6 years and their prevalence (%) of fascioliasis were found 47.83%, 41.37% and 41.17% respectively. The higher prevalence of fascioliasis was recorded in female 52.83% followed by male 33.33%. Among the study population, the highest prevalence was recorded in cross breed (60%) cattle than local or indigenous (42.68%) cattle (Howlader *et al.,* 2016).

The main objectives of a study were to determine the prevalence of fascioliasis infections in cattle and buffaloes, slaughtered in El-Kharga city slaughterhouse at New Valley Governorate. The slaughtered animals were daily inspected for liver fascioliasis all over 2016. Macroscopic fascioliasis was detected from a total of 2251 basing on animals specie, sex, season, and Fasciola spp. in addition to microscopic examination of blood, fecal samples which collected from female cattle and buffalo (50 each). The total prevalence rate of Fasciola sp. infection occurs in the study area were about 695/2251 (30.88%) from the total cattle and bovine slaughtered carcasses. The incidence of fascioliasis was 4/12 (33.33%) and 678/2200 (30.82%) for females and males cattle carcasses, respectively, while the infection rate in buffalo carcasses was 1/4 (25.00%) and 12/35 (34.29%) for females and males buffalo carcasses, respectively. The moderate fasciolosis infection in cattle and buffaloes slaughtered at the municipal abattoir of El-Kharga, Egypt. The highest fascioliasis infection was recorded during winter and autumn. It constitutes a major cause of economic losses at El-Kharga abattoir and threat public health (Elshraway & Mahmoud, 2016).

In adult cattle, the infection usually takes a chronic course, with no obvious clinical signs. Significant production losses occur in the herds having a prevalence of *F. hepatica* infection of 25 % or above (Vercruysse and Claerebout, 2001). The cercariae of liver flukes were observed from a pond first time by Otto Muller in 1773 (Andrews, 1999). A large variety of animals, such as sheep, goats, cattle, buffalo, horses, donkeys, camels and, rabbits, show infection rates that may reach 90% in some areas (Farag, 1998). The snail usually habitat along the edges of stagnant ponds, marshy lands and ditches which may be a reason of increased prevalence of Fascioliasis in the animals bathing in stagnant water (Ulmer, 1971; Saladin, 1979). Both *F. hepatica* and *F. gigantic* are prevalent in Pakistan (Maqbool *et al*., 1994, 2002; Siddiqui and Shah, 1984; Chaudhry and Niaz, 1984; Masud and Majid, 1984; Sahar, 1996).

The prevalence of fascioliasis in cattle slaughtered in the Sokoto metropolitan abattoir was investigated. Faeces and bile samples were collected and processed using formal ether concentration technique. Gross lesions from 224 out of 1,313 slaughtered cattle were randomly selected and examined. Out of the 224 cattle examined, 95 (42.41%) were males and 129 (57.59%) were females. Out of 95 male cattle examined, 27 (28.42%) were infected and out of 129 females 35 (27.13%) were infected. Based on breed, infection rates were 31 (31.0%), and 31 (25.2%) for breeds of Sokoto Gudali and Red Bororo respectively. No infection was recorded in White Fulani breed. Lesions observed were more in males than in females and more in Red Bororo than in Sokoto Gudali. Overall, prevalence of infection with Fasciola was 27.68%. There was no statistically significant association between infection and breed and between infection and sex of the animals sampled ($P > 0.05$), Regular treatment of all animals with an effective flukicide, as well as snail habitat control, tracing source of animals, public enlightenment about the disease, proper abattoir inspection, adequate and clean water supply to animals, and payment of compensation of condemned tissues and organs infested with the parasite by government were suggested (Magaji *et al.*, 2014).

2.5 Pathogenesis

Pathogenesis in fascioliasis commences with juvenile fluke entering the hepatic tissues. The course of pathogenesis in different hosts is similar but may vary in severity with the number of metacercariae ingested, the species involved and the stage of the parasitic development. The juvenile flukes migrate in the parenchyma extensively and cause traumatic lesions with haemorrhage and inflammation. Lesions produced by *F. gigantica* are more severe with fewer flukes as compared to *F. hepatica*, which may be attributed to the longer duration of migration in hepatic parenchyma, larger size and spines present all over the tegument of the former species (Ogunrinade, & Anosa, 1981).

2.6 Pathology

The disease fascioliasis is by most authors clinically described to occur in three phases, viz., acute, sub-acute and the chronic. The acute phase commences between 2-6 weeks after ingestion of substantial number of metacercariae and the species involved. In *F. hepatica* it may be 2000 metacercariae for sheep while for *F. gigantica* 300 metacercariae can produce similar condition. In large ruminants, buffalo and cattle, ingestion of 1000 metacercariae of *F. gigantica* produces acute disease clinically. The acute phase is characterized by severe haemorrhage caused by the migrating juvenile flukes in the hepatic tissue rupturing the blood vessels. The liver parenchyma, particularly the ventral lobe associated with gall bladder, is severely damaged assuming an uneven surface covered with blood clots. In late stage between 11-25 weeks of infection depending on the species of parasite and host involved, the animal squats in a specific posture on its right abdomen resting the ground and head turned to opposite direction. The animal disinclines to stand or move and feed. Between 3-7 days the condition worsens and the animal lays flat with extended body, blood froth may ooze out through mouth and nostrils before death. At necropsy, the liver is enlarged, haemorrhagic covered with fibrous clots and necrotic tunnels with migrating flukes. The ventral lobe is covered with fibrous exudates and from sub capsular haemorrhage the blood stained fluid pass into abdominal cavity with ascitis. In small ruminants multiple adhesions of adjacent organs with liver are present. Sheep may die suddenly without exhibiting any clinical manifestations, or following weakness, anorexia and pain. (Freeland, 1976).

