

I

INTRODUCTION

Human societies have relied for centuries on animal agriculture to provide food, fiber, power, fertilizer and employment. This is more apparent in developing countries when human activities are interwoven with the raising of livestock. Agriculture dominates the economy of Nepal with livestock revenues accounting for 30 percent of the Agriculture Gross Domestic Product (AGDP). Nepal has a strong animal agriculture with 14 percent of the national gross domestic product desired from livestock. More than 90 percent of the farmers in rural areas supplement their income by the sale of products from domestic animals. Furthermore, domestic animals are idolized and scarified in really every religious ritual. Thus, the significant economical cultural importance of these animals to the Nepalese society deserves special attention in the planning of policies and national strategies.

Babulus bubalis (buffalo) also known as Asian animal play an important role in farmers economy. It is one of the most important species of domestic livestock as a source of dairy, meat, manure and drought power in Nepal. Buffaloes are substantially distributed in more than 25 tropical countries stretching from Southern Europe through India and China to the full of south-east Asia. Buffaloes in Nepal are widely distributed from sub-humid regions of subtropics in Terai to cool temperate regions in the hills and mountains (Shah, 1981, Shree G., 1982). Epstein (1977) has classified Nepalese buffaloes in four regional types: terai, hilly, midland and himalayan mountain. These types are distinguished by size shape, curvature and length of their horns and the colour of their coats.

Buffalo farming is an integral part of Nepalese livestock industry. Buffaloes are multipurpose and economically the most important livestock species raised under the farming system of Nepal. They are better converter of poor quality roughage and crop residue into protein rich human food.

There are about 42,04,887 buffaloes in Nepal in year 2005/06 with an average of 0.32 buffaloes per household (VEC: national information, 2006). The number of buffaloes increased upto 43,66,813 in 2006/07 (MOAC, 2007). Nepal exported about 5, 720 buffaloes during 2002/2003 and 7,423 buffaloes in the year 2003/2004 to India and abroad through the main routes Kakarvitta (IELA and LP, Nepal 2002/03-2003/04).

Nepal produced about 1,42,040 metric tons, buffalo meat and 9,26,950 metric tons buffalo milk in the year 2005/06 (VEC: National information, 2006) and the animal production of meat per household was 51.54 kg and milk per household was 308.5 kg in the year 2005/06. The number of milking buffaloes in the year 2005/06 was 10,84,764 buffaloes (VEC: National Information, 2006). According to livestock production, Nepal has produced 1,47,031 metric tons buffalo meat and 9,58,603 metric tons buffalo milk in the year 2006/07 and the number of milking buffaloes was 11,24,454 in 2006/07 (VEC: national information, 2007). The annual production of milk was 311.2 kg per household and meat 52.64 kg per household in the year 2006/07 (MOAC, 2007).

Buffaloes diseases have been identified as one of the major factor which have disrupted the development of the industry in Asia and have caused substantial economic loss to the poor subsistent farmers in the developing countries (Othman and Baker, 1981). The parasitic diseases are not less important in buffaloes than other infectious diseases. These

mainly include gastro-intestinal helminthosis, coccidiosis, fascioliasis and mange (Othman and Baker, 1981).

Trematode species play vital roles which are parasitic in livestock. All the trematode species which are parasitic in livestock belong to the subclass Digenea. In general, these trematodes (known commonly as flukes) are dorso-ventrally flattened, some being leaf shaped and some long and narrow, the gastro intestinal flukes have thick fleshy bodies. The *Schistosomes*, which also belong to this group, are elongated and almost roundworm like in appearance. The flukes that parasitize livestock are hermaphrodites (except the *Schistosomes*) but they require an intermediate host to complete their life cycles. Most flukes are very discriminating in their choice of snail as intermediate host and the geographic distribution of trematode species is dependent on the distribution of suitable species of snails.

Liver fluke disease or fascioliasis is a parasitic disease in herbivorous by a flat worm (*Fasciola* spp) world wide. A large variety of animals such as cattle, buffaloes show infection rate that varies from 70% to 90% in some areas. The different local names of this disease such as namle, mate, lew etc., in different regions, are proof of its continued existence for many years in the animal population of the country.

Infection of domestic ruminants with *Fasciola hepatica* and *Fasciola gigantica* cause significant loss estimated at over US \$ 2000 million per annum to the agriculture sector world wide with over 600 million animals affected (Boray, 1985, Hillyer and Apt, 1997).

Economical losses caused by liver fluke consist mainly in the decrease in meat and milk production. Depending on the degree of parasitism *F. hepatica* may cause meat production decrease upto 20

percent in cattle. The economic loss due to fascioliasis in Nepal was estimated to be Rs. 14.2 crore (Lohani and Rasaili, 1995). Severe infection may cause death in young animals. Fascioliasis causes decreased fertility and it increases susceptibility of infected animals to other diseases such as salmonellas. The prevalence of fascioliasis ranging between 50% to 90% has been reported in buffaloes (Mahato, 1993). In addition Fascioliasis is now recognized as an emerging human disease.

Previous study at Lumle Agricultural Research Centre (LARC) has identified the disease prevalence by mainly affected by the availability of Khetland, paddy cultivation and permanent water sources rather than altitude (Joshi, 1988). Rice straw which is the major feed for livestock during winter months has been reported as the potential source of infection for fascioliasis (Joshi, 1987, Mahoto 1993). Green grasses from near permanent water sources or water lodging areas in Monsoon are another potential source of *Fasciola* infection. Therefore in the Nepalese hills, the major risk period of *Fasciola* infection is during post monsoon and winter months (September to February).

Dicrocoeliasis is the disease caused by *Dicrocoelium* spp. Clinical signs and loss of production are rare in cattle, however older cattle may suffer reduced milk production and loss of weight.

Dicrocoelium spp was first time reported by Mukhia in 2007 in buffaloes where prevalence rate was found upto 20.61%.

Schistosoma spp are the only trematodes that live in the blood stream of warm-blooded hosts. The blood stream is rich in glucose and amino acids. So along with the plasma and blood cells, it represents an environment which is suitable for egg producing trematodes. *Schistosoma* spp causes disease called Schistosomiasis or Bilharzia and is the most

important of helminth disease. The infections are often manifest by acute intestinal signs, the mucosa of the intestine is severally damage and the animal develops profuse sometimes bloody diarrhoea, dehydration and loss of appetite. Over 200 million people are infected in at least 75 countries with 500 million or more people exposed to infection (Arcari, 2000). Most of the species like *Schistosoma spindalis*, *Schistosoma japonicum* and *Schistosoma bovis* has been reported among buffaloes from Surkhet district (Ghimire, 1987). In Satungal, Kathmandu prevalence rate of *Schistosoma* spp was found 46.9%. (Mukhia, 2007).

Significance of the study:

Many trematode species have been recorded in other part of globe and in Nepal. In Nepal majority of work has been done only on *Fasciola* spp and a little work has been done regarding other genera of trematodes. An attempt has been undertaken to study the present topic entitled “Prevalence of eggs of three trematode genera (*Fasciola* spp, *Dicrocoelium* spp and *Schistosoma* spp) in buffloes of Satungal slaughter house, Kathmandu,” This study will help to find out the prevalence rate of trematode infections in buffaloes.

Moreover, the present study may help to the future investigator to advance their knowledge. The present study throws lights on different problems found by public and butcher notifying the burden of infection of the trematodes. Less attention has been paid toward the study on parasitic zoonotic diseases in relation to buffalo. The next steps to be considered are the most practical economic way by which goal can be achieved. Continuous research is vitally important.

II LITERATURE REVIEW

Upto the middle of the 17th century, knowledge of parasitology was limited to a few external parasites such as lice and flies, and some internal parasites as roundworms, pinworm, tapeworm, but they were considered as natural products of human bodies.

