1. INTRODUCTION

1.1 Background Information

Trematodes are parasite of many kinds of vertebrate including birds, wildilife, domestic animals and human. They are commonly called flukes on account of their flat leaf like form, with two attachment organs, called oral and ventral suckers. The fluke parasites in man habitually infest the intestine, liver, lungs, blood vessels, muscles, spleen, brain and many other organs. In other animals, some species inhabit the eustachian tubes of frogs, the frontal sinuses of polecats, the eye sockets of birds, cysts in the skin of birds etc. All trematodes are parasitic, some externally on aquatic animals, others internally on aquatic or land animals. The trematodes that live as external parasites of aquatic animals have a simple life cycle and other that live as internal parasites have a complex life cycle including two, three sexual generations and involving two, three or even four different hosts (Chandler, 1961).

The variety of snails and bivalves are an intermediate host of flukes, but snails are involved as host for the sporocyst and redia generation of fluke parasite in man and domestic animals. These includes several families of freshwater snails which have gills and can close their shells by means of an operculum (order Prosobranchia) and both freshwater and landsnails (order Pulmonata) which have respiratory sac instead of gills and no operculum. Prosobranchia included families like Hydrobiidae, Thiaridae (formely Melaniidae), Potamidae and Pulmonata included Planorbidae, Lymneidae, Bulinidae etc .

Trematode infections are a serious public and livestock health problems in several parts of the world. It is estimated that considering the number about 40 million people are affected worldwide (WHO, 1995). Considering the number of people affected the most important parasites are those belonging to genera *Schistosoma* (Blood fluke), *Paragonimus* (Lung fluke), *Fasciola* (Liver fluke), *Dicrocelium, Clonorchis, Paramphistomum ,Echinostoma, Strigea* etc.

Fascioliasis (commonly called liver fluke disease) is caused by *Fasciola* sp. (Linnaeus, 1758). It was worldwide in distribution causing severe economic loss in many countries including Nepal. The adult worm normally inhabit in the bile ducts and gall bladders of a wide range of domestic animal sheeps, goats, cattle and buffaloes. A freshwater snail of genus *Lymnaea* is the intermediate host which are found in permanent, slow flowing and clear water with abundant vegetations. Explosive outbreaks of fatal fasciolopsis were documented in sheep in Europe as early as the 18th century (Ollerenshaw and Smith, 1969). In Nepal *Fasciola* sp. affecting all species of ruminants including Yaks of the Himalayas (Joshi and Tiwari, 1975). Singh et al. (1973) reported an infection rate of 50%-90%in animals in area below 1800 meter and estimated an overall annual loss of Rs 200 million due to fasciolopsis in Nepal. Mahato et al. (1995) reported a greater economic loss, i.e. Rs 1680 million only due to the decrease milk and meat production of buffaloes. The fasciolopsis causes weight loss, decrease in milk production, reduction in conception rate, decrease in feed intake, indigestion and finally cause death of animals in severe infection.

Schistosomiasis (commonly called blood fluke disease) is cause by *Schistosoma* sp. (Bilhartz, 1852). It was distributed throughout the various parts of Africa, middle East and South America, Asia (i.e. oriental regions) including Japan, Philippines, Celebes and India. It is common parasite of man, monkeys, various kinds of rodents, cattle, goats, pigs, cats, aquatic birds (ducks) among domestic as well as in the wild animals. The adult *Schistosoma* normally inhabit in visceral and prostrate venous plexes especially on blood passage of human being and other animals. The intermediate hosts are belong to the families of snail Planorbidae, Hydrobiidae and Physidae (or Bulinidae).

It is widely distributed including oriental regions of Asia where 46,000,000 were infected with schistosomes in the orient, out of 114,000,000 people infected in the world (Stoll, 1947). Maegraith (1958) regarded schistosomiasis as the most serious parasitic disease in Red China and estimated that there were more than 11,000,000 infected persons in the country. Instead of 6,000,000 schistosome infection in Egypt, there are probably more like 10,000,000. Hunters et al. (1952) estimated an annual loss of $ 3,000,000 in wages and treatment costs in just one of the five endemic areas in Japan.

Paragonimiasis (commonly called Lung fluke disease) is cause by *Paragonimus* sp. (Kerbert,1878). It is distributed in Japan, Korea, Formosa, China, India including Nepal and some parts of Africa and South America. The adult worms live in the respiratory tract (lung) of man and domestic as well as wild animals A freshwater snail of the genus *Melania* (Thiaridae) is the 1st intermediate host and a freshwater cray fish or a crab is the 2nd intermediate host (Chatterjee, 2002).

Paramphistomiasis (commonly called intestinal fluke disease) confined to Africa, Asia, Australia, Eastern Europe and Russia caused by massive infection of the small intestine of the vertebrate hosts sheeps, goats cattle and water buffaloes, *Paramphistomum* spp. infection acquired by the ingestion of small numbers of metacercariae on one or several occasions cause to harm to the host mature rapidly and serve as a source of infection for successive generation of snails . The parasites does not generally cause severe clinical disease but heavy infection affects in animals production and growth (Padungtod et al., 2001 cited in Perry et al., 2002)

Clonorchiasis , the disease caused by *Clonorchis sinensis* (Cobbold ,1875) is endemic in East Asia , viz. China, Hongkong, Macao, Republic of Korea , Laos and Vietnam (Malek, 1980., WHO, 1995). While Opisthorchiasis caused by *Opisthorchis viverrini* and *O. felineus* is endemic in Thailand, Loas and the Russian Federation, Ukraine and Kazakhstan (Peng et al., 1993).

The stregeid are common parasites of aquatic birds, fish, frog -eating mammals and water snakes. The biology of this group of flukes has been extensively reviewed and studied by Dubois (Cheng, 2006). Man is rarely parasitized by stregeids on heavy infections. The Egyptian species *Prohemistomum vivax*, a parasite of kites but also common in dogs and cats which eat or are feed raw nile fishes or tadpoles (Nasr,1941). A typical life cycle has been illustrated by the Hunters (1935). It causes a dysentric symptoms in human beings.

1.2 Statement of problems

This study was under taken to establish the baseline information of different cercariae larva retained from aquatic snails and eggs of ruminant trematodes in Ramgram Municipality, Parasi, Nawalparasi.

In spite of successful study among different locations and fields. There were some problems, which was faced during the field survey, collection of snails, observing in the lab, screening of samples, these problem are given below:

a) Some times microscope was not available due to busy of lab staff, holidays etc.

b) Lack of sufficient resources including man power, fine and well accessible equipment , sufficient budget etc.

c) Lack of access of baseline information , research work and articles representing the host snails and cercarial larva of trematode parasite.

1.3 Rationals and Objectives

In spite of the considerable economic impact and endemic nature , there is a shortage of information about the pathological pattern of trematode cercariae in relation to malacology (the science of molluscs) in the different climatic and agro-ecological conditions and its relation with the various animal management systems practiced in the country . The trematode cercariae have been identified as disease causing parasites in different parts of Nepal . They cause different diseases among human beings as well as in other animals by which, Nepal Government convey or expenditure many budget in every year to face the health problem in every year . The experience of other countries indicates that the health problem is possible to reduce to a minimum level if control and awareness programme are based on the sound knowledge of different aspects of diseases and are implemented strategically in an organized and well planned .

