PREVALENCE OF GASTROINTESTINAL PARASITES IN DOMESTIC PIGS (*Sus scrofa domesticus*, Carl Linnaeus, 1758) OF JALTHAL VDC, JHAPA, NEPAL

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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

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Institute of Science and Technology
Tribhuvan University
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DECLARATION

I hereby declare that the work presented in this thesis has been done by myself, and has not been submitted elsewhere for the award of any degree. All sources of information have been specifically acknowledged by reference to the authors or institution(s).

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RECOMMENDATION

This is to recommend that the thesis entitled **" Prevalence of gastrointestinal parasites in domestic pigs of Jalthal VDC"** has been carried out by Sugandha Shah 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 **Mr. Janak Raj Subedi** this thesis submitted by Sugandha Shah entitled **" Prevalence of gastrointestinal parasites in domestic pigs of Jalthal VDC"** 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 Sugandha Shah entitled **"Prevalence of gastrointestinal parasites in domestic pigs of Jalthal VDC"** has been accepted by the partial fulfillment for the requirements of Master's of Science in Zoology with special paper Parasitology .

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LIST OF ABBREVIATIONS

Abbreviated form	Details of abbreviations
AAAE	Addis Ababa Abattoris Enterprise
ABPSD	Agri-Business Promotion and Statistics Division
CBS	Central Bureau of Statistics
CDR	Central Development Region
CI	Confidence Interval
df	Degree Of Freedom
ELISA	Enzyme-Linked Immune Sorbent Assay
FAO	Food and Agriculture Organization
GDP	Gross Domestic Product
GI	Gastrointestinal
GIP	Gastrointestinal Parasite
GIT	Gastrointestinal Tract
gm	Gram
Km	Kilometre
Km^2	Kilometre Square
mm	Millimetre
ml	Milliliter
NZFHRC	National Zoonoses and Food Hygiene Research Centre
P value	Probability value
rpm	Revolutions per minute
sp.	Species
TU	Tribhuvan University
VDCs	Village Development Committees

ABSTRACT

The domestic pigs (*Sus scrofa domesticus* or *Sus domesticus*), often called swine or hogs are collectively grouped under the genus *Suis* within the suidae family. Gastrointestinal parasitic infections in pigs are one of the major challenges in pig production in Nepal. Pigs heavily infected with gastrointestinal parasites are more susceptible to disease, the resulting diseases being major causes of zoonosis and economic loss. The present study was conducted from May to September, 2017 to determine the prevalence of gastrointestinal parasites in pigs. A total of 150 faecal samples were collected from two different locations (Kali-Jhoda and Meche Gaun) of Jathal village development committee of Jhapa, Nepal. The collected faecal samples were microscopically examined by differentiation floatation and sedimentation techniques for isolation of parasitic eggs and oocysts. Out of 150 faecal samples examined, 136 samples (90.67%) were found to be positive for gastrointestinal parasites including ten genus of parasites. Out of 136 total positive samples, 38 samples (27.94%), 69 samples (50.74%) and 29 samples (21.32%) were found to be positive for protozoan, nematode and trematode infections respectively. Among the examined samples, *Ascaris* sp. (46%) showed the highest prevalence followed by *Strongyloides* sp. (38.67%) *Ancyclostoma* sp. (32.67%) *Eimeria* sp. (25.33%), *Fasciola* sp. (19.33%), *Balantidium coli* (9.33%), *Trichuris* sp. (8.67%), *Trichostrongylus* sp. (2%), while *Isospora* sp. (1.33%) and *Metastrongylus* sp. (1.33%) showed the lowest prevalence. The study had also revealed that 107 (78.68%) samples and 29 (21.32%) samples were found to have mixed and single infection respectively. Statistically, the differences in the prevalence of single and mixed infections were found to be insignificant (χ^2=29.776, P>0.05). Location wise, the highest prevalence was found in Kali-jhoda (93.33%) while Meche gaun (88%) showed the lowest prevalence. Statistically, the difference in the prevalence of gastrointestinal parasitic infection in relation to location were found to be insignificant (χ^2=0.0169, P>0.05). This study revealed that gastrointestinal parasites were the major biological constraints contributing to the low productivity of pig and hampered the economic benefit obtained from the sector. Therefore, further detailed investigations are needed to formulate appropriate and cost-effective strategies for the control of gastrointestinal parasites in pigs in Jalthal village development committee.

1. INTRODUCTION

1.1 Background

Pigs are collectively grouped under the genus *Suis* within the suidae family. The domestic pig (*Sus scrofa domesticus* or *Sus domesticus*), often called swine or hog is a large even-toed ungulate. It is most often considered to be a sub-species of the wild boar, which was given the name *Sus scrofa* by Carl Linnaeus in 1758; following from this the formal name of domestic pig is *Sus scrofa domesticus* (Gentry *et al*., 2004). Pigs are omnivores and consume both plants and animals. Traditionally pigs are most neglected domestic animals which have been domesticated as a source of food, leather and similar products since ancient times. More recently, they have been involved in biochemical, research and treatment (Adebisi, 2008).

Nepal is an agricultural country with poor economy. The livestock sector contributes 30% of the agricultural GDP (FAO, 2005). About 92% of the rural households benefits from this sector (CBS, 2004). The livestock population of the country namely cattle, buffalo, goat, pig and fowl are (in millions) 7.0, 4.2, 7.4, 0.9 and 23.2 respectively (ABPSD, 2006). Pig husbandry and pork production in Nepal is an early stage of development compared to other livestock systems. Traditionally, pigs have been associated with low social groups, and so these animals have been neglected in improvement programmes. The pig production in Nepal concentrates in the hill zones (mostly in the eastern hills) because the resident ethnic groups (Rai, Limbu, Magar, Tamang, Gurung and Tharu) have no reservations to keep and eat pork. Total population of pig in Nepal is 1190138 (CBS, 2014). The pig population is 58% in the mid hills, 11% in the mountains and 31% in the plain territories (Sharma, 2003). In Nepal, three main types of indigenous pig are kept, these are Bampudke, Chwanche and Hurra (Shaha and Joshi, 2003). Bampudke pigs are found in lower hills, Chwanche in the middle mountains and Hurra pigs are in the plain territories. It is estimated that 58% of the pigs are Chwanche, 23% Hurra and 19% are improved breeds. The exotic breeds Hampshire, Landrace, Tamworth, Saddleback and Fauyen, are introduced in Nepal with a view to upgrade native swine.

In Nepal rearing of pigs have been found in different places including rural and urban areas. However, the management is mainly extensive whereby pigs are allowed to scavenge on household wastes at backyards and municipal garbage dumping sites. Such extensive pig husbandry with poor environmental hygiene and pigs edacious feeding behavior may render infection of the animals with helminth and GIT in Nepal. In addition, pigs can harbor a range of parasites and diseases that can be transmitted to humans. These include trichinellosis, taeniasis, cysticercosis and brucellosis. The disease in human and pigs is an ancient parasitic disease that has been rooted in developing countries and is now emerging as a major health problem of global dimensions (Sciutto *et al*., 2000). Infection of pigs with gastrointestinal parasites is widely reported from all corners of the world and shown to be influenced by the type of pig management practiced. Poor environmental hygiene coupled with extensive managements is reported as risk factors of infection of pig with GIT parasites (Nansen and Roepstorff, 1999). The common practices of feeding offal and kitchen waste in the backyards to the pigs contributes to the transmission of parasitic diseases in Nepal (Joshi *et al*., 2005).