Linnaeus gave another view about these internal parasites that they originated from accidentally swallowed free living organisms. However, this belief was erased in the later half of the 17th century by Francisco Redi, the grandfather of parasitology. He demonstrated development of maggots from eggs of flies. He also proved that Ascariasis had males and females and produced eggs. At the same time, Leeuwenhoek perfected microscope and discovered protozoan parasites.

Literature Review in Context of World

In 1379, Brie de was the first to describe trematode *Fasciola*.

In 1818, Rudolphi was the first to report *Dicrocoelium*.

In 1851, Bilharz was the first to demonstrate the adult worm of *Schistosoma* in mesenteric veins of a man in Cairo.

Blamire *et al.*, (1970), through the 1960, the condemnation rate for fascioliasis in adult cattle in England and Wales ranged between 22% and 35%. It then fell steadily from 20% in 1970 to 6% in 1978 (Blamire *et al.*, 1980). In 1978, the rate for fascioliasis in England and Wales was 5.6% (MAFF, 1979) and in 1980 6.5% (MAFF, 1980).

Dumag *et al.*, (1975) in a survey of 522 water buffaloes in an endemic province an incidence of 94.2% was found positive for *Fasciola*

eggs. In some areas the incidence of liverfluke infection varied from 95%-100%.

Kenneth J. Lameta (1981), of the 330 cattle, *Fasciola* spp was found in 3.64%. *F. gigantica* was recovered in 3.03% of the 330 cattles. *F.hepatica* was found only in mixed infection with *F. gigantica* in 0.60% of cattle.

Molif. Rafig, Matto *et al.*, (1989) found 42.07%, 35% and 35% buffaloes infected with *Fasciola gigantica* at Bareilly, Ludhiana and Patna respectively. *Schistosoma indicum* was detected in the liver of only one buffalo at Bareilly.

Anwar and Gill (1990) reported prevalence of *Schistosmes* of veterinary importance in Pakistan and India. A total of 20,000 examined animals from different localities of Punjab province of Pakistan; 13% cattle and buffaloes infected with *S. indicum* and *S. spindalis*.

WHO (1993) reported *Schistosoma* spp in about 25% of cow, water buffalo, dog and pigs in Philippines.

WHO (1993) in China, 40 species of wild and domestic animals have been found naturally infected with *S.japonicum*.The main animal reservoirs are cattle,buffalo,pig and dogs.

Marques and Seroferneker (1999) conducted a study for *F. hepatica* infection in buffaloes in the state of Rio Grande Do Sul Brazil. They examined 105 slaughtered buffaloes at a meat packing plant between April 1999 and Nov. 1999, in Viamao in Rio Grande Do Sul in Southern Brazil. The occurrence rate of *Fasciola hepatica* was 20% for buffaloes.

Iassan *et al.*, (2000) conducted a study, on prevalence of fascioliasis in buffaloes in Giza Governorate. They collected 1,042 buffaloes faecal samples. Their coprological examination revealed that 16.46% of the examined buffaloes were harbouring *Fasciola* spp.

Oliveria *et al.*, (2002) reported hepatic fascioliasis in buffaloes in Abattoir of Vale Do Ribeira, Sao Paulo - Brazil. Hepatic fascioliasis among 130 slaughtered buffaloes was studied at the Cajati Abbatoir from September 2002 to December 2002. The Incidence of livers infected by *Fasciola hepatica* was 75%.

Magbool *et al.*, (2002) undertook as epidemiological studies at slaughter houses, livestock, farms, veterinary hospitals and on household buffaloes under the different climatic condition existing in Punjab province. Infection rate was 25.59% at slaughter houses, 20.16% at livestock farms, 13.7% at veterinary hospitals and 10.5% at household buffaloes. Overall highest 24% sexual prevalence in all types of buffalo was recorded during autumn, spring and winter. It was found that a higher infection rate was recorded in older buffalo than in youngsters (below 2 years of age). Buffalo of either sex were equally affected.

Lezeriuc *et al.*, (2002) between 1995 and 2001, 28,878 cattle were slaughtered in the abattoir at Bacau, Romania. During this period 2,220 cattle were diagnosed with fascioliasis and 5,120 cattle with dicrocoeliasis. The prevalence of parasitic infections in cattle was higher in 2001 compared to 1995, especially in the case of bovine fascioliasis which increased from 4.0-14% and bovine dicrocoeliasis which increased from 3.8-37.1%.

Haridy *et al.*, (2002) Fascioliasis in Dakahlia Centres based on parasitological examination of buffaloes and the rate of infection was

9.73%. The mean egg per gram stool was 13.6 for buffaloes. The mean number of *Fasciola* works was 62.7. The highly infected buffaloes were in Manzalla (19.29%) and the lowest was in Mit Ghamr (4.93%).

Akhtar and Mohammad (2003) conducted a study on prevalence of helminthiasis in buffaloes in colony, Hyderabad. The prevalence of helminthiasis was found to be 15.2% in buffaloes. Out of 500 samples examined, 9.2% were infected with nematodes and 5.4% were infected with trematodes respectively. The chief trematodes identified was *Fasciola gigantica* 3.2% and *Fasciola hepatica* 2.2%.

Magboub and Sayed (2003) studied on relationship between one system of climatic condition on the helminthic infection rate at middle delta, Egypt. 1,178 buffaloes owned by farmers from 160 herds belong to 32 villages in Egypt were randomly chosen to study factors which influence the infection with gastro-intestinal parasites. Relationship between number of parasites, herd - size resources of water and season of the year were investigated. The main results showed (i) *Fasciola gigantica* infection recorded the highest percentage 48.04% followed by *Neosaris vitulorum* and *Eimera sp.* (ii) percentage rates of parasitic infection (single, double or triple) in each animal were 62.82%, 29.43 and 7.77% respectively. (iii) infection rate tended to increase with herd size in most cases. (iv) resource of water had highly, significant effect on infection rate.

Yadav *et al.*, (2003) reported the prevalence rate of fascioliasis high in terai region of India followed by hills and plains respectively. Buffaloes were the most susceptible hosts followed by cattle and sheep.

SESA (2003) in Equador the prevalence of *F.hepatica* in livestock remains uncertain because most data were collected by sanitary

inspectors from slaughter houses. The Andean region was the endemic area of fascioliasis where prevalence of infection in livestock ranges from 20.60%.

Bory (2003) prevalence of liver fluke in buffaloes and beef cattle slaughtered at VISSAN. The infection in buffaloes and beet cattle were 14.83% and 22.92% respectively. *F. gigantica* infected both buffaloes and cattle where *F. hepatica* infected only buffaloes.

Basu *et al.*, (2003) reported *Fasciola hepatica* and *F. gigantica* to be the major parasites involved, and causing economic losses to livestock in E. Africa. *F. hepatica* has shown to be the most important fluke species in Ethiopian livestock with distribution over 3 quarters of the nation except in the arid north-east and east of the country. The distribution of *F. gigantica* was mainly localized in the western humid zone of the country. The prevalence of bovine fascioliasis has shown range from 11.5% to 87%. A rough estimate of the economic loss due to fascioliasis in bovine is about 350 million birr per year.

Swai *et al.*, (2004) surveyed of the prevalence of gastro - intestinal (GI) parasite in grazing cattle in pastoral farming area during the period of March 2004. Data were gathered from 17 herds /farms with a total of 90 cattle in five wards (Sale, Pinying, Malambo, Sapiro and Digodigo) of Ngorongoro district. Trematode infections were found in 56.6% of the cattle. Most farms (94%) had trematode infections. Trematode infections were influenced by level of tick infection and the location of farm.

Banglapedia (1988) : *Schistosoma spindalis* and *S. indicum* was prevalent among cattle all over Bangladesh. Mostly, adult cattle above 3 years of age were severely affected upto 25% incidence. *S.nasalis* was widespread among cattles and buffaloes all over the country. Its

occurrence was very high (60%) and was very common in the southern districts of Bangladesh.