There are considerable variations in the studying and documentations regarding the importance of disease and method of transmission through the intermediate host snails among different parts of Nepal So there was great problem to find out the articles related to the present study . It is essential to find out the trematode parasites in different areas of Nepal because of the existence of wide variations in physiographic and agro-ecological conditions .

Considering the above points, the study has been taken with following aims and objectives :

1. To prepare the checklist of aquatic snails used as intermediate hosts by trematode cercariae
2. To determine the prevalence rate of trematode parasite in aquatic snails collected from the study area .
3. To document larval trematode cercariae retained (sorted) from host snails
4. To determine the monthly variation of trematode cercariae during four months
5. To determine the monthly prevalence of fasciolopsis in domestic buffaloes .

1.5 Limitations

The study was carried out during the months of July 2008 to October 2008 in different parts of Ramgarm Municipality, Nawalparasi district. The samples were collected from both sides of metalled road, unmetalled road, cart track temporary ditches, pools and drainages.

In this study many obstacles were raised being a student research, resources were limited so that it was difficult to conduct a research in the beginning. Due to lack of sufficient resources including manpower fine and well laboratory with accessible equipments and sufficient budget so that the whole villages of sample site of the study couldn't be covered.

1.6 Significance of the study

This study will help to know the relationship between the intermediate host (especially freshwater snails) and definitive hosts including human beings as well as domestic animals. These are geographically distributed throughout low and mid part of the Nepal on where Nepal Government conveys and expenditure a lot of cost to control and prevent these diseases yearly. The present study will help the policymaker and to explore knowledge about digenetic trematode infections among human beings as well as domestic animals.

2. DEVELOPMENTAL STAGES AND LIFE CYCLE OF DIGENETIC TREMATODE PARASITES

2.1 Development and Hatching To Trematode Eggs

Under suitable moisture and temperature digenetic flukes produce eggs often by tens of thousands which escape from the host's body with the help of feces urine of sputum, according to the habits of the adults. Either before or after the eggs have escaped from he host ciliated embryos develop within them, these hatch either in water or in the intestine of molluse which serve as intermediate host. The ciliated embryos, called miracidia are developed and hatch further into cercaria larva. But the eggs are unable to develop and hatch during drought and cool temperature. During summer, development and hatching times also increases in stagnant pools water with plenty of organic matter.

2.2 Infection of Snail host by Miracidium Larva

The host finding ability of trematode miracidia plays a vital role in the transimission of the disease. The host finding process is a complex interplay between the physiological and behavioural activities of the snail and miracidium. The initial attachment and pentration of the epithelial cells of the snail host is performed by the miracidium. The larvae, which penetrate the epithelial cells of mental cavity, causes much damage to the underlying connective tissue but less extencsive damage on the columnar epithelium of the body or foot.

Many miracidia have eye spots, but some are blind. The miracidia donot feed and they die in 24 hrs or less if unsuccessful in finding a proper molluscan host.

2.3 **Development of Larval Stage in the Snail**

After penetration in the snail by miracidium larva, it enters into the snail and changes into young sporocyst larva. The shape of sporocyst is elongated, sac-like and covered with cuticle. A single sporocyst gives rise to many cercariae but it depends upon the availability of food and space in the digestive gland of the snail hosts. The cercariae possesses either fork tail or single long or short tail. The tail functions as an adhesive organ and swimming appendages. The damage is done by cercariae at the time of emergence, which is accompanied by trauma and this is the reason for mortality of snail populations. The larvae of trematode cause retarded growth shells are abnormally transparent and lacking in pigmentation, shell whorls are distorted and the snails themselves appear lethargic.

2.4 **Survival and Emergence of Cercariae Larva**

The pattern of emergence and survival of trematode cercariae are greatly influenced by presence of absence of definitive host , intermediate host that serve as the distribution of parasitic organisms flora and vegetations the serve as food and shaelter for parasites. Animal excreta (i.e. tool) requires source of water in which cercariae migrate and reach towards its respective host .The other external factors such as velocity of water current change in temperature , depletion of oxygen , an increase in the concentration of carbon dioxide and oxygen change in the hydrogen ion concentration (i.e. pH value of water salinity and mineral contents of water also influenced the number and survival rates of intermediate host the longevity of free swimming miracidia and cercariae of certain trematode parasites .

2.5 Some Variations in life Cycle of Digenetic Trematodes

Digenean trematode have very comjplicated life cycle involving several non sexual generations which always develop in snails or bivalve mollusks except two species cercara loossi and cercaria hartmanae , develops in annelids .

It requires two types of hosts definitive host used by adult form and intermediate host used for larval development .Sometimes a second intermediate host (fish or crab) is required for encasement .

The eggs liberated by definitive host gain access into the water. A free swimming ciliated embro miracidium develops and hatches out of the egg. The miracidium gains access to its proper intermediate host (snail of bivalve mollusc) and localizes in the gut for further development . the miracidium is transferred in to a sac like structure called sporocyst further multiplication does not occur except in schistosomes where a second generation of sporocyst is formed . The sporocyst changes into a redia having an oral sucker a pharynx , a sac-like intestine and a birth pore through which the larvae escapes , either daughter rediae (a second generation rediae) of into cercariae . The cercaria represent the final stage of larval development in the snail and is infective to definitive host. Itpossesses a tail by means of which it propels itself in wate. On maturation of cercariae escape from the snail into the water and may remain free in water of encyst (metacercaria) either in water-plants of in another intermediate host a freshwater fish of crab. Only a rare cases returming ot the either sane of another mollusk host in which it developed. Definitive host vertebrate is infected either by drinking contaminated water of ingesting encysted cercariae in the water plant fish or crab of in some cases free cercariae can enter directly in the definitive host (schistosoma)to grow in to adult worms after reaching their site of localization form where adult parasite repeat their further life cycle .

Figure 1: Some Variations in Life Cycle of Digenetic Trematodes

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Figure 1: Some Variations in Life Cycle of Digenetic Trematodes .

Sources: (Cable, 1963., Parker and Haswll ,1995 Chang 1964 Dogiel 1964 Chatterjee, 2002)

3. LITERATURE REVIEW

The Eber's Papyrus of Egypt (1600B.C) Described a worm probably the beef tapeworm (Taenia saginata), as pathogenic in man The Greeks of Aristotle's day recognized tapeworms. Hippocrates diagnosed echinococcus diseade and desease and described a techindque for removal of the cyst from the human body. The Persian physicians Avicenna (981-1037) described worms probably referable to Ascaris lumbricoides taenia saginata enterobius vermicularis and Ancyostoma duodenale listed the symptoms produced by them and prescribed remedies hoeppli (1956) has given an information general review of the early knowledge of parasites and parasitic infection upto the 17^{th} century .

In the 17^{th} century , several studied on parasites were conducted and evolved some views, out of these one view says that internal parasites originating from accidently swallowed free-living organisms. Other says that maggots developed from the eggs of flies and Ascaris had a male and female which developed by producing eggs (Francesco Redia , the father of parasitology cited in chandler 1961) .

About a century later, recognized five classes of worms named by Rudolphi ,as Nematoidea , Acanthocephala Nematoda , Cestoda and Cystica (Zeder , 1800) . Nearly two century later recognized as the first protozoan human parasite was Balantidium coli (malmstem, 1858).

