GASTRO-INTESTINAL PARASITES OF SHEEP (*Ovis aries* Linnaeus, 1758) IN LAXMIPUR VDC, DANG, NEPAL

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T.U. Registration No: 5-2-54-12-2010

T.U. Examination Roll No: 116

Batch: 2071/072

A thesis submitted

In partial fulfillment of the requirements for the award of the degree of Master of Science in Zoology with special paper Parasitology

Submitted to

Central Department of Zoology

Institute of Science and Technology

Tribhuvan University

Kirtipur, Kathmandu

Nepal

February, 2018

DECLARATION

I hereby declare that the work presented in this thesis entitled **"Gastro-intestinal parasites of sheep (*Ovis aries* Linnaeus, 1758) in Laxmipur VDC, Dang, Nepal"** has been done by myself, and has not been submitted elsewhere for the award of any degree. All the sources of the information have been specifically acknowledged by reference to the author(s) or institution(s).

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RECOMMENDATION

This is to recommend that the thesis entitled **"Gastro-intestinal parasites of sheep (*Ovis aries* Linnaeus, 1758) in Laxmipur VDC, Dang, Nepal"** has been carried out by **Miss Gitanjali Rana** for the partial fulfillment of Master's Degree of Science in Zoology with special paper 'Parasitology'. This is her original work and has been carried out under my supervision. To the best of my knowledge, this thesis work has not been submitted for any other degree in any institutions.

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LETTER OF APPROVAL

On the recommendation of supervisor **Lecturer Mr. Janak Raj Subedi**, this thesis submitted by **Miss Gitanjali Rana** entitled **"Gastro-intestinal parasites of sheep (*Ovis aries* Linnaeus, 1758) in Laxmipur VDC, Dang, Nepal"** is approved for the examination and submitted to the Tribhuvan University in partial fulfillment of the requirements for Master's Degree of Science in Zoology with special paper Parasitology.

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CERTIFICATE OF ACCEPTANCE

This thesis work submitted by **Miss Gitanjali Rana** entitled **"Gastro-intestinal parasites of sheep (*Ovis aries* Linnaeus, 1758) in Laxmipur VDC, Dang, Nepal"** has been accepted as a partial fulfillment for the requirements of Master's Degree of Science in Zoology with special paper Parasitology.

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ACKNOWLEDGEMENTS

I would like to express my sincere gratitude to my supervisor Mr. Janak Raj Subedi, lecturer, Central Department of Zoology, Tribhuvan University for supervision, guidance and invaluable suggestion throughout my study. I am highly grateful to Prof. Dr. Ranjana Gupta, Head of the Central Department of Zoology for providing necessary facilities required for this assignment. I would like to express my gratitude to Prof. Dr. Kumar Sapkota, Acting Head of Central Department of Zoology for his valuable suggestions. In addition to this, I express my thanks and best regards to all my respected teachers and staffs of Central Department of Zoology, T.U., Kirtipur.

I am thankful to the inhabitants of Laxmipur VDC for their co-operation during field work. I would also like to thank my respected working staffs of Laboratory for proper guidance and providing necessary equipments to proceed my research.

I would like to extend my deepest gratitude goes to my parents for their support and inspiration in my whole academic career. Lastly, I would like to thank my friends for their kind support throughout my dissertation work.

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LIST OF ABBREVIATIONS

VDC	-	Village development committee
GIT	-	Gastro-intestinal tract
GI	-	Gastro-intestinal
FAO	-	Food and Agriculture Organization
FY	-	Fiscal year
asl	-	above sea level
RBC	-	Red blood cells
PCV	-	Packed cell volume
ml	-	milliliter
mm	-	millimeter
rpm	-	revolutions per minute
sp.	-	species
TU	-	Tribhuvan University
P-value	-	Probability value
^{0}C	-	Degree Celsius

ABSTRACT

Sheep (*Ovis aries*), a member of Bovidae family is supposed to be one of the oldest farm animal domesticated, meant as an important livestock. Gastrointestinal parasite causes significant impact on health of sheep. Present study was conducted to assess the prevalence, seasonal variation and concurrency of gastro-intestinal parasites of sheep in Laxmipur Village Development Committee, Dang. A total of 110 sheep were randomly selected from the study area by visiting to farmer's bawn. 55 faecal samples were collected during the month of April 2017 and rest 55 during the month of August 2017 and examined by direct smear and concentration methods (sedimentation and floatation techniques). The study revealed overall 80% prevalence of gastro intestinal parasites including total 15 genera belonging to 5 classes. The study demonstrated *Haemonchus* sp. as predominant species having the prevalence rate of 33.64%, followed by *Strongyloides* sp. (30.91%), *Strongyle* sp. (25.45%), *Trichostrongylus* sp. (14.55%), *Oesophagostomum* sp. (13.64%), *Fasciola* sp. (10.91%), *Bunostomum* sp. (10%), *Trichuris* sp. (8.18%), *Paramphistomum* sp. (7.27%), *Moniezia* sp. (6.36%), *Nematodirus* sp. (5.45%), *Chabertia* sp. (4.54%), *Eimeria* sp. (3.64%), *Entamoeba* sp. (2.73%), and *Ascaris* sp. (1.82%). Higher infection of gastro-intestinal parasites in sheep was observed in rainy season (92.93%) than in summer season (67.27%). Statistically there was significant difference ($p<0.05$) in the seasonal prevalence of gastro-intestinal parasites in two seasons. Multiple infection was observed in 80.68% and single infection was 19.32% of sheep. Management practices, grazing area, nutritional factors, traditional rearing system, climatic conditions, environmental and geographical factors were considered as important factors which influences the prevalence of gastro-intestinal parasites.

1. INTRODUCTION

1.1 Background:

Ruminants are characterized by the presence of four-compartment stomach which are rumen, reticulum, omasum and abomasum. The rumen is a large storage space for consumed food then later regurgitated, re-chewed, and re-swallowed in a process called cud chewing. The cud is a food bolus that has been regurgitated. Rumination or cud chewing occurs primarily when the animal is resting and not eating. There are about 150 different domestic and wild ruminant species. Some of them include cattle, sheep, goats, giraffes, antelopes, camels, etc.

Sheep (*Ovis aries*) is a domesticated quadrupedal ungulate ruminant mammal that belongs to family Bovidae, specially reared for wool and meat. Antelopes, gazelles, goats, sheep and other relatives fall in this family. Current chromosomal and archeological evidence indicates that the divergence occurred about 9000-11000 years ago and the first sheep domesticated were from the wild species; mouflon *(Ovis musimon*) flocks from Sardinia and Corsica (Grzimek, 1990). Sheep are supposed to be the first farm animals domesticated.

Sheep are globally distributed, widely adapted to different climates and are found in all production systems but mainly found in arid and semi- arid areas. They are primarily grazing animals. They are especially tamed for meat and manure. Besides meat, they provide a vast range of products such as milk, hide and skin, wool, horns, bones, and medicine. Sheep meat production is widely distributed throughout the world. China dominates with a 24% share in 2013, based on data from FAO. Australia and New Zealand are the next largest producers, accounting for 8% and 5% respectively of global production (Colby, 2016). Sheep have been estimated to provide up to 30% of the meat and 15% of the milk supplies in sub-Saharan Africa (Tefera *et al.*, 2009). Sheep and goats represent an important component of the farming system because they require smaller investment; have shorter production cycles, faster growth rate and greater environmental adaptability than cattle (Lebbie, 2004; Anon, 2005). Sheep have become adapted to a range of environments from the cool alpine climate of the mountains to the hot and arid pastoral areas of the lowlands (Mirkana, 2010). They also have lower feed requirements compared to cattle because of their small body size and low metabolic requirements and maintenance needed in arid and semi-arid areas (Bikila *et al*., 2013). This allows easy integration of small ruminants into different farming systems. In the subsistence sector farmers and pastoralists depend on sheep and goats (Hirpa and Abebe, 2008) for much of their livelihood (Kenea *et al*., 2015). They are also called poor man cow. They also play a major role in the food security and social well-being of rural populations living under conditions of extreme poverty (Duguma *et al.*, 2010). Unlike goats, sheep hardly damage any tree. Sheep have the ability to produce prime carcasses on roughage alone, thus they are well adapted to many areas unable to produce grain profitably. Sheep dung is a valuable fertilizer, and since they are grazed on sub-marginal lands, their droppings are the only means of improving the growth of plants in such areas. Domestic sheep are sometimes used in medical research, particularly for researching cardiovascular physiology, in areas such as hypertension and heart failure (Recchia and Lionetti, 2007).

Pregnant sheep are also a useful model for human pregnancy (Barry and Anthony, 2008) and have been used to investigate the effects on fetal development of malnutrition and hypoxia (Vuguin, 2007). In behavioral sciences, sheep have been used in isolated cases for the study of facial recognition, as their mental process of recognition is qualitatively similar to humans (Peirce *et al*., 2001). Sheep are one of the few animals where the molecular basis of the diversity of male sexual preferences has been examined (Roselli *et al*., 2004). Lamb intestine has been formed into surgical sutures, as well as strings for musical instruments and tennis racket. Sheep droppings, which are high in cellulose, have even been sterilized and mixed with traditional pulp materials to make paper. Of all sheep byproducts, perhaps the most valuable is lanolin: the waterproof, fatty substance found naturally in sheep's wool and used as a base for innumerable cosmetics and other products. At the smallholder level, sheep are the major source of food security serving a diverse function including cash income, savings, fertilizer, socio-cultural functions. Sheep are particularly important for the pastoralist/agro pastoralist and for farmers in the sub-alpine highlands where crop production is unreliable (Getachew *et al*., 2016).

In Nepal, buffaloes contribute about 64% of meat consumed, followed by goat meat 20%, pork 7%, chicken 6% and sheep 3% (Joshi *et al.*, 2001). Sheep are an important livestock commodity in the country, particularly for the hills and mountains. Traditionally, in Nepal sheep were used as means of transport before the development of vehicles. Still in the mountain regions where transportation facility is not available, sheep are in use to carry goods. Sheep are known to have flocking instinct so single shepherd can control large herd. Sheep and goat husbandry is an important component of mixed subsistence agriculture systems. Resource poor farmers in the hills, who cannot invest large sums of money in cattle and buffalo, prefer goat and sheep keeping. There are 8,00,658 sheep in the country and dang tames 34,091 of it. Jumla comprises the highest number of sheep population and Jhapa has the least number. In the FY 2001/02 the sheep population was 8,40,141 then after by continuously decreasing every year the population has become 8,00,658 in the FY 2015/2016 (SIONA, 2015/16). Five to ten percent of this is exotic (pure or crossbred) and rest is indigenous suggesting more contributions comes from indigenous breeds. There are four indigenous breeds of sheep namely; Bhyanglung (4%), Bharuwal (63%), Kage (21%), Lampuchhre (12%). Bhyanglung are the only indigenous sheep breed that can produce wool suitable for carpet. They have been reared in situ condition by farmers themselves. Their population is declining and needs attention for conservation. Bharuwal sheep are the principal sheep breed in the country and are good for wool production. They are located in hilly region of the country. Their wool is suitable for making radi/pakhi (blankets), pullovers and shawls. They are normal and are not at risk for conservation. They have good grazing instinct and suitable for migratory system. The breed is usually used as pack animal transporting goods from one lower place to higher mountain. Each can carry at least 8 kg load. Kage is the smallest among the indigenous sheep breeds with better prolificacy (18% twinning) and are found in low to mid hills (subtropical) and are mainly used for wool. Lampuchhre sheep are found in terai region (tropical and subtropical climate) of the country and good for meat and fighting specially reared in western Terai belt by nomadic people (Regmi, 2015). Transhumant sheep farming is a traditional practice in the northern high altitudinal regions of Nepal. Among the total sheep population available in the country, 60% of the sheep are reared under the transhumance system and Bharuwal is one of the native sheep breeds of Nepal kept in transhumance as a multi-purpose small ruminant (Joshi *et al*., 2004). Usually the upward migration starts at April and the flocks were allowed to graze on three to five pastures of varying altitudes ranging from 2,000 to 4,500m. The duration of stay at pastures depends upon the weather, pasture condition and availability and the herd size. At high pastures, the grazing competition basically occurs with yaks and their hybrids (Barsila *et al*., 2014). Sheep and goat meat is valued as a source of main animal protein for ethnic group of population. There is also a tradition of providing sheep and goat as gift (Pewa) to their sister and daughter in some community of the country (Khakural *et al*., 2005).

There are multidimensional problems, complex in nature due to which productivity and production of sheep and goats declined over time. The animals suffer from a variety of infectious and non-infectious diseases, particularly that of parasitic origin (Akhter and Arshad, 2006). Parasitism is a problem to farmers that can affect their sheep causing financial loss and welfare concerns. Parasites are classified as ectoparasites and endoparasites on the basis where they live. Ectoparasites are those parasites that live on or in the skin but not within the body. Fleas and lice are ectoparasites. Infestation with an ectoparasite is called an ectoparasitosis. Endoparasites are those organisms living in their hosts, in the gut, body cavity, liver, lungs, gall bladder, blood and within the intestinal, tissues or cell of the host.

Gastrointestinal parasites are those parasites that infect the gastrointestinal tracts of animals. Domesticated animals are susceptible to many diseases like coccidiosis, fasciolopsis, schistosomiasis, nematodiasis, etc. Generally, ruminants are infected by various common intestinal parasites.