Carabin *et al.*, (2003) conducted a study to estimate the sensitivity and specificity of the Danish bilharziasis in buffaloes in Philippines. Faecal samples from animals were collected on five consecutive days in four villages between January to July 2003.

Literature Review in Context of Nepal

Parajuli (1967-1992): a total of 37 faecal samples of buffaloes were examined. Sample positive for flukes were 31 (83.78%) in buffaloes and sample positive for *Fasciola* were 21 (56.75%).

Lolani and Jaeckle (1981-82) conducted a study to identify *Fasciola* spp in Palpa. Livestocks specimens were collected from five slaughtering places of Tansen in the last week of July and beginning of August 1981. Identification was done by Hoerning Institute of Parasitology, University of Berna and results were mixed infections with predominance of *Fasciola gigantica*.

Ghimire (1987) conducted a study on incidence of common diseases of cattle and buffaloes in Surkhet district. The endoparasitic infections recorded were fascioliasis, toxocariasis, paramphistomiasis, internal schistosomiasis and monieziensis.

Mahato (1993) reported the prevalence of fascioliasis ranging between 50-90% in buffaloes of Nepal.

Mahato (1993) reported *Fasciola* prevalence of 57.9% in buffalo in the hills and 4.3% in the terai.

Acharya (1996) conducted a study on efficiency of Trichobendazole and Oxyclozanide against fascioliasis of lactating buffaloes and cattle. The study was conducted between January 1996 and March 1996. Of the 317 lactating cow and buffaloes examined 21.6% cows and 30% buffaloes were positive for *Fasciola* infection. However, buffaloes did not response in either treatment groups as effectively as cows.

Joshi *et al.*, (1997) reported more than 50% *Fasciola* spp in cattle and buffaloes in different parts of Nepal.

Mahato *et al.*, (1997, 2000) reported an epidemiological basis of the control of fascioliasis in Nepal. Despite increased awareness of the disease and massive increase in the use of anthelmintics, they found no impact on the prevalence of the disease in the last two decades. Failure to control the disease here mainly due to lack of information about its epidemiology in country.

Shrestha and Joshi (1997) carried out a study to evaluate the effectiveness of a strategic drenching against fascioliasis in cattle in the western hills of Nepal. Faecal samples were collected at monthly intervals and were examined by standard sedimentation method for the presence of *Fasciola* eggs. The strategic drenching reduced the overall infection in treated animals.

Sharma (1998-99) conducted a study on parasitic infection in animals of Panchthar district. fascioliasis was found in 40.12%.

Regmi *et al.*, (1999) conducted a study to know the fascioliasis prevalence in Thuladihi VDC of Syanja district. Coprological

examination revealed that 67.66% buffaloes and 62.10% cattle were affected with fascioliasis.

Ghimire and Karki (1999) conducted a coprological study and it revealed a higher prevalence of fascioliasis in the adult buffalo population of rice between area of Nagleblure and Lapsiphedhi VDCs of Kathmandu district.

Pandey, Mahato and Gupta (2002) studied prevalence of *Fasciola* infection in *Lymnea* snails and buffaloes in Devbhumi Baluwa VDC of Kavre District. The infection rate in rice field was found 1.67% springs 1.40% and in irrigation channels 0.99%.

CVL (2002/2003) conducted a study on the prevalence and diversity of *Fasciola* spp in buffaloes in area of Kathmandu valley. 92 faecal samples of buffaloes were analyzed, where 56 (61%) were found positive for *Fasciola* spp.

Adhikari *et al.*, (2003) conducted a study on the prevalence and diversity of *Fasciola* spp in buffaloes and cattle in areas of Kathmandu valley from April 2003 to 30 June 2003. The prevalence of *Fasciola* spp was found to be 36 percent in cattle and 61 percent in buffaloes respectively.

Mukhia, G. (2007) conducted a study and reported 90.90 percent samples positive for trematodes in buffaloes of Satungal VDC, Kathmandu. *Schistosoma* spp was found in 46.94 percent followed by *Fasciola* spp 32.6 percent and *Dicrocoelium* spp 20.61 percent.

III OBJECTIVES

Ñ **General Objective:**

To study the prevalence of eggs of three trematode genera in the buffaloes of Satungal, live meat purpose Buffalo Market.

Ñ **Specific Objectives:**

- To determine the general prevalence of trematodes .
- To determine the species wise and age-wise prevalence of trematode.
- To determine the overall load of eggs per gram of faeces.

IV MATERIALS AND METHOD

A survey was conducted to find out the prevalence of the trematodes in buffaloes of Satungal slaughter house of Kathmandu. The study was carried out at Central Veterinary Laboratory, CVL and Animal Disease Control Section, Tripureshwor, Kathmandu.

The essential materials required in this research study are mentioned below.

Ñ **Materials and Equipments**

1. Plastic bags	5. Slides	9. Eye-dropper
2. Gloves	6. Coverslips	10. Cotton
3. Microscope	7. Beaker	
4. Refrigerator	8. Small flat top tubes	

Ñ **Chemicals**

1. 10% formalin
2. Methylene blue

Ñ **Study Area**

Nepal is a South Asian independent land locked country which lies in between China and India having the total area of 5,60,136 square miles i.e. 1,47,181 square km. Geographically, it is 80⁰4" east to 88⁰12 East longitude and 26⁰ 22" North to 30⁰ 27" North latitude. The northern part of the country is separated from Tibet autonomous region of China by the Himalayan peaks forming natural border whereas east, west and south is surrounded by west Bengal, Bihar and other north states of India. Only 17% of the total area is occupied by plains i.e., terai, whereas 68% and 15% of the area are occupied by hills and mountains respectively.

PSEUDOPARASITES

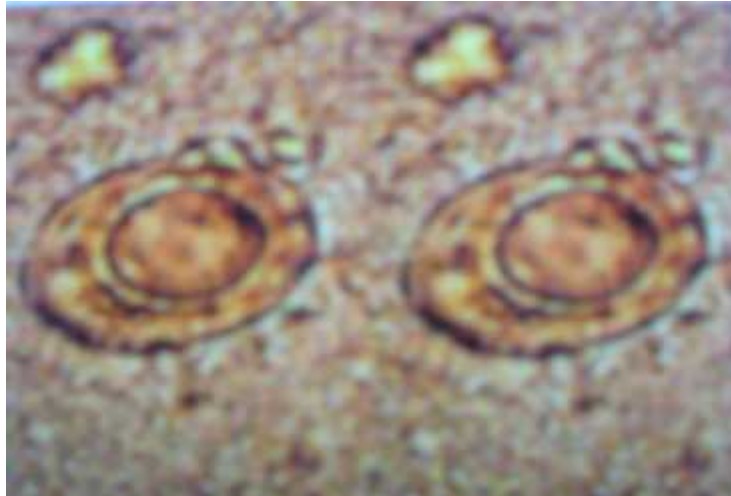


Plate 10: Pollen grain

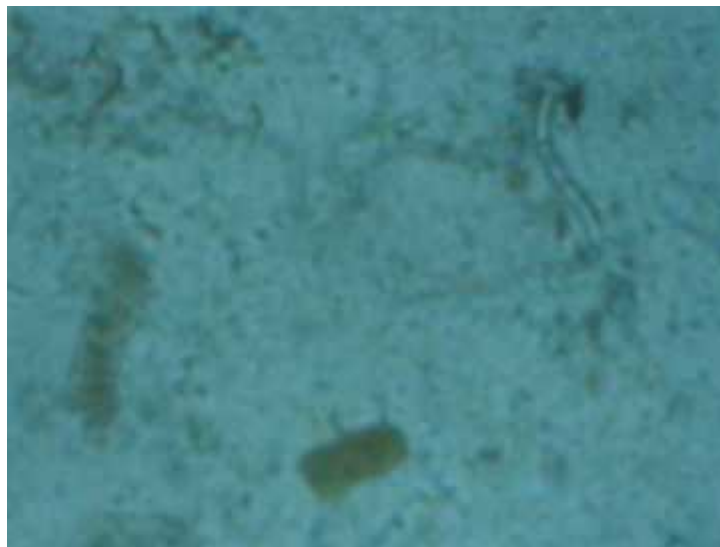


Plate 11: Plant hair

Plate 12: Air bubble

EGGS OF TREMATODES



Plate 7: *Fasciola* spp



Plate 8: *Dicrocoelium* spp



Plate 9: *Schistosoma* spp



Plate1: Preparing for sediment technique



Plate 2: Preparing for sediment technique



Plate 3: Examining under microscope



Plate 4: Buffaloes kept for slaughter purpose



Plate 5: Collecting stool samples



Plate 6: Asking questions with the dealer

The Kathmandu valley is located in Nepal and is the capital city. It stands at an elevation of approximately 4,265 feet (1300m). There are three cities in Kathmandu valley-Kathmandu, Patan and Bhaktapur.