3.1 Literature review in the context of the world

Price (1953) examined fasciola species from various parts of the United states and found that some areas had species identical to fasciola gigantica from old world , where as other area had fasciola gigantica

Banglapedia (1988) reported the prevalence of Schistosoma spindalis and Sindicum among cattle all over Bangladesh and result was found that the adult cattle above 3 years of age were severely affected up to 25% incidence.

Chaudhari et al.(1993) collected a faecal sample of cattle and buffaloes for over a period from January 1986 to December 1991 in Eastern Haryana, India High incidence of fasciola gigantica infection was observed in cattle and buffaloes during winter (November to February) Prevalence was greater n buffaloes than cattle fasciola gigantica cereariae were found in high percentage in Lymnaea auricular The percentage of infection was high during rainy season (July to October)and the highest percentage of excreted eggs was during winter .

Nwosu and Shrivastava (1993)collected faecal samples form live and livers of slaughtered zebu cattle for a period form may 1989 to April 1990 in Savar Dhaka A total of 1080 Faecal samples and 144 livers were examined of which 19.7 percent faecal samples and 43.7 percent livers were found to be positive for fasciola gigantica . The prevalence was higher in old and female animals than young and male animals

EL- Bahy (1997) collected and examined faecal sample from june 1995 to june 1996 in Egypt Faecal sample positive fom fasciola were 14.5 percent of 1225 buffaloes 26.6 percent of 3500 sheeps 12.5 percent of 708 donkeys and 6 percent of 1800 human fasciola infection rates highest during the summer. The highest prevalence being buffaloes over three (3) years old .

Nguyen at.(1999) done a survey of 955 swan buffaloes lives in Vietnam form January to july1998 . Out of 955 only 46.07 percent were infected with fasciola species. The buffaloes infected with F gigantica were 14.14 percent and both F hepatica and f. gigantica infected with 32.04 percent .

Mekround et at (2004) the natural infections of fasciola hepatic were carried from cattle and sheep slaughtered at constatine and jijel of Algeria from 31 cattle and sheep breeding farms and from four (4) populations of galba truncatula snail the higher prevalences of fascioliasis were foun in the cattle and sheeps from jijel as compares with cattle from Constantine (27.0%) in slaughtered cattle and 27.3 percent in cattle from farms compared to 9.1 percent and 6.3 percent respectively)

Ruppel et al.(2004) recorded only schistosoma japonicum was considered by virtue of its remarkable speed and success of migration as well as by specific biochemical and immunological features than the schistosoma mansoni and also schistosome japonicum have a typical enzyme that facilitate skin penetration .

Kornas et al. (2005) the investigation carried out on the basis of necropsy material (i.e livers of cattle) taken form slaughter houses in the Bochnia region near Krakow . A total of 218 heads of cattle livers were examined and 62 heads of cattle livers (28.44%) showed fascioliasis infection. It was most often evident (71%) in the livers of oldest animals i.c. over 12 years old. Liver bile duct examinaiotn showed the presence of adult flukes in only 29 liver (46.77%) The most cattle (18 heads were infected at 6-11 yeats of age .

Ravindra et al.(2007) studied on domestic ruminants slaughtered in wayanad located in Koeral India and fournd 57.3 percent 50 and 4.7 percent of cattle buffaloes and goats respectively has visceral schistosomiasis upon slaughtered ruminants.

Gupta et al.(1987) recorded nine (9) species of aquatic snails and cercarial types in Eastern Haryana , India. The incidence of cercaria in Lyknaea luteolaf ovalis,

L.acuminata and Gyraulus convexiusculas were seasonal .indoplaplanorbis exiutus and digonistoma pulchella reveald high incidence during july to October and june to september respectively.lymena lutuola f.typica and indoplanorbis exustus accountd

For 60 percent of total snail population examined Fasciola cercariae were only recorded from lymnaea acuminate in small numbers throughout the year.

Wamae and Cheruiyot (1990) collected and dissected four hundered fourteen (414) snail over a year in Kenya. Out of them 31.4 percent were harboured with larvae of Fasciola and 60.9 percent were harboured with chaetogaster limnei

Hurley et al. (1994) studied the larval trematodes in freshwater snails constituting Autropeplea lessoni Gyraulus giberti Ameriann corinata and Thiara balannensis with emphasis on Trichobilharzia spp. (causative agents of swimmer's itch) Authors suggested both A lessoni (4.5%) and G GILBERTI (1.8%) act as intermediate host of Trichobilharzia sp. They also reported the thiara balonnensis were infected with trematode cerariae representing six (6) families.

Hussain et al.(1996) studied the prevalence of lymnaea snails identification of infected and non-infected snail with intermediate stages of liver fluke . The prevalence of Lymnaea snails were found to be 42 percent of fasciola hepatica and infection of lymnaea snails with intermddiate stages of fasciola hepatica ranged between 38 percent and 69 percent in five habitats .

Rondelaud and Dreyfuss (1997) carriage out field investigations over a four (4) years period in three (3) farm of Haute –Vienne Department (France) in order to determine the prevalence of F hepatica infection in annual generations of lymnaea truncatula Fasciola infection of Lymnaea truncatula originationg from spring and summer generations was more efficient than from the winter generation of snails .

Chingwena et al (2002) collected a total of 13,789 freshwater snails representing 10 species from 21 sites from the highveld and lowveld areas of Zimbabwe to determine the occurrence of larval trematodes and recorded that 916 (6.6%) harboured patent trematode infection representing 8 morphologically distinct types of cercariae . They reported Bulinus tropicus had the highest overall prevalence o infection (13.1%) and the echinostome was the most common type of cercariae contributing 38.2 percent of al infections .

Chingwena et al. (2002) examined the susceptibility of Bulinus tropicus B Globosus biomphalaria pfeifferi Lymnaea natalensis and melanoides tuberculata to calicophoron microbothrium and reported that Bulinus tropicus had the highest prevalence (65.0%) followed by Biomphalaria Pfeifferi (37.5%) Bulinus globosus (6.8%) and Melanoides tuberculata (5.9%)

Briers Roberts A (2003) studied the prevalence of trematode parasitism in population of freshwater gastropod snail Lymnaea stagnalis and found the like lihood of trematode infection increased with snail size . The changes in abiotic factors to wards the range limit may influence snail life history .

Hassan et al. (2003) the host finding behaviour of miracidia of 2 strains of schistosoma mansoni from Egypt and Brazil was studied by recording their responses to snail – Conditional water (SCW) from the Egytptian sympatric snail Bomphalaria alexandrina physa acuta Lymnaea cailliueli Bulinus truncates as well as form

B. Arabica and B glabrata Miracidia of the Egylptian strain strain significanty preferred SCW Form their compatible hosts B alexanadrian and Arabica and showed no or a weak response to SCW From other sympatric species Where as miracidia of the Brazilian strain did not differentiate between SCW From different snail species .

Prepelitchi Lucila et al .(2003) a sample of 601 snails was collected in may 2003 from Paraguay Brazil and Urugary Among 500 examined snails, 44 (8.8%) were exclusively infected with fasciola hepatica

Be;faoza et al (2004) investigated double sporocyst infections with normal development of redial generation in 43.9 percent of infected snails out of 296 found single sporocyst infection with normal development of redial generation in 53.7 percent and abnormal development in 2.4 percent for successive redial generations were found in long – surviving snails (more than 90 days).