1.2 Gastro-intestinal parasites:

Among different parasitic infections, gastrointestinal (GI) parasitism is considered one of the most serious and underestimated problems, which hinders sheep productivity. Despite the remarkable achievement in the discovery of gastrointestinal parasites drugs with ever increasing spectra of activity, nematode parasitic infestation and disease remains one of the greatest limiting factors in the production of sheep worldwide (Perry and Randolph, 1999). Moreover, studies have shown that some of sheep gastro-intestinal parasites are of public health importance and they were incriminated in zoonotic transmission to human either by direct contact with sheep manure or indirectly through ingestion of contaminated food or water (Feng and Xiao, 2011; Ralph *et al.*, 2006). The diseases are usually transmitted by the ingestion of the infective eggs/oocyst or larvae or by its penetration through the skin (Ibrahim *et al*., 2008). The prevalence of gastrointestinal helminthes is related to the agro-climatic conditions like quantity and quality of pasture, temperature, humidity and grazing behavior of the host (Pal and Qayyum, 1993).Changes in season, prevalence and relative burden are the key factors to control the parasitic diseases effectively (Malczewski *et al.*, 1996). Gastrointestinal nematode infections are the main prevalent parasitic diseases affecting small ruminant productivity worldwide, especially in tropics and sub-tropics (Torres-Acosta and Hoste, 2008; Calvete *et al*., 2014). Small ruminants reared under intensive and extensive production systems are extremely susceptible to the effects of wide range of endo-parasites which were reported to equivalent to the combined effects of other ill health problems (Biffa *et al*., 2006; Melaku *et al*., 2013).

Environmental factors and vector abundance have been incriminated in the distribution of most parasitic diseases. Gastrointestinal parasites are common in both temperate and tropical countries, but more prevalent in warm countries where sanitation is poor and standard of living is low (Schmidt *et al*., 2000). The continuous drive to increase meat production for the protein needs of the ever increasing world population is faced with a number of constraints (Boes *et al*., 2000), among which are diseases caused by intestinal helminthes and protozoa (Aliaga-Leyton *et al*., 2011). Stomach and intestinal worms occur in all species of animals. Young and malnourished animals of both sexes and lactating animals are most suitable to these parasites. Stomach worms affect specially camels, goats and sheep. Different types of worms are transmitted when an animal eats grass or drinks water contaminated with larva or eggs. The problem is high in the rainy/wet season (Tefera *et al*., 2009) when infection is as high as 100% in ruminants (Adedepo *et al*., 2005).

Gastrointestinal helminthes are major contributors to reduced productivity and can lower the production of meat, milk and wool (Coop and Holmes, 1996). Also the gastrointestinal nematode infestation resulted in negative influence on both the average daily gain of body and the rate of growth of wool (Niezen *et al*., 1995). Gastrointestinal nematodes are an important problem in sheep and are responsible for heavy loses especially in tropical and subtropical areas (Mendoza *et al*., 1998). Diseases caused by helminth parasites in livestock continue to be a major productivity constraint; sheep and goats under intensive and extensive production systems are extremely susceptible to the effects of wide range of helminth endoparasites (Kenea *et al*., 2015). The direct losses caused by these parasites are attributed to acute illness and death, premature slaughter and rejection of some parts at meat inspection. Indirect losses include the diminution of productive potential such as decreased growth rate, weight loss in young growing animals and late maturity of slaughter stock (Abouzeid *et al*., 2010).

Various risk factors play an important role in the onset of gastro-intestinal nematode infections, due to host and environment. The major factor that influences gastrointestinal parasites infestation in small ruminants, especially sheep is exposure to faecal contamination of grazing land (Pathak, 2011). Environmental factors include agro-ecological conditions, animal husbandry practices such as housing system, deworming intervals and pasture management (Ratanapob *et al*., 2012). These largely determine the type, incidence and severity of various parasitic diseases (Badran *et al*., 2012). Other risk factors such as the host species, sex of the animal, season, age, body condition and breed/genotype (Badaso and Addis, 2015), parasite species and intensity of the worm population, have an effect on the development of gastrointestinal parasitic infections (Tariq *et al*., 2010). Poorly nourished animals maintained on poor quality roughage which are deficient in protein, energy, minerals and vitamins are also susceptible to parasitic infection (Pathak and Pal, 2008). The degree of nematode infection varies according to the host immune response to these worms (Greer, 2008).

Damage to the host occurs when parasites either attach to the lining of the gastrointestinal tract and cause it to become inflamed, or they live in the lumen (open area) of gastro-intestinal tract and have access to ingested feed nutrients before the host can digest them. This can result in impaired ability of the host animal to absorb nutrients, causing poor body condition (thinness), poor growth rates, low milk production, and/or poor hair coat or fleece growth. Some parasites cause a reduction in appetite by the host animal. Parasitic infections especially gastro-intestinal nematode and trematode pose a serious threat and limit the productivity of livestock due to the associated morbidity, mortality, cost of treatment and control measures (Nwosu *et al*., 2007; Raza *et al.,* 2010; Lashari and Tasawar, 2011), decreased production of meat, milk, enhanced susceptibility to diseases, losses resulting from condemnation of carcasses and organs (Rajakaruna and Warnakulasooria, 2011).The prevalence of helminthes of small ruminants results in low productivity due to poor weight gain and poor feed utilization, stunted growth, reduced work capacity, cost of treatment and control of helminthosis (Pedreira *et al*., 2006; Odoie *et al*., 2007; Chaudhary *et al*., 2007).

Protozoa are ubiquitous single-celled organisms. Protozoan parasites that are commonly found in intestinal tract of sheep *Eimeria* species, *Cryptosporidum parvum,* and *Toxoplasma gondii*. Helminth parasite is the main disease that affect sheep and goat productivity in the world (Gadahi *et al*., 2009). Helminthiasis adversely affects ruminants, causing hematological, biochemical disturbances (Ijaz *et al*., 2009), anorexia, weight loss, poor reproductive performance, and even death of lambs (Khajuria and Kapoor, 2003). Commonly occurring helminth parasites are *Fasciola* sp., *Haemonchus* sp., *Ostertagia* sp., *Strongylus* sp., *Chabertia* sp., *Trichostrongylus* sp., *Cooperia* sp. (Abouzeid *et al*., 2010), *Strongyloides* sp., *Trichuris* sp., *Oesophagostomum* sp. (Yaro *et al*., 2015), *Bunostomum phlebotomum, Cotylophora cotylophorum, Dicrocoelium dendriticum, Echinococcus granulosus, Moniezia benedeni, Moniezia expansa, Nematodirus* sp., *Paramphistomum cervi* (Raza *et al*., 2014; Getachew *et al*., 2017). The blood sucking parasite *Haemonchus contortus* which is found in the abomasum of the sheep and goat causes significant blood loss; each worm removes 0.05 ml blood per day so that sheep with 500 *H. contortus* may lose about 250 ml per day (Urquhart *et al*., 1987) resulting in decrease in erythrocytes, lymphocytes, hemoglobin, PCV, body weight and wool growth (Rasool *et al*., 1995). Commonly occurring gastrointestinal parasitic diseases in sheep are Haemonchosis, Ostertagiasis, Strongyloidosis, Oesophagostomiasis, Bunostomiasis and Trichostrongylosis (Singh *et al*., 2013).

1.3 Objectives of the study:

1.3.1 General Objective:

- Prevalence of gastro-intestinal parasites of sheep in Laxmipur VDC Dang.

1.3.2 Specific objectives:

- To study the prevalence of gastrointestinal parasites of sheep.
- To compare the seasonal prevalence of gastro-intestinal parasites of sheep.
- To determine the concurrency of parasites in sheep.

1.4 Significance of the study:

Sheep is the major source of wool and they work as means of transport in hilly and mountain regions but it is believed to be declining the sheep population over generations. Sheep are suffering from different parasitic infection which renders sheep productivity. Very few researches have been carried about gastro-intestinal parasites of sheep in the country and no research has been carried in Dang district. The population of sheep has been declined due to different reasons like lack of manpower to care, mountain shifting, etc. Sheep are rearing individually by farmers in Laxmipur VDC. The current study is aimed to identify and investigate the prevalence and seasonal fluctuation of various parasites of sheep in Laxmipur VDC. This is my first research on gastro-intestinal parasites of sheep in this area. The finding of this study is essential for the development of a better understanding of the gastrointestinal parasites and seasonal prevalence of sheep and for the prevention and control of the spread of infectious parasitic diseases among domesticated animals. Thus, a study regarding parasite prevalence seems inevitable and launched for health related diseases in Laxmipur VDC Dang. The data generated would be helpful to formulate effective control strategies against gastro-intestinal parasites in sheep population of this region and development of strategic deworming program.

2. LITERATURE REVIEW

Parasitism is of a major importance in many agro-ecological zones and still a serious threat to the livestock economy worldwide. The gastro-intestinal parasites cause more than 20 percent loss in the production and productivity of animal (Tripathi and Subedi, 2015). Sheep are known to suffer from various endoparasites of which helminth infection are of great importance (Diaz *et al*., 2007). Gastrointestinal parasitism is associated with economic losses, lowered productivity, reduced animal performance (Badran *et al.*, 2012), mortality and morbidity (Negasi *et al.*, 2012). In general, severe gastro-intestinal nematode pathogenesis has been attributed to the migration of the infective larvae after ingestion rather than the adult worms in the gut.

Globally the most common nematode species known to affect small ruminants are *Haemonchus contortus*, *Trichostrongylus colubriformis*, *Teladorsagia circumcincta* and some species such as *Nematodirus* sp. (Bishop and Morris, 2007). Helminth parasites *Fasciola* sp. and *Paramphistomum* sp. has been recorded (Pathak, 2011; Neupane, 2012; Bashir, 2009). *Paramphistomum* sp. is one of the common parasite of sheep and goats, light infection doesn't cause serious damage to the animals but when present in massive number it can migrate through intestinal tract causing acute parasitic gastroenteritits with high morbidity and mortality rates particularly in young animals (Melaku and Addis, 2012). Infection with *Paramphistomum* sp. or *Amphistomum* sp. as single infection or in combination with liverfluke has been reported from various ruminants of Vietnam, France, Gambia, Turkey, etc (Holland *et al*., 2000; Szmidt-Adjide *et al*., 2000; Fritsche *et al*., 1993; Ozdal *et al*., 2010) while infection with *Dicrocoelium* sp. in ruminants was found less than *Fasciola* sp. and *Paramphistomum* sp. These parasites have been reported from Switzerland, Spain, etc (Schweizer *et al*., 2003; Diaz *et al*., 2007). *Moniezia* sp. and *Taenia* in sheep and goats has been reported from India (Bandyopadhyay *et al*., 2010), Iran (Gharekhani *et al*., 2015). *Moniezia* sp. and *Avitellina* sp. double cestode infection in goats and sheep has been reported in India (Pathak and Pal, 2008). *Strongyle*-type worms were the ascendant parasite amongst all three seasons with a high prevalence rate in rainy season (Bansal *et al*., 2015). *Haemonchus contortus* is one of the most fecund strongylid nematodes; individual females are capable of producing thousands of eggs per day, which can lead to rapid larval pasture contamination and associated outbreaks of haemonchosis. *Nematodirus battus* is of particular pathogenic significance in some temperate areas. Trichostrongyle nematodes are considered among the most pathogenic and economically important parasites of small ruminants (Jurasek *et al*., 2010). Some trichostrongyle nematodes cause anaemia due to their ability to remove RBCs as well as proteins, which can lead to ill-thrift in animals (Zvinorova *et al*., 2016).

Trichostrongylus sp. occur in the small intestine and mainly exert their pathogenic effects in lambs and weaners, but have also been reported to cause significant depression of wool growth in older sheep (Roeber *et al*., 2013). *Cooperia curticei*, *Nematodirus spathiger*, *N. fillicollis*, *N. abnormalis* and *Oesophagostomum venulosum* are common parasites of the small and/or large intestine, whilst *Chabertia ovina* and the hookworm, *Bunostomum trigonocephalum* are less common (Zajac, 2006).

2.1 In global context:

Many researches have been done regarding the sheep world widely. In the survey conducted in Magadi division, South-Western Kenya, 80% prevalence of gastro-intestinal parasites of sheep had been observed by using McMaster method in the faecal samples of 20 sheep and the overall prevalence of coccidial oocysts in sheep was 44% (Maichomo *et al*., 2004). 39.34% prevalence of gastrointestinal parasites in sheep has been reported from Southern Taluks of Central dry zone of Karnataka during drought period where *Strongyles* were the most common nematode. *Strongyle* infection in livestock was found higher during southwest monsoon (Muraleedharan, 2005). 100% prevalence had been observed in a coprological survey done in Galicia (NW Spain) in 1710 faecal samples of sheep and the genera identified were *Chabertia* sp, *Cooperia* sp, *Haemonchus* sp, *Nematodirus* sp, *Oesophagostomum* sp, *Teladorsagia* sp, *Trichostrongylus* sp and *Trichuris* sp (Pedreira *et al*., 2006). Out of 245 faecal sampes examination, 83.24% gastro-intestinal parasite infection had been observed in sheep from R.S. Pura, Bishnah and Samba tehsils of Jammu district, India where *Strongyles* (44.62%) were predominant followed by *Amphistomes* (8.07%), *Eimeria* sp. (6.73%), *Fasciola* sp. (3.08%), *Trichuris* sp. (3.08%), *Dicrocoelium* sp. (1.92%), *Strongyloides* sp. (1.15%) and *Moniezia* sp. (0.96%). Mixed infection with one or more gastro-intestinal ova was also detected in 13.46% of animals and seasonal variation was highest during rainy season (88.54%) followed by summer (83.15%) and winter (76.01%) (Yadav *et al*., 2006).