The sub acute form of disease results when the host ingests moderate doses of metacercariae over a longer period and there in influx of different migratory stages of flukes in the liver. While some have reached bile ducts and caused cholangitis other are still migrating tissues like that of acute disease but of less severity. Liver shows enlargement and haemorrhagic tracks all over the surface and in substance. Rupture of sub capsular haemorrhage is rare. The animals show weight loss and there is accumulation of fluid in the abdomen causing ascites with submandibular and facial oedema. Other clinical manifestations include anaemia, hypoalbuminaemia, eosinophilia and elevated alanine aminotransferase (ALT) and aspartate tgransaminase (ASL) serum levels. Animals, particularly sheep harbouring *Clostridium novyi* in liver, after invasion with juvenile fluke can lead to necrotic *hepatitis* called 'black disease' with fatal consequences. Chronic fascioliasis is manifestation caused by infection with moderate number of metacercariae. It is the most common form of infection in animals and also man. Liver pathology consists of progressive biliary cirrhosis; bile ducts are prominent, thicken, fibrous and may be calcified. Fibrosis is as a result of repair of migratory tracks and cholangitis. Bile ducts containing flukes are dilated, filled with fluke eggs, blood and tissue cells. The fluke spines embed in the epithelium resulting in hyperplasia. In later stage, encrustation of calcium form complete cast of bile duct. At necropsy the liver is pale and hard with irregular outline. The liver pathology is characterised by hepatic fibrosis and hyperplastic cholangitis. There is progressive loss of condition. Anaemia and hypoalbuminaemia results in submandibular oedema and ascites. Anaemia is hyppchromic and macrocytic accompanied with eosinophilia. Fasciola eggs are demonstrated in faeces. In milder infections the disease may go un-noticed but effect of production could be significant due to inappetance and effect on post absorptive metabolism of protein, carbohydrate and minerals. Acute Fasciolosis causes huge economic loses as directly or indirectly in terms of anemia due to its ability to suck blood to the extent of 0.2-0.5 ml per day and decrease in the total proteins especially albumin (Soulsby, 1987)

2.7 Immunology

A number of studies indicated the mechanism of resistance at the gut wall is thymus-independent and that non-specific and hypersensitivity reactions may play a role. The juvenile flukes migrating through intestinal wall and peritoneum induce infiltration of eosinophils, IgG1 and IgG2 antibodies around the parasites. Protection to *F. hepatica* in rats has also been associated with accumulation of eosinophils and IgE sensitised cells in the gut wall and juvenile antigenspecific IgG1, induced in early infection (Tliba *et al.*, 2000).

Contrarily, sheep seems to show no resistance to reinfection with *F. hepatica* despite of large infiltration of white blood cells in liver and production of antibodies to the parasite and fibrosis in the liver8. In cattle, elimination of primary *F. hepatica* infection at 20-28 weeks coincided with highest level of homocytotropic antibodies whereas drug abbreviated *F. hepatica* infection induced resistance resulting in insignificant liver fibrosis (Hoyle *et al.*, 2003). Study knowledge about prevalence, diagnosis, treatment and control of Fascioliasis has been reviewed. This article evaluates more recent work along with previous studies. The fecal egg count, signs / symptoms and specific antibodies in serum were the only diagnostic tools in the past however now for detection of *F. hepatica* specific copro-antigen has been developed and commercialized. An indirect enzyme linked immunosorbent, assay (IEA) allows an early diagnosis. The tracer animals and snail studies have widened the existing knowledge. The treatment has been carried out mainly with Fasciolicides (Albendazole, Oxyclozanide and Triclabendazole), however resistance has been developed. Most of the recommended treatments are not feasible. Seasonal deworming is essential. Pasture management by creating bio competent environment with snail predators can be very effective in reducing the rate of incidence and controlling the problem. Vaccine is there but is not frequently used due to incompatible immune response. The studies on reduction in milk yield due to Fascioliasis are still lacking and require extensive research/ investigations (Rana *et al.,* 2014).

Suggesting protective immunological response reducing migration of flukes into the hepatic tissue and bile ducts. Further studies have suggested a correlation between the IgG2 response to fluke proteinases (cathepsin L) vaccine and protection in cattle, whereas the IgG1 response was correlated with susceptibility and positively regulated by IL4. The elevated parasite-specific IgG1 but low IgG2 levels in infected cattle is consistently observed (Estes DM, Brown WC 2002).

Fasciola evade the host immune attack by frequently sloughing the tegumental associated glycocalyx layer and target of host antibody-mediated eosinophils, neutrophils or macrophages attacks (Piedrafita *et al.,* 2001).

Other Fasciola isoenzymes viz., cysteine proteases (cathepsin L and B) are considered to play main role in tissue invasion and immune evasion (Mulcahy & Dalton, 2001). The cathepsin Ls can 1) degrade both extra cellular matrix (fibrillar types I and II collagen) and basement membrane (type IV collagen) aiding in parasite tissue invasions; 2) degrade haemoglobulins rendering it to the parasite for nutritional purposes and help in parasite survival, and 3) cleave immunoglobulins in the hinge region (Berasain *et al.*, 2000) preventing antibody mediated attachment of eosinophils, neutrophils and macrophages. Recently secretory products of Fasciola rich in cathepsins has been shown to suppress T cell proliferation in sheep in vitro (Prowse *et al.*, 2002).