Hunilczewska et al (2005) examined the effects of the presence of sporcysts rediae and cercariae of fasciola hepatica on the lipid content in the digestive gland of lymnaea trunactula as well as lipid levels in the tissues of the parasited by means of histochemical and cytoplasmometric techniques. The snails digestive gland lipid level was found to be almost halved in 20 days post infection .

Okere and odaibo (2005) the study on the effect of indoplanorbis exustus ,a planorbid sanil and possible competitor snail of Biomphalaria pfeifferi on the fecundity and growth rate showed a significant difference in the growth rates and fecundity of indoplanorbis exustus and Biomphalaria pfeifferi . The was direct relationship between fecundity and growth rates .

Zbikowska et al (2005) conducted a study by taking shells of naturally infected and un infected snail of Lymnaea stagnalis from 25 different lakes in the central part of Poland. The height of shell the height of the spiral and the width of the shell were measured. It was found that snail infected with exhinopyphium aconiatum Echinostoma revolutum Diplostomum pseudospathaceum and opisthiolglpheranae differed in shell shalpe compared with uninfected individuals snails infected with plagiorchis elegans did not differ from un infected individuals .De kocl K. N. and C . T . Wolmarans (2007) collected 736 localities currently on record in the database of the National Frehwater snails Collection (NFSC) of south Africa which were yielded b rivers streams and dams of freshwater cotaining stony substrate and aquatic vegetations . They concluded from this survey that altitude temperature and water bodies were the most important factors determined the geographical distribution of snails .

Dreyfuss (2006) of the four groups of snail population studied and revaled the ablest snails to sustain a complete larval development of f hepatica originationig form population living in siliceous soils at 600 meter and more in altitude then living alnong river banks on siliceous soil (due to poor characteristic of snail infection high snail mortality low prevalence of snail infection and low number of cercariae produced) except for these last populations shed less than 300 cercariae ecen if a maximum of 1,772

Cercariae were obtained from a single snail .

Vignoles et al (2006) They studied under laboratory and found that cercaria shedding snails were in low number and their shell height at day 60 p.e was significantly greater than that of numerous infected snails that died without cercarial shedding. The rediae number increased with increasing shell heights of infected snails. Metacercaria were only found 9mm of shell height and were in low numbers.

Caron et al (2007) Radix labiata and Radix balthica snail were collected in Belgium and identified by and based techniques. Microscopy releaved the presence of larval stages in 78.3 percent 465 percent and 6.25 percent of galba truncatula Radix labiata shed fewer metacercariae than galba truncatula. This study demonstrated that radix labiata may act as incidental intermediate host for fasciola hepatica in Belgium.

Dechruksa et al.(2007) studied at five different of water souses in Thiland between December 2004 and October 2005 A total of 9,568 snail samples comprised of 14 species five species of snail were susceptible to trematode infection. They were tarebia granifera (26.1%) Melanoides tuberculata (33.3%)

Thiara scabra (1.2%) paracrostoma paludiformis paludiformis (0.3%) and Brotia wykoffi (0.7%) The cercarial infection rates in the above 5 species were 0.1 percent (5/9,568),0.2 percent (15/9,568),0.3 percent (24/9,568), 0.4 percent (37/9,568) and 0.1 percent (5/9,568) respectively .

Gerard et al.(2007) studied 13,280 gastropods belonging to 17 species from the lake over 10 years of time series . they demonstrated the trematode larvae belonging to 11 morphotypes of cercariae in 15 of the 17 species of gastropods and had a total prevalence of infection was 29 percent .

Kleiman et al.(2007)study conducted in farm between December 1998 and February 2002 and found overall prevalence in lymnaea viatrix was 0.6 percent (range : 0.9-14%) and infection was detected in summer and autumn. When infected snail were present transmission is maximum in summer-autumn and other in early spring due to over wintering metacercariae .

Phiri et al. (2007) studied the trematode infections in freshwater snails and cattle of Zambia They collected a total of 984 snails, comprising 9 species from 6 areas between August to October 2003 and found 135 (13.7%) were positive Authors recorded the most trematode infections from L ymnaea natalensis (42.8%) which harboured 4 of the 5 morphologically different cercariae. No Trematode infections were recorded form Bellamya capital Biomphalaria pfeifferi Melanoides tuberculatus physa acuta and Cleopatra nswendweensis.

Schweizer et al.(2007) studied the prevalence of fasciola hepatica in the intermediate host snail Lymnaea truncatula and the infection risk to the definitive host snails collection from 130 different habitats. A total of 4733 snails were examined of which 331 (7.0%) were found to be infected with fasciola hepatica the risk of infection of lymnaea truncatula with fasciola hepatica was significantly higher in population origination from spring swamps wells and reeds compared to population from streams .

Faltynkova et al.(2008), They studied of 7,628 snails of 12 species examined between 1998 to 2006 from central Europe (Austria, Germany, Hungary, Poland and the Slovak Republic). A total of 34 Trematode larval stages comprising cercaria of 28 species and metacercaria of seven (7) species (one species occurred both as cercaria and metacercaria) of nine (9) families were found in 898 (11.5%) snail of eight (8) species the dominant were recorded form families Echinostomatidae plagiorchiidae and omphalometridae and less dominant cercariae form families Diplostomidae and strigeidae all occurring in planorbarius cornesus (Linnaeus). The most heavily infected snail species was planorbarius corneus followed by planorbis planorbis (Linnaeus) and Segmentina nitida (Muller). Fourty Two (42) cerariae identified to the species level belonging to 15 families were reported from 11 species of planorbids **.**

3.2 Literature review in the context of Nepal

Lohani and jaeckele (1981-1982) identified the fasciola species in palpa. Out of twenty four (24) animals four (4) animals were infected with fasciola gigantica five (5) animals with both F. hepatica and F gigantica and fifteen (15) animals with F. hepatica f. gigantica and intermediate form between them. The animals infected with F hepatica were not found .

Dhakal and Nepali (1983) tested one hundred thirty (130) faecal samples form different age groups of cattle and buffaloes at IAAS, Rampur chitwan from hundred thiry (130) faecal samples nine (9) samples were of calves twenty three (23) of young animals and ninty eight (98) of adults. The positivity for fasciola infection was 11.1 percent, 13.1 percent and 20.4 percent respectively .

Mahota et al (1991) collected faecal sample directly form the calves and examined for fasciola eggs in Dhankuta. In the low altitude (below 1100 meter),50 percent and 48 percent faecal samples of cattle and buffaloes respectively were positive for fasciola . In mid altitudes (1100-1700 meter), 28.6 percent faecal samples of cattle and 50 percent faecal samples of buffaloes were positive for fasciola species positive. In high altitudes 20 percent and 52. percent faecal samples of cattle and buffaloes were positive of fasciola respectively .

Singh and shah (1992-1993) conducted a survey in the low lands of IAAS Rampur, Chitwan to investigate the relationship between snail population and liver fluke infection in ruminants. The liver fluke infection was 48.57% percent 28.57 percent 25 percent and 21.28 percent in buffaloes, cattle sheeps and goats respectively.The highest infection rate of liver fluke was observed in September (50%) follwed by the month October (43.75%) and January (35%). And The snail population was negatively associated to the rainfall .There was also negative correlation between liver fluke infection and snail population .