In an irrigated area of lower Punjab, Pakistan, overall 62%prevalence of helminthiasis was recorded in sheep with nematodes being the most common helminthes (Raza *et al*., 2007). Overall thirteen species belonging to nine genera of gastro-intestinal nematodes and four species belonging to three genera of trematodes were recorded from 655 sheep in the semi-arid zone of eastern Ethiopia with highest burdens around the middle of the rainy season during May 2003 to April 2005 (Sissay *et al*., 2007). Out of 102 sheep examination by the modified McMaster technique in the semi-arid zone of North-Eastern Nigeria, 43.1% sheep had been observed to harbor gastro-intestinal nematodes in them where three nematode egg types were recovered with *Strongyle* egg type (22.5%) being the most prevalent followed respectively by *Trichuris* (5.9%) and *Strongyloides* (4.9%) egg types. Counts of *Strongyle* egg type increased with the rains and reached peak levels at about the peak of the rainy season in September while other egg types did not show much variation with the season of the year (Nwosu *et al*., 2007). Out of 86 fecal samples of sheep examined in and around Rawalpindi and Islamabad, Pakistan, 72% were found positive for gastrointestinal parasites and *Haemonchus* sp. (80.64%) was in highest prevalence (Asif *et al.*, 2008). From the study among 79 sheep in Tulus Area, Sudan, seven nematode species (99.8%) and *Moniezia expansa* (0.2%) were identified where *Haemonchus contortus* was highest prevalent nematode (53.4%) followed by *Strongyloides papillosus* (26.2%), *Trichostrongylus colubriformis* (14.7%), *Cooperia pectinata* (3.1%), *Oesophagostomum columbianum* (2.2%), *Skrjabinema ovis* (0.3%) and *Trichuris globulosa* (0.1%). The total worm burden was least during dry season and increased gradually during the rainy season (Almalaik *et al*., 2008). Out of 1533 sheep examined in Kashmir valley, 61.64% were found infected with GIT nematodes where the prevalence rate were *Haemonchus contortus* (59.6%), *Ostertagia circumcincta* (38.0%), *Bunostomum trigonocephalum* (37.7%), *Chabertia ovina* (37.7%), *Trichostrongylus* sp. (33.9%), *Nematodirus spathiger* (29.4%), *Oesophagostomum columbianum* (28.4%), *Trichuris ovis* (23.5%) and *Marshallagia marshalli* (22.1%). The maximum nematode infection was observed in summer season and lowest in winter (Tariq *et al*., 2008). Coprological examination in Patiala and its adjoining areas has revealed total prevalence of gastrointestinal parasites to be 87.05% in sheep where the different GI parasites detected were *Trichostrongylus* sp. (78.57%), *Haemonchus* sp. (64.29%), *Strongyloides* sp. (57.14%), C*ryptosporidium* sp. (57.14%), *Oesophagostomum* sp. (42.86%), *Moniezia* sp. (21.43%) and *Dicrocoelium dendriticum* (14.29%) and mixed helminth and protozoan infection was 85.71% (Kaur and Kaur, 2008). In Rawalpindi and Islamabad of Pakistan, 48 samples (53.33%) were detected positive in sheep for gastrointestinal parasites from 90 faecal samples examination in which *Trichuris* sp. was found in highest intensity of infection (40%) and different species like *Haemonchus* sp., *Coccidia* sp., *Nematodirus* sp., and *Fasciola* sp., were found with prevalence of 28.88%, 27.77%, 11.11% and 4.44% respectively (Gadahi *et al*., 2009).

Out of 219 faecal samples in and around Bedelle, South-Western Ethiopia 91.32% sheep were found to harbor eggs of GIT helminthes where eight genera of nematodes were found with maximum prevalence rate of 67.5% by *Haemonchus* sp. followed by *Trichuris* sp. (46.1%), *Trichostrongylus* sp. (48.8%), *Oesophagostomum* sp. (48.8%), *Bunostomum* sp. (30.3%), *Ostertagia* sp. (25.6%), *Chabertia* sp. (20.9%) and *Strongyloid* sp. (16.3%) in sheep (Tefera *et al*., 2009). From the district Toba Tek Singh, Punjab, Pakistan, 44.17% sheep had been reported to harbor gastro-intestinal parasites from the faecal samples of 840 sheep (Khan *et al*., 2010). Eighteen sheep (10.4%) were infected with eight different species of nematodes in 173 slaughtered sheep in Nile Delta, Egypt where the prevalence rates of detected nematode parasites were; *Haemonchus contortus* (3.5%), *Haemonchus placei* (1.7%), *Trichuris ovis* (5.8%), *Parabronema skrjabini* (2.9%), *Ostertagia trifurcate* (1.2%), *Chabertia ovina* (0.6%), *Strongyloides papillosus* (0.6%) and *Graphidiops* species (2.9%). The seasonal prevalence of the infection with the nematode parasites was highest during autumn (15.2%) followed by summer (11.1%) and winter (9.4%) while the lowest rate was during spring (5.6%) (Khalafalla *et al*., 2011). Out of 523 faecal samples of sheep examined in Southern Punjab, Pakistan, overall 240(46.33%) were found to contain various gastro-intestinalparasites where the prevalence of *Fasciolia hepatica, Avitellina centripunctata, Haemonchus contortus* and *Trichuris globulosa* was 21.41%, 12.23%, 6.50% and 5.73%, respectively (Lashari and Tasawar, 2011).

Out of 550 faecal samples examined in Sulamani province, the prevalence of gastro-intestinal parasites was found 34.36% where 5% showed multiple infection and the parasites observed includes*; Nemarodirus, Strongylus, Marshalagia, Trichuris* and Coccidia with a percentage of 29.8%, 19.8%, 9.9%, 6% and 21% respectively (Nassrullah, 2011). In Thessaly region of Greece, helminth eggs were detected in 44 (7.9%) samples out of 557 faecal samples where the prevalence rates of detected parasites were *Strongyle*-type eggs (3.4%), *Nematodirus* sp. (1.1%), *Trichuris* sp. (2.9%), *Fasciola hepatica* (0.5%), *Dicrocoelium dendriticum* (0.2%) and Coccidian oocysts (6.5%) (Kantzoura *et al*., 2012). A total of 190 sheep were examined at Tangail district, Bangladesh of which 154 (81.1%) were positive for one or more species of helminth parasites where seven species of helminthes were identified, of them three species were trematodes, namely, *Fasciola gigantica* (8.4%), *Paramphistomum* sp. (44.2%) and *Schistosoma indicum* (3.7%); four species were nematodes, namely, *Bunostomum* sp. (19.0%), *Trichuris* sp. (2.1%), *Strongyles* (62.6%) and *Strongyloides* sp. (9.5%) (Sangma *et al*., 2012). Out of 384 faecal samples 259 (67.75%) sheep harbor one or more genera of helminthes with nematodes (59.89%) being the most prevalent helminthes in and around Haramaya, Southeastern Ethiopia. The study revealed significantly higher ($p<0.05$) prevalence of helminthes in wet season than drier ones. (Zeryehun, 2012). Among 500 fecal samples of sheep examined in Kashmir valley of India by direct microscopic examination, floatation and sedimentation techniques, overall prevalence rate was 62.9% where most commonly encountered parasites were *Strongyle* sp., *Strongyloides* sp., *Eimeria* sp., *Nematodirus* sp. and *Monezia* sp. with the prevalence rate of 24.61%, 15.5%, 9.8%, 9.0% and 3.3%, respectively. The highest prevalence of GI parasites was recorded during monsoon season (March-May) followed by summer season (June-August) whereas the lowest prevalence was recorded during winter season (Bhat *et al*., 2012). Out of the 408 samples examination, 278 (68.1%) had been observed to be infected with gastrointestinal nematode from five different sites in and around Asella, South Eastern Ethiopia where *Strongyles* were the most frequently (64.0%) recovered nematode eggs followed by *Strongyloides* (7.4%) and *Trichuris* species (3.7%). The higher infection prevalence was recorded in months during wet seasons whereas the lower was in months during dry seasons (Lemma and Abera, 2013).

Out of 83 faecal samples of sheep examined by direct wet smear in southern Rajasthan, India, 55.42 % sheep were found to be infected with different species of gastrointestinal parasites. A total of 8 species of gastrointestinal parasites were encountered, among these parasites *Fasciola hepatica* was the commonest (15.18 %) followed by *Haemonchus contortus* (11.32 %), *Ancylostoma duodenale* (10.36 %), *Trichuris trichiura* (9.15 %), *Amphistome* species (7.95 %), *Moniezia expansa* (6.98 %), *Strongyloides stercoralis* (4.57 %) and *Balantidium coli* (3.37 %). The highest prevalence rate was found in rainy season (84.21 %) followed by winter (73.9 %) and summer (52.8 %) (Choubisa and Jaroli, 2013). A total of 257 sheep were examined in Jammu area of Jammu and Kashmir State in which 115 (44.74%) were found positive for various types of helminthic infections and the identified nematodes are *Trichuris ovis*, *Haemonchus contortus*, *Dictyocaulus filarae* and *Chabertia ovina*. Seasonal prevalence of infections indicated that the nematode infection was highest in spring (57.14%), and summer months (56.94%) followed by winter (37.09%) and autumn (28.35%) (Mir *et al*., 2013). In Papua New Guinea, out of 110 faecal samples examined, 72% of sheep were found to be infected with one or more species of gastrointestinal parasites by using modified McMaster technique where the identified parasites and their prevalence were *Strongyle* (67.3%), *Eimeria* (17.3%), *Strongyloides* (8.2%), *Fasciola* (5.5%), *Trichuris* (1.8%) and *Nematodirus* (1.8%) (Koinari *et al*., 2013). Out of 960 faecal samples examined, 68.54% sheep were positive for helminthic infections in the middle agro-climatic zone of Jammu where *Strongyles* (50.1%), *Trichuris* (12.1%), *Strongyloides* sp. (4.2%), *Amphistomes* (8.3 %), *Fasciola* sp. (8.2 %) and *Dicrocoelium* sp. (5.4 %) had been observed and significantly ($p < 0.05$) higher infection was observed in monsoon as compared to winter by *Strongyles.* Coproculture studies revealed that *Haemonchus contortus* (61.18%) predominated during all the seasons, followed by *Trichostrongylus* sp. (13.67%), *Ostertagia* sp. (12.17%), *Strongyloides* sp. (4.14%), *Oesophagostomum* sp. (3.84%) and *Bunostomum* sp. (3.83%) (Khajuria *et al.,* 2013). Among 50 fecal samples of sheep examined in and around Mathura, India 34 samples were found to be positive with prevalence rate 68% where 16(47%) harboured bursate worms (*Haemonchus*), 9(26.47%) harboured coccidian worms and 9(26.47%) had mixed infections (Singh *et al.*, 2013). In and around Bako town, Western Ethiopia,77.4% of the sheep were found to harbor eggs of gastro-intestinal parasites from a total of 424 faecal samples where 65.3% were infected with *Strongyle* sp., 6.1% with *Strongyloides* sp., 7.3% with *Trichuris* sp., 30.9% with E*meria* sp., 7.3% with *Monezia* sp., 9% with *Fasciola* sp. and 3.1% with *Paramphistomum* sp. (Gizachew *et al*., 2014). A total of 315 sheep were examined from Dembia district, Northwest Ethiopia using standard parasitological procedure where 132 (41.49%) sheep were detected positive for gastrointestinal nematode parasites (Muluneh *et al.,* 2014). Out of 214 sheep examined coprologically in and around Jimma town, Western Ethiopia, 191(89.3%) sheep were found to harbor one or more gastrointestinal parasites where the prevalence of various types of parasites in sheep were *Fasciola* sp. 19.6%, *Paramphistomum* sp. 22.4%, *Haemonchus* sp. 37.4%, *Trichostrongylus* sp. 26.2%, *Strongyloid* 20.1%, *Ostertagia* sp. 16.8%, *Oesophagastamum* sp. 9.3%, *Trichuris* sp. 7.9%, *Chabertia* sp. 4.2%, *Bonustomum* sp. 2.3%, *Monezia* 13.1% and *Emeria* sp. 11.7% (Ibrahim *et al*., 2014).