2.8 Diagnosis

The diagnosis of Fasciolosis is based on the detection of eggs in feces or *F. hepatica* specific antibodies in serum. Recently, a method based on detection of *F. hepatica* specific copro-antigen has been developed and commercialized (Mezo *et al.*, 2004). The sensitivity (Se) and specificity (Sp) of these tests have been determined after experimental infection (Cornelissen *et al.*, 2001) or by using two distinct populations, a positive population selected from an enzootic area and a negative population from a fluke free area. (Ibarra *et al.*, 1998; Mezo *et al.*, 2004; Salimi- Bejestani *et al.*, 2005). The current diagnostic tests for Fasciolosis in cattle are qualitative only, yet the level of infection is considered an important factor in determining production losses. (Dargie, 1987; Vercruysse and Claerebout, 2001).

The current trend in the diagnosis of more common bovine diseases, including Fascioliasis is to use the same samples of milk that are collected on farm for routine monitoring of animals productivity and quality of milk (Hill *et al.,* 2010; Mars *et al*., 2010), reducing the associated costs and disturbance to animals as a result of handling sampling. The MM3-SERO ELISA is a sensitive and highly specific test for the sero-diagnosis of cattle Fasciolosis and can be reliable to use with milk samples. It is an excellent method of estimating with in herd prevalence of infection when used with bulk samples (Mezo *et al*., 2009, 2010). The immune enzymatic techniques as indirect ELISA have been found very suitable for the diagnosis of Fasciolosis due to their high sensitivity and the possibility of many sera samples (Arriaga *et al*., 1989). These techniques based on detection of antibodies have been successfully utilized to detect early infection (Oldham, 1985; Hillyer and Sole, 1991; Poitou *et al*., 1993; Paz *et al*., 1998).

Long persistence of high levels of immunoglobulin, even though animals have been successfully treated, makes interpretation more difficult (Ibarra *et al*., 1998), (Langley and Hillyer, 1989) in detected antigenemia as early as 2 weeks after infection in cows. It is concluded that it is very important and useful to combine two enzymatic assays, indirect and direct ELISA, to achieve a more reliable knowledge of the real infection status of the host. Results of iELISA using different antigens of *F. gegantica* for detecting antibodies against Fasciola in sera may be used in cattle. The diagnostic sensitivity, specificity and accuracy of the assay can be calculated according to Timmreck, 1994 and Smith, 1995**.** During migratory phase of infection, *F. hepatica* antigens are available to the immune system, and it is possible to detect them by serologic probes as sandwich-enzymelinked immunosorbent assay SEA (Langley and Hillyer, 1989).

When the parasite is established in the bile ducts less antigen is there available to the immune system, and its detection must be directed to fecal or bile samples. It has been demonstrated that most of pathological damage takes place when flukes are migrating through peritoneal cavity and liver parenchyma before their establishment in the bile ducts. It is very important to use early diagnostic techniques to reduce the great losses in cattle. An indirect-enzyme immune-linked immunosorbent assay IEA allows an early diagnosis of *Fasciolosis*. *F. hepatica* antibodies can first be detected by indirect-ELISA between 3 and 6 weeks after infection during the liver migratory phase of immature worms (Marin, 1992).

2.9 Treatment

Several drugs are effective for fascioliasis, both in humans and in domestic animals. The drug of choice in the treatment of fasciolosis is triclabendazole, a member of the benzimidazole family of anthelmintics (Savioli *et al.*, 1999). The drug works by preventing the polymerization of the molecule tubulin into the cytoskeletal structures, microtubules. Resistance of *F. hepatica* to triclabendazole has been recorded in Australia in 1995 (Overend & Bowen, 1995) and Ireland in 1998 (Mulcahy & Dalton, 1998). Praziquantel treatment is ineffective (Schubert & Phetsouvanh, 1990). There are case reports of nitazoxanide being successfully used in human fasciolosis treatment in Mexico (Rossignol *et al.*, 1998).

2.10 Prevention and Control

Hygienic feeding habits are the best preventive measures for keeping parasitic infections. So is true for fascioliasis. Besides promoting public health awareness through education in endemic areas are proving to be increasingly effective in prevention of fascioliasis diseases these days. So far fascioliasis is concerned, avoiding of watercress plants, water chestnuts and vegetation grown in aquatic environment, consuming raw has far reaching consequences in endemic areas. Thorough washing of edible aquatic vegetation with running water can get rid of metacercariae to great extent and treating vegetation with 1% citric acid, 1% acetic acid (12% commercial vinegar), liquid soap (1.2%) or potassium permanganate solution (24 ppm) kills and detaches all metacercariae(Pan American Health Organization, 2003).

In country like Bangladesh and elsewhere too, providing animals' clean water to drink and prevent these enter into natural water reservoirs infested with snails shall greatly reduce the risk of acquiring fascioliasis. Ensiling of animal feed such as paddy straw likely to be infested with metacercariae kills not only the deposited metacercariae (Gupta & Kamra, 1987). But also increase the digestibility and nutritive quality of the feed. Elimination of snails from environment shall break the life cycle of the parasite. This can be achieved by application of molluscicides like copper sulphate, bayluside, trifenmorf with caution because this is not always possible due to environmental objections. Cleaning of ponds, ditches, canals etc., of unwanted aquatic vegetation can drastically reduce the mollusc population and almost eliminate all the molluscan eggs. Plant origin molluscicides, which degrade into simpler components within short period of application, may also be fruitful (Gupta *et al.*, 1988).

Biological methods using predators (carnivore fishes, ducks, etc.), parasites of snails can also be used effectively. The other measure for control of parasitic infections is vaccination. Since success achieved in development of vaccine against the helminth Dictyocaulus viviparous, the lung worm of cattle, in 1960s, researchers have been engaged to explore the field with other helminth parasites also. The approach for vaccine development against fascioliasis has not been much different so far. Initial trials consisted of vaccine using radiation-attenuated metacercariae followed by crude somatic parasite extracts and mixtures of secreted parasite proteins (Haroun & Hillyer, 1986).