Parajuli (1993-1995) stutied the prevalence of fasciola and paramphistome in cattle and buffaloes in surkhet. Sixty percent cattle and 83.8 percent buffaloes were found to be positive for flukes. In cattle prevalence of fascioliases and parasphistomiasis were 42 percent and 25.4 percent respectively, whereas that for buffaloes were 56.8 percent and 35.1 percent respectively

Regmi et al.(1999) examined the faecal sample of cattle and buffaloes in syangja district during june to October 1998 coprological examination revealed 67.8 percent buffaloes and 62.1 percent cattle were positive of fascioliases

Pandey (2001) studied prevalence of fasciola sp. Infection in lymnaea snail and duffaloes of Devbhumi baluwa VDC OF Kavre district revealed fasciola larvae infection in lymnaea snail were most prevalence in rice field (1.67%) and was absent in streams. Out of 182 faecal sample examined 50 percent sample was positive for fascioliasis in buffaloes. He reported the highest prevalence rate of fasciola larvae in November (5.35%) and no larvae were found in March, April and May .

CVL (2002/2003) Conducted a study of 92 faecal samples of buffaloes taken from kathmandu valley and analysed 56 (61%) cases were positive for fasciola spp infection.

Adhikari et al. (2003) conducted astudy on the prevalence and diversity of fasciola sp. In buffaloes and cattle in areas of kathmandu valley and found to be 36 percent in cattle and 61 percent in buffaloes infected with fascioliasis.

CVL (2004/2005) Collected a total of 1,633 faecal samples of cattle examined and reported that fasciola sp. Was the highest positive cases 430 (43.65%) as compared with paramphistomiasis 168 (17.05%) infections.

Mukhia, G. (2007) reported 90.90 percent samples positive for trematodes in buffaloes of satungal VDC of kathamandu district. Out of these Schistosoma spp.infection was found to be in 46.94 percent, Fasciola spp. In 32.6 percent and Dicrocoelium spp. In 2061 percent.

Singh et al. (1973) collected snails from different regions of Nepal and found Lymnaea auricularia race rufescens and L. viridis to be intermediate hosts of Fasciola species which are wide spread in different regions of Nepal.

Morel and Mahato (1987) determined the epidemiological cycle of Fasciola h epatica in the intermediate and definitive hosts . Lymnaea auricularia race rufescens and luteola were responsible for the transmission of F gigantica in the koshi hills of Nepal . The prevalence of the disease in catte was the higest during june to September and again during January to February .

Mahato (1993) identified four (4) different species of lymnaed as l auriclaria race rufescens l auricularia sensu strictu l viridis and l luteola in Nepal the high snail population density was found during dry period and decline with the onset of monsoon .

Devkota (2008) collected 25,25 freshwater snails representing 10 species from six (6) habitats of Chitwan and Nawalparasi districts of Nepal reported 89 (3.25%) harboured parent trematode infections. Gabia orcula had the highest overall prevalence of infection (6.5%) and no infection was recorded in Bellamya bengalensis Brotia costula pila globosa and segmentina spp. The highest percentage of infection was found in small temporary ponds (6.23%). He recorded double infection (Ampistome and schistosome) only from the indoplanorbis exustus snail This study also reported the presence of he eggs of schistosoma spp . and fasciola spp. In captive elephants of chitwan

4. METHODS

4.1 Study area

Nawalparasi is a Western Terai, Inner Terai and Mountaneous district situated between $83^{0}36'$ East to $84^{0}36'$. East longitudes and $27^{0}21'$ North to $27^{0}47'$ North latitudes. Elevation ranges from 100 to 1,636 meter from the sea level. It covers nearly 98 Km East-West Mahendra Highway. It has border with districts as Chitwan in East Tanahu and Palpa in North Rupendehi in West and U.P India in South. River Narayani flows along the border with Chitwan. There are 73 V.D.Cs. and one municipality in the district. Rainfall occurs from July to October. November to March is usually dry with no rainfall. The temperature ranges between minimum 5^{0}C to maximum 44^{0}C.

This study was conducted in the Ramgram Municipality. It has been made by 4 V.D.Cs. including Parasi bazaar (head quarter of Nawalparasi district) named as Parasi Adarsh, Manghariya, Unwach and Jamuward. It has different ethnic groups comprising Tharu Brahmin Chamar (Harijan) Yadav etc. Most people have Bhojpuri, Nepali language to communicate each other. There are 4,166 households in the Ramgram and area is 3120.28 hectare, out of this 2,558.76 hectare used by people as agriculture fields.

Major occupation of the people of this municipality are paddy cultivation, wheat and maize growing, cattle faming and daily wage jobs. Rice is grown in the irrigated and rain fed fields. [Source : Bulletin of Ramgram Municipality, Parasi, Nawal Parasi, 2007]

4.2 Date Collection

The related data or information were collected either from primary sources or secondary sources. The primary data which are in the original form were collected through field visits, personal interviews to specify the location on where people grazed their cattle. Dung were collected for examination in the lab. The secondary data were collected either published or unpublished articles, reports, newspapers, journals, books, dissertations (thesis), internets and other reports of INGOs., NGOs.

4.3 Preliminary Field Survey

A preliminary field survey was carried out to ensure the presence of aquatic snails in the study site before actual field work doing. It has been some difficulty for road and traveling due to the rainy season so that it was ensure by discussing with local people and others. There are total 41 sites which were selected for the study during July to October (4 months). A total of 2,921 snails population have been recorded and studied.

4.4 Snail Collection

It was carried out for a period of 4 months from July to October in 2008 by visiting road side temporary ditches and drainages. The general features of the water bodies in the visited areas were rich with aquatic vegetations like as grasses, water lilies, hyacinth and Ipomea sp. All the sites in the study area were examined for the presence of snail. Snail were collected with the help of scoop, hands and forceps. The samples were immediately brought to room for preservation.

4.5 Laboratory Examination

4.5.1 Snail and Larval Examination

Collected snails were rinsed in chlorine free tap water to remove mud and plants. Each snails were kept in well-plates with small amount of clean water. The well-plates containing fresh snail sample were covered and kept near window with enough light or under tube-light.

A well-plate is a plastic device with different number of cavities in it. In this study the well-plates with six (diameter = 3.5cm), twelve (diameter = 2.2 cm) and twenty four (diameter = 1.6 cm) cavities were used. The length and breadth of the all types of well-plate and the depth of the cavities were same (length = 12.8 cm, breadth = 8.5 cm and depth of the cavity = 1.8 cm.).

All snails kept in well-plates were observed stereomicroscope after placing for one full night (i.e. 24 hrs). It was brought at the Lab of Prithvi Chandra Hospital, Parasi, Nawalparasi for screening. As the snails found to be infected, the cercariae were clearly seen swimming in a well cavity. A detail morphology of a cercariae were observed under high magnification compound microscope on a slide. The cercariae were photographed by using a camera and identified by using an introductory guide to the identification of cercariae and key to cercariae from African freshwater snails with special reference to cercariae of trematode species of medical and veterinary importance .

4.5.2 Faecal Examination

The different animal stools (dung) were collected in the District Veterinary Office, Parasi as daily, which were brought by farmers for testing the parasitic infections. About 3 gm of stool were taken and put into mortar and broken up with some tap water making a well mixed suspension and it with the help of course to very fine mesh sizes. The filtrate was put undisturbed to sedimentation for 20-30 minutes. After that the supernatant was poured away and from the sediment, kept 1 to 2 drops on a neat and clean slide on which one drop of methyl blue (a type of stain) has been kept and cover it with cover slip. The slid was observed under the stereomicroscope to locate the presence of the trematode eggs. The eggs of *Fasciola* sp. were operculated and golden brown in colour with methyl blue stain which can be easily distinguished from the eggs of other amphistomes as they are clear and contained large granules. The eggs were identified in lab by Dr.Hari Bahadur Kunwar, Veterinary Doctor, District Veterinary Office, Nawalparasi.