1.2 Intestinal Parasites of Pig

Gastrointestinal parasites are parasites that live within the host's gastrointestinal tract extending from the mouth through the oesophagus, stomach, small and large intestine down to the rectum mainly helminth and protozoan parasites (Junaidu and Adamu, 1997). The animals suffer from a variety of infectious and non-infectious diseases, particularly that of parasitic origin (Iqbal *et al*., 2000; Akhter and Arshad, 2006). Internal parasites devitalize pigs by robbing them of essential nutrients and injuring vital organs (Myer and Walker, 1999). Pigs heavily parasitized are more susceptible to disease, the resulting diseases being major causes of zoonosis and economic loss (Bernard *et al*., 2015). The most commonly encountered gastrointestinal parasites are the large round worm *Ascaris suum*, the thread worm *Strongyloides ransomi*, the whipworm *Trichuris suis*, the nodular worm *Oesophagostomum dentatum* and the *coccidia* especially *Isospora suis* and *Cryptosporidium parvum* in neonates and *Eimeria* sp. at weaning. Gastrointestinal parasite of pigs can result in loss of appetite, poor growth rate, poor feed conversion efficiency and potentiation of other pathogens or even death (Stewart and Hoyt, 2006). These consequently lead to significant economic losses like decreased litter size, poor growth rate, reduced weight gain and their potential to infect humans have recently become a major issue among the public because of recent out breaks of water-borne parasitic diseases such as Giardiasis and cryptosporidiosis (Olson and Guselle, 2000).

1.2.1 Intestinal protozoan parasites

The protozoan parasites are microscopic, unicellular organism in which the various activities of metabolism, locomotion, etc. are carried out by organelles of the cell (Soulsby, 1982). The widely prevalent intestinal protozoan parasites commonly found in pigs include *Eimeria* sp., *Entamoeba* sp., *Isospora* sp., *Cryptosporidium* sp., *Balantidium coli* etc.

Eimeria, *Isospora* and *Cryptosporidia* are three types of coccidia that live and multiply inside the host cells, mainly in the intestinal tract and causes disease called coccidiosis. *Isospora suis* is the most pathogenic of the three types of coccidia. Disease is common and widespread in suckling piglets and occasionally in pigs up to 15 weeks of age. The main clinical sign is severe diarrhea, anorexia and weight loss (Tomass *et al*., 2013).

Entamoeba polecki is a cosmopolitan intestinal protozoan which is best known for its infection in pigs. This parasite is found commonly in wild and domestic pigs all around the world. Infection with *Entamoeba polecki* is almost always asymptomatic in humans, but debate remains about the possibility of non specific symptoms such as diarrhea, bloody stool, fever, nausea, vomiting, abdominal cramps, inspiratory restriction and weight loss (Solaymani-Mohammadi and Petri Jr, 2006).

Balantidium coli is the only ciliated protozoan capable of causing disease in humans. Pigs are the reservoir for human infection and is transmitted by the ingestion of *B.coli* cysts from pig faeces through water and food intake. *B.coli* cysts are ovoid to spherical and measure 40-60μm. They are faintly yellowish in colour (Soulsby, 1982).

1.2.2 Intestinal helminth parasites

The helminthes are multicellular, bilaterally symmetrical, elongated, and flat or round bodied organisms (Soulsby, 1982). Three major assemblages of parasitic helminthes are recognized: the Nemathelminthes (nematodes) and the Platyhelminthes (flatworms), the latter being subdivided into the cestoda (tapeworms) and the Trematoda (flukes).

1.2.2.1 Nematodes

Nematodes (roundworms) are free-living or parasitic, unsegmented worms, usually cylindrical and elongate in shape and tapering at the extremities. They have a fluid-filled internal body cavity (pseudocoelum) which acts as a hydrostatic skeleton providing rigidity. Adult worms form separate sexes with well-developed reproductive systems (Soulsby, 1982). The widely prevalent nematodes are *Ascaris suum*, *Strongyloides ransomi*, *Oesophagostomum* sp., *Trichuris suis*, *Hyostronglus rubidus* etc.

Ascaris suum (large roundworm) is the most important gastrointestinal worm of pigs. It is more common in growing pigs than in adult pigs. Adults are large and pinkish-yellow. The males measure 15-25cm by about 3mm and the females up to 41cm by 5mm. The eggs are oval, measuring 50-75 by 40-50μm. They have thick shells, the albuminous layer bears prominent projections and they are brownish-yellow in colour. Ascariasis in pigs depend on the severity of the infection. Newborn pigs which become heavily infected may show signs of pneumonia, especially a cough and exudate into the lungs. In less severe cases the animals cough and their growth is stunted. Heavy infections with adult worms produce diarrhoea which has a marked effect on their growth rate (Soulsby, 1982).

Strongyloides ransomi (intestinal threadworm) is more common in the warmer climatic regions, where it is an important parasite of suckling pigs. *Strongyloides ransomi* occurs in the small intestine of the pig. Adults are practically microscopic, measuring about 3.33-4.49mm in length. The eggs measure 45-55 by 26-35μm. Larvae enter the pig by penetrating the skin or mucous membranes of the mouth and are transported by the blood to the lungs, coughed up and swallowed. They then develop to maturity in the small intestine. The infective larvae can also cross the placenta or be excreted by the colostrums and therefore infect piglets within 24 hours of birth. The prenatal period is from 3-7 days. Migration of the larvae causes considerable damage and result in coughing, stiffness, pain, vomiting and bloody diarrhoea particularly from 10-14 days of age (Soulsby, 1982).

Strongyle worms are the most important gastrointestinal worms of pigs. There are two important strongyle worms in the pig. *Hyostrongylus rubidus*, the red stomach worm and *Oesophagostomum dentatum*, the nodular worm which lives in the large intestine.

Hyostrongylus rubidus (red stomach worm) occurs in the stomach of the pig in many countries. The male is 4-7mm long and the female 5-10mm. The worms are slender and reddish when they are fresh. The body cuticle is transversely striated and also bears 40-45 longitudinal striations. The bursa is well developed, but the dorsal lobe is small. The spicules are 0.13 mm long. The vulva is situated 1.3-1.7 mm anterior to the anus. The eggs measure 71-78 by 35-42 μm. The eggs hatch, at ordinary temperatures, in 39 hours and the larvae develop to the infective stage in seven days. The parasites burrow into the gastric mucosa and suck blood which may be present in small numbers without causing any ill effects, but often their presence is associated with a marked gastritis and in some cases marked ulceration. The animals lose condition rapidly and became weak. Diarrhoea occurs and the faeces may be dark in colour (Soulsby, 1982).

Oesophagostomum dentatum (nodular worm) occurs in the large intestine of pigs and peccaries throughout the world. The males are 8-10 mm long and the females 11-14 mm. The cephalic vesicle is prominent, but cervical alae are practically absent. The cervical papillae are towards the posterior end of the oesophagus. The spicules are 1.15-1.3 mm long. The eggs measure 35-45 by 60-80μm. The infective larvae exsheath in the small intestine and enter the mucosa of the large intestine, causing small nodules. Larvae re-enter the lumen of the large intestine six to seven days later, having moulted to the fourth-stage at four days after infection (Soulsby, 1982).