The total population of animal that could contribute to national meat production is comprised of 3.4 million buffaloes (CBS, 2002). It was estimated that the total meat consumption in the country during 2002 was about 2, 03,000 metric tons with per capital consumption of 8.6 kg/annum (CBS, 2002). On analysis the contribution of different animal species in national meat supply, it was evident that buffaloes contribute about 57.4% of total meat supply (Joshi *et al.*, 2003).

Satungal VDC of Kathmandu metropolitan city is surrounded by Gurjadhara and Naikap from its east and west respectively. The total area of Satungal VDC is approximately 2 km. The study was covered out in ward no. 9 of Satungal VDC. The dealing and distribution of buffalo is done at Satungal for the entire places of Kathmandu. Daily 2-3 trucks carrying buffaloes are brought to Satungal for slaughter purpose but in 2 days of a week i.e. Monday and Wednesday the number of trucks carrying buffaloes are 10-12. The number of buffaloes in each truck are according to their age and size. The number of big size buffaloes are 18-20 in one truck and 30-32 are the number of small size buffaloes in one truck. The buffaloes are brought to Satungal from many places such as Nepalgunj, Jamuniya, Jitpur, Nawalparasi, Sakuwa are the main places from where maximum number of buffaloes are brought to Satungal for slaughter purpose whereas Janakpur, Kailali, Guankhel, etc. are second places.

Ñ **Faecal Sampling**

210 faecal samples were collected from Satungal, Kathmandu.

- Fresh faecal samples for parasitological examination were collected.
- 10% formalin was added into the faeces to preserve the parasite eggs.
- Collected samples were dispatched as soon as possible to a laboratory in plastic bags.
- Samples were immediately stored in the refrigerator (4⁰c) until they were processed.

Ñ **Stool Examination**

The stool samples were examined by sedimentation technique.

Ñ **Sedimentation Technique**

The sedimentation technique is a qualitative method for detecting trematodes eggs in the faeces. Most trematodes eggs are relatively large and heavy. This technique concentrates them in a sediment.

Ñ **Method**

3 gm of stool sample was taken in a beaker and 50ml of water was added. With the help of motor and pistle, the sample was grinded highly and filtered with a tea strainer. The filtered sample was poured into plastic tube of 15ml and centrifuged at 1000 rpm for 15 minutes. The tube was taken out and removed the water with the help of pipette. A drop of sediment was taken with the pipette and placed on the slides, added drop of methylene blue into it and was covered with a cover slip.

Ñ **Microscopical Examination of Prepared Sample**

The prepared samples on microslides from the sedimentation method were examined under a microscope at the magnification level of 4x and 10x.

Ñ **Faecal examination using the stroll egg counting method:**

According to Dr. Tom Nolan, University of Pennsylvania (2004) it is the easiest quantitative method to count the number of eggs present in the field without the help of McMaster.

3 gm of faeces was taken in a beaker and 42 ml of water was added. Using a tongue depressor, 3 gm of faeces was pushed through a sieve into the water. Then the sieve was lift and was hold over the dish. Then the remaining water was push out from the faeces. After stirring the water-faeces mixture 0.15 of the suspension was taken and spread over 2 slides. Each slide was cover with a long coverslip. Then both slips were examined for eggs. The total amount of eggs counted X 100 represents the number of eggs per gram of faeces.

V

RESULTS

A total of 210 stool samples were collected from the study area ward no. 9 Satungal VDC where buffaloes are brought from out of the valley and kept for sale for slaughter purpose.

In the present study three different types of trematodes were examined with the help of sedimentation technique.

Ñ **General Prevalence of Trematodes in Buffaloes**

Out of 210 samples examined for trematode, 130 samples were found positive for trematodes.

Therefore, the general prevalence of trematodes in buffaloes was found to be 61.90 percent.

Ñ Prevalence of Trematode Genera in Buffaloes

Out of 61.90 percent trematodes, three different genera viz. *Fasciola* spp, *Dicrocoelium* spp and *Schistosoma* spp were observed 38.57 percent samples was found positive for *Fasciola* spp, 28.10 percent was found positive for *Schistosoma* spp and 18.10 percent was found positive for *Dicrocoelium* spp. The difference in the prevalence of different genus of trematodes was found statistically to be significant in buffaloes ($\chi^2 = 16, P < 0.05, d.f = 2$)

Table No.1: Prevalence of trematode genera

Total No. of samples examined	Trematode Genus	No. of positive samples	Prevalence %
210	<i>Fasciola</i> spp	81	38.57
	<i>Dicrocoelium</i> spp	38	18.10
	<i>Schistosoma</i> spp	59	28.10

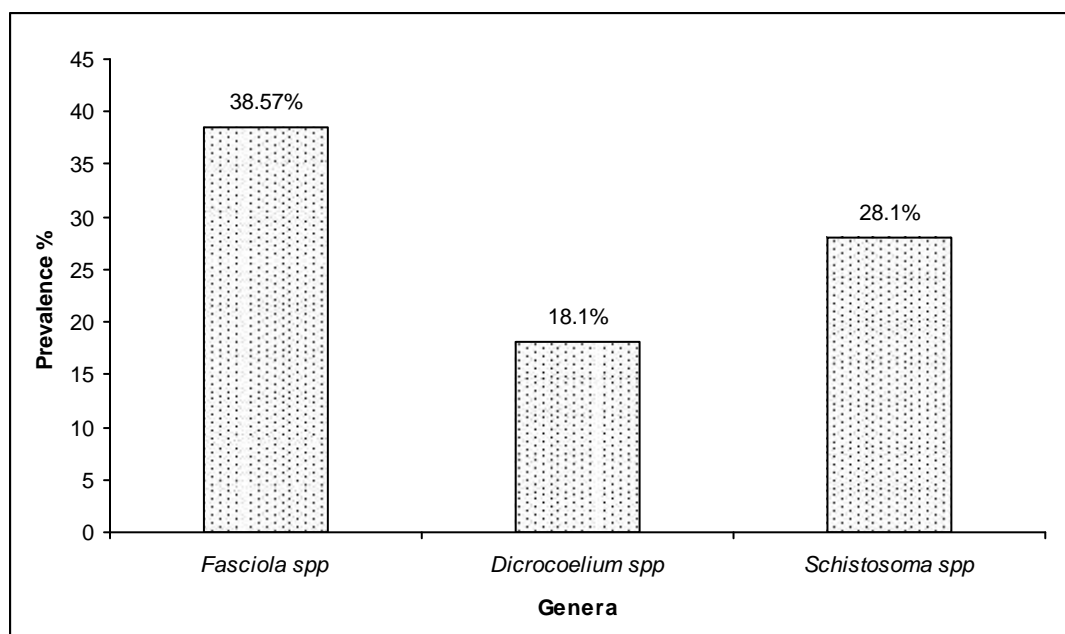


Figure 1: Prevalence of trematode genera

Ñ Prevalence of Single Infection of Trematode Genus in Buffaloes

Among 210 samples examined single infection was found the highest of *Fasciola* spp 5.24% followed by *Schistosoma* spp 2.38% and *Dicrocoelium* spp 0.95 % (Table 2). The prevalence of difference of single infection of trematodes was found statistically to be significant in buffaloes ($\chi^2 = 7$, $P < 0.05$, d.f. = 2).