Out of 135 faecal samples five species of gastrointestinal parasites eggs were discovered from Maiduguri, Borno state, West Africa which are *Strongyle, Strongloides*, Coccidian occyst, *Moniezia*, and *Trichuris* by using the Modified Mcmaster technique with significant differences ($P<0.05$) between the mean gastrointestinal parasites egg counts among sheep in different seasons of the year where the raining season had higher overall mean parasites egg counts and prevalence than the dry season (Alade and Bwala, 2015). Out of 445 faecal samples examination, overall prevalence of gastrointestinal helminthes 90.11 % had been observed in sheep from Mhow, Indore where the highest prevalence was of *Strongyles* (85.40 %) followed by *Amphistomes* (21.78 %), *Trichuris* sp. (21.70%), *Strongyloides* sp. (12.24 %), *Moniezia* sp. (5.77 %) and *Fasciola* sp. (4.56 %) and in monsoon season maximum prevalence (92.96 %) was recorded followed by winter (89.20 %) and summer (87.76 %) (Bansal *et al*., 2015). Of the total 299 faecal sample examined in 2012 and 127 examined in 2013, 177(59.2%) were positive for gastrointestinal parasites in 2012, whereas, 105 (82.7%) were positive for gastrointestinal parasites in 2013 in Minna, North Central, Nigeria. Out of the 53 positive samples in sheep in 2012, coccidian sp. had the highest prevalence of 47.1%, followed by *Fasciola* sp., *Haemonchu*s sp., *Ascaris* sp., *Strongyle* sp., *Bunostomum* sp., *Oesophagostomum* sp., *Schistosoma* sp. and *Taenia* sp. with 11.3%, 15%, 9.4% 5.6% 1.8%, 3.7%, 1.8% and 3.7% respectively in 2012 whereas in 2013 *Coccidia* sp. had the highest prevalence of 58.2% , followed by *Haemonchus* sp., *Monieza expansa* and *Strongyle* sp. with 23.5%, 11.7% and 5.8% prevalence respectively (Ibukun and Oludunsin, 2015). From 492 examined faecal sample of sheep in Kaffa and Bench Maji zones, Southwest Ethiopia, 17.7% sheep had mixed infection and identified genera are *Bunostomum* (18.69%), *Chabertia* (3.54%), *Cooperia* (17.68%), *Haemonchus* (18.69%), *Oesophagostomum* (4.55%), *Teladorsagia* (6.57%), *Trichostrongylus* (30.30%) (Kenea *et al*., 2015). Out of 600 faecal samples 66.33% sheep were positive for gastrointestinal parasites in TamilNadu where *Haemonchus* sp. was found to be predominant. The results of season wise analysis indicates that the overall infection percentage was higher in rainy season (68.36%) followed by winter (60.84%) than in summer (55.30%) (Varadharajan and Vijayalakshmi, 2015). From the total 229 different sheep examined, the overall prevalence of one or more gastro-intestinal parasites was 86.6% and predominant worm species were *Strongyle* (71.9%), *Trichuris* (1.5%) and 26% of the positive specimens were harboring multiple worm species (Getachew *et al*., 2016). Out of 160 faecal samples examined, 96(60%) of sheep were found infected with one or more species of helminth parasites in different localities at Bardarash district, Duhok, Iraqi Kurdistan where 11 helminth species were diagnosed and the recovered species were identified as *Marshallagia marshalli, Nematodirus* sp., *Haemonchus contortus, Fasciola hepatica, Moniezia benedeni , Trichuris ovis, Moniezia expanza, Chabertia ovina, Bunostomum trigonocephalum, Teladorsagia circuntcincta* and *Cooperia* sp. (Mohammedameen, 2016). Out of 224 fecal samples of sheep examined from Nile-Delta, Egypt 50% individuals had been infected by gastrointestinal parasites of which 29.02% were protozoan parasites and 37.05% were helminth parasites and the parasites identified include *Strongyle*-group (19.21%) *Paramphistomes* (9.38%), *Trichuris* sp. (2.68%), *Moniezia* sp. (0.89%), *Nematodirus* sp. (0.45%), *Eimeria* sp. (16.52%) *Strongyloides* papillosus (4.02%), *Entamoeba* sp. (10.27%), *Giardia duodenalis (0.45%)* and *Balantidium coli* cyst (1.79%) (Sultan *et al.,* 2016). In Sherpur, Bangaladesh out of 106 samples, 67.9% (72/106) were positive for ova of different helminthes associated with *Fasciola gigantica* (11.3%), *Paramphistomes* (13.2%), *Schistosoma indicum* (3.8%), *Moniezia* sp. (3.8%), Strongyle-type (24.5%), hook worm (6.6%), *Strongyloides* sp. (12.3%) and *Trichuris* sp. (1.9%) (Poddar *et al*., 2017). Out of the 450 samples of sheep of different breeds examined in Lafia, North Central Nigeria, 320(71.11%) were positive for gastrointestinal parasites and the parasites observed were *Eimeria* sp., *Haemonchus* sp., *Strongyle* sp., *Oesophagostomum* sp. and *Trichuris* sp. (Muhammed *et al*., 2017).

2.2 In context of Nepal:

Very few researches have been done regarding gastro-intestinal parasites of sheep in the case of our country, Nepal. In Karnali, Nepal, 77 samples (87.5%) were detected positive for gastro-intestinal parasites from 88 faecal samples of sheep (Khakural *et al*., 2005).

Out of 83 fecal samples of sheep examined both qualitatively and quantitatively by the sedimentation and floatation methods, 78.31% samples were positive for parasites in Ghanpokhara, Lamjung at an altitude of 2200 meters asl where *Strongyle* (78%) was predominant folllowed by *Trichostrongyl*e (44%), *Moniezia* (12%), *Trichuris* (8%) and *Coccidia* (3%) (Acharya, 2017).

3. MATERIALS AND METHODS

3.1 Study Area:

Dang district is located in the Mid-Western Region of Nepal, which occupies the area of 2,955 km^2 with a population of 548,141 (CBS, 2012). Dang valley is the largest valley of Asia; this district is also known as inner terai. It lies on the geographical coordinates of latitude 27^0 37′ to 28^0 39′ North and longitude 82^0 2′ to 82^0 54′ East. The maximum and minimum temperatures of this district are around 34^0C and 14^0C respectively and rainfall averages more than 1300mm annually. It is located at 410 km west of the capital city Kathmandu. Ghorahi (formerly Tribhuvannagar) is the district's administrative center and largest city. The major cities of Dang district are Ghorahi, Thulsipur, and Lamahi. (https://en.wikipedia.org/wiki/Dang_Deukhuri_District, 2017).

North of Deukhuri valley, the Dang range rises as high as 1,000 meters with pass at about 700 meters then upslope to the crest of Mahabharats at 1,500 to 1,700 meters elevation. It has two mountain range Chure and Mahabharat Mountain range. The climate is Tropical and Subtropical. Through historic times Dang and Deukhuri valleys were home to indigenous Tharu people (https://en.wikipedia.org/wiki/Dang_Deukhuri_District, 2017).

Dang district has majority of people from Tharu ethnics, Bahuns and Chettri had come from the southern mountains. Kumal (potter) ethnic group is also semi-indigenous. Since the early 1990s activist groups have been attempting to eradicate the practice of indentured servitude among Tharu, many of whom sold young daughters to wealthy families in urban areas (https://en.wikipedia.org/wiki/Dang_Deukhuri_District, 2017).

Present study was carried out in sheep of Laxmipur VDC, Dang. Laxmipur VDC lies approximately 8 km south-east from Ghorahi, the administeative center of Dang. The area is characterized by Tropical climate. According to administrative and political division, this VDC is divided into nine wards. It is good place for the animal husbandry like fish, chicken, goat etc. The main crops produced are paddy, wheat, maze etc., cash crops are sugarcane, mustard. Sheep farming is carried out individually by Chaudhary (Tharu) groups in this area. There is no sheep farming carried out by other ethnic groups or any institutions. Almost every household rear sheep. They rear sheep for meat purpose. Grass, fodder, hay and agricultural byproducts includes the sheep diet. Sheep get parasitic infection when they are grazing in contaminated open grass fiels.

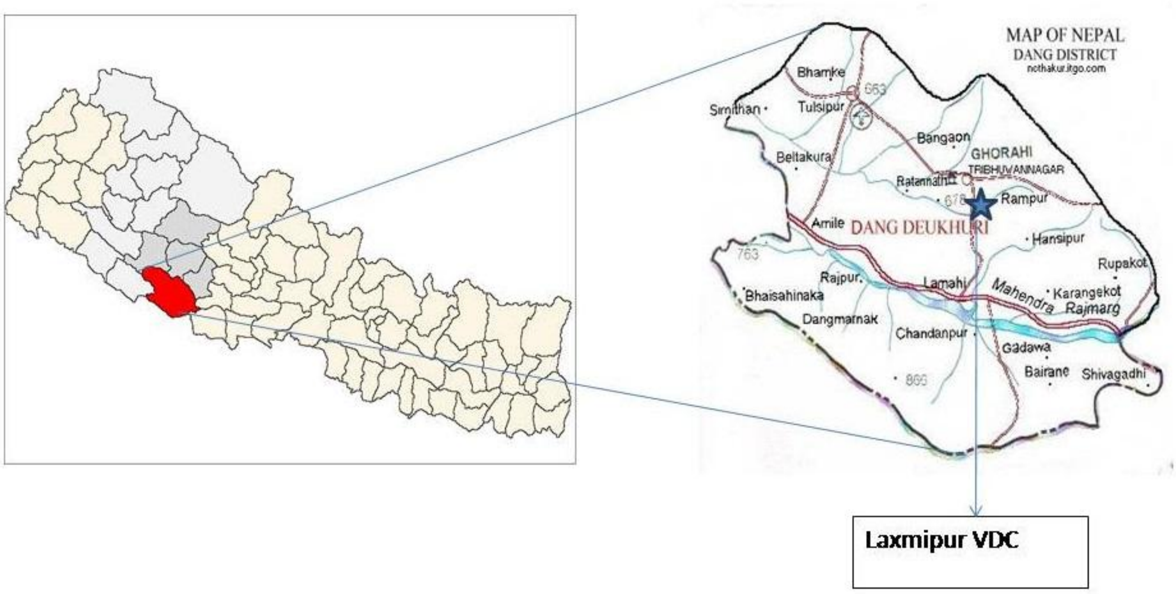

Figure 1: Location of study area.(Source: https://www.google.com.np)

3.2 Materials used:

3.2.1 Materials for field:

I. Sterile vials
II. Gloves

3.2.2 Materials for laboratory:

I. Electric microscope
II. Ocular micrometer
III. Stage micrometer
IV. Centrifuge machine
V. Centrifuge tubes
VI. Volumetric flask
VII. Test-tube rack
VIII. Cavity blog
IX. Tea strainer
X. Beakers
XI. Glass rod
XII. Slides
XIII. Cover-slips
XIV. Dropper
XV. Gloves and masks
XVI.Tooth picks
XVII. Watch glass
XVIII. Spatula

3.2.3 Chemicals:

I. Potassium dichromate (2.5%)
II. Normal saline
III. Methylene blue
IV. Distilled water
V. Saturated Nacl solution
VI. Iodine solution.

3.3 Study Design:

The study was designed to assess the gastro-intestinal parasitic infection in sheep of Laxmipur VDC. The study design includes:

Selection of study area (Laxmipur VDC)

↓

Collection of 10 gram fresh fecal samples in separate sterile vials of each sheep

↓

Preservation of fecal samples in Potassium dichromate ($K_2Cr_2O_7$(2.5%))

↓

Microscopic examination of fecal samples using saline wet mount and concentration techniques

↓

Identification and measurement of eggs, cysts and larva of parasites.

↓

Analysis

Figure 2: Flow chart of research outline

3.3.1 Sample size:

A total of 110 sheep were randomly selected from the study area by visiting to farmer's bawn. One sheep was sampled from one bawn. Samples were collected in two divided time periods. 55 faecal samples were collected during the month of April 2017 and rest 55 during the month of August 2017.

3.3.2 Collection of sample:

Faecal samples were collected at early in the morning between 5 to 6:30 am. Fresh fecal samples were collected immediately after voiding. About 10 gm of faeces were collected from each sheep by hand picking method with the help of disposable gloves and transferred inside the sterile plastic vials applying appropriate precautions. The same process was repeated for all collected faecal samples. Necessary information were recorded clearly such as faecal samples collection date, number.

3.3.3 Preservation of sample:

Collected faecal samples of sheep were preserved in 2.5% potassium dichromate that helps in maintaining the morphology of protozoan and helminth parasites.

3.3.4 Laboratory examination:

The faecal samples were brought to laboratory of Central Department of Zoology, Tribhuvan University, Kirtipur, Kathmandu, Nepal. The faecal samples were examined by using saline wet mount and concentration (differential floatation and sedimentation) techniques.

3.3.4.1 Saline wet mount:

Approximately 2 gram of faecal sample was taken in a watch glass and added 5 ml of saturated sodium chloride solution then the sample was grinded lightly with the help of spatula. One drop of Iodine solution was placed on cover slip then the crushed faecal sample was placed on that iodine with the help of tooth pick and stirred it well then cover slip was placed on it (Soulsby, 1982).

3.3.4.2 Differential floatation technique:

This technique is used widely for detecting eggs of nematodes and cestodes. As their eggs are lighter and small, they can float in the floatation liquid.

Approximately 3 gram of faecal sample was taken in a beaker and added 15 ml of saturated sodium chloride solution then the sample was grinded lightly with the help of spatula and filtered the solution by the tea strainer. The filtrate solution was poured into a centrifuge tube of 15 ml and tube was filled with more sodium chloride solution and centrifuged at 1,000 rpm for 5 minutes.

After centrifuge more saturated sodium chloride was added to develop convex surface at the top of the tube and one drop of methylene blue (to stain) was added and a cover slip was placed for 5 minutes and then cover slip was removed and placed on a slide and examined at 10X × 10X and 10X × 40X objectives. The photographs of eggs, cysts and larva of parasites were taken and identified based on colour, shape and size (morphometry) (Soulsby, 1982).

3.3.4.3 Sedimentation technique:

This technique is used for detecting trematode eggs. It provides good results as the eggs of trematodes are a bit heavier than the other, where sediments of centrifuged contents is taken for eggs detection.

Saturated salt solution was removed gently from the test tube after examining the floatation portion and poured the sediment content into watch glass and stirred the content gently to mix it. One drop from the mixture was taken to prepare a second slide. The specimen was stained with iodine wet mounts solution.

In this way, two slides were prepared from one sample (one from floatation and one from sedimentation) were examined under 10X × 10X and 10X × 40X objectives of microscopes to detect eggs of protozoan, helminthes or cysts of gastro-intestinal parasites (Soulsby, 1982).

3.3.5 Measurement:

Eggs and cyst were measured by using ocular and stage micrometer.

3.3.6 Eggs, cysts and larva identification:

Identification was done on the basis of shape and size of published literature journals and books (Soulsby, 1982), (Indre *et al*., 2010), (Sangma *et al*., 2012) and (Poddar *et al*., 2017) and other published and unpublished articles and internet sources. The differentiation of all species of nematode eggs was done only based on morphological characteristics of eggs. Differentiation based on larvae culture could not be done due to technical problem.

3.3.7 Data analysis:

For this study, prevalence was measured as the percentage of host individuals infected with a particular parasite. The data were coded and entered into Microsoft Excel spread sheet. Data were statistically analyzed by using "R", version 3.3.1 sofware packages. In all cases 95% confidence interval (CI) and $p<0.05$ was considered for statistically significant difference.

Photographs during lab work and field work:

	
Photo 1: Examining under microscope	Photo 2: Running centrifuge machine
	
Photo 3: Faeces of sheep	Photo 4: Sheep in byre

4. RESULTS

4.1 Overall prevalence of GI parasites in sheep:

Out of 110 samples examined, 88 (80%) samples were found to be positive for parasitic egg, cyst and larvae.