Trichuris suis (whip worm) lives in the large bowel and causes local damage to the intestinal wall. These worms do not migrate around the body. It is cosmopolitan in distribution. The male is 30-50 mm long and female 35-50 mm. The anterior portion forms about two-thirds of the total length. The spicule is 2-3.35 mm long, with a blunt tip, and its sheath is variable in shape. The eggs measure 50-60 by 21-25μm. They are readily recognized through a worm egg examination through their bipolar egg shape (Soulsby, 1982).

Trichinella spiralis is an important parasite of the pig as man may become infected resulting in severe muscular pains and swelling of the face. The adult worm lives in the small intestine of pigs. The males measure about 1.4-1.6mm and the female 3-4mm in length. The body is slender and the oesophageal portion is not markedly longer than the posterior part. The hind end of the male bears a pair of lateral flaps on either side of the cloacal opening, with two pairs of papillae behind them. There is neither a spicule nor a sheath. The vulva is situated near the middle of the oesophageal region. The eggs measure 40 by 30μm and contain fully developed embryos when in the uterus of the female (Soulsby, 1982).

1.2.2.2 Cestodes

Cestodes (tapeworms) are hermaphrodite, endoparasitic worms with an elongate flat ribbon-like bodies with a single anterior holdfast organ (scolex) and without a body cavity or alimentary canal. They may be a few millimeters to several metres in length. The body consists of a head or scolex. This is usually followed by a short unsegmented portion called the neck, and in general, the remainder of the body or strobila consists of a number of segments or proglottids which are separated by transverse constrictions and vary considerably in shape and size. Each proglottid usually contains one or two sets of reproductive organs. The scolex is usually globular (Soulsby, 1982). The major cestode parasite of pig is *Taenia solium.*

Taenia solium is the pork tapeworm belonging to cyclophyllid cestodes in the family Taeniidae. The adult worm occurs in the small intestine of man and has a flat, ribbon-like body, which is white in color and measures 3-5 m or up to 8 m long and can survive for up to 25 years. Its distinct head, the scolex, bears a rostellum which has two rows of hooks. The main body, the strobila, consists of a chain of segments known as proglottids. The gravid proglottids are 10-12 mm long by 5-6 mm wide and the uterus has seven to 16 lateral branches. The eggs are 26-34 m in diameter. The pig and wild boar are the main hosts of the metacestode. It completes its lifecycle in humans as definitive host and pigs as intermediate host. It is transmitted to pigs through human faeces or contaminated fodder, and to humans through uncooked or undercooked pork. Pigs ingest embryonated eggs, morula, which develop into larvae, the oncospheres, and ultimately into infective larvae, cysticerci. A cysticercus grows into an adult worm in human small intestines (Soulsby, 1982).

1.2.2.3 Trematodes

Trematodes or flukes are dorsoventrally flattened and are unsegmented and leaflike. All the organs are embedded in a parenchyma, no body cavity being present. Suckers, hooks or clamps attach these species to the exterior or the internal organs of their hosts. A mouth and an alimentary canal are present, but usually there is no anus (Soulsby, 1982). The major trematode parasites of pigs are *Fasciola* sp., *Schistosoma suis*, *Dicrocoelium* sp. etc.

1.3 Objectives

1.3.1 General Objective

- Prevalence of gastrointestinal parasites in domestic pigs *(Sus scrofa domesticus)* of Jalthal VDC, Jhapa, Nepal.

1.3.2 Specific Objectives

- To determine the prevalence of gastrointestinal parasites in pigs.
- To compare location-wise prevalence of gastrointestinal parasites in pigs.
- To determine the concurrency and intensity of gastrointestinal parasites in pigs.

1.4 Significance of the study

Pig production in Nepal is one of the rapidly growing livestock business for meat as well as for leather. Although pig contributes the second major source of meat, the proper growth of the farming is not seen as expected (Thapa Shrestha *et al*., 2014). Poor environmental hygiene coupled with extensive managements is reported as risk factors of infection of pig with GIT parasites (Nansen and Roepstorff, 1999). Gastrointestinal parasitic infections in Pig’s cause economic losses due to condemnation of liver, reduce growth rate and feed conversion (Stephenson *et al*., 1980 and Hale *et al*., 1985). In global context a lot of research has been carried out regarding gastrointestinal parasites of pigs. However, in context of Nepal it appears that the literatures regarding gastrointestinal parasites in pigs are very much scanty. The lack of gastro-intestinal parasitic information in pigs in Nepal, thus motivate this study. Hence, such studies are essential and the present study may provide information and suggestive guidelines for further research, researches and investigation regarding various protozoan and helminth parasites of pigs.

1.5 Limitations of the study

This study was designed to determine the prevalence of gastrointestinal parasites in pigs. The identification of parasites was based on morphology and size of parasitic eggs/oocyst and larva. The study doesn't revealed why some parasites were more predominant and others were not. This study was only limited to certain parameters related to the topic due to cost and time constraints. This study had been carried out for the partial fulfillment of the requirements for the Master's Degree in Zoology at Tribhuvan University, Kathmandu, Nepal.

2. LITERATURE REVIEW

Parasites play a major role in ecosystems (Esch and Fernandez, 1993), host population growth and regulation (Hochachka and Dhondt 2000, Hudson *et al*., 1998) and community biodiversity (Hudson *et al*., 2002). Many studies have been carried out regarding the intestinal parasites affecting pigs in different countries. Parasitic diseases are a major obstacle to the growth of the pig industry. The main negative effects on animals are reflected in economic losses for producers, such as: reduced feed conversion, reduced fertility, low number of piglets born and weaned low weight piglets at birth and at weaning as well as losses relating to viscera of high condemnation rate in slaughter houses (Nansen and Roepstorff, 1999; Roepstorff *et al*., 1998). In addition, pigs are considered the main reservoir of *Balantidium coli* and *Entamoeba polecki*, which can infect humans, especially farm laborers (Solaymani-Mohammadi and Petri Jr, 2006). Pigs can be infected by different parasites including protozoans, trematodes, cestodes and nematodes. Prevalence studies on intestinal parasites affecting pigs have been undertaken worldwide.

2.1 In Global context

Several surveys have been conducted to determine the occurrence of internal parasites in pigs. The numbers of species present and the prevalence rate are high when production systems are traditional (Roberts, 1940; Ajayi and Arabs, 1988), however, many recent studies have shown that superior housing and hygienic in combination with routine anthelmintics usage have lead to decreasing prevalence rates of many parasites (Pattison *et al*., 1980, Morris *et al*., 1984, Kennedy *et al*., 1988, Mercy *et al*., 1989). Generally, pigs are infected by various common intestinal parasites. Gastrointestinal parasites of pigs include both protozoan and helminth parasites. Nearly all of the most common intestinal parasites of pigs have been shown to be unevenly distributed among different age groups (Jacobs and Dunn, 1969, Pattison *et al*., 1980, Morris *et al*., 1984, Mercy *et al*., 1989). *Isospora suis* and *Strongyloides ransomi* are most common in piglets, *Ascaris suum* and *Trichuris suis* in growing pigs and *Oesophagostomum* sp., *Hyostrongylus rubidus* and *Eimeria* sp. are most common in adult pigs. These characteristics age-dependent distributions are most probably caused by different host-parasite relationships, especially the immunogenicity of the parasite (Pattison *et al*., 1980, Murell, 1986, Roepstorff and Nansen, 1994).