Table No.2: Prevalence of single infection of trematode genus

Total No. of samples examined	Trematode	No. of positive samples with single infection	Prevalence %
210	<i>Fasciola</i> spp	11	5.24
	<i>Dicrocoelium</i> spp	2	0.95
	<i>Schistosoma</i> spp	5	2.38

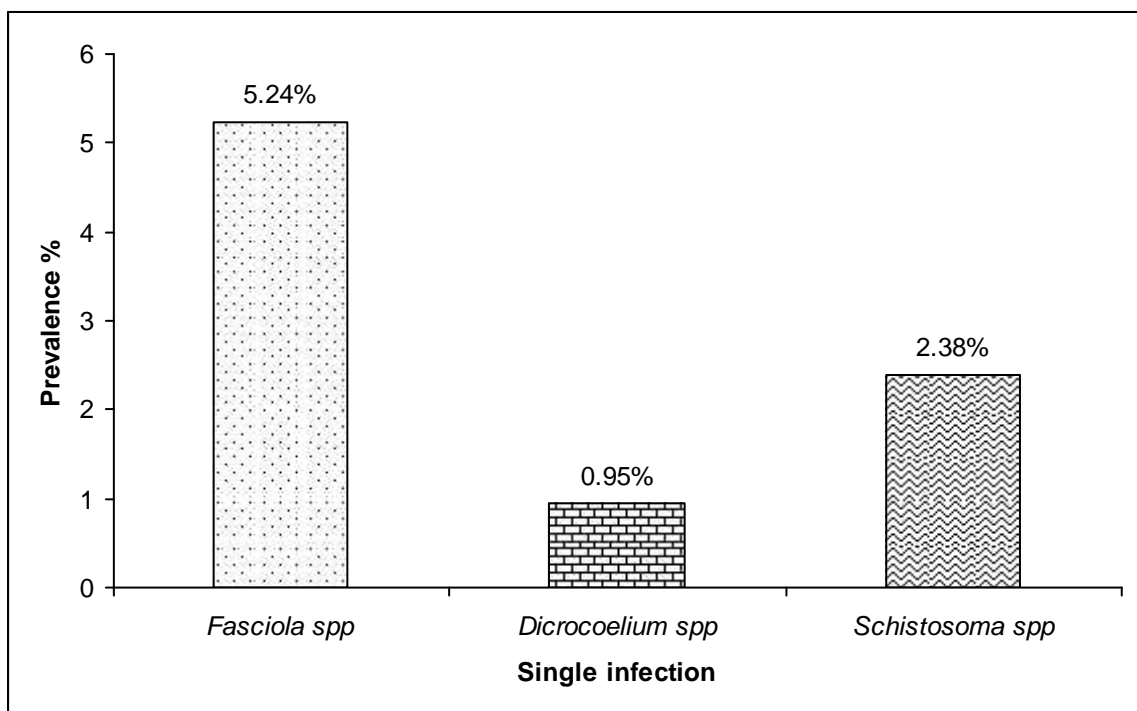


Figure 2: Prevalence of single infection of trematode genus

Ñ Prevalence of Mixed Infection of Trematodes

Out of 210 samples examined, mixed infection of *Fasciola* and *Schistosoma* was found to be in the highest with 4.76%, followed by *Fasciola* and *Dicrocoelium* 3.81%, then by *Fasciola*, *Dicrocoelium* and *Schistosoma* 3.33% and lastly by *Dicrocoelium* and *Schistosoma* 2.38%.

Total no. of samples examined	Mixed parasites	No. of mixed genera positive slides	Prevalence %
210	<i>Fasciola</i> + <i>Dicrocoelium</i> + <i>Schistosoma</i>	7	3.33
	<i>Fasciola</i> + <i>Dicrocoelium</i>	8	3.81
	<i>Fasciola</i> + <i>Schistosoma</i>	10	4.76
	<i>Dicrocoelium</i> + <i>Schistosoma</i>	5	2.38

Table No.3: Prevalence of mixed infection of trematodes

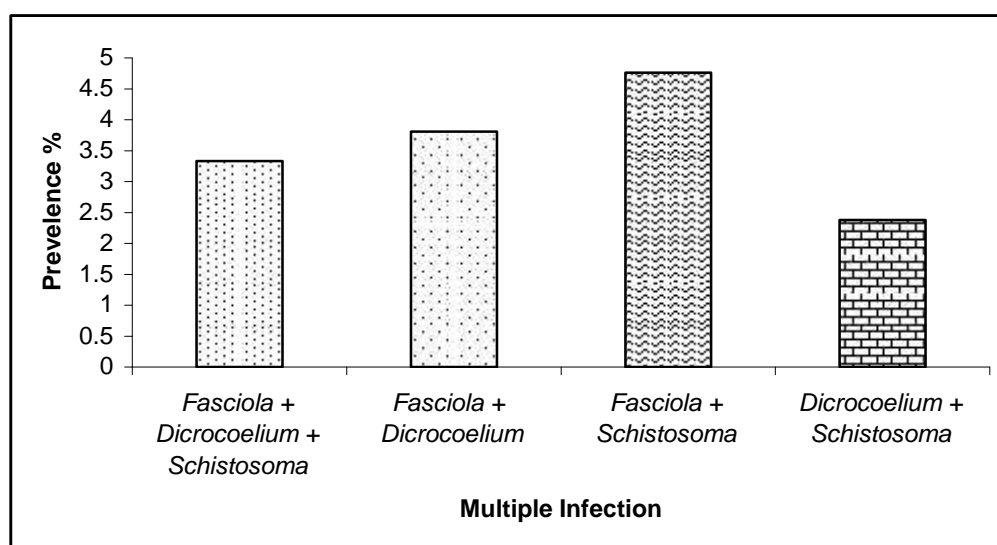


Figure 3: Prevalence of mixed infection of trematodes

Ñ Age-wise Distribution of Trematodes

According to age-wise distribution of trematodes, buffaloes below 2 years (200-300kg) were found to be less infected (46.99%) than buffaloes above 2 years (above 300 kg) with 71.65 %. Age-wise prevalence of different genus of trematodes results statistically significant in buffaloes ($\chi^2=21$, $P<0.05$, d.f. = 1)

Age of buffaloes	Total samples examined	No. of positive samples	Prevalence %
Below 2 years (200-300kg)	83	39	46.99
Above 2 years (Above 300kg)	127	91	71.65