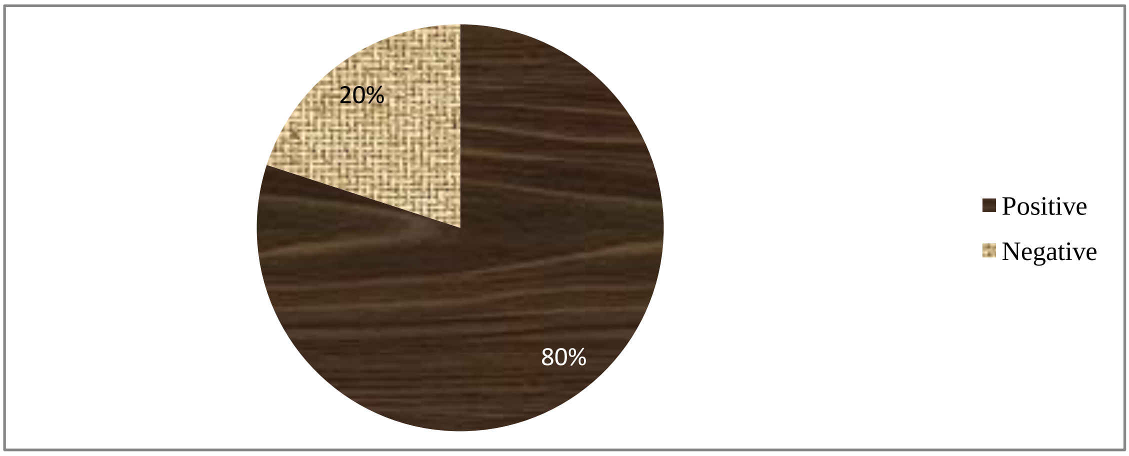

Figure 3: Overall prevalence of gastro-intestinal parasites.

4.2 Prevalence of GI parasites in sheep:

Out of 110 faecal samples examined, 15 genera of parasites were identified and the identified genera are *Eimeria* sp. 4(3.64%), *Entamoeba* sp. 3(2.73%), *Haemonchus* sp. 37 (33.64%), *Bunostomum* sp. 11 (10%), *Oesophagostomum* sp. 15 (13.64%), *Strongyle* sp. 28 (25.45%), *Strongyloides* sp. 34 (30.91%), *Trichostrongylus* sp. 16 (14.55%), *Nematodirus* sp. 6 (5.45%), *Trichuris* sp. 9 (8.18%), *Ascaris* sp. 2 (1.82%), *Chabertia* sp. 5 (4.54%), *Fasciola* sp. 12 (10.91%), *Paramphistomum* sp. 8 (7.27%) and *Moniezia* sp. 7(6.36%).

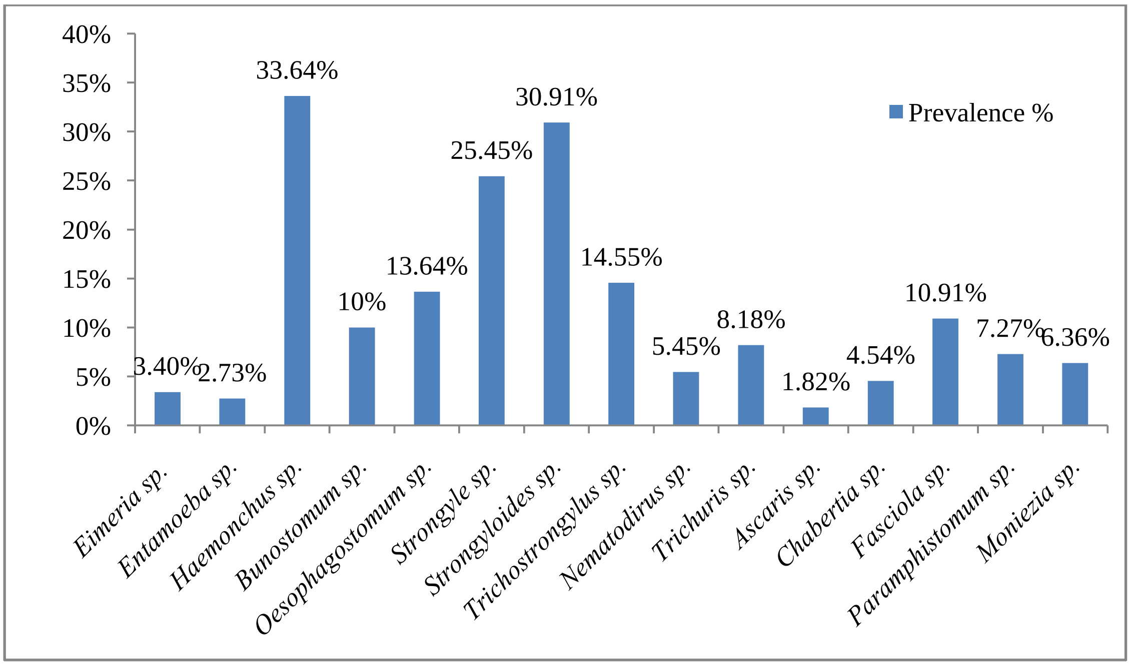

Figure 4: Prevalence of gastro-intestinal parasites in sheep.

4.2.1 Protozoan parasites:

Overall, sheep were found to harbor protozoan parasites belonging to two classes. 3.64% were found to be positive for *Eimeria* sp. and 2.73% were found to be positive for *Entamoeba* sp. There was not significant difference in the distribution of *Eimeria* sp. and *Entamoeba* sp. (χ^2=1.127, df=1, p-value=1).

Table 1: Protozoan parasites in sheep:

S.N.	Class	Parasite Name	No. of positive	Prevalence (%)
1.	Sporozoa	*Eimeria* sp.	4	3.64%
2.	Sarcodina	*Entamoeba* sp.	3	2.73%

4.2.2 Helminth parasites:

Sheep were found to be infected with helminth parasites belonging to three classes. Among helminth parasites, maximum no. of sheep were found to be infected from nematode parasites belonging to ten genera compared to cestodes and trematodes belonging to two and one genera respectively. *Haemonchus* sp. of helminth parasites appeared to be predominant parasite species having the prevalence rate of 33.64% and *Ascaris* sp. showed the lowest prevalence with 1.82% of sheep of Laxmipur VDC, Dang.

Table 2: Overall helminth parasites in sheep:

S.N.	Class	Parasite Name	No. of positive	Prevalence (%)
1.	Nematoda	*Haemonchus* sp.	37	33.64%
2.		*Bunostomum* sp.	11	10%
3.		*Oesophagostomum* sp.	15	13.64%
4.		*Strongyle* sp.	28	25.45%
5.		*Strongyloides* sp.	34	30.91%
6.		*Trichostrongylus* sp.	16	14.55%
7.		*Nematodirus* sp.	6	5.45%
8.		*Trichuris* sp.	9	8.18%
9.		*Ascaris* sp.	2	1.82%
10.		*Chabertia* sp.	5	4.54%
11.	Trematoda	*Fasciola* sp.	12	10.91%
12.		*Paramphistomum* sp.	8	7.27%
13.	Cestoda	*Moniezia* sp.	7	6.36%

4.2.3 Trematode parasites:

Out of 110 samples, prevalence of trematode parasites was observed in 17 (15.45%) samples. Among them, *Fasciola* sp. was observed in 12 (10.91%) samples and *Paramphistomum* sp. was observed in 8 (7.27%) samples. Distribution of *Fasciola* sp. and *Paramphistomum* sp. was not found significant (χ^2=0.495, df=1, p-value=0.482).

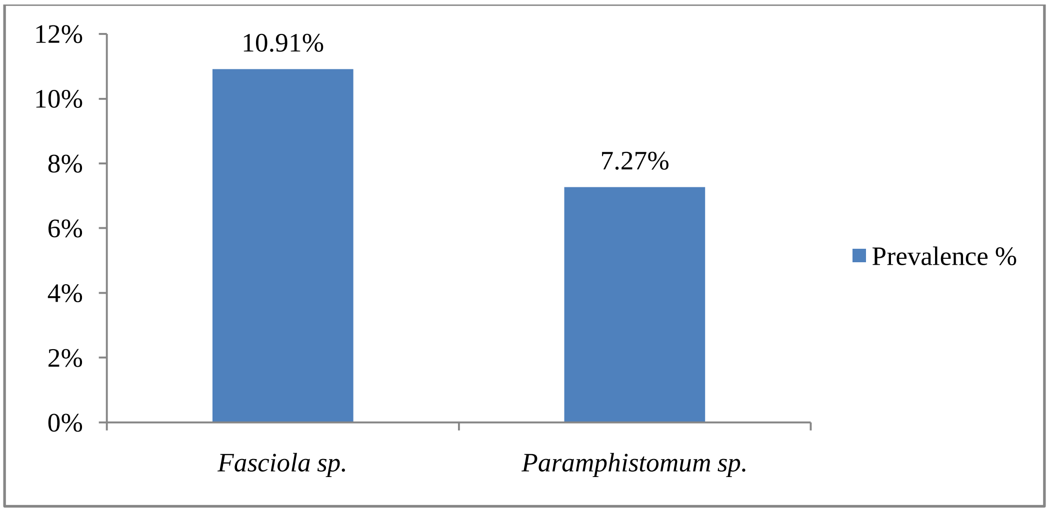

Figure 5: Prevalence of trematode parasites.

4.2.4 Cestode parasites:

Among 110 faecal samples examined, prevalence of cestode parasites was observed in 7 (6.36%) samples which is *Moniezia* sp.

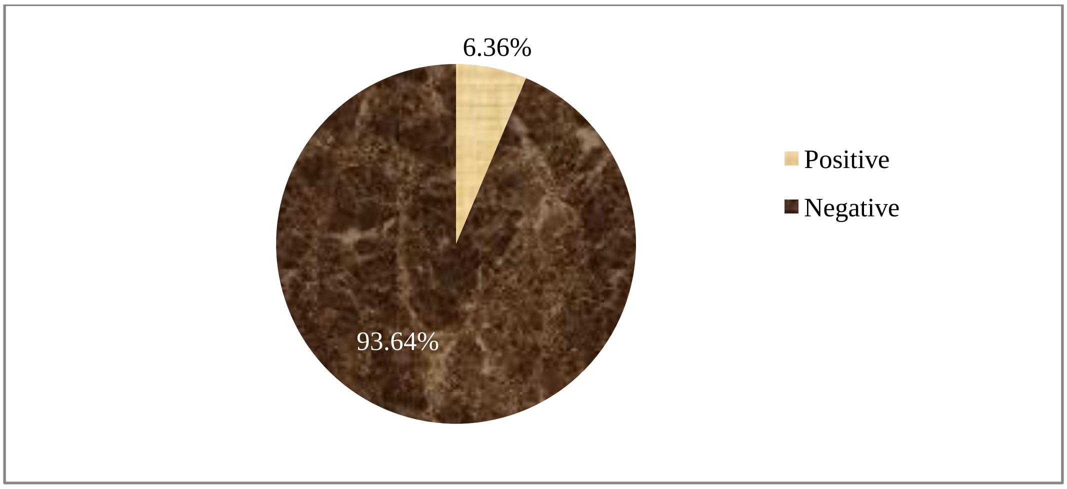

Figure 6: Prevalence of cestode parasites.

4.2.5 Nematode parasites:

Out of 110 faecal samples examined, 73 (66.36%) samples were found to be positive for nematode parasites. A total of ten genera of nematodes were identified. Among them *Haemonchus* sp. showed the highest prevalence (33.64%) in Laxmipur VDC, Dang followed by *Strongyloides* sp. (30.91%), *Strongyle* sp. (25.45%), *Trichostrongylus* sp. (14.55%), *Oesophagostomum* sp. (13.64%), *Bunostomum* sp. (10%), *Trichuris* sp. (8.18%), *Moniezia* sp. (6.36%), *Nematodirus* sp. (5.45%), *Chabertia* sp. (4.54%) and *Ascaris* sp. (1.82%). Statistically, distribution of nematode parasites in sheep was not found significant (χ^2=100.84, df=9, p-value=2.2).

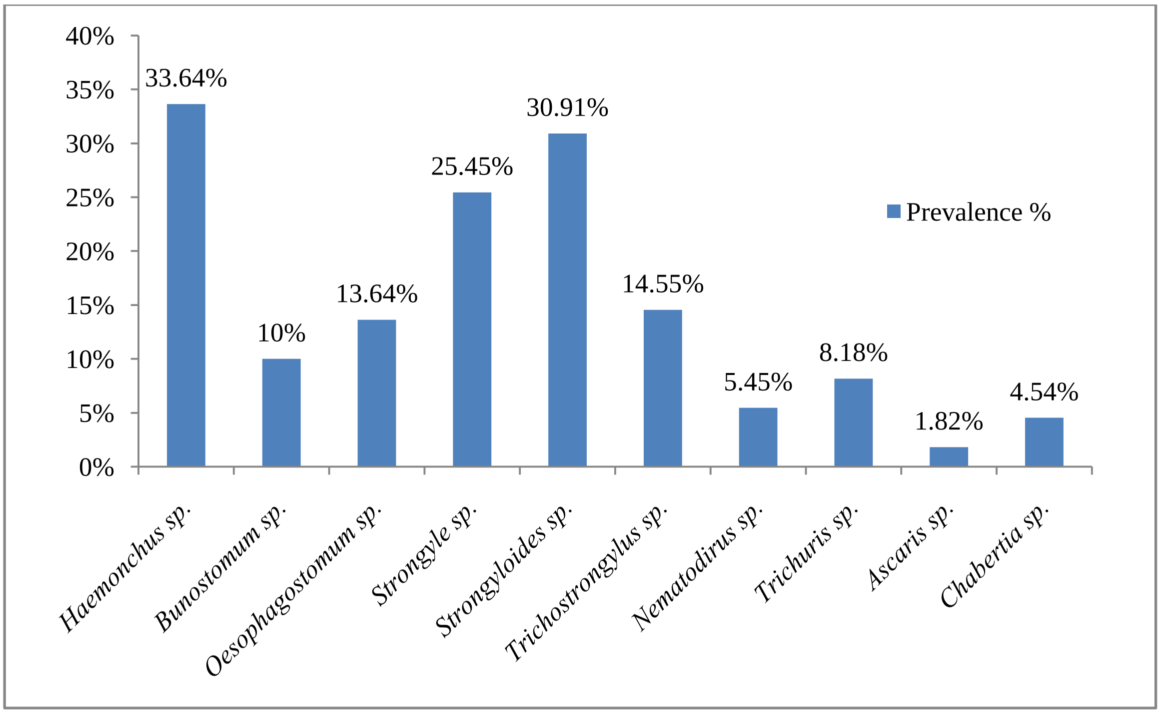

Figure 7: Prevalence of nematode parasites.