Several research work have been carried out in Asia regarding the gastrointestinal parasites in pigs. Various gastrointestinal parasites particularly, *Coccicia*, *Cryptosporidium* sp., *Balantidium coli*, *Ascaris suum*, *Trichuris suis*, *Strongyles*, *Oesophagostomum* sp., *Metastrongylus* sp., *Strongyloides* sp., *Fasciola* sp., *Dicrocoelium* sp., *Schistosoma suis* etc has been reported in pigs from India, Malaysia, China, Japan and Bangladesh (Borkotoky *et al*., 2014, Edmund *et al*., 2005, Weng *et al*., 2005, Matsubayashi *et al*., 2009, Dey *et al*., 2014). Shaikh and Huq (1984) examined six faecal samples from pigs and recorded *Ascaris lumbricoides* (33.3%), *Ancyclostoma duodenale* (16.7%), *Trichuris suis* (16.7%) and *Fasciolopsis buski* (16.7%) for the first time in Bangladesh. However, Edmund *et al.* (2005) reported *Balantidium coli* (53.3%), *Trichuris suis* (17.5%), *Ascaris suum* (15.8%), *Coccidia* (11.6%), *Strongyles* (8.3%), *Entamoeba suis* (2.5%) and *Echinostoma malayum* (1.7%) in Shah Alam abattoir in Selangor, Malaysia. However, Dadas *et al.* (2016) reported six parasites species namely, *Ascaris suum*, *Balantidium coli*, *Trichuris suis*, *Isospora suis*, *Strongyloides ransomi* and *Globocephalus urosubulatus* in feral as well as domesticated pigs of Mumbai region.

Prevalence rate of parasitic infection was found to be different in different country. Ananda *et al.* (2014) revealed 64.6% prevalence of parasitic infection in pigs of Shimoga region in Karnataka, India. While in the same country the parasitic prevalence 51.11% was reported in feral as well as domesticated pigs of Mumbai region (Dadas *et al*., 2016). While another study was carried out in four states of Northern Eastern Region of India and examination revealed overall prevalence of 37.77% in pigs (Laha *et al*., 2014). Whereas in rural farms of Chungcheongnam-do Korea, pigs were found to be infected by intestinal parasites with the prevalence of 73.5% and found three types of parasites namely *Balantidium coli*, *Ascaris suum* and *Entamoeba* sp. (Ismail *et al*., 2010). While in another research the examination revealed 96.4% prevalence of endoparasites in pigs from different areas of Mymensingh, Bangladesh and identified twelve types of endoparasites (Dey *et al*., 2014). Similarly, in the same country Azam *et al.* (2015) conducted three month cross-sectional study on gastrointestinal parasitism of pigs in two upazillas of Dinapur District and the investigation revealed 65% overall prevalence of gastrointestinal parasitic infections. However, Padilla and Ducusin (2017) reported 29.1% overall prevalence of gastrointestinal parasitic infection in pigs from 65 small holder farms in Cavite and Batangas, Philippines.

Single infection of GIT parasites has been reported highest than double infection from results obtained by Padilla and Ducusin, (2017) in Phillipines (single 23%, mixed 6.1%), Ananda *et al.* (2014) in Karnataka (single 52%, mixed 12.6%), Ismail *et al.* (2010) in Korea (single 63.2%, mixed 10.3%).

Among the reported gastrointestinal parasites in pigs, *Ascaris suum* has been reported with the highest prevalence from various studies (Laha *et al*., 2014 in India, Borkotoky *et al*., 2014 in India, Dadas *et al*., 2016 in India, Kaur *et al*., 2017 in India, Junhui *et al*., 2013 in China, Dey *et al*., 2014 in Bangladesh, Azam *et al*., 2015 in Bangladesh). While in few studies, *Balantidium coli* has been reported as the most prevalent parasite (Edmund *et al*., 2005 in Malaysia and Ismail *et al*., 2010 in Korea). However, Weng *et al.* (2005) reported *Trichuris suis* as the most prevalent parasite in intensive pigs farms in Guangdong Province, China. While the study carried out from free range pigs of shimoga city in India revealed *Strongyle* eggs was the most prevalent parasite (Ananda *et al*., 2014). Whereas, Matsubayashi *et al.* (2009) reported *Eimeria* sp. as the most prevalent parasite followed by *Trichuris suis*, *Ascaris suum* and *Metastrongylus* sp. in Osaka, Japan. In the same way another research revealed *Hyostrongylus rubidus* as the most prevalent parasites followed by *Trichuris suis*, *Stephanurus dentatus*, *Ascaris suum* and *Eimeria* sp. (Padilla and Ducusin, 2017).

Several studies have been conducted into the prevalence and economic importance of gastrointestinal parasites in pigs in some African countries. Several research work in African countries showed Intestinal parasites in pigs are still a major problem. Gastrointestinal helminthes including *Oesophagostomum dentatum, Trichuris suis, Ascaris suum, Oesophagostomum quadrispinulatum, Trichostrongylus axei*, *Strongyloides ransomi*, *Hyostrongylus rubidus* and *Physocephalus sexatus* have been identified in pigs raised under extensive production in Kenya, Ghana and Burkina Faso (Nganga *et al*., 2008, Permin *et al*., 1999, Tamboura *et al*., 2006). Semi-extensively managed pigs were reported to harbor *Taenia solium* in Nigeria (Gweba *et al*., 2010). In addition to helminths, extensively managed pigs were also reported to harbor intestinal protozoans including *Cryptosporidium* sp., *Giardia lamblia*, *Balantidium coli* and *Eimeria* sp. in developing countries (Usyal *et al*., 2009). Salifu *et al.* (1990) in Nigeria reported *Ascaris suum* (53.1%), *Hyostrongylus rubidus* (13.1%), *Strongyloides ransomi* (87.7%), *Ancyclostoma duodenale* (83.2%), *Oesophagostomum dentatum* (50%), *Metastrongylus salmi* (3.7%) and *Eimeria* sp. (3.6%). Similarly, Jarvis *et al.* (2007) investigated viscera of one hundred wild boars (*Sus scrofa)* and obtained seven helminth species namely, *Metastrongylus pudendotectus, M. salmi, M. elongatus, Ascaris suum, Trichuris suis, Dicrocoelium dendriticum* and *Taenia hydatigena* larva. In a survey conducted by Nwoha and Ekwurike (2011) reported *Globocephalus* sp., *Fasciolopsis* sp., *Ascarops* sp., *Stephanurus* sp., *Oesophagostomum* sp., *Trichuris trichuria* and *Necator* sp. in Umuahia city of Abia state.