5. RESULTS

5.1 Types of Snails Observed

A total of 2921 individual aquatic snails were collected from the study area. These all snails were related to eight (8) different species belonging to the families Ampullariidae, Bithyniidae, Lymneidae, Planorbidae, Pleuroceridae and Viviparidae. Out of eight (8) species, *Gyraulus* spp. was found to be in more percentage 1202 (41.15%) of the total snails observed as compared to others. *Gabia orcula* was found to be in less percentage of the total snails observed as 48 (1.64%) which is shown in the Table no. 1.

Table no. 1: Types of Snails Observed

S.No.	Family	Genus/ Species	Total no. of snails observed	% of total snails observed
1	Ampullariidae	*Pila globosa*	91	3.11
2	Bithyniidae	*Gabia orcula*	48	1.64
3	Lymneidae	*Lymnaea* spp.	809	27.69
4	Planorbidae	*Indoplanorbis*	299	10.24
5	Planorbidae	*Gyraulus* spp.	1202	41.15
6	Pleuroceridae	*Segmentina* sp.	468	16.02
7	Viviparidae	*Bellamya*	126	4.31
8	Viviparidae	*Bellamya dissimilis*	66	2.26
Total			2921	100.00

5.2 Types of Snails and Cercarial Infection Rate

Five different species of snails namely *Gabia orcula*, *Lymnaea* spp., *Indoplanordis exustus*, *Gyraulus* spp. and *Segmentina* sp. were found to harbour patent trematode infections. Among these, the highest percentage of infection was recorded in *Lymnaea* sp. (7.26%), followed by *Indoplanorbis exustus* (3.08%). The overall infection rate was recorded from 92 individuals as 3.15%. Three species of snail were devoid of any infection named as *Pila globosa*, *Bellamya bengalensis* and *Bellamya dissimilis.* There was significance positive correlation between the total number of particular snails species examined and the total number of infected snails, (r_{xy}=0.85, t_{cal}=3.92, d.f.=6, p<0.05). This infection rate is shown in Table no. 2.

Table No. 2: Types of Snails and Cercarial Infection Rate

S.No.	Types of Snails	Total no. of snail observed	Total no. of infected Snail	Percentage of Infection
1	*Bellamya bengalensis*	91	00	0.00
2	*Bellamya dissimilis*	48	00	0.00
3	*Gabia orcula*	809	13	1.61
4	*Gyraulus* spp.	299	06	2.01
5	*Indoplanorbis exustus*	1202	37	3.08
6	*Lymnaea* spp.	468	34	7.26
7	*Pila globosa*	126	00	0.00
8	*Segmentina* sp.	66	02	3.03
Total		2921	92	3.15

5.3 Monthly Variation of Trematodes in Fresh Water Snails

The present survey was done in the months of July, August, September and October, 2008. It was found that 2829 snails were non-infected and the remaining 92 were infected (out of total 2921 snails). The highest prevalence of trematode cercariae was reported in August as 4.88% followed by September (4.30%), October (2.29%) and July (1.55%) which is shown in the Table no. 3.

Table No. 3. Monthly Variation of Trematode Cercariae

S. No.	Months	No. of snails		% of infection
		Examined	**Infected**	
1	July	835	13	1.55
2	August	573	28	4.88
3	September	814	35	4.30
4	October	699	16	2.29
Total		2921	92	3.15

5.4 Freshwater Host Snails and Their Trematode Cercariae

Altogether six (6) different morphotypes of cercariae were reported during the study period. The most commonly occurring cercariae were Longifurcate-pharyngeate cercaria (*Strigea*), Brevifurcate-pharyngeate cercaria (*Clinostomoid*), Brevifurcate-apharyngeate cercaria (*Schistosoma*) and Amphistome cercaria found in the *Indoplanorbis exustus* snail host. The Longifurcate-pharyngeate cercaria and Gymnocephalous cercaria (*Fasciola*) found in the *Lymnaea* spp. snail host. Amphistome cercaria and Xiphidiocercariae were found in the *Gabia orcula* snail host. Amphistome cercaria was found in the *Gyraulus* spp. snail and Amphistome cercaria was found in *Segmentina* spp. snail. Amphistome cercaria was found in four (4) types of snails named as *Indoplanorbis exustus*, *Lymnaea* spp., *Gyraulus* spp., and *Segmentina* spp. after that, *Strigea* and *Schistosoma* are found in *Indoplanorbis exustus* and *Lymnaea* spp., Xiphidiocercaria is found in *Gabia orcula* and Gymnocephalous cercaria is found in *Lymnaea* spp. This is shown in Table no. 4.

Table no.4: Freshwater Host Snails and Their Trematode Cercariae

S. No.	Hosts	Cercarial Types					
		LPC	**BPC**	**BAC**	**AC**	**XC**	**GC**
1	*Indoplanorbis exustus*	+	+	+	+	–	–
2	*Gabia orcula*	–	–	–	+	+	–
3	*Lymnaea* spp.	+	–	–	–	–	+
4	*Gyraulus* spp.	–	–	–	+	–	–
5	*Segmentina* spp.	–	–	–	+	–	–

(+ve shows presence of cercaria and -ve shows absence of cercaria)

Note - LPC = Longifurcate - pharyngeate cercaria (*Strigea*)

BPC = Brevifurcate - pharyngeate cercaria (*Clinostomoid*)

BAC = Brevifurcate - apharyngeate cercaria (*Schistosoma*)

AC = Amphistome cercaria

XC = Xiphidiocercaria

GC = Gymnocephalous cercaria (*Fasciola*)

5.5 Intestinal Helminth Infections in Domestic Buffaloes

5.5.1 Monthwise Prevalence of Helminth Infections in Faecal Samples

Altogether 735 faecal samples of domestic buffaloes of Ramgram municipality were collected from the local farmers. The Laboratory observations showed that 189 (25.71%) of total samples were positive for helminth infection. The prevalence of helminth infection is more in the month of September 72 (38.09%) out of 200, followed by August 51 (26.98%) out of 207, October 36 (19.01%) out of 145 and July 30 (15.87%) out of 183 which is shown in the Table no. 5.

Table no. 5: Prevalence of Infected Feces During the Study Period

S. No.	Months	Total no. of fecal examination	Total no. of helminth infected samples
1	July	183	30 (15.87%)
2	August	207	51 (26.98%)
3	September	200	72 (38.09%)
4	October	145	36 (19.01%)
Total		735	189 (25.71%)

5.5.2 Monthwise Prevalence of *Fasciola* Eggs in Buffaloes

Out of 735 total examined samples, 146 (19.86%) cases were found to be positive for trematode eggs of *Fasciola* species. The prevalence of *Fasciola* eggs in faecal samples was maximum in the month of September 62 (31.00%) followed by October 29 (20.00%), August 31 (14.97%) and July 24 (13.11%).