4.3 Season-wise prevalence of parasite in sheep:

The seasonal occurrence of parasitic infection in sheep shows higher infection of gastro-intestinal parasites in rainy season (92.93%) than in summer season (67.27%). Statistically, the distribution of gastro-intestinal parasites in two seasons seems significant (p-value=0.00194).

Table 3. Overall season-wise prevalence of gastro-intestinal parasite in sheep:

Seasons	No. of examined	No. of positives	Prevalence	χ^2	p-value
Summer	55	37	67.27%	9.6023	0.00194
Rainy	55	51	92.93%		

4.3.1 Seasonal prevalence of protozoan parasites:

Species-wise prevalence of protozoan parasites in different seasons demonstrated that the prevalence of *Entamoeba* sp. increased in relation to season but *Eimeria* sp. remained constant. Statistically there was not significant difference in distribution of protozoan parasites in summer season and rainy season.

Table 4. Seasonal prevalence of protozoan parasites:

S.N.	Parasite Name	Summer season (April)		Rainy season (August)		$\chi2$	p-value
		No. of positive	Prevalence (%)	No. of positive	Prevalence (%)		
1.	*Eimeria* sp.	2	3.63	2	3.63	0	1
2.	*Entamoeba* sp.	1	1.82	2	3.63	0	1

4.3.2 Seasonal prevalence of helminth parasites:

Prevalence of helminth parasites in relation to season showed that the parasites build up in rainy season and occur in low prevalence in summer season. *Ascaris* sp. and *Nematodirus* sp. are lacking in summer season. Distribution of *Nematodirus* sp. showed significant difference between summer season and rainy season (χ^2=4.407, p-value<0.05) while the distribution of other parasites did not show significant difference.

Table 5. Seasonal prevalence of helminth parasites:

S.N.	Parasite Name	Summer season (April)		Rainy season (August)		χ^2	p-value
		No. of positive	Prevalence (%)	No. of positive	Prevalence (%)		
1	*Haemonchus* sp.	18	32.73	19	34.55	0	1
2.	*Bunostomum* sp.	5	9.09	6	10.91	1.121	1
3.	*Oesophagostomum* sp.	6	10.91	9	16.36	0.309	0.578
4.	*Strongyle* sp.	11	20	17	30.91	1.198	0.274
5.	*Strongyloides* sp.	12	21.82	22	20	3.448	0.06
6.	*Trichostrongylus*	7	12.73	9	16.36	0.073	0.787
7.	*Nematodirus* sp.	-	-	6	10.91	4.407	0.036
8.	*Trichuris* sp.	2	3.63	7	12.73	1.936	0.164
9.	*Ascaris* sp.	-	-	2	3.63	0.509	0.476
10.	*Chabertia* sp.	2	3.63	3	5.45	0	1
11.	*Fasciola* sp.	5	9.09	7	12.73	0.093	0.76
12.	*Paramphistomum* sp.	3	5.45	5	9.09	0.135	0.713
13.	*Moniezia* sp.	3	5.45	4	7.27	1.127	1

4.4 Concurrency of gastro-intestinal parasites:

In this study from one host more than one type of genera of helminthes was identified in sheep. Out of 88 (80%) positive samples, 71 (80.68%) samples were detected to have mixed infection with two or more type of genera and 17 (19.32%) samples were detected to have single infection with only one type of genus in each microscopic field. There isn't significant difference between single infection and mixed infection (χ^2 =63.84, p-value=1.349).

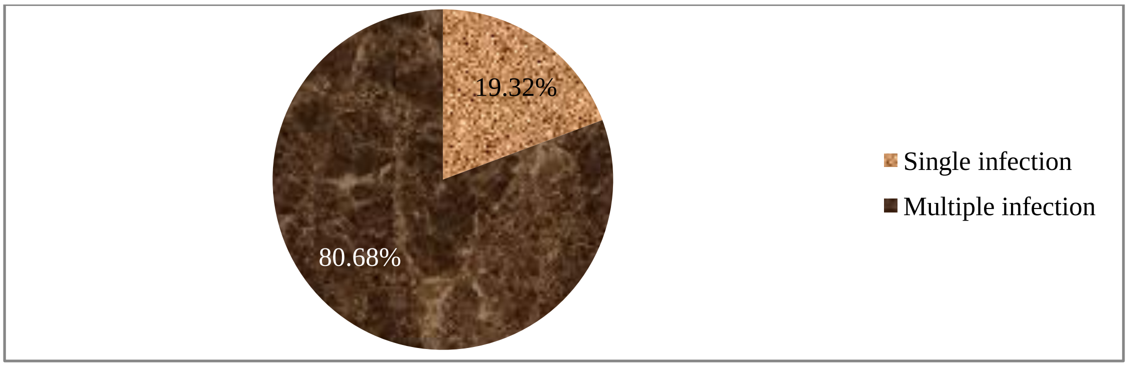

Figure 8: Overall concurrency of gastro-intestinal parasites.

4.5 Season-wise concurrency of gastro-intestinal parasites:

Sheep were found to be infected with single, double, triple and multiple gastro-intestinal parasites infection. In summer season out of 37 positive samples, the highest prevalence was noted for double infection (19 samples; 51.35%), followed by single (10 samples; 27.03%) and triple infection (8 samples; 21.62%) while in rainy season out of 51 positive samples, highest prevalence was noted for double infection (26 samples; 50.98%) followed by triple infection (13 samples; 25.49%), single infection (7 samples; 13.73%) and multiple infection (5 samples; 9.8%). Multiple infection was not observed in summer season. Statistically, concurrency of gastro-intestinal parasites in sheep was not found significant between summer season and rainy season.

Table 6: Season-wise concurrency of gastro-intestinal parasites in sheep:

Types of infection	Seasons		χ^2	p-value
	Summer (April)	Rainy(August)		
Single	27.03%	13.73%	1.656	0.198
Double	51.35%	50.98%	3.963	1
Triple	21.62%	25.49%	0.028	0.867
Multiple	-	9.8%	2.234	0.135

Photographs of identified eggs/cysts/larva of parasites found in sheep with 10X × 40X and 10X × 10X (Annex. 1).

Photo 5: *Eimeria* sp. (41µm)

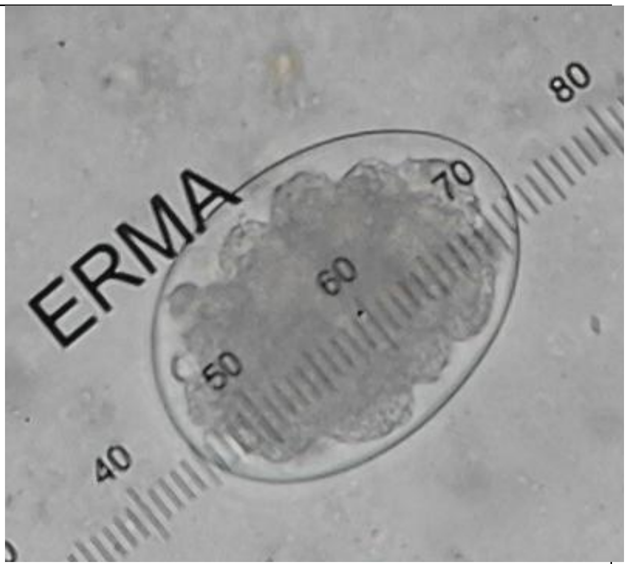

Photo 6: *Haemonchus* sp. (73 × 48 µm)

Photo 7: *Chabertia* sp. (92 ×51 µm)

Photo 8: *Bunostomum* sp. (95 × 46µm)

Photo 9: *Fasciola* sp. (140 × 73µm)

Photo 10*: Paramphostomum* sp. (149 × 80µm)

Photo 11: *Moniezia* sp. (59µm)

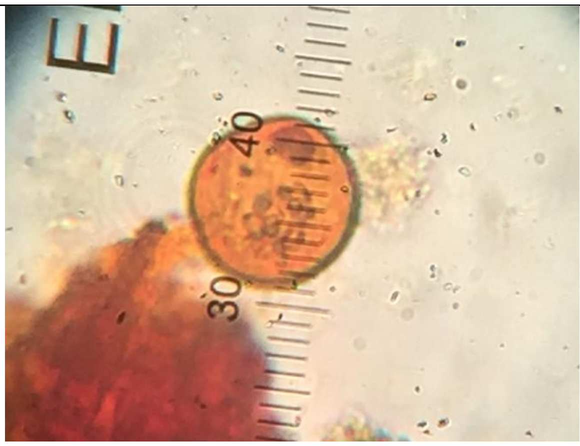

Photo 12: *Entamoeba* sp. (30µm)

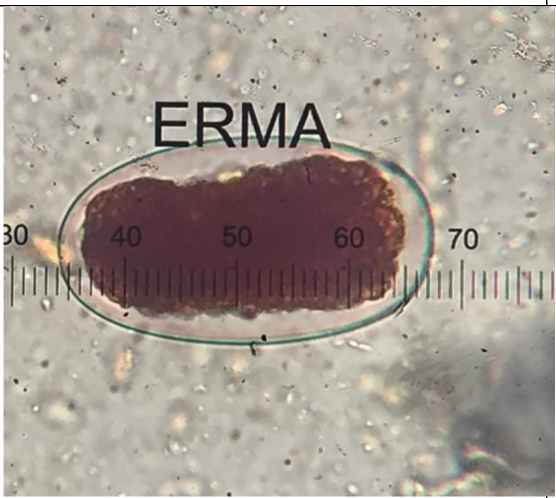

Photo 13: *Strongyle* sp. (89 × 49µm)

Photo 14: *Oesophagostomum* sp.(86 × 51 µm)

Photo 15: *Nematodirus* sp. (150 × 68 µm)

Photo 16: *Ascaris* sp. (40 µm)

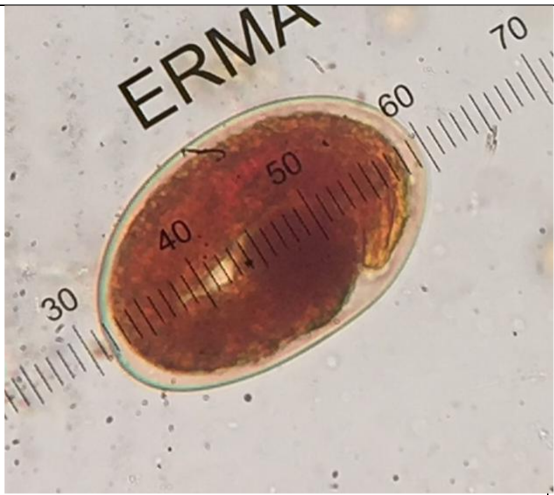

Photo 17: *Strongyloides* sp. (62 × 40 µm)

Photo 18: *Trichostrongylus* sp.(92 × 40 µm)

Photo 19: *Trichuris* sp. (78 × 41 µm)

Photo 20: *Strongyloides* larva

Unidentified eggs:

Photo 21: Unidentified egg (140 × 57 µm)

Photo 22: Unidentified egg (59 ×43 µm)

5. DISCUSSION

Parasites are cosmopolitan in distribution and no animal is free from parasites. Plenty of researches have been done in sheep in global context but hardly few in national context. Prevalence of gastro-intestinal parasites is considerably influenced by the climatic and geographical factors. Generally the warm and humid conditions, continuous high rainfall, decrease of grazing area, contaminated pasture, nutritional factors and traditional rearing system provide good condition for many gastro-intestinal parasites to flourish. Sheep shed parasites mainly by defecation and urination and other sheep get infection from the contaminated area. The gastrointestinal nematodes of sheep are one of the important parasitic diseases that obviously result in reduced productivity of sheep raised by smallholders using traditional husbandry management system. Nematode parasites cause death of sheep when they are severely infected. In the present study, the prevalence of gastro-intestinal parasites of sheep (*Ovis aries*) has been carried out for the first time in Dang district and it is also among very few research in Nepal. The research work is mostly compared with the research work done in other countries and very few with the national context.

The analysis of faecal samples revealed that among the 110 faecal samples examined, 88 were found positive for gastro-intestinal parasites infection with an overall prevalence of 80% in Laxmipur VDC, Dang which is a high prevalence. Total 15 genera belonging to 5 classes of gastro-intestinal parasites were observed during faecal examination. Among identified parasites, *Haemonchus* sp. showed the highest prevalence (33.64%), followed by *Strongyloides* sp. (30.91%), *Strongyle* sp. (25.45%), *Trichostrongylus* sp. (14.55%), *Oesophagostomum* sp. (13.64%), *Fasciola* sp. (10.91%), *Bunostomum* sp. (10%), *Trichuris* sp. (8.18%), *Paramphistomum* sp. (7.27%), *Moniezia* sp. (6.36%), *Nematodiru*s sp. (5.45%), *Chabetria* sp. (4.54%), *Eimeria* sp. (3.64%), *Entamoeba* sp. (2.73%), and *Ascaris* sp. (1.82%). It was found evident that Haemonchus sp was the dominant species affecting sheep. Next to it, the sheep were affected with the *Strongyloides* sp.