Table No.4:Age-wise distribution of trematodes

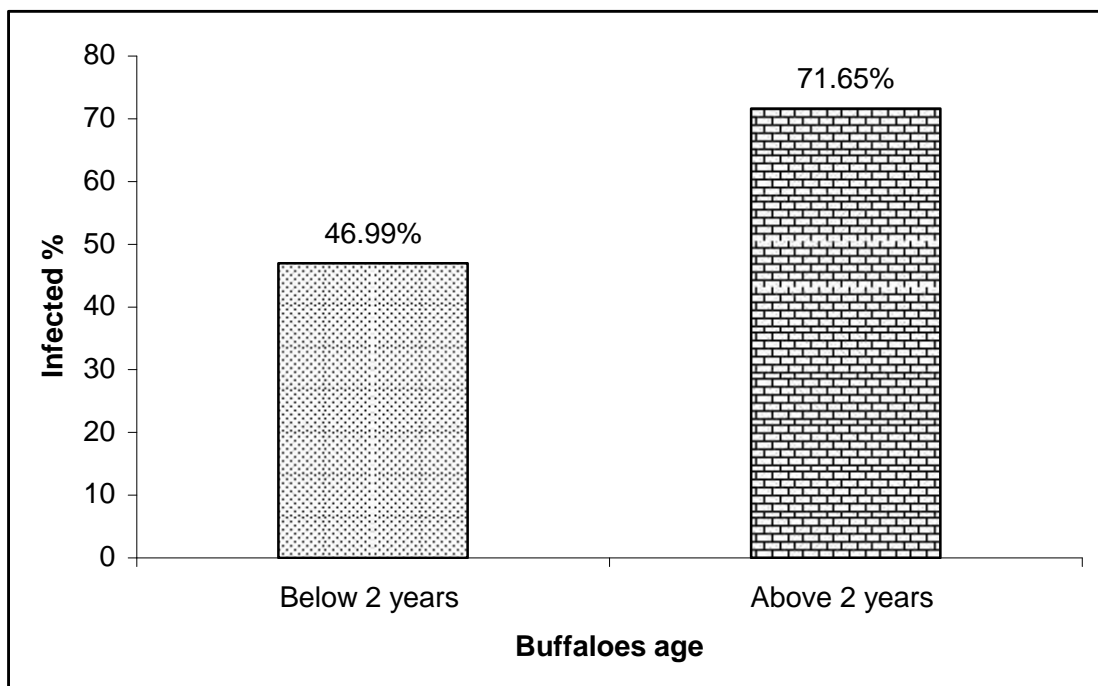


Figure 4: Age-wise distribution of trematodes

Ñ Genus-wise Prevalence of Trematodes in Buffaloes Below 2 Years

83 samples of buffaloes below 2 years were examined. The highest prevalence was found of *Fasciola* spp (30.12%), then *Schistosoma* spp (18.07%) and at last *Dicrocoelium* spp (13.25%).

Total samples examined	Trematodes	No. of Positive samples	Prevalence %
83	<i>Fasciola</i> spp	25	30.12
	<i>Dicrocoelium</i> spp	11	13.25
	<i>Schistosoma</i> spp	15	18.07

TableNo.5: Prevalence of trematode genera in buffaloes below 2 years

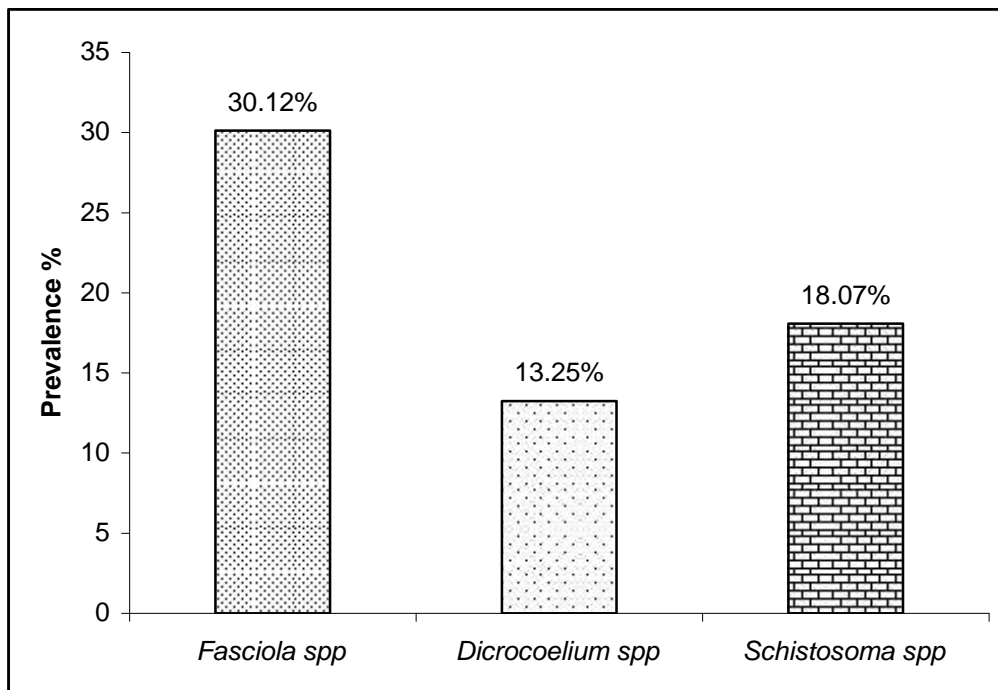


Figure 5: Prevalence of trematode genera in buffaloes below 2 years

Ñ Genus-wise Prevalence of Trematodes in Buffaloes Above 2 Years

Among 127 samples tested of buffaloes above 2 years, positive sample for *Fasciola* spp was found highest with 44.09%, followed by *Schistosoma* spp 34.65% and *Dicrocoelium* spp 21.26% respectively.

Total samples examined	Trematodes	No. of Positive Sample	Prevalence %
127	<i>Fasciola</i> spp	56	44.09
	<i>Dicrocoelium</i> spp	27	21.26
	<i>Schistosoma</i> spp	44	34.65

Table No.6: Prevalence of trematode genera in buffaloes above 2 years

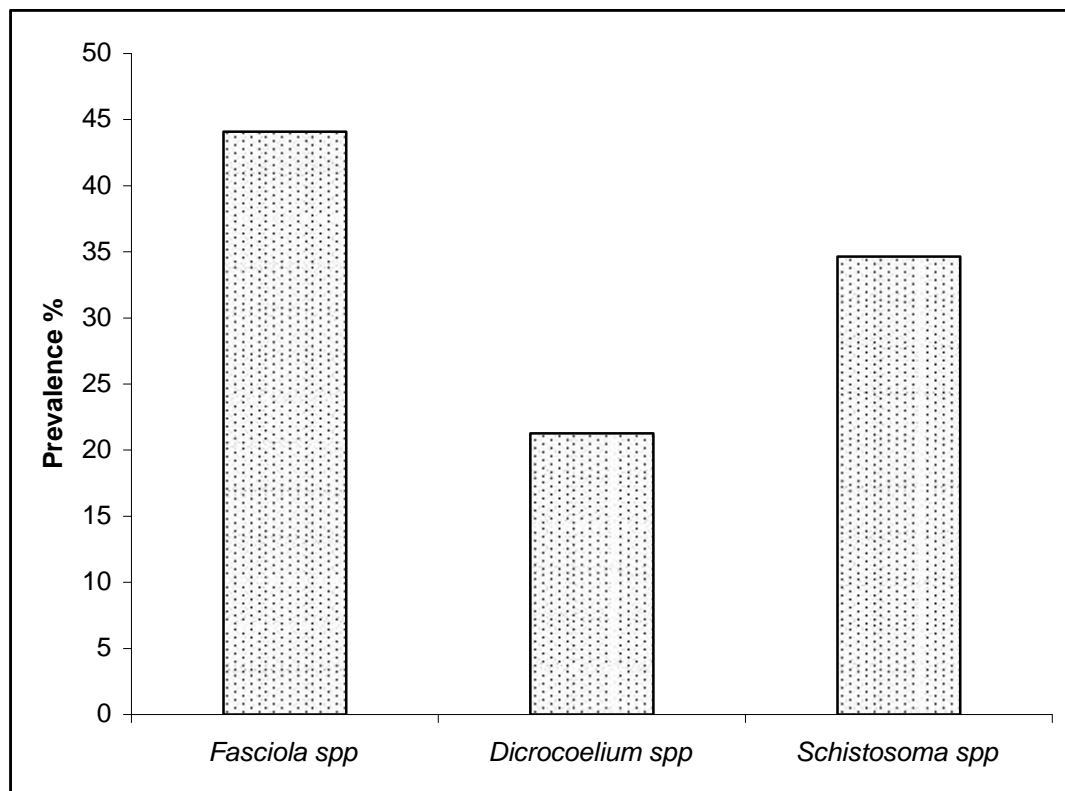


Figure 6: Prevalence of trematode genera in buffaloes above 2 years

Ñ Age-wise Single Infection of Trematodes in Buffaloes

Among 210 samples examined, single infection show high prevalence in buffaloes above 2 years than among buffaloes below 2 years of age.