Table no. 6: Showing the (+)ve Cases of *Fasciola* Eggs in Buffaloes

S. No.	Months	Total no. of helminth infected samples	(+)ve cases of *Fasciola* eggs
1	July	30*	24 (13.11%)
2	August	51*	31 (14.97%)
3	September	72*	62 (31.00%)
4	October	36*	29 (20.00%)
Total		189*	146 (19.86%)

* Note: Faecal samples containing eggs of helminth parasites. All eggs other than *Fasciola* were not confirmed and not specified in this case.

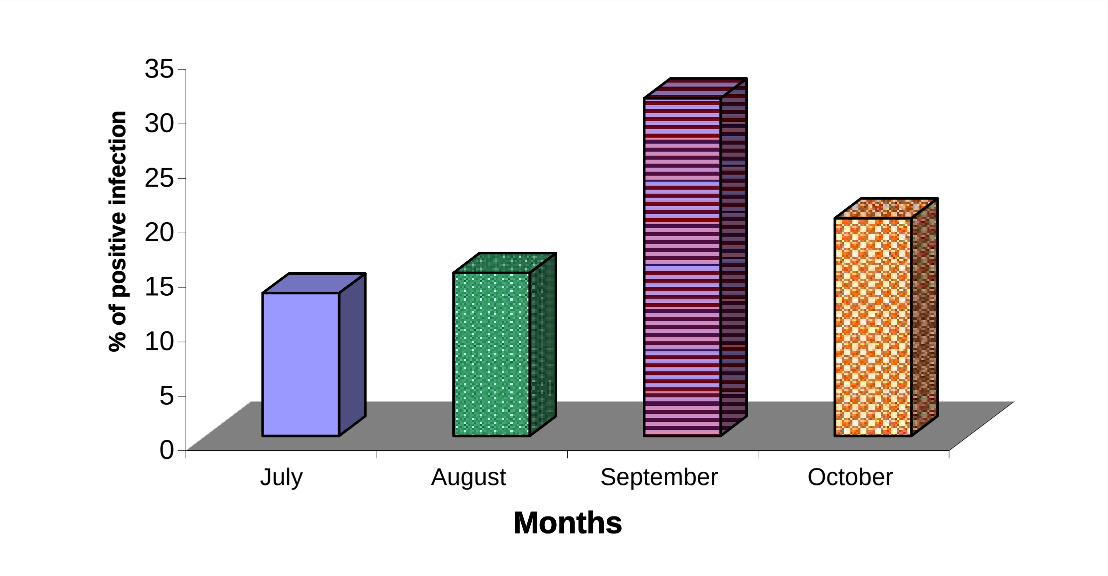

Figure 6: Showing the (+)ve Cases of *Fasciola* Egg

6. DISCUSSION

The major group of helminths which exist in the world today nearly among 2,200 million people , out of these ,occurred 72 million cestode,148 million trematode and over 2,000 million nematode infections(stoll, 1947). All trematodes and parasites of all kind of vertebrates especially of man domestic as well as wild animals . The termatode parasites enter towards their power definitive and intermediate host through several ways onwhere they grow and continue thir life cycle.

Trematode infections are a serious public and livestock problems in several parts of the world .It was estimated that about 40 million people are affected worldwide(WHO,1995).

In the present study site , the rainy season started during July , which changed the environmentaltemperature and humidity so as to favour the emergence ceraraie from the freshwater snails . The carcaria release from snails and from metacercaria into the aqatic vegetation needs ingestion by ruminant animals(buffaloes, sheep, goats etc).

The freshwater snails are not found in the habitat where it is used by the people for bathing and washing purposes and with no aquatic vegetations. According to Matato (1993), road side pools appeared to be the most important habitat in the Terai.These pools are daily contaminated with the faeces of domestic animals , which graze and wallow around and in them.

The present study showed that the most commonly infected snail host was *Lymnaea* spp. (7.26%) followed by *Indoplanorbis exustus* (3.08%), *Segmentia* spp.(3.03%), *Gyraulus* spp (2.01%) and *Gabia orcula(*1.61%). This record compared with the Swell (1922) reported upto 50 percent trematode infection in *Indoplanorbis exustus* , 6.4 percent to 50 percent in *Gyraulus euphraticus*, in India. Devkota (2008)reported the most commonly infected snail host was *Gabia orcula* (6.5) followed by *Indoplanorbis exustus* (4.5%), *Gyralus spp*. (3.1%), *Lymnea spp*. (2.92%), *Melanoids tuberculants* (1.82%) and Thiara spp. (0.63%). The rate of parasite infection varies greately with different host snails like as *Indoplanorbis exustus*. Contained more i.e. four cercarial larva compared with other host snails due to less resistance power toward the miracidia infection.

The most heavily infected snail host species was *Planorbarius corneus* followed by *Planorbis Planorbis* (Linnaeus) and *Segmentina nitida* (Muller) reported from Central Europe. Forty-two (42) cercariae identified to the species level belonging to 15 families were reported from 11 specices of planorbids. (Flaynkova et al., 2008).

The high rate of prevalence of infection occurred due to high parasite preassure gained by faecal contamination in water-animal contacts. The low rate of prevalence of infection occurred due to low parasite preassure and developed acquired resistance to an infection. The type of infection depends on the shell size and distrubtion of snail hosts. Snail host do not become infected with cercariae till they have reached manturity, older snails are more likely to be infected with parasite as compare with juvenile and younger snails since they have had most chronic exposure to infective miracidia (Muira and Chiba, 2007).

No infection was recorded in *Bellamya bengalensis*, *Bellamya dissimilis* and *Pila globosa*, which was comparable with the records from *Bipmphalaria capillata, Bimphalaria pfeifferi, Melanoides tuberculata, Physa acuta and Cloepatra, nswendweensis* in the wetland of Zambia (phiri et al., 2007). No trematode infection was recorded from *Biomphalaria capillata, Lanstes ovum or Gyraulus costulatus* (Chingwena et al., 2002). Similarly no termatode infection was recorded from *Bellamya bengalensis, Brotia costula, Pila globosa and Segmentina* spp. From Chitwan, Nepal (Dakota, 2008). Some snails showed no trematode infection it is because of the snails resistant to trematode infection which play a role in determining infection rates (Bayne and Yoshino, 1989).

The study was conducted in July, August, September, and October, 2008 and recorded in following trematode infection level, July (1.55%), August (4.88%), September (4.30%) and October (2.29%). Pandey (2001) examined 2,040 individuals of Lymnaea spp.from Kavre district of Nepal and found Fasciola larva infectin level as August (1.49%), September (3.30%) and November (5.35%). Similarly Devkota (2008) examined several freshwater snails from chitwan and Nawalparasi districts of Nepal and recorded trematode infection rate in Augest (2.16%), September (4.16%) and November (2.02%).

Six (6) morphologically different types of larvel trematode infection of fresh-water snails from Nawalparasi district were recorded in the present study which showed similar records of eight (8) trematode cerecariae found from Zimbabwe (Chingwena et al., 2002), six (6) diffrente families in Noorthern Australia (Hurley et al., 1994), eight (8) trematode species in *Batillaria atttamentaria* from Japan (Muira and Chiba, 2007), five (5) morphologically diffterent types of larval trematode infection in freshwater snails from Kafua wetlend of Zambia (Phiri et al., 2007), five (5) morphologically different types of trematode larval infection in freshwater snails recorded from Nepal (Devkota, 2008), 42 cercariae indentified to the species level belonging to 15 families were reported from 11 species of Planorbids (Faltynkova et al., 2008).