The prevalence of gastrointestinal parasite is related to the agro-climatic conditions like quantity and quality of pasture, temperature, humidity and grazing behaviour of the host (Muhammed *et al*., 2017). The rate of helminthes infection in sheep varies in different parts of the world. A variety of factors like standard of management, anthelmintic used, level of education and economic capacity of the farmers can also influence the prevalence of helminthes. Prevailing agro-climatic conditions like overstocking of animals, grazing of young and adult animals together with poorly drained land provide an ideal condition for the transmission of endoparasites to build up clinical infestation of the host (Gadahi *et al*., 2009).

The high prevalence of infection observed in the study (80%) agrees with the results of Maichomo *et al*. (2004), Yadav *et al*. (2006), Asif *et al.* (2008), Kaur and Kaur (2008), Sangma *et al*. (2012), Gizachew *et al*. (2014), Getachew *et al*. (2016), Khakural *et al*. (2005) and Acharya (2017) who reported 80%, 83.24%, 72%, 82%, 81.1%, 77.4%, 86.6%, 87.5% and 78.31% from Magadi division, south-western Kenya, Jammu district, India, Rawalpindi and Islamabad, Pakistan, Patiala and its adjoining areas, Tangail district, Bangladesh, Bako town, Western Ethiopia, Areka, Ethiopia, Karnali, Nepal, and

Ghanpokhara, Lamjung respectively who have also reported almost same prevalence in sheep with minor differences. The maintenance of high infection rates of GIT helminthes in sheep in the study areas was associated with the presence of favorable environmental conditions for the existence and development of the parasitic GIT helminthes larvae (Tefera *et al*., 2009). Active movement may increase the frequency of exposure to contaminated drinking water and food (Choubisa and Jaroli, 2013). The higher susceptibility in sheep may be associated to their foraging behavior (Bansal *et al*., 2015). The prevalence of parasites in these sheep supports the notion that parasitic infections are among the main health problems in small ruminants globally (Mbuh *et al*., 2008; Kantzoura, 2012; Muhammed *et al*., 2017).

The prevalence in sheep observed in the present study (80%) is lower than the study carried out by Pedreira *et al*. (2006), Almalaik *et al*. (2008), Tefera *et al.* (2009), Ibrahim *et al*. (2014) and Bansal *et al*. (2015) who reported 100%, 99.9%, 91.32%, 89.3% and 90.11% in Galicia, NW Spain, Tulus Area, Sudan, Bedelle, South-Western Ethiopia, Jimma town, Western Ethiopia and Mhow, Indore respectively. This difference might be due to the difference in age of examined animals and geographical and environmental location of the area. (Tefera *et al*., 2009). The higher prevalence observed in different parts of countries could be ascribed to over stocking, poor nutrition (starvation), poor management practice of the animals (lack of sanitation) and frequent exposure to the communal grazing lands that have been contaminated (Gizachew *et al*., 2014; Getachew *et al*., 2016).

Present study recorded higher prevalence in sheep compared with the study carried by Muraleedharan (2005), Raza *et al*. (2007), Nwosu *et al*. (2007), Tariq *et al*. (2008), Gadahi *et al.* (2009), Khan *et al*. (2010), Khalafalla *et al*. (2011), Nassrullah, (2011), Lashari and Tasawar (2011), Bhat, *et al*. (2012), Kantzoura *et al*. (2012), Zeryehun (2012), Lemma and Aber (2013), Choubisa and Jaroli (2013), Mir *et al.* (2013), Khajuria *et al*. (2013), Singh *et al.* (2013), Kenea *et al*. (2015), Varadharajan and Vijayalakshmi (2015), Mohammedameen (2016), Sultan *et al.* (2016) and Poddar *et al*. (2017) who reported 39.34%, 62%, 43.1%, 61.64%, 53.33%, 44.17%, 10.4%, 34.36%, 46.33%, 62.9%, 7.9%, 67.75%, 68.1%, 55.42%, 44.74%, 68.54%, 68%, 54.1%, 66.33%, 60%, 50% and 67.9% respectively from Southern Taluks of Central dry zone of Karnataka, Lower Punjab, Pakistan, North-Eastern Nigeria, Kashmir valley, Rawalpindi and Islamabad of Pakistan, Toba Tek Singh, Punjab, Pakistan, Nile Delta, Egypt, Sulamani province, Southern Punjab, Pakistan, Kashmir valley of India, Thessaly region of Greece, Haramaya, Southeastern Ethiopia, Asella, South Eastern Ethiopia, Southern Rajasthan, India, Jammu area of Jammu and Kashmir State, Jammu, Mathura, India, Kaffa and Bench Maji zones, Southwest Ethiopia, TamilNadu, India, Bardarash district, Duhok, Iraqi Kurdistan, Nile-Delta, Egypt and Sherpur, Bangladesh respectively.

The above authors reported relatively lower prevalence than the present findings. This difference might be attributed to the difference in animal species, and environmental conditions for the parasitic stage of larvae growth. The decrease in the prevalence in the present study compared with other studies could be due to the existence of unfavourable climatic or environmental factors that could support prolonged survival and development of the infective larval stage of most nematodes (Muhammed *et al*., 2017). This difference in prevalence rate in different parts of the country might be attributed to the difference in agro ecology and variation in management practice of animals (Gizachew *et al*., 2014).

In the current study, *Haemonchus* sp. (33.64%) was in highest prevalence which is almost similar to the work of Almalaik *et al*. (2008), Asif *et al*. (2008), Tariq *et al*. (2008), Tefera *et al.* (2009), Khajuria *et al*. (2013), Singh *et al.* (2013) and Varadharajan and Vijayalakshmi (2015) who also reported *Haemonchus* sp. as predominant species from Tulus Area, Sudan, Rawalpindi and Islamabad, Pakistan, Kashmir valley, Bedelle, South-Western Ethiopia, Jammu, India, Mathura, India, and TamilNadu, India respectively although the present result is less than the finding of Almalaik *et al*. (2008), Asif *et al*. (2008), Tariq *et al*. (2008), Tefera *et al.* (2009) and Khajuria *et al*. (2013) who reported *Haemonchus* sp. with the prevalence rate of 53.4%, 80.64%, 59.6%, 67.5% and 61.18% respectively. The present study confirmed that *Haemonchus* sp. is the most predominant parasite of parasite in Laxmipur Dang. Gastrointestinal nematodes rank highest on global index with *Haemonchus contortus* on top (Perry *et al*., 2002). The higher prevalence of *Haemonchus* sp. in the present study could be the high biotic potential of this nematode parasite to acquire resistant faster than others. It could also be due to poor management practices, grazing area, traditional rearing system and nutritional factors.

Strongyloides sp. is another common nematode parasite in sheep. The prevalence of *Strongyloides* sp. was 30.91% which is in close agreement with the study of Almalaik *et al*. (2008) who reported 26.2% from Tulus Area, Sudan and second prevalent parasite after *Haemonchus* sp. The study indicated that *Strongyloides* sp. also is one of the most prevalent species in sheep. Present result (30.91%) is lower than the research conducted by Kaur and Kaur (2008) in Patiala and it's adjoining areas who reported 57.14%. The highest prevalence of *Strongyloides* sp. in sheep may be due to increased resistance against *Haemonchu*s sp. in these hosts. It could also be due to lack of sanitation, difference in agro ecology and variation in management practice of animals.

The prevalence of *Strongyle* sp. (25.45%) reported in present study is in close agreement with the study of Nwosu *et al*. (2007) and Poddar *et al*. (2017) study conducted in North-eastern Nigeria and Sherpur, Bangladesh who reported 22.5% and 24.5% respectively. *Strongyle* sp. has been reported as predominant sp. having the higher prevalence rate from the study of Muraleedharan (2005), Yadav *et al*. (2006), Nwosu *et al*. (2007), Koinari *et al*. (2013), Khajuria *et al*. (2013), Bansal *et al*. (2015), Getachew *et al*. (2016), Poddar *et al*. (2017) and Acharya (2017) conducted in Southern Taluks of Central dry zone of Karnataka, R.S. Pura, Bishnah and Samba tehsils of Jammu district, India, North-eastern Nigeria, Papua New Guinea, middle agro-climatic zone of Jammu, Mhow, Indore, Areka, Ethiopia, Sherpur, Bangladesh, and Ghanpokhara, Lamjung which mismatches the present result. This could be due to climatic conditions and geographical factors between countries. This variation in prevalence of parasitic infestation depends upon difference in agro climatic condition and availability of susceptible host (Singh *et al.*, 2013).

Trichostrongylus sp. (14.55%) reported in present study is in close agreement with the finding of Khajuria *et al*. (2013) who also reported 13.67% prevalence rate of this species from middle agro-climatic zone of Jammu. *Trichostrongylus* sp. reported in the present study is less than the findings of Tefera *et al*. (2009), Ibrahim *et al*. (2014)), Kenea *et al*. (2015) and Acharya (2017) who reported 48.8%, 26.2%, 30.30% and 44% from Bedelle, South-Western Ethiopia, Jimma town, Western Ethiopia, Kaffa and Bench Maji zones, Southwest Ethiopia and Ghanpokhara Lamjung Nepal respectively and is very less than finding of Kaur and Kaur (2008) who reported 78.57% from Patiala and its adjoining areas. The difference in prevalence rate could be due to climatic condition, altitudinal and geographical variances.

Oesophagostomum sp. (13.64%) also has wide distribution in this study area. This result is higher than the finding of Almalaik *et al*. (2008), Khajuria *et al*. (2013), Ibrahim *et al*. (2014) and Kenea *et al*. (2015) who reported 2.2%, 3.84, 9.3% and 4.55% from Tulus Area, Sudan, middle agro-climatic zone of Jammu and Jimma town, Western Ethiopia and Kaffa and Bench Maji zones, Southwest Ethiopia respectively and lower than the finding of Tariq et al. (2008), Kaur and Kaur (2008) and Tefera *et al*. (2009) who reported 28.4%, 42.86% and 30.3% from Kashmir valley, Patiala and its adjoining areas and Bedelle, South-Western Ethiopia respectively. This difference could be due to grazing habits, standard of management, anthelmintic used and frequent exposure to the communal grazing lands that have been contaminated.

The prevalence of 10.91% of *Fasciola* sp. recorded in the present study is in close agreement with the report of Sangma *et al*. (2012), Khajuria *et al*. (2013) and Gizachew *et al*. (2014) who reported 8.4%, 8.2% and 9% in Tangail district, Bangladesh, middle agro-climatic zone of Jammu, Bako town Western Ethiopia respectively and higher than the report of Yadav *et al*. (2006), Gadahi *et al.* (2009), Kantzoura *et al*. (2012) and Bansal *et al*. (2015) who reported 3.08%, 4.44%, 0.5% and 4.56% from R.S. Pura, Bishnah and Samba tehsils of Jammu district, India, Rawalpindi and Islamabad of Pakistan, Thessaly region of Greece and Mhow, Indore respectively and lower than the finding of Lashari and Tasawar (2011), Choubisa and Jaroli (2013) and Ibrahim *et al*. (2014) who detected 21.41%, 15.18% and 19.6% in Southern Punjab, Pakistan, southern Rajasthan, India and Jimma town, Western Ethiopia respectively. This could be due to agro-climatic conditions like quantity and quality of pasture, temperature, humidity and environmental conditions.

The prevalence of *Bunostomum* sp. was 10% in the present study which is less than the report of Tariq *et al*. (2008), Tefera *et al.* (2009), Sangma *et al*. (2012) and Kenea *et al.* (2015) who reported 37.7%, 25.6%, 19.09% and 18.69% from Kashmir valley, Bedelle, South-western Ethiopia, Tangail district, Bangladesh and Kaffa and Bench Maji zones, Southwest Ethiopia respectively and higher than the report of Khajuria *et al*. (2013), Ibrahim *et al*. (2014) and Ibukun and Oludunsin (2015) who reported 3.83%, 2.3% and 1.8% from middle agro-climatic zone of Jammu, Jimma town, Western Ethiopia and Minna, North Central, Nigeria respectively. This variation could be due to preference of the parasite to cold climate than warm.

The prevalence rate of 8.18% of *Trichuris* sp. recorded in the present study is higher than the report of Sangma *et al*. (2012), Lemma and Aber (2013), Koinari *et al*. (2013), Getachew *et al*. (2016), Sultan *et al.* (2016) and Poddar *et al*. (2017) who reported 2.1%, 3.7%, 1.8%, 1.5%, 2.68% and 1.9% from Tangail district, Bangladesh, Asella, South Eastern Ethiopia, Papua New Guinea, Areka, Ethiopia, Nile-Delta, Egypt and Sherpur, Bangladesh respectively, almost similar with the report of Nassrullah (2011), Choubisa and Jaroli (2013), Gizachew *et al*. (2014), Ibrahim *et al*. (2014), and Acharya (2017) who reported 6%, 9.15%, 7.3%, 7.9% and 8% from Sulamani province, Southern Rajasthan, India, Bako town, Western Ethiopia, Jimma town, Western Ethiopia and Ghanpokhara, Lamjung respectively, less than the report of Tariq *et al.* (2008), Khajuria *et al*. (2013) and Bansal *et al.* (2015), who reported 23.5%, 12.1% and 21.70% from Kashmir valley, middle agro-climatic zone of Jammu and Mhow, Indore respectively and very less than the report of Gadahi *et al.* (2009) and Tefera *et al*. (2009), who reported 40% and 48.8%, from Rawalpindi and Islamabad of Pakistan and Bedelle, South-Western Ethiopia respectively. The difference in prevalence of *Trichuris* sp. in different reports in different countries is conflicting this could be due to different management practice of the animals and environmental factors.