Total samples examined	Trematodes	Total prevalence (single infection)	Below 2 yrs prevalence Nos.	Above 2 yrs prevalence Nos.
210	<i>Fasciola</i> spp	11	4 (1.90%)	7 (3.33%)
	<i>Dicrocoelium</i> spp	2	0 (0%)	2(0.95%)
	<i>Schistosoma</i> spp	5	1 (0.48%)	4 (1.90%)

Table No.7:Age-wise single infection of trematodes

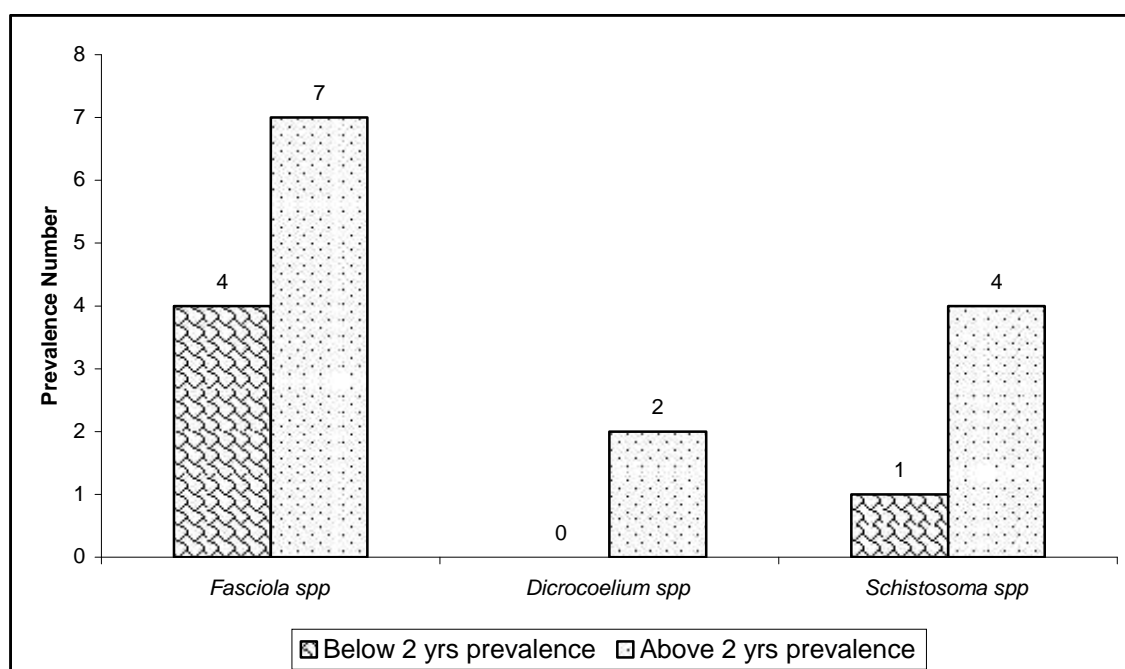


Figure 7: Age-wise single infection of trematodes

Ñ Intensity of Infection of Trematode Genera in Buffaloes

Among 210 samples examined, light infection was found the highest in all the genera of trematodes. Comparatively all the three types i.e. light, mild and heavy infections were found the highest in *Fasciola* spp followed by *Schistosoma* spp and *Dicrocoelium* spp (Table no. 8).

Total samples examined	Trematodes	Infection rate(Nos.)		
		Light	Mild	Heavy
210	<i>Fasciola</i> spp	22	22	7
	<i>Dicrocoelium</i> spp	25	8	4
	<i>Schistosoma</i> spp	28	15	6

TableNo.8: Intensity of infection of trematode genera

Light infection = less than 2 ova per field

Mild infection = 2-5 ova per field

Heavy infection = 5 or more ova per field

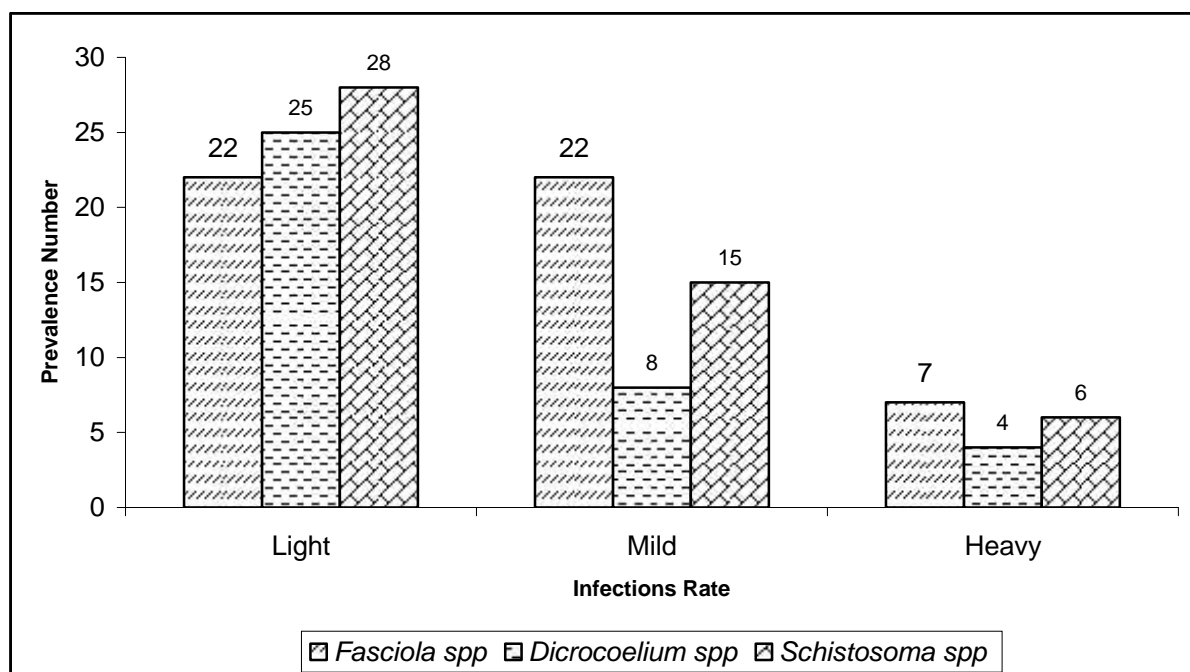


Figure 8: Intensity of infection of trematode genera

VI

DISCUSSION AND CONCLUSION

The aim of the study was to investigate the prevalence of three genera of trematodes (*Fasciola* spp, *Dicrocoelium* spp and *Schistosoma* spp) in buffaloes. The investigation was conducted in buffaloes and the area of study was ward no. 9 Satungal VDC, Kathmandu. The area selected for the study was the main centre where buffaloes are brought for slaughter purpose from various places like Nepalgunj, Jitpur, Gaur, Jamunia, Janakpur, etc.

During the study period a total of 210 faecal samples were examined. The present study revealed that 130 (61.90%) of the buffaloes were infected by three different kinds of trematodes. In the present study three different types of trematodes were examined *Fasciola* spp (38.57%), *Schistosoma* spp (28.10%) and *Dicrocoelium* spp (18.10%).

Identification of the eggs of trematodes:

210 stool samples were examined with the help of sedimentation technique, 130 (61.90%) samples were found to be positive for trematodes. The overall prevalence of different species of trematodes result statistically significant ($\chi^2 = 1024$, $P < 0.05$, d.f. = 29.88).