Three (3) species of lymneids were found through out the most of Mazandaran (Iran). *Lymnaea (Stagnalis) Palustris,* a secondary intermediate host of *Fasciola hepatica, Galba truncatula* , the main intermediate host of *Fasciola hepatica* and *Radix grdrosiana,* a member of the *auricularia* complex transmitting *Fasciola gigantica.* The distribution of the three (3) Lymneids agree with the distribution of human and animal fascioliasis (Moghaddam et al., 2004). The prevalence of natutal fasciolopsis in cattle, sheep and the host snail (*Galba truncatula)* showed that the higher prevalences of fasciolopsis were found in the cattle and sheep (27.0% in slaughtered cattle and 27.3% in cattle farms) (Mekeound et al., 2004). Among 500 examined *Lymnaea columella* (Say, 1817) snails, 44 (8.8%) were exclusively infected with *Fasciola hepatica.* (Prepelitchi et al., 2003).
Month wise analysis of fascioliasis by Singh and Shah (1992) showed the higher incidence of liver fiuke occurred durring the month of September in rainy season. The present finding also agree with Singh and Shah. The trend of the prevalence of fasciolopsis in domestic buffaloes showed increase in the month of September (31.00%) and decrease in July (13.11%). The finding agrees with the Pandey (2001) as the highest infection rate of liver fluke was observed in September (50%). The release of *Fasciola eggs* from faeces and contamination of adjacent habitats was not happening because of faecal pats were dried quickly by sunlight. The Infection of snail with rediae and cercariae were seen during the months of rainy season.

The present study showed that the prevalence of *Fasciola* species detected by faecal output in domestic buffaloes was 146 (19.86%). This finding is in close agreement with many data given by others. At the Regional Veterinary Diagnostic Laboratory Surkhet, the samples positive for *Fasciola* were 56.75 percent (Parajuli, 1992). The faecal sample positive for *Fasciola* eggs were 44.96 percent (Dhakal and Kharel, 1988), 48.57 percent in Chitwan (Singh and Shah, 1992-1993), 46 percent in Far Western development Region (NARC research highlight), 68.3 percent in rural Kathmandu (Ghimire et al., 1996).Joshi (1991) found prevalence of fasciolopsis to be 38.8 percent, 27 percent and 75.5 percent in mid, high and low hills respectively. Mahato (1993) reported that the prevalence of fasciolopsis was 60 percent in eastern part of Nepal. Joshi (1988) reported from the 262 numbers of faecal sample examined was 78 percent in western part of Nepal. In some places found the higher rate of infection because of domestic ruminants faeces were fallen and collected along the roadside which were contaminate the water bodies and also due to the continuous cattle grazing and livestocks come to water contact to in the study areas.

7. CONCLUSIONS

From the result of the present study, it would be concluded that:

- Altogether 2,921 individual snails representing eight (8) species from temporary roadside drainages and ditches were collected and studied for the presence of trematode cercariae.
- Out of 2,921 snails, 92 (3.15%) individual snails harboured by patent trematode infections.
- The over all prevalence infection (7.26%) was recorded in *Lymnaea* spp.
- The prevalence of different cercariae larvae were found to be more in the month of August (4.89%)
- Out of eight (8) species of aquatic snails, only five (5) species were infected by trematode cercariae namely, *Indoplanorbis exustus*, *Lymnaea* spp.,*Gabia orcula*.,*Gyraulus* spp. and *Segmentina* spp.
- No infection was recorded from *Bellamya bengalensis, Bellamya dissimilis* and *Pila globosa*
- The Amphistome cercaria was the most common occurring cercaria found in four (4) types of snails named as *Indoplanorbis exustus,Lymnaea* spp.*Gyraulus* spp. and *Segmentina* spp.
- Out of 735 total faecal samples,189 (25.71%) cases were found to be positive for faecal infection.
- The prevalence of *Fasciola* egg infection is more in the month of September (38.09%) and less in the month of July (15.87%).
- Out of 189,146 (19.86%)cases were found to be positive for trematode (*Fasciola* sp.) egg infection.

8. RECOMMENDATIONS

- The introduction of large number of ducks and other anquatic birds which consume them as food, helps to reduce the number of snails.
- Well cooked snails should be consumed by local people as food.
- Control the buffaloes and other cattle from grazing in the infected areas.
- Livestock mainly ruminants should be drenched at least twice a year to control the pasture contamination.
- Unnecessary drainages, ditches, pools etc. should be filled with soil and others.
- Awareness about trematode diseases and its transmission should be given to local people. Due to the lack of awareness, they use contaminated water for washing cloths with bare foot and bathing with naked body through which some cercariae (esp.*Schistosoma*) have been more chance to penetrate through the skin and can cause disease.

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10. ANNEX I

T-test for significance of an observed sample with correlation coefficient.

S.No	Total No. of examined snails (X)	Total No. of infected snails (Y)	X^2	Y^2	XY
1	91	0	8281	0	0
2	48	0	2304	0	0
3	809	13	654481	169	10517
4	299	06	89401	36	1794
5	1202	37	1444804	1369	44474
6	468	34	129024	1156	15912
7	126	0	15876	0	0
8	66	02	4356	4	132
Total	$\sum X = 2921$	$\sum Y = 92$	$\sum X^2=2348527$	$\sum Y^2=2734$	$\sum XY = 72829$

We have correlation formula,

$$(r_{xy}) = \frac{n\sum XY - \sum X.\sum Y}{\sqrt{[n.\sum x^2-(\sum X)^2][n\sum Y^2-(\sum Y)^2]}}$$

$$= \frac{582632-268732}{\sqrt{[18788216-(2921)^2][21872-(92)^2]}}$$

$$= \frac{313900}{\sqrt{18788216-8532241}}$$

$$= \frac{313900}{\sqrt{10255975 \, x \, 13408}}$$

$$= \frac{313900}{370809.92}$$

$\therefore (r_{xy}) = 0.85.$

This means, that the number of individual snails examined and the number of individual snails infected are positively Co-related with each other.

Test of Significant of the value of r_{xy}.

Ho: $r_{xy} = 0$ (value of r_{xy} is not significant).

Ha: $r_{xy} > 0$ (value r_{xy} is significant).

Again,

$r_{xy} = 0.85$.

n=8

Test Statistic]

Under null Hypothesis (Ho), the test statistic is given by;

$$t = \frac{r\sqrt{(n-2)}}{\sqrt{(1-r^2)}} \quad \text{with (n-2) d.f.}$$

$$= \frac{0.85\sqrt{(8-2)}}{\sqrt{1-(0.85)^2}} \quad \text{with (8-2) d.f.}$$

$$= \frac{2.082}{\sqrt{1-0.72}} \quad \text{with 6 d.f.}$$

$$= \frac{2.082}{\sqrt{0.28}} \quad \text{with 6 d.f.}$$

$$= \frac{2.08}{0.53}$$

$$= 3.92$$

The calculated value of ‘t’ is 3.92 for 6 d.f. The Tabulated value of ‘t’ is 2.45 for the Confidence level 95% (0.05). Here the calculated value of ‘t’ is more then the tabulated value at 95% confidence level. So, that the value of ‘t’ is significant, i.e. Alternative Hypothesis Ha: $r_{xy} > 0$, is accepted and

Null Hypothesis (Ho): $r_{xy} < 0$, is rejected.