The finding of *Paramphistomum* sp. (7.27%) in the present study is less than the finding of Poddar *et al*. (2017) who reported 13.2% from Sherpur, Bangladesh, very less than the report of Sangma *et al*. (2012) and Ibrahim *et al*. (2014) who reported 44.2% and 22.4% from Tangail district, Bangladesh and Jimma town, Western Ethiopia respectively, higher than the report of Gizachew *et al*. (2014) who reported 3.1% from Bako town, Western Ethiopia and is in agreement with the report of Sultan *et al.* (2016) who reported 9.38% from Nile-Delta, Egypt. It may be due to different climatic, geographic area.

The prevalence rate of 6.36% of *Moniezia* sp. is in harmony with the report of Choubisa and Jaroli (2013), Gizachew *et al*. (2014) and Bansal *et al*. (2015) who reported 6.98%, 7.3% and 5.77% from southern Rajasthan, India, Bako town, Western Ethiopia and Mhow, Indore respectively, higher than the report of Yadav *et al*. (2006), Bhat, *et al*. (2012), Sultan *et al.* (2016) and Poddar *et al*. (2017) who reported 0.96%, 3.3%, 0.89% and 3.8% from R.S. Pura, Bishnah and Samba tehsils of Jammu district, India, Kashmir valley of India, Nile-Delta, Egypt and Sherpur, Bangladesh respectively and less than the report of Kaur and Kaur, (2008), Ibrahim *et al*. (2014), Ibukun and Oludunsin (2015) and Acharya (2017)who reported 21.43%, 13.1%, 11.7% and 12% from Patiala and its adjoining areas, Jimma town, Western Ethiopia, Minna, North Central, Nigeria and Ghanpokhara, Lamjung respectively. This difference could be related to the agro-climatic conditions like quantity and quality of pasture, temperature, humidity.

The prevalence rate of *Nematodiru*s sp. (5.45%) in the present study is less than the report of Tariq *et al*. (2008), Gadahi *et al,* (2009), Nassrullah (2011) and Bhat *et al*. (2012) who recorded 29.4%, 11.11%, 29.8% and 9.0% from Kashmir valley, Rawalpindi and Islamabad of Pakistan, Sulamani province and Kashmir valley of India respectively and higher than the report of Kantzoura *et al*. (2012), Koinari *et al*. (2013) and Sultan *et al.* (2016) who reported 1.1%, 1.8% and 0.45% from Thessaly region of Greece, Papua New Guinea and Nile-Delta, Egypt respectively. This variation could be due to agro-climatic conditions of different countries.

The prevalence rate of *Chabertia* sp. (4.54%) in the present study is very less than the report of Tariq *et al.* (2008) and Tefera *et al*. (2009) who reported 37.7% and 20.9% from Kashmir valley and in and around Bedelle, South-Western Ethiopia respectively and almost similar to the report of Khalafalla *et al.* (2011), Ibrahim *et al*. (2014) and Kenea *et al.* (2015) who reported 0.6%, 4.2% and 3.54% from Nile Delta-Egypt, Jimma town, Western Ethiopia and Kaffa and Bench Maji zones, Southwest Ethiopia respectively.

The prevalence rate of 3.64% of *Eimeria* sp. is almost similar with the report of Yadav *et al*. (2006) who reported 6.73%, from R.S. Pura, Bishnah and Samba tehsils of Jammu district, India, less than the report of Bhat *et al*. (2012), Koinari *et al*. (2013), Ibrahim *et al*. (2014) and Sultan *et al.* (2016) who reported 9.8%, 17.3%, 11.7% and 16.52% from Kashmir valley of India, Papua New Guinea, Jimma town, Western Ethiopia and Nile-Delta, Egypt respectively and very less than the report of Gizachew *et al*. (2014) who reported 30.9% from Bako town, Western Ethiopia. From these results, it was seen that cooler and temperate climate is suitable for development of *Eimeria* sp. than warm and tropical climates.

Only few studies have been done regarding protozoan parasites. It was difficult to discuss about the prevalence rate of *Entamoeba* sp. (2.73%) of present study due to lack of species finding regarding the parasite but Sultan *et al.* (2016) has reported 10.27% prevalence of the parasite in Nile-Delta Egypt which is higher than the present result. The low prevalence rate of *Entamoeba* sp. from present finding indicates that the study area is less contaminated with parasite.

The prevalence rate of *Ascaris* sp. (1.82%) in the present study is less than the report of Ibukun and Oludunsin (2015) who reported 9.4% from Minna, North Central, Nigeria. The low prevalence rate of *Ascaris* sp. from present finding indicates that the parasite has less biotic potential at this study area and is less contaminated with parasite.

In the present study the seasonal occurrence of parasitic infection in sheep depicted higher infection of gastro-intestinal parasites in rainy season (92.93%) than in summer season (67.27%). The season wise infection percentage for different seasons was displayed in table number 3. The highest prevalence of parasites in rainy season is in consent with the research carried by Yadav *et al*. (2006), Sissay *et al.* (2007), Nwosu *et al*. (2007), Almalaik *et al*. (2008), Zeryehun (2012), Bhat *et al*. (2012), Lemma and Aber (2013), Khajuria *et al*. (2013), Choubisa and Jaroli (2013), Alade and Bwala (2015), Bansal *et al*. (2015) and Varadharajan and Vijayalakshmi (2015) in R.S. Pura, Bishnah and Samba tehsils of Jammu district, India, Semi-arid zone of Eastern Ethiopia, Semi-arid zone of north-Eastern Nigeria, Tulus Area, Sudan, Haramaya, Southeastern Ethiopia, Kashmir valley of India, Asella, South Eastern Ethiopia, Middle agro-climatic zone of Jammu, Southern Rajasthan, India, Maiduguri, Borno state, West Africa, Mhow, Indore and Coastal areas of TamilNadu.

On the contrary few workers have also observed and reported a high incidence of these parasitic infections in summer (Tariq *et al*., 2008), autumn (Khalafalla *et al*., 2011) and spring (Mir *et al*., 2013) season which is not in agreement with the present study but the reason is not clear yet.

During rainfall the number of infective larval stage of nematodes are increased, thereby making the animal more susceptible to infestation whereas the infestation is generally less during the dry season (Kantzoura *et al*., 2012). The rainy season which starts in the spring and earlier in summer made the environmental conditions more favorable for the development and survival of parasitic stages and led to increased availability of infective larvae in the rainy and post rainy season. It is well documented that gastrointestinal parasitism in grazing animals is directly related to the availability of larvae on pasture and seasonal pasture contamination. Heavy rainfall and high relative humidity predisposed to heavy parasitic infection in rainy/wet season. Climatic factors also influence dispersion of larvae in the herbage which increased the chance of contact between host and larvae. Higher infection during rainy season may also be attributed to suitable molarity of salt present in soil, an important factor for ecdysis (Soulsby, 1982; Varadharajan and Vijayalakshmi, 2015). In rainy season drinking water and food/fodder of animals gets contaminated. Therefore, in rainy season a high prevalence of protozoan and helminthic infection is possible (Choubisa and Jaroli, 2013). Rainfall favors the development of preparasitic larval stages. The prevalence of these parasites depends on the availability of suitable intermediate hosts which varies with different seasons. Generally snails are predominant during monsoon season (Bansal *et al*., 2015) which are the vectors of different endoparasites. Moisture is one of the bionomic factor that support the development of the infective stage of most parasites (Zeryehun, 2012). Rainy season is the most favourable period for larval development, oocyst sporulation and survival of the infective stage larval coupled with sufficient moisture and temperature (Faizal and Rajapakse, 2001; Braimah, 2015; Muhammed *et al*., 2017). Availability of the browse and a longer browsing time increases the chances of contact between the host and parasites (Quattara and Dorchies, 2001).

In this study from one host more than one type of genera of helminthes was identified in 71 (80.68%) samples. This finding is in harmony with reports of previous studies of Yadav *et al*. (2006), Almalaik *et al*. (2008), Kaur and Kaur (2008), Singh *et al.* (2013), Raza *et al*. (2014) and Getachew *et al*. (2016) conducted in R.S. Pura, Bishnah and Samba tehsils of Jammu district, India, Tulus Area, Sudan, Patiala and its adjoining areas, Mathura, India, Cholistan desert of Pakistan and Areka, Ethiopia who also detected mixed infection in their studies.

In present study, mixed infection were detected in 80.68% samples which is higher with the finding of previous studies conducted by Yadav *et al*. (2006), Singh *et al.* (2013), Kenea *et al*. (2015) and Getachew *et al*. (2016) who reported 13.46%, 26.47%, 17.7% and 26% from R.S. Pura, Bishnah and Samba tehsils of Jammu district, India , Mathura, India, Kaffa and Bench Maji zones, Southwest Ethiopia and Areka, Ethiopia respectively. These lower result than present study could be due to use of anti-parasitic drugs and de-worming programs in their studied area. But present study is almost similar with the study conducted in Patiala and its adjoining areas (Kaur and Kaur, 2008) who reported mixed infection in 85.71% samples. This higher mixed infection than the current study could be due to lack of sheep de-worming, very less use of anti-parasitic drugs, dietary deficiency, poor management system, lack of knowledge for sanitation and climatic and geographical variation.

These polyparasitism has been suggested to be an important cause of morbidity and loss of production in sheep (Kumsa *et al*., 2010). Moreover, the presence of interaction and compromization of the immune system of the host by polyparasitism has been described to increase their susceptibility to other diseases or parasites (Wang *et al*., 2006). Hence, polyparasitism is an important problem of sheep production in the current study area.

6.CONCLUSION AND RECOMMENDATIONS

6.1 Conclusion:

The present study conducted on sheep gastro-intestinal parasites in Laxmipur VDC, Dang shows that gastro-intestinal parasites of sheep are the most prevalent disease in the area affecting the well being of the animal. The study demonstrates the overall prevalence of gastro-intestinal infection in sheep of Laxmipur VDC, Dang was 80% using floatation and sedimentation technique. The total number of genera observed during faecal examination were 15 in number. The study demonstrated *Haemonchus* sp. as predominant species having the prevalence rate of 33.64%, followed by *Strongyloides* sp. (30.91%), *Strongyle* sp. (25.45%), *Trichostrongylus* sp. (14.55%), *Oesophagostomum* sp. (13.64%), *Fasciola* sp. (10.91%), *Bunostomum* sp. (10%), *Trichuris* sp. (8.18%), *Paramphistomum* sp. (7.27%), *Moniezia* sp. (6.36%), *Nematodiru*s sp. (5.45%), *Chabertia* sp. (4.54%), *Eimeria* sp. (3.64%), *Entamoeba* sp. (2.73%), and *Ascaris* sp. (1.82%).

The present study revealed infection rate higher in rainy season (92.93) than summer season (67.27%). The study showed sheep were more infected with mixed infection (80.68%) than single infection (19.32%). The prevalence is higher in the study area as a normal and has been characterized by poor husbandry practices due to lack of veterinarian information and lack of awareness about gastro-intestinal diseases in sheep. The study confirmed that sheep were found to be most susceptible and infested by various gastro-intestinal parasites and the predominant parasites were *Haemonchu*s sp., *Strongyloides* sp. and *Trichostrongylus* sp. in the study area. Management practices, grazing area, nutritional factors, traditional rearing system, climatic conditions and geographical factors can be considered as important factors which influence the prevalence of gastro-intestinal parasites. The higher prevalence of present showed an alarming situation in the study area. This finding might be useful while designing control strategies of gastro-intestinal parasites in sheep and other ruminants as well.

6.2 Recommendations:

Based on the conclusion, it was recommended that:

- Molecular techniques should be applied for species level identification.
- Intensity of gastro-intestinal parasites should be studied.
- High prevalence was seen in rainy season so appropriate strategic treatment with broad spectrum anthelmintic should be practised during the start and end of rainy season.
- Good husbandry practices should be employed.
- The field veterinarians and stockowners should be aware of the importance and burden of GIT helminthosis in sheep and other domestic animals because sheep have possibility of infecting from other animals.

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ANNEX. 1

Identified eggs and cysts of parasites found in sheep:

Name of parasites	Range of diameter of eggs and cysts (in µm)		Morphological characters	Reference values (Soulsby, 1982; Poddar *et al*., 2017)
	Length	Width		
Eimeria sp.	33	-	Cysts are small, rounded or spherical with four nucleus.	15-55µm
Entamoeba sp.	22	-	Cysts are elongated oval shaped, having prominent cyst wall with micropyle and protoplasmic mass is spherical in centrally or sub-centrally.	20-35 µm
Haemonchus sp.	73	48	Embryo divided into 16-32 cells when passed in faeces.	70-85µm × 41-48µm
Bunostomum sp.	95	46	Bluntly rounded, embryonic cells darkly granulated	79-97µm × 47-50µm
Oesophagostomum sp.	78	52	Thin shells, ovular, laid in eight to sixteen cell stage	73-89µm × 34-45µm
Strongyle sp.	84	43	Oval, thin-shelled, segmented, similar or nearly similar pole, strongly barrel-shaped side-walls, smooth surface.	75-92µm ×41-54µm
Strongyloides sp.	54	28	Blunt ends, thin cells, contain fully developed embryos or larva	40-60µm × 20-40µm
Trichostrongylus sp.	70	30	Irregular, ellipse dissimilar, kidney-shaped not very wide poles, one of which was more rounded than the other, dissimilar side-walls.	70-108µm ×30-40µm
Nematodirus sp.	150	68	Brown shell, thin, parallel sides	152-182µm × 67-77 µm
Trichuris sp.	80	40	Brown,barrel-shaped, with a transparent plug at either pole	70-80µm × 30-42µm
Ascaris sp.	40	-	Oval, thick shells, brownish-yellow in colour, albuminous layer bear prominent projections	50-75µm ×40-50µm
Chabertia sp.	91	50	Unembryonated, typical *Strongyle* type	90-105µm ×50-55µm
Fasciola sp.	138	75	Thin shells, operculated,brown	130-150µm × 63-90µm
Paramphistomum sp.	114	73	Operculated, gets stained in methylene blue	114-176µm ×73-100µm
Moniezia sp.	59		Triangular, with pyriform apparatus	56-67µm