Prevalence rate of parasitic infection in pigs was found to be different in different country of African continent. Sowemimo *et al.* (2012) showed an overall prevalence of 35.8% and identified five types of parasites namely *Trichuris suis* (12.2%), *Ascaris suum* (11.1%), human hookworm (5.9%), *S.dentatus* (1.1%) and *Isospora suis* (6.3%) the only protozoan. However, Nganga *et al.* (2008) reported 67.8% prevalence of gastrointestinal parasites with 31.3% mixed infection and 36.52% single infection and detected parasites were *Oesophagostomum dentatum* (39.1%), *Trichuris suis* (32.2%), *Ascaris suum* (28.7%), *Oesophagostomum quadrispinulatum* (14.8%), *Trichostrongylus colubriformis* (10.4%), *Trichostrongylus axei* (4.3%), *Strongyloides ransomi* (4.3%), *Hyostrongylus rubidus* (1.7%), *Ascarops strongylina* (1.7%) and *Physocephalus sexalutus* (0.9%) in pigs (61 growers and 54 adult) of Kenya. While another study carried out by Tamboura *et al.* (2006) found 91% to be infected and reported six different helminthes species namely, *Ascaris suum* (40%), *Strongyloides ransomi* (21%), *Oesophagostomum* sp. (18%), *Hyostrongylus rubidus* (11%), *Globocephalus* sp. (10%) and

Trichuris suis (1%). However, study carried out by Obonyo *et al.* (2012) examined 372 faecal samples of free range pigs in Homabay District, Kenya and found that the prevalence was 83%. The identified parasites were *Strongyles* (75%), *Strongyloides* sp. (26.6%), *Trichuris* sp. (7.8%), *Ascaris* sp. (5.4%), *Metastrongylus* sp. (0.3%), *Oesophagostomum* sp. (74%), *Hyostrongylus rubidus* (22%) and *Trichostrongylus* sp. (4%).

In a study carried out by Waiswa *et al.* 2007 reported 94.8% overall prevalence of gastrointestinal parasitic infection and the endoparasitic infections were recorded either as single species infection (19.7%) or mixed infections (80.3%) in pigs from South Eastern Uganda. Similarly, Tomass *et al.* (2013) revealed 27.3% prevalence of parasitic infection in extensively managed pigs in Mekelle and urban areas of Southern zone of Tigray region, Northern Ethiopia. However, cross-sectional study carried out by Geresu *et al.* (2015) reported 61.8% overall prevalence of gastrointestinal parasites and identified six GIT parasites namely, *Strongyloides* sp. (16.2%), *Ascaris suum* (12.6%), *Trichuris suis* (6.9%), *Oesophagostomum* sp. (3.9%), *Coccidia* sp. (11.8%) and *Fasciola hepatica* (10.5%) in pigs slaughtered at Addis Ababa Abattoirs Enterprise, Ethiopia. The study had also revealed that about 11.28% and 37.69% pigs had harbored mixed and single infection, respectively. Another study carried out by Jufare *et al.* (2015) reported 25% overall prevalence of GIT parasites and revealed the four different gastrointestinal parasites namely, *Coccidia* (12%), *Strongyles* (5.2%), *Ascaris suum* (4.9%) and *Trichuris suis* (2.9%). Nonga and Paulo (2015) observed parasitic infection in 83% of slaughter pigs at sanawari slaughter slab in Arusha city and recorded two types of parasites, helmiths (79%) and Coccidia (19%). Similarly, Atawalna *et al.* (2016) identified four parasites, *Coccidia* sp. (14.5%), *Strongyle* sp. (11%), *Ascaris suum* (2%) and *Trichuris suis* (0.5%).

Among the reported gastrointestinal parasites in pigs of African continent, *Ascaris suum* had been reported with the highest prevalence from various studies (Salifu *et al*., 1990, Tamboura *et al*., 2006, Tomass *et al*., 2013, Bernard *et al*., 2015). While the study conducted by Nonga and Paulo (2015) reported *Strongyles* eggs as the most prevalent in pigs. However, *Strongyloides* sp. has been reported with the highest prevalence from studies by Geresu *et al.* 2015. According to the study carried out by Nganga *et al.* (2008) revealed *Oesophagostomum* sp. as the most prevalent species in adult pigs while *Trichuris suis* as the most prevalent species in growers. However, Sowemimo *et al.* (2012) reported *Trichuris suis* having a highest prevalence in faecal samples of pigs in Ibadan, South west Nigeria. Whereas, Coccidia has been reported as the most prevalent in study carried out by Jufare *et al.* (2015) in pigs from two privately owned intensive farms in Bishoftu, Ethiopia.

Some of the studies has been found which focuses upon a Cysticercosis and Taeniasis of porcine origin. Githigia *et al.* (2005) studied the prevalence of *Cysticercus cellulosae* and the risk factors by lingual examination while Mutua *et al.* (2007) estimated the prevalence of palpable lingual cysts at 9.8% from 316 randomly selected small scale pig farmers in Western Kenya as a possible indicator of porcine cysticercosis. Similarly, Zirintunda and Ekou (2015) examined 178 pigs to establish the prevalence of porcine cysticercosis in free-ranging pigs delivered to slaughter points in Arapai, Soroti district, Uganda and reported a prevalence of 18%.

Some of the studies has been carried out regarding intestinal parasitic infection in pigs of European countries. Pilarczyk *et al.* (2004) reported four coccidian species and four nematode species, namely *E. deblecki*, *E. suis*, *E. perminuta*, *E. scabra*, *Oesophagostomum dentatum*, *Metastrongylus* sp., *Trichuris suis* and *Ascaris suum* respectively. However, Mossini (2002) examined 232 visceral sample of pig in Italy and found 52% were infected by *Ascaris suum.* While the study carried out by Karamon *et al.* (2007) demonstrated the high prevalence of *Isospora suis* in suckling piglets on the large swine farms in Poland. Similarly, Weiler *et al.* (2001) reported the prevalence of *Isospora suis* and *Cryptosporidium parvum* in suckling and weaned piglets from 24 farms in Southern Germany. Also, the survey conducted by Mundt *et al.* (2005) in 324 farms of Germany, Austria and Switzerland reported 93.5% with *Isospora suis* in piglets.

2.2 In national context

Nepal, being a developing country depends on agriculture and animal husbandry for its bulk of economy. Stating that, pig rearing is becoming an important live stock business in Nepal for meat as well as for leather. Pig farming and marketing have increased dramatically in the country due to increased consumer demand for pork. Besides this, pigs are the intermediate hosts of the cestode parasite *Taenia solium* in which the metacestode stages develop in different parts of the muscle forming cysticercus cellulosae. Gastrointestinal parasitic infections in Pig's cause economic losses due to condemnation of liver, reduce growth rate and feed conversion (Stephenson *et al*., 1980 and Hale *et al*., 1985). Practically very little work has been carried out on the topic related to pigs in Nepal. It appears that the literatures regarding gastrointestinal parasites of pigs are very much scanty. In a national context, a few researches in pigs have been done and similar such type of works has been performed by different authors at different parts of Nepal.

Sharma (2006) surveyed about 437 households and pig farming area in 3VDCs in syanja district. He found that the contamination of soil and water contributes greatly for the parasitic infestation in pigs. Most of the pigs were kept inside the house and were fed on kitchen wastes and excreta which was the important factor that was co-related with the high prevalence of parasitic infestations like Taeniasis in pigs and humans. However, Joshi (1991) observed *Taenia* cysts in pig meat slaughtered in Kangeswari, Kathmandu. Similarly, Joshi *et al.* (2001) examined 250 slaughtered pigs for meat purpose, in various localities of Kathmandu Metropolitan City and Dharan Municipality for *Taenia* cyst to study the awareness of taeniasis/cysticercosis and 34

(13.6%) were found positive for cysticercosis. The result also revealed that sexwise prevalence rate of infection was 8.77% in male pigs and 24.05% in female pigs. Similarly, Sapkota (2008) conducted a cross-sectional study to find the prevalence of porcine cysticercosis in slaughtered pigs sampled from four major slaughter slabs in Kathmandu valley of Nepal and found prevalence of 0.99%. However, another study conducted by Shakya (2009) found 4% positive for *Taenia* cyst in 150 pigs slaughtered for meat in Kirtipur municipality and also study revealed that females (4.44%) were found to be more infected than the males (3.33%). Similarly, Chaulagain *et al.* (2017) carried out cross-sectional study on prevalence of porcine cysticercosis among 384 pigs by using serum samples in Kathmandu valley and found 8.59% positive for cysticercosis. However, Rana *et al.* (2016) examined 400 samples to determine the prevalence of cysticercosis of swine in Nepal and revealed that 46(11.5%) swine were infected with *Taenia solium* and *Ascaris suum*.