Since then a number of parasite proteins identified and characterized from both *F. hepatica* and *F. gigantica* have been tested in vaccine trials with partial success in different laboratory and natural animal hosts. The current vaccine strategy implies the immunological mechanism by which the parasite metabolic products (enzymes) variously termed as ‘defined, natural and conventional’ antigens aiding in invasion, survival and evasion of host immune responses are neutralized rendering the parasite helpless. Such parasite proteolytic enzyme agents known as defined molecular candidate vaccine agents are being looked forward against Fasciola with bright prospects. Vaccination of cattle with same enzyme resulted in 50% reduction of worm burden but when the candidate vaccine agent was combined with fluke haemoglobin the protection was enhanced to 70%, and 100% failure of eggs to embryonate shed by such vaccinated animals. Contrarily, cathepsin L from *F. gigantica* failed to provide similar protection in cattle to homologous infection. The basic concept is that if such vaccination is successful it can reduce parasite transmission enormously. Lastly but not the least, prevention of human infection by avoiding consuming metacercariae is much discussed above. Measures are many but to follow these require patience and concern. Otherwise, human infection is like meeting a fatal accident as a result of careless driving. Treatment of animals with broad-spectrum fasciolicide would prevent contamination of environment with parasite eggs and subsequently infection of intermediate snail host and contamination of environment with metacercariae. On the basis of strategies used to identify these defined candidate vaccine molecules of *F. hepatica* and *F. gigantica*, three prototype antigens have been identified These are: (1) The fatty acid binding proteins (FABP), a cross protective antigen of *F. hepatica* recognized by cross reactive antibodies raised against the trematode Schistosoma mansoni, rendering cross protection against S. mansoni. (2) The glutathione stransferase (GST), a Fasciola molecule homologous to antigen previously shown to protect animals against S. mansoni and S. japonicum infection. (3) Cathepsin-L, the essential Fasciola molecules, constituting of cysteine proteinases and predicted to perform functions essential for parasite growth and survival. Under this category, *F. hepatica* cysteine proteinase recognized by the host post-infection inducing high levels of protection of up to 70% in sheep vaccinated with the purified cystiene proteinases (Spithill *et al.*, 1997).

CHAPTER III

MATERIALS AND METHODS

CHAPTER III

MATERIALS AND METHODS

3.1 Study Area

This study was design to focus on the prevalence of fascioliasis in cattle at Dinajpur district of Bangladesh; the experiment plan was prepared in the Department of Pathology and Parasitology, Faculty of Veterinary and Animal Sciences, Hajee Mohammed Danesh Science and Technology University (HSTU), Dinajpur, Bangladesh.

Figure 2: Map of the Area study

3.2 Duration of the study

This study was conducted during the period of July to December, 2018 in parasitology Laboratory, at Hajee Mohammad Danesh Science and Technology University.

3.3 The major work of the present study

- Collection of faecal sample directly from the rectum by using hand in selected cattle at the area of the study
- Preservation and immediate shipment of collected samples to the Laboratory.
- Investigation of the prevalence of cattle fascioliasis in relation to Age, Sex, and nutritional status.

3.4 Test material

- Protective apron, gloves and mask.
- Fresh faecal sample from rectum.
- Vial
- Mark pen
- Formalin
- Note Book
- Pen
- Water or Flotation Solution
- Beakers containers
- Pasteur pipettes
- Test tubes
- Centrifuge Machine
- Slides, Cover slips & Microscope

3.5 Selection and grouping of animal

A total of 100 cattle were selected purposefully from Uttar Sadipur, Nandoir, Kornai, and Vatapara in Dinajpur district under Northern region of Bangladesh. Those animals were considered for the present study as study population. Study population was divided into three age groups i.e. young (6 month-1year); adult (1-2 years); old (2- above years) on the basis of owner record and dental formula. Their sex was divided into (62 Male and 38 Female) and also their nutritional status was divided into two healthy or poor healthy on the basis of body condition score (BCS), under BCS cachectic, poor and overweight considered as poor healthy, while medium and slightly fatty considered as healthy ones. Epidemiological data were recorded after history, physical, clinical and coprological examination.

3.6 History, Physical and Clinical Examination

History along with other necessary information's was taken from individual farmers by cross questioning. History included Age, sex, breed, clinical signs and location of cattle pen and previous history of fascioliasis. Physical examination was done for each cattle in the study area. Simultaneously faecal sample was collected directly from the rectum.

3.7 Coprological Examination

About 10-20 grams of feces were collected from each animal. Collected samples were transferred to a vial containing 10% formalin and labeled properly. The fecal sample was examined using standard direct smear, floatation and sedimentation methods of faecal sample examinations.

3.7.1 Direct smear method

- Small amount of faeces was placed on clean slide
- Thin smear preparation adding 1-2 drop of water
- Separating and discarding of coarse particles
- Placing a cover slip on prepared smear
- Observation under microscope

(Soulsby, 1965)

3.7.2 Sedimentation method

- A small amount of faeces was mixed with water in a flask
- Separation of coarse particles with a sieve
- Keeping for sedimentation
- A small amount of sediment was placed on a clean slide
- Put a cover slip on the drop
- Observed under microscope

(Soulsby, 1965)

3.7.3 Floatation method

- A small amount of faeces was mixed with saturated salt solution in a test tube
- Adding additional solution upto tip of the tube
- Keeping the tube in standing position for few minutes
- Touching a clean, dried slide to top of the solution
- Put a cover slip on the slide
- Observed under microscope

(Soulsby, 1965)

3.8 Prevalence study

3.8.1 Overall prevalence

Overall prevalence was calculated by following formula:

Prevalence = No. of infected cattle ÷ No. of observed cattle x 100

3.8.2 Age-wise prevalence

The age were divided into three young (6 months – 1 year), adult (1-2 years) and old (2 and above years).