Altogether 3 genera of trematodes were examined. The eggs of trematodes have been identified according to their characters and morphology.

Description of eggs

*** *Fasciola* spp**

Eggs are 130µm by 150µm by 63-90µm in size, yellowish in colour, consist of embryonic mass and shell, operculum usually indistinct.

*** *Dicrocoelium* spp**

Eggs are 36.45µm by 20-30µm in size, operculated and thick shelled, brown in colour.

*** *Schistosoma* spp**

Eggs are 200µm by 70-90µm in size, spindle shaped, flattened at one side, greatly elongated with straight slender terminal spine.

Trematodes (*Fasciola* spp, *Dicrocoelium* spp and *Schistosoma* spp) are parasitic zoonoses and produce significant mortality and morbidity in the human and are responsible for major economic losses by affecting the animal health. Therefore, trematodes are also found in other host in great number.

Discussion

Ñ *Fasciola* spp

In 1758; Linnaeus, reported *F. hepatica* from the bile ducts of the sheep and other ruminants.

In 1978, Caple, reported *Fasciola* spp in Asian elephant.

In 1980, Fredrick and Reece, reported *Fasciola hepatica* in cow in Solomon Islands.

In 1981, Sprall, reported *F. hepatica* in Marsupial in Southeastern Australia.

In 1996, Nooruddin and Islam, reported *F. gigantica* in goat population of Bangladesh.

In 2001-2007, Michigan DNR wildlife disease laboratory reported *F. magna* in deer in Peninsula.

In 2001, Valero reported *F. hepatica* in murid rodents on the Island of Corsica.

In 2003, Esteban, reported human fascioliasis caused by *F. hepatica* and *F. gigantica* in the Andean highlands of South America.

In 2005, Villaricecio, reported *Lymnaea cousini* naturally infected with *Fasciola hepatica* in Ecuador.

In 2005, Marcos, reported high prevalence of human fascioliasis.

In 2006, Ayele, reported *Fasciola* spp in donkeys in Dugda Bora, district, Ethiopia.

From Nepal,

In 1967-92 Mainali, reported *Fasciola* spp from lulu cattle.

In 1970, Singh reported *F. hepatica* in sheep and goat from middle terai.

In 1975, Joshi and Tiwari, reported *Fasciola* spp in yaks and yakows of the Himalayans.

In 1978, Joshi, reported *F. hepatica* in goats from Jamunapari of Nuwakot districts.

In 1981/82, Lohani and Jackle reported *Fasciola* spp from Palpa district.

In 1987, Karki, reported *Fasciola* spp in sheep.

In 1987, Ghimire, reported *Fasciola* spp in cattle, buffaloes and goats from Surkhet district.

In 1997-98, Sharma reported *Fasciola* infection 40.12% in animals from Panchthar district.

In 2003, Rabwin, Joshi and Chettri reported *Fasciola* spp in yaks from Chandanbari, Langtang.

In 2007, Karki, reported *Fasciola* spp in elephants of Nepal.

*** *Dicrocoelium* spp**

In 1899, Looss reported *D. lancaetum* from the bile duct of the sheep, goat and cattle.

In 1990, Sheikh reported *Dicrocoelium* eggs in human beings in Saudi Arabia.

In 1991, Schuster reported *Dicrocoelium* metacercariae in ants of Germany.

In 1996, Jithendran and Bhatt reported *D.dentricum* in sheep and goats in hilly areas of India.

In 1997-2003, Thomas R.G., reported *Dicrocoelium* spp in Colombus monkey of Uganda.

In 1998-2002, Thomas, reported *Dicrocoelium* spp in guenons of Uganda.

From Nepal,

In 2007, Mukia reported *Dicrocoelium lancentum* in buffaloes of Nepal.

In 2007, Karki, reported *Dicrocoelium* spp in elephants of Nepal.

Ñ *Schistosoma* spp

In 1851, Bilharz reported the adult worm of *Schistosoma* in mesenteric veins of a man in Cairo.

In 1876, Sonsino reported *S. bovis* from the portal and mesenteric viens of cattle and sheep.

In 1904, Katsurade reported *S. japonicum* from the portal and mesenteric veins of both man and animals

In 1954, S.R. Roa, reported, *Schistosoma* spp in elephants in Bombay State. India.

In 1966, Montgomery reported *S. spindalis* from the mesenteric veins of ruminants.

In 1993, WHO, reported 40 species of wild and domestic animals have been found infected with *S. japonicum* in China.

In 1993, WHO, reported *Schistosoma* spp in cow, water buffalo and dog and pigs in Philippiness.

In 2004,Agatsuma, reported *Schistosoma* spp in elephant in Sri lanka.

From Nepal,

In 2007, Mukhia reported *Schistosoma* spp in buffaloes of Kathmandu valley.

In 2007, Karki, reported *Schistosoma* spp in buffaloes of Nepal.

From the above discussion it is seen that most of the works have been done on *Fasciola* spp and a little work or study are done on *Dicrocoelium* spp and *Schistosoma* spp in Nepal and outside Nepal. High prevalence of *Fasciola hepatica* i.e., 67.66% has been reported from Thuladihi, Syangia among buffaloes (Regmi *et al.*, 1999) followed by 56.75% *Fasciola* infection from Surkhet district (Parajuli, 1967-92) and 40.12% infection in animals from Panchthar district (Sharma, 1997-98) which were the highest infection among buffaloes comparing to the present study i.e., 38.57 % . Almost similar type of prevalence of the *Fasciola* has been reported from Sau Paulo-Brazil in buffaloes (Oliveira *et al.*, 2002).

Regarding the age-group of buffaloes the present study showed adult buffaloes above 2 years were more infected with *Fasciola*, *Dicrocoelium* and *Schistosoma* (71.65%) than young buffaloes below 2 years (46.99%). A report from Pakistan which showed high infection rate in older buffaloes than in young buffaloes (Maqbool *et al.*, 2002).

Most of the species like *Schistosoma spindalis*, *Schistosoma japonicum* and *Schistosoma bovis* has been reported among buffalo from Surkhet district (Ghimire, 1987).

High prevalence of *Schistosoma* spp ie. 46.94% has been reported from Satungal, Kathmandu among buffaloes (Mukhia, 2007) which was higher among buffaloes comparing to present study i.e. 28.10%. Similar type of high rate prevalence of *Schistosoma* spp has been reported from Bangladesh (Banglapedia).

Dicrocoelium spp (20.61%) reported from Satungal, Kathmandu (Mukhia, 2007) is found to be higher than the present study i.e. 18.10 %.

In the present study, sedimentation technique was used for detecting trematode eggs in the faeces. In several regions of Vietnam where equipment for faecal examination such as a microscope is not readily available Agar Gel Diffusion Test (AGDT) was used to detect *Fasciola* egg (Thuy *et al.*, 2003).

In the present study mixed infection of *Fasciola* spp, *Dicrocoelium* spp and *Schistosoma* spp was also observed which was found to be 14.76%. Pseudo-parasites were also observed in this study. About 10.95% pseudo-parasites were found. Pollen was found in highest number 8(3.80%), followed by crystal 7(3.33%), plant hair 6 (2.85%) and at last air bubbles 2 (0.9%). No work regarding the pseudo-parasites was found.

VII RECOMMENDATIONS

- Proper management of domestic animal including buffaloes has become a must.
- The practical application of biological and epidemiological knowledge should be provided through mass media like radio, television, etc.
- The program for awareness of the meat borne disease and zoonotic disease to the public and butcher should be developed.
- Anthelmintic treatment should be encouraged to the livestock farmers to eliminate the parasites from the host.
- Biological control of snail, the intermediate host should be practiced.
- The research work on helminth parasites of livestock should be encouraged.

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