Shrestha *et al.* (2008) carried out an study for the prevalence of Brucellosis in pigs where 153 serum samples of pigs from Itahari were tested by using the Brewer Diagnostic Card and 7.18% were found to be positive for brucellosis. Similarly, another study carried out by Rana (2005) revealed 21.58% of the total serum samples tested positive for brucellosis in 190 slaughtered pigs in Koteshwor and Talcchikhel areas in the Kathmandu valley. While the study conducted by Poudel *et al.* (2014) reported 13.59% positive for brucellosis in pigs in 6 VDCs of Rupandehi district. However, the study carried out by Sharma *et al.* (2017) to determine the prevalence of brucellosis in pigs of Bhaktapur, Kavre and Banke districts of Nepal found 3.90% of samples to have sero-positivity towards brucellosis by using ELISA test.

Karn (2007) conducted an epidemiological cross-sectional study of *Trichinella* sp. in pigs in CDR, Nepal using pepsin digestion and ELISA serology and revealed that *Trichinella* sp. in this region had a low detection level with a very low prevalence. Similarly, Devleesschauwer *et al.* (2013) performed a cross-sectional study in the Kathmandu valley to determine the infection status of slaughtered pigs with regard to three of the most important parasites transmitted through pork consumption and reported 0.1% positive for *Trichinella* infection, 13.8% for *Taenia solium* cysticercosis and 11.7% for *Toxoplasma* infection. However, the study carried out by Kathayat *et al.* (2015) to find out the seroprevalence of *Trichinella* sp. in pigs of eastern and Mid-western regions of Nepal revealed prevalence rate of 3.8%.

Thapa Shrestha *et al.* (2014) carried out comparative study of parasitic infection in pig population from two climatic zones; Kathmandu valley and Terai. The study revealed that 96% samples were found to contain intestinal parasites while all samples from Terai were found to be infected. The parasites identified were *Blastocystis hominis* (81.33%), *Eimeria* sp. (76.67%), *Isospora* sp., *Hookworm* (10%), *Ascaris suum* (12%).

3. MATERIALS AND METHODS

3.1 Study Area

Jhapa is an inner-terai district in Mechi zone of the Eastern Development Region of Nepal. Under the new federal division, the district lies in Province No. 1. The district covers 1,606 square kilometers. Geographically, the district is located in the latitude of 26^o 20’ to 26^o 50’N and the longitude of 87^o 39’ to 88^o 12’ E. The 2011 Nepal census puts the total population of the district at 812,650. It borders with Ilam districts in the north, West Bengal and Bihar state of India in the east, Morang districts in the West, Bihar state of India in the south. Jhapa lies in tropical region and is mostly affected by monsoon effect. High temperature up to 42 degree Celsius can be observed during summer time whereas minimum temperature of 5 degree Celsius occurs during winter time. Minimum rainfall occurs in winter season and 80% of total rainfall occurs during April to September. Average annual rainfall varies in between 1840 mm to 2200 mm. The head-quarter of the district is Bhadrapur Municipality. It is divided into 42 village development committees and 7 municipalities. The study was conducted in two different locations of Jalthal VDC of Jhapa district in the Eastern Development Region of Nepal.

Jalthal is a village development committee in Jhapa District in the Mechi Zone of South eastern Nepal. Its geographic location co-ordinates 26.50^o N to 88.01^o E. At the time of the 1991 Nepal census it had a population of 13,132. People of different cast were found living together and sharing their cultures and supporting each other. Majority of people living in these locations are Meche, Rai, Limbu, Magar, Tamang, Bahun and chhetri. In the study area it was noticed that pigs were kept under poor unhygienic conditions such as dirty isolated lots or pens and also poor quality of husbandry practices by their rearers was seen during the study which encourages helminthosis.

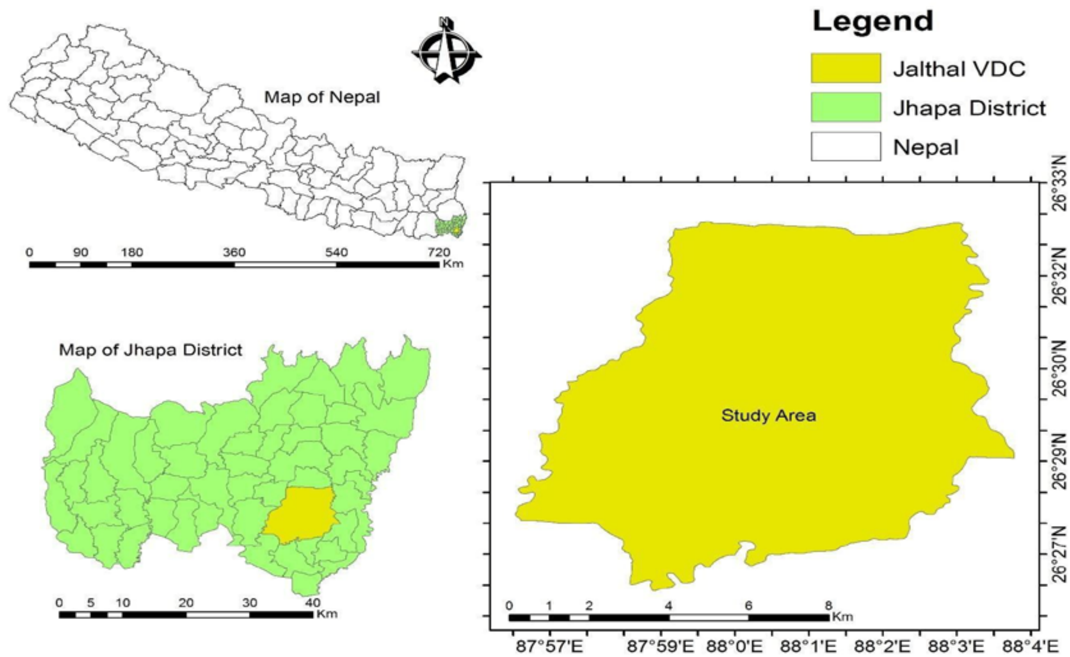

Figure 1: Map of Jhapa district showing study area

3.2 Materials

During the study the materials used have been listed below:

3.2.1 Laboratory materials

Beakers, Mortar/Pistil, Glass rod, Slides, coverslips, Conical flask, Dropper, Toothpicks, Electric microscope, Gloves and masks, stage micrometer, ocular micrometer, , Centrifuge machine, Tea strainer, Test tubes.

3.2.2 Field materials

Sterile vials, Gloves, Camera.

3.2.3 Chemicals

Potassium dichromate, Distilled water, Normal saline, Iodine solution, Saturated salt solution, Methylene blue.

3.3 Methods

3.3.1 Study Period

The study was carried out from May 2017 to September 2017.