3.8.3 Sex-wise prevalence

62 were male and 38 were female

3.8.4 Prevalence based on nutritional status

Nutritional status divided into poor healthy and healthy on the basis of body condition score (BCS)

3.9 Experimental layout

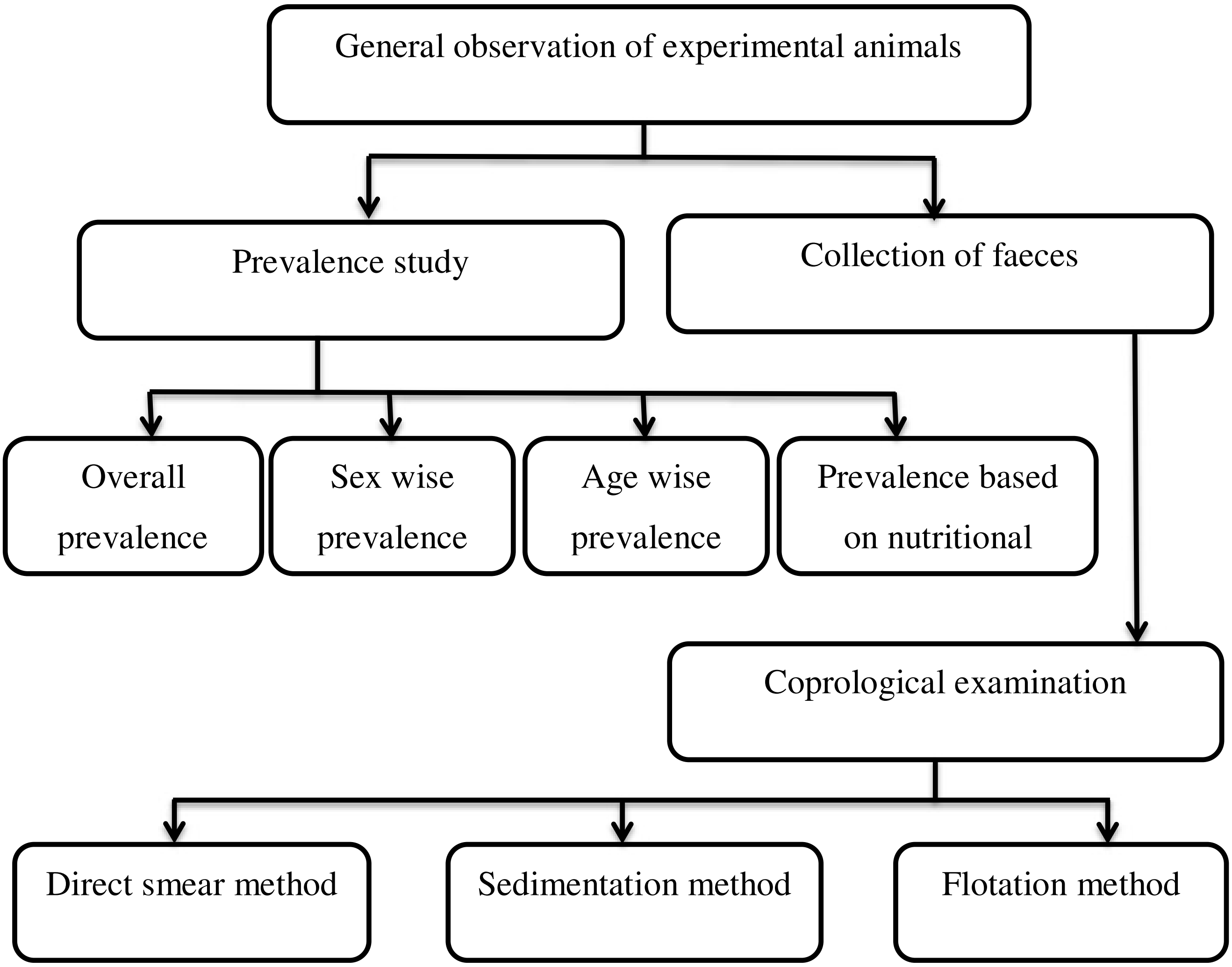


Figure 3: Flow chart

3.10 Collecting faecal samples from the rectum

Figure 4: Collecting feacal sample directly from the rectum

3.11 Parasitological Findings (*Fasiola egg present in the faeces*)

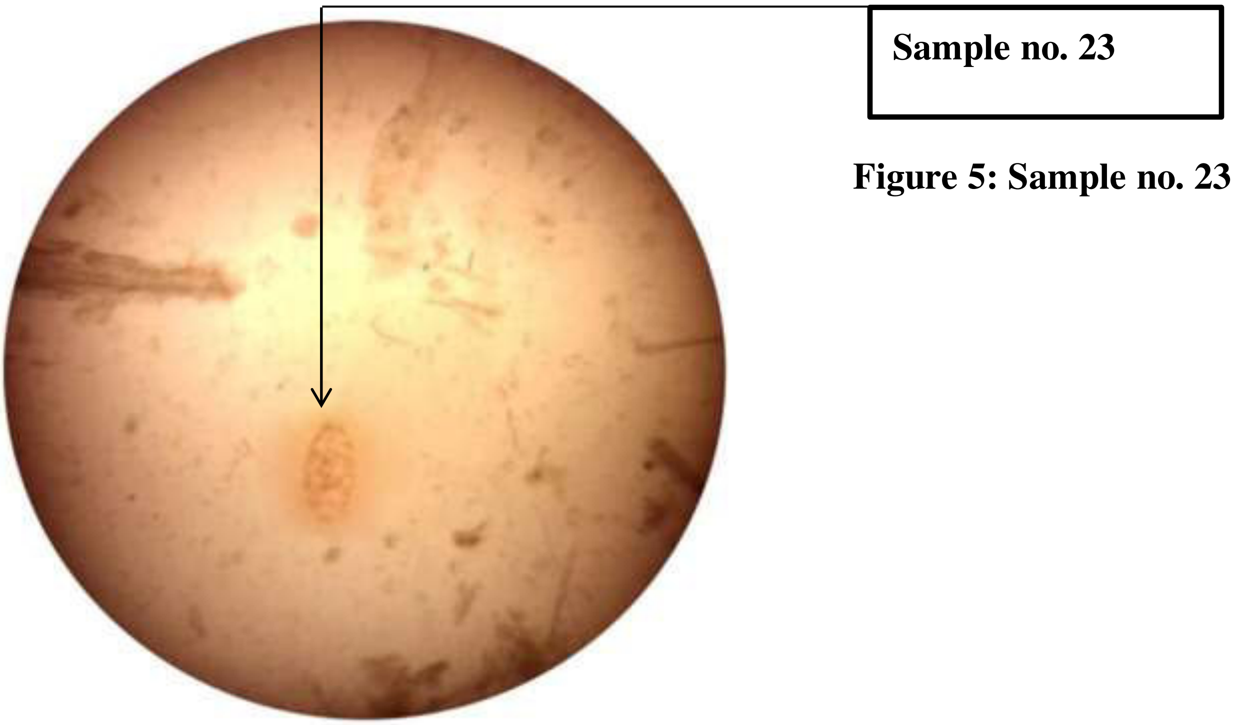


Figure 5: Sample no. 23

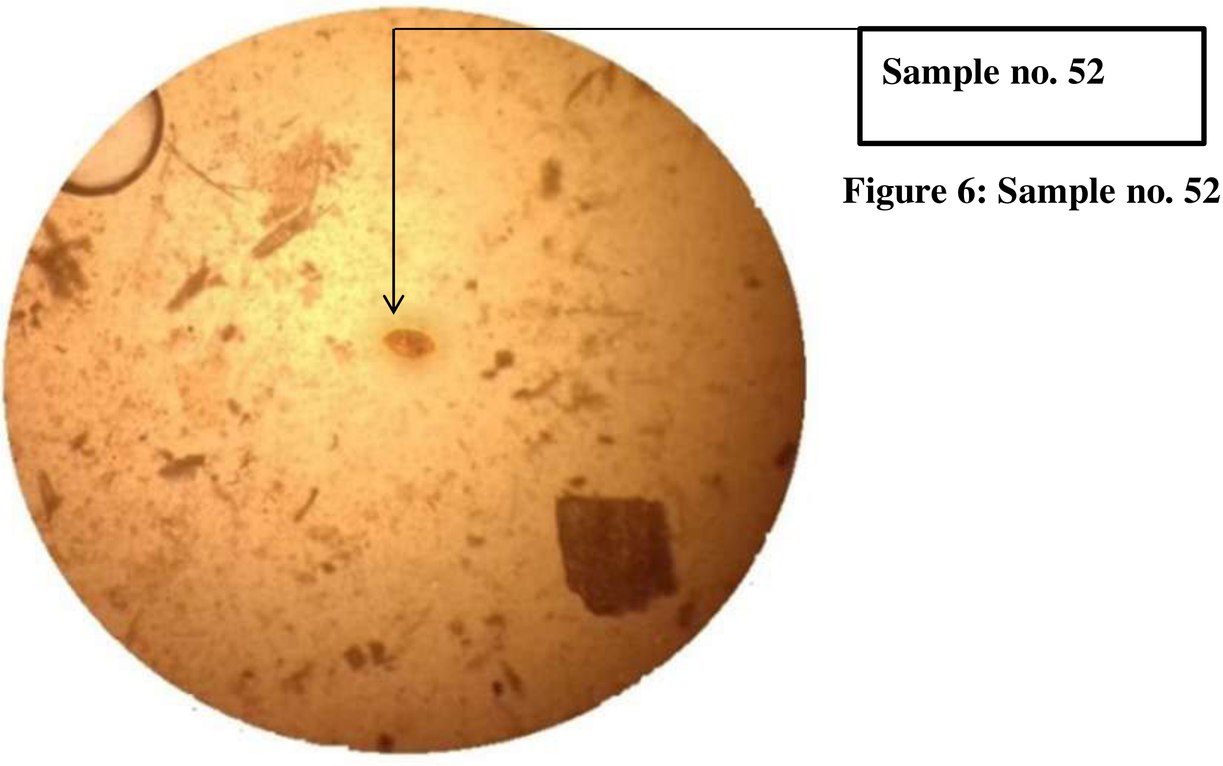


Figure 6: Sample no. 52

3.12 Diagnosis of fascioliasis

Tentative diagnosis was made on the basis of history, physical and clinical signs (depression, dullness, in appetite, rough hair coat, diarrhea, emaciation, bottle Jaw appearance formation). Confirmatory diagnosis was made on the basis of coprological examination findings; Fasciola eggs were confirmed by the characteristics of oval shaped, eccentric morulla, operculum present, yellow brown in color (Valero *et al*., 2009). Presence of single eggs of *Fasciola sp*. in one microscopic focus during coprological examination was recorded as positive for fascioliasis.

3.13 Statistical analysis

The data was recorded systematically and analyzed statistically using SPSS version 20.