3.3.2 Faecal samples collection and preservation

Fresh faecal samples of the pigs were collected in wide mouthed, clean, leak-proof sterile vials. Immediately, after collection 2.5 potassium dichromate solution was added in the vials containing faeces for the preservation of the parasite present in the faeces. Morning samples was collected in order to get good results.

3.3.3 Sample size

A total of 150 faecal samples of pigs were collected from two different locations of Jalthal VDC of Jhapa. Out of 150 samples, 75 samples were collected from Kali-jhoda and 75 samples from Meche gaun.

3.4 Laboratory Examination

The collected samples were brought and examined in the laboratory of the Central Department of Zoology, Kirtipur.

3.4.1 Unstained smear preparation of faeces

A drop of (0.85%) normal saline was placed at the centre of a clean glass slide and a small portion of the stool was picked up with the help of an applicator stick and smear was made in the drop. It was then covered with a coverslip (Soulsby, 1982).

3.4.2 Stained smear preparation of faeces

A drop of 1% iodine was placed at the centre of a clean glass slide and a small portion of stool specimen was emulsified in the drop using applicator sticks. The smear was then covered with a coverslip (Soulsby, 1982).

3.4.3 Concentration Method

There are two types of concentration procedures, sedimentation and floatation, both of which are designed to separate protozoan organisms and helminth eggs and larvae from faecal debris. Eggs/cysts were often low number in faeces that they were difficult to be detected in direct smear. Therefore, faecal samples were examined using flotation and sedimentation techniques.

3.4.3.1 Floatation technique

This technique is widely used for detecting eggs of nematodes, cestodes and Coccidia oocysts in the faeces. Approximately, 3gm of each faecal sample was taken in a beaker and added 15ml of water then the sample was mixed using mortar and pistil. The resulting faecal suspension was poured through a tea strainer into the beaker. The filtrate solution was poured from the beaker into a centrifuge tube of 15ml and centrifuged at 1000 rpm for 5 minutes. The solution was replaced with saturated sodium chloride solution and again centrifuged. After centrifuge more saturated sodium chloride solution was added to develop convex meniscus at the top of the tube and one drop of methylene blue (to stain) was added and coverslip was placed over it and let to stand for 20 minutes. Then the coverslip was carefully lifted from the test tube and immediately placed on a glass slide and examined at 10X and 40X. Photographs of cyst, eggs and parasites were taken and identified based on egg's color, shape, and size (Soulsby, 1982).

3.4.3.2 Sedimentation technique

To detect helminth eggs which do not float well in the sodium chloride solution such as *Fasciola* sp., simple sedimentation technique was carried out. Approximately, 3g of faeces was taken in a beaker and mixed with 15ml of water. The mixture was then sieved through a tea strainer into a beaker, transferred into a centrifuge tube and centrifuged at 2000 rotation per minutes for 15 minutes. The tube was taken out and suspension was removed with the help of pipette. The sediment was taken out from the tube with the help of pipette and placed in the slide, The specimen was stained with Iodine wet mount's solution and examined under the microscope at 10X and 40X (soulsby, 1982).

3.5 Eggs, cysts and larva size measurement

By using ocular and stage micrometer, the length and breadth of parasites (eggs, cysts and larvae) was measured with calibration.

3.6 Eggs, cysts and larva size identification

The identification of eggs, cyst and larva of different parasites was done based on structural and morphometric criteria by using books of Soulsby (1982), and other published and unpublished article and also from internet sources.

3.7 Intensity of infection

Intensity of parasitic infection has been calculated based upon the number of eggs/oocyst and larvae found per field. The intensity of infection of gastrointestinal parasites was categorized into 4 groups, i.e. light infection, mild infection, moderate infection and heavy infection. Light infection was determined by the occurrence of less than 2 egg/cyst/larva of the same species per field. Similarly mild, moderate and heavy parasitic infection were determined by the occurrence of 2-4 egg/cyst/larva, 4-6egg/cyst/larva, 6 or more egg/cyst/larva of the same species per field respectively.

3.8 Data analysis

The collected data were coded and entered into Microsoft Excel spread sheet. Data were statistically analyzed using Pearson’s Chi-square test with Yates’ continuity correction, performed by “R”, version 3.3.1 software packages. Percentage was used to calculate prevalence. Data were statistically analyzed using Chi-square. In all cases 95% confidence interval (CI) and $p<0.05$ was considered for statistically significant difference.

Photographs during field and lab work

Photo 1: Pig of study area

Photo 2: Collecting faecal sample

Photo 3: Faecal sample

Photo 4: Faecal sample preservation

Photo 5: Samples in floatation and sedimentation techniques

Photo 6: Lab work at T.U., Kirtipur

Photo 7: Microscopic observation of slide

4. RESULTS

4.1 General prevalence of gastrointestinal parasites in pigs

During the study period, a total of 150 faecal samples were collected from two different locations (Kali-jhoda and Meche gaun) of Jalthal VDC of Jhapa and microscopically examined by using floatation and sedimentation technique. Out of 150 samples examined, 136 samples (90.67%) were found to be positive for gastrointestinal parasites while 14 samples (9.33%) were found to be negative.

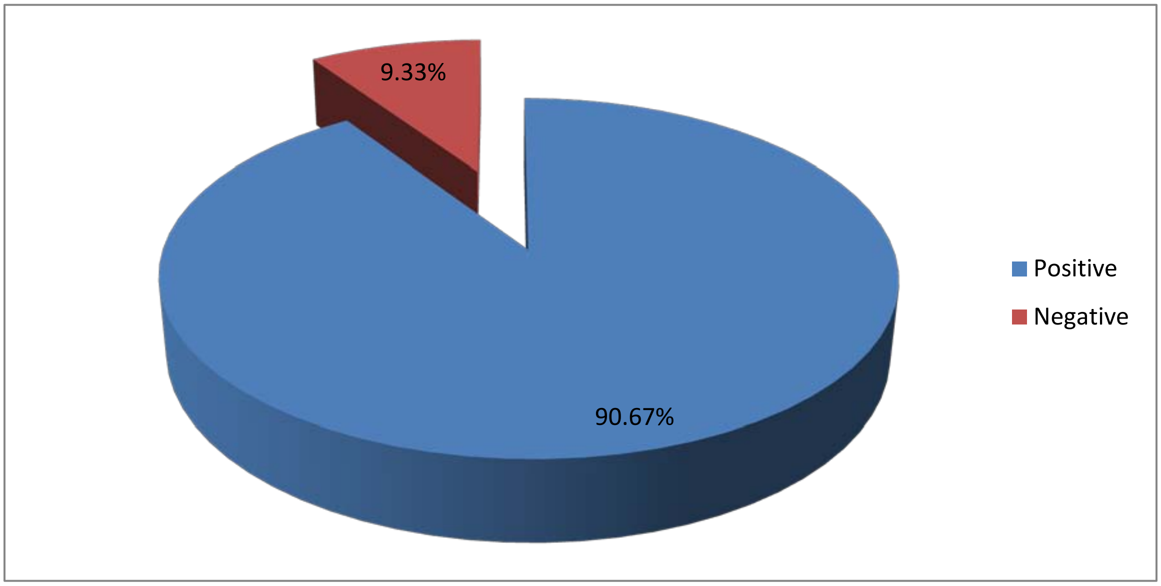

Figure 2: General prevalence of gastrointestinal parasites in pigs

4.2 Prevalence of specific GI parasites in Pigs :

Among 150 samples examined, ten genus of parasites were identified including protozoans and helminth parasites. Among protozoan parasites, *Eimeria* sp. 38 (25.33%), *Isospora* sp. 2 (1.33%) and *Balantidium coli* 14 (9.33%) were found. Similarly, among helminthes one genus of trematode i.e. *Fasciola* sp. 29 (19.33%) and six genus of nematodes i.e. *Ascaris* sp. 69 (46%), *Strongyloides* sp. 58 (38.67%), *Ancyclostoma* sp. 49 (32.67%), *Trichuris* sp. 13 (8.67%), *Trichostrongylus* sp. 3 (2%) and *Metastrongylus* sp. 2 (1.33%) were observed in the study. Among the identified parasites, *Ascaris* sp. (46%) showed the highest prevalence followed by *Strongyloides* sp. (38.67%) *Ancyclostoma* sp. (32.67%) *Eimeria* sp. (25.33%), *Fasciola* sp.