CHAPTER IV

RESULTS

CHAPTER IV

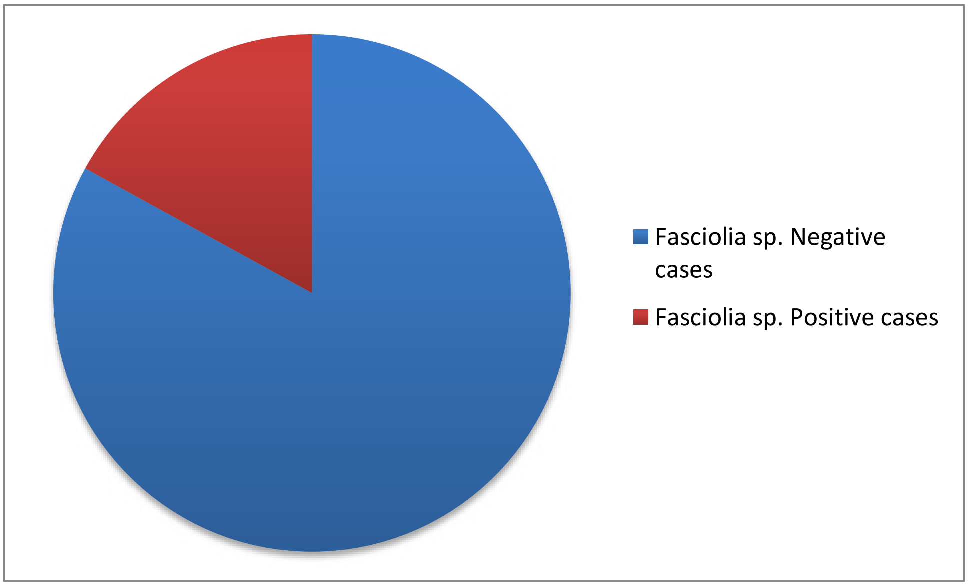
RESULTS

4.1 Overall prevalence of fascioliasis

A total number of 100 faecal samples were examined and out of 100 samples, 17 samples were positive. The result of the present study revealed that the overall prevalence of fascioliasis was 17%. (Table 1).

Table 1: Overall prevalence of fascioliasis in cattle

No. of cases recorded	*Fasciolia* sp. Negative cases	*Fasciolia* sp. Positive cases	Prevalence (%)
100	83	17	17



Graph 1: Overall prevalence of fascioliasis in cattle

4.2 Prevalence of fascioliasis on the basis of age

Study population of cattle was divided into three age groups i.e. young (6 month-1year), adult (1-2 years) and old (2 and above years). It is found that age had significant ($P < 0.05$) effect on the prevalence of fascioliasis in cattle. The highest prevalecne of fascioliasis was observed in the cattle group ages among old (24.14%) followed by adults (8.7%) and young (5.26%). In the present study, the odd ratio of Adult vs. Young (1.5) implied that the adult was 1.5 times more susceptible than young. Also the odd ratio of Old vs. Young (5.1) implied that the old was 5.1 times more susceptible than young. And also the odd ratio of Old vs. Adult (3.34) implied that the old was 3.34 times more susceptible than adult. (Table 2).

Table 2: Age wise prevalence of fascioliasis

Age groups	No. of Observed	No. of infected	Prevalence (%)	Odd ratio
Young (6 month-1year)	19	1	5.26	Adult vs. Young=1.5 Old vs. Young = 5.1 Old vs. Adult = 3.34
Adult (1-2 years)	23	2	8.7	
Old (2 and above years)	58	14	24.14	
Total	**100**	**17**	**17**	
Chi-square Value	**8.244**			
P-value	**0.016***			

* means significant at 5% of level of significance ($P<0.05$)

4.3 Prevalence of fascioliasis on the basis of sex

The present study revealed that the prevalence of fascioliasis was significantly (P<0.05) affected by sex. In the present study the prevalence of fascioliasis of cattle in female was higher (28.95%) than in males (9.68%). In the present study, the odd ratio of female vs. male (3.80) implied that the female was 3.8 times more susceptible than male. (Table 3).

Table 3: Sex wise prevalence of fascioliasis in cattle

Sex group	No. of observed	No. of infected	Prevalence (%)	Odd Ratio
Male	62	6	9.68	Female vs. Male=3.80
Female	38	11	28.95	
Total	**100**	**17**	**17**	
Chi-square Value	**6.20**			
P-value	**0.013***			

* means significant at 5% of level of significance (P<0.05)

4.4 Prevalence of fascioliasis on the basis of nutritional status.

Study population of cattle was divided into two nutritional status groups (healthy and poor healthy). The present study revealed that the prevalence of fascioliasis was significantly ($P<0.05$) affected by nutritional status. The prevalence of fascioliasis in poor healthy was higher (22.39%) than in healthy (6.06%). In the present study, the odd ratio of poor healthy vs. healthy (4.47) implied that the poor healthy were 4.47 times more susceptible than healthy (Table 4).

Table 4: Nutritional status wise prevalence of fascioliasis in cattle

Nutritional status groups	No. of observed	No. of infected	Prevalence (%)	Odd Ratio
Healthy	33	2	6.06	Poor vs Healthy=4.47
Poor healthy	67	15	22.39	
Total	**100**	**17**	**17**	
Chi-square Value	**4.177**			
P-value	**0.041***			

* means significant at 5% of level of significance ($P<0.05$)

CHAPTER V
DISCUSSIONS

CHAPTER V

DISCUSSIONS

5.1 Overall prevalence of fascioliasis

After examination of 100 cattle, 17 samples were positive. The result of the present study revealed that the overall prevalence of fascioliasis was 17%.

The overall prevalence of fascioliasis was 14.8% (Chakraborty and Prodhan, 2015); the overall prevalence of fascioliasis in the Nile Delta region of Egypt was 9.77% (Abdelgawad *et al.,* 2017); the overall prevalence of fascioliasis in cow was 25% (Sumbal Haleem *et al.,* 2016); the overall prevalence of fascioliasis in cattle was 51.0% (Yadav *et al.,* 2015); the prevalence of fascioliasis in cattle was 44.8% (Abraham *et al.,* 2014) and the overall prevalence of fascioliasis was 66.14% (Karim *et al.,* 2015).

This findings is nearly supported by Chakraborty and Prodhan (2015), Abdelgawad *et al.,* (2017) and Sumbal Haleem *et al.* (2016). But Yadav *et al.* (2015), Abraham *et al.* (2014), and Karim *et al.* (2015). Those results disagree with the present study results.

The variation with the findings of the present study was very high; it might be due to location, use of anthelmintic, session and duration of the study. Prevalence of fasciolosis in cattle is attributed by multi-factorial risk factors which comprise host, parasite and environmental effects. High-rainfall areas favour development and survival of both the intermediate host snail and the developmental stages of the parasite Affroze S. *et al.* (2013). This variation might be due to the variation on sample size and sampling, nutritional status, geographical location such as grazing on low lying areas is an important predisposing cause of Fasciola infestation Khatun *et al.* (2015); Tembely *et al.* (1995).

5.2 Fascioliasis associated with age

In the present study the cattle age was divided into three age groups i.e. young (6 month-1year), adult (1-2 years) and old (2 and above years). It is found that age had significant ($P < 0.05$) effect on the prevalence of fascioliasis in cattle. The highest prevalence of fascioliasis was observed in the old (24.14%) followed by adults (8.7%) and young (5.26%).

The bovine fasciolosis was significantly ($p<0.01$) higher in old cattle (76.43%) compared to adult (68.69%) and young (48.62%), (Karim *et al.,* 2015); The highest level of infection was found in older group i.e., above 6 years (62.62%) followed by in age groups of 4-6 years (57.28%), 2-4 years (42.56%) and up to 2 year (17.87%), (Bhutto *et al.,* 2012); The prevalence of *Fasciola gigantica* were highest in cattle of more than 36 months of age and lowest in the age of less than12 months. Khandaker *et al.* (1993); The prevalence of *F. gigantica* was 7.2% in adult cattle; where 3.9% in young (Sumbal Haleem *et al.,* 2016); Young (6 to 18 months) are more infected compared to adult animals (Nath *et al.,* 2016) And the highest prevalence was observed in <2 age group (10.91%), and the lowest was >3 age groups (8.35%), (Abdelgawad *et al.,* 2017).

The result of the present study was nearly similar to the study of Karim *et al.* (2015), Bhutto *et al.* (2012), Khandaker *et al.* (1993), and Sumbal Haleem *et al.* (2016). All of those agree that the old cattle is higher prevalence than young and adult. But the results of Nath *et al.* (2016), and Abdelgawad S. *et al,* (2017) disagree the present study.

The findings of the present study were varying from previous study findings. In present study old (2 and above years) cattle was found more susceptible to fascioliasis, this might be due to old are comparatively more susceptible than the young and adult to be infected with intestinal parasite, it might because old cattle are frequently graze on the field so they have much more exposure on circulating circariae and metacercariae.

5.3 Fascioliasis associated with sex

This study revealed that the prevalence of fascioliasis of cattle in female was higher (28.95%) than in males (9.68%).

The higher prevalence of fascioliasis in female 52.83% followed by male 33.33% (Howlader *et al.,* 2017); The female cattle 41.36% are highly susceptible than male 13.85% (Affroze *et al.,* 2013); The female cattle 70.3% are highly susceptible than male 55.23% (Karim *et al.,* 2015); The infestation of fasciola sp is more in female cattle 52.2% than male 47.8% (Nath *et al.,* 2016); and the male 14% is highly susceptible than females 9.8% (Sumbal Haleem *et al.,* 2016).

The present study is similar to the findings of Howlader *et al.,*(2017); Affroze *et al.,* (2013); Karim *et al.,* (2015); Nath *et al.,* (2016). All of those agree that the female is higher prevalence than male. But Sumbal Haleem *et al.,* (2016) disagree with the present findings.

Females cattle were more succeptible to fasciola infection than males, the exact cause of this is still beyond quentionable, but females are physically and immunologically weaker than male cattle probably make them more prone to fasciola infection Molina *et al.* (2005) and Chowdhury *et al.* (1994).

5.4 Fascioliasis associated with nutritional status

Nutritional status was divided in to two groups (healthy and poor healthy). The prevalence of fascioliasis in poor healthy was higher (22.39%) than in healthy (6.06%). The present study revealed that the prevalence of fascioliasis was significantly ($P<0.05$) affected by nutritional status

It was difficult to get enough secondary data related to this parameter, since many researchers talk to this in many ways, which may be mostly related to humans but not in cattle. The relationship between fascioliasis and nutritional status in 400 Mexican schoolchildren was investigated. More than half of the children in the study showed fascioliasis. The prevalence of fascioliasis infections was significantly ($P < 0.05$) affected by nutritional status (Quihui-Cota *et al.*, 2004).

Mostly poor healthy can cause the cattle to become thin, lose muscle and be prone to infection, and also poor health can lead to many symptoms, including skin rash, depression, hair loss, tiredness, brittle bones and bleeding gums which makes the poor healthy cattle more susceptible than healthy.

CHAPTER VI

CONCLUSION

CHAPTER VI

CONCLUSION

The present study was conducted to determine the overall prevalence of fascioliasis in cattle based on physical and coprological examination and to compare the prevalence of fascioliasis on the basis of age, sex and nutritional status. The presence of fascioliasis is one of the major constrain for cattle rearing in this region of Bangladesh. Although, the present study results have some limitation due to small sample size, large study area and low duration of study may lead improper diagnosis. So, further detail epidemiological study is strongly recommended for proper diagnosis and control strategy of bovine fascioliasis in that area.

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