(19.33%), *Balantidium coli* (9.33%), *Trichuris* sp. (8.67%), *Trichostrongylus* sp. (2%), while *Isospora* sp. (1.33%) and *Metastrongylus* sp. (1.33%) showed the lowest prevalence. There was insignificant difference in the prevalence of specific GI parasites in pigs (χ^2=163.56, P>0.05).

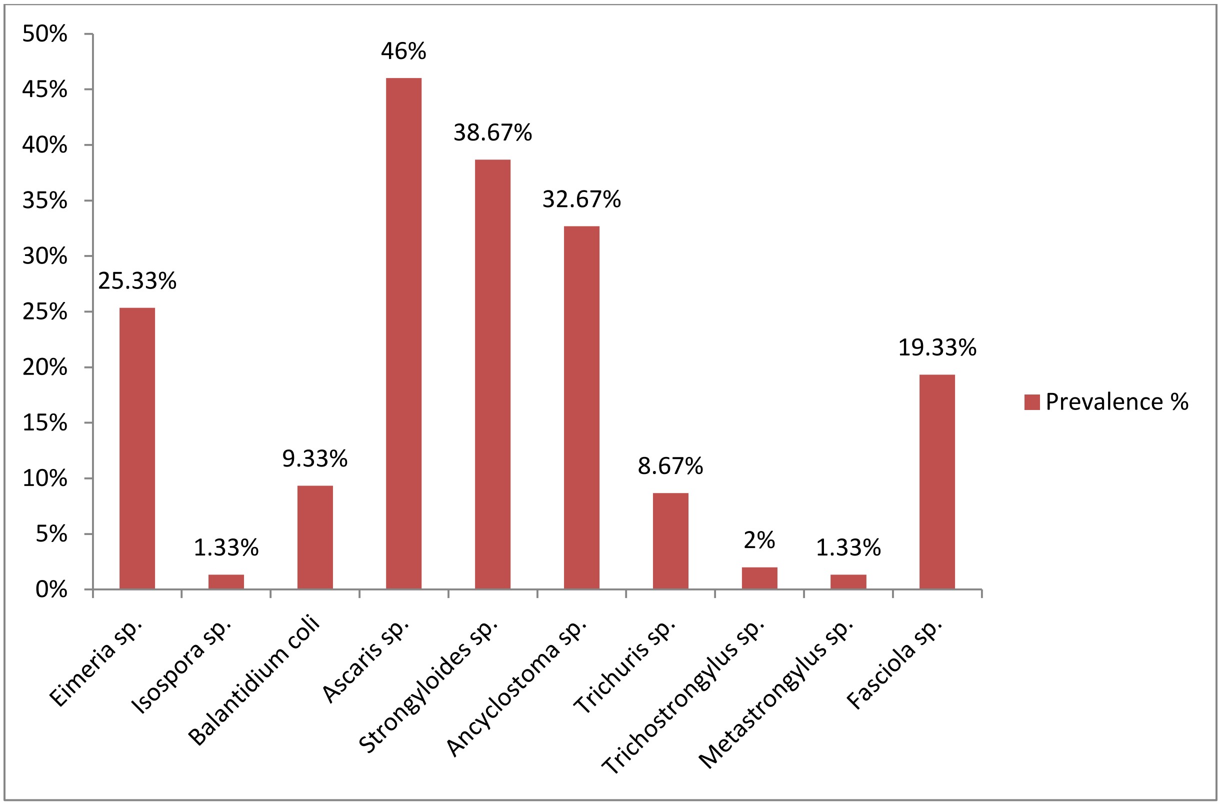

Figure 3 : Prevalence of specific gastrointestinal parasites in pigs.

4.2.1 Class-wise prevalence of gastrointestinal parasites in pigs:

Out of 136 total positive samples, 38 samples (27.94%) were found to be positive for protozoans whereas 69 (50.74%) and 29 (21.32%) were found to be positive for nematodes and trematodes respectively. There was statistically significant difference in the prevalence of GI parasitic infection among protozoans and helminthes (χ^2=8.1053, P<0.05).

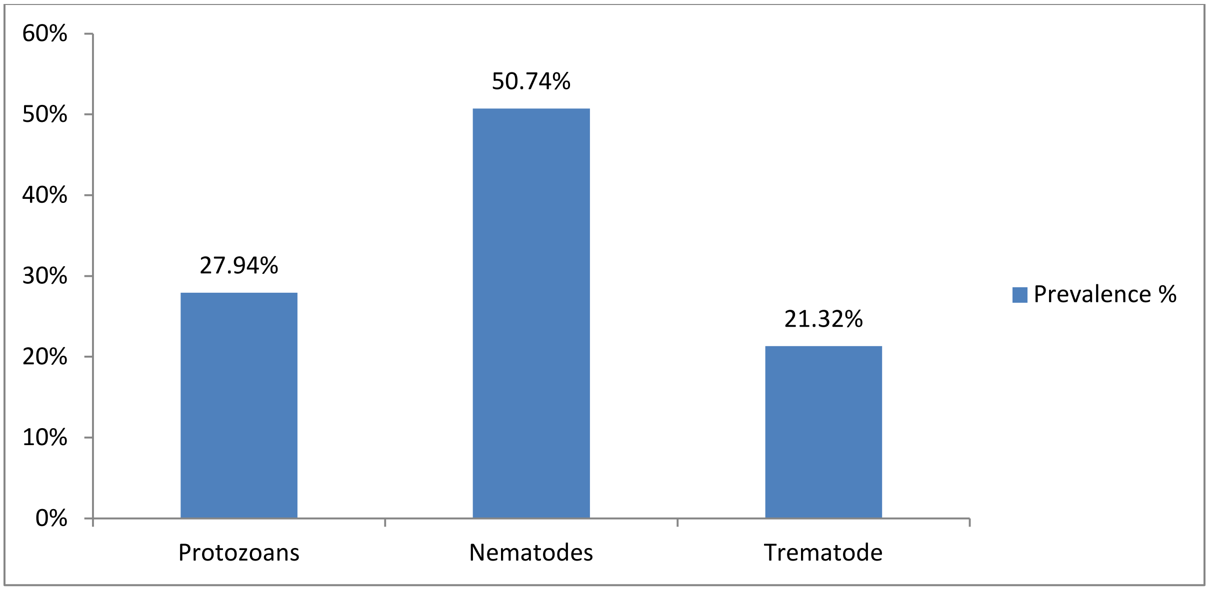

Figure 4: Class-wise prevalence of GI parasites in pigs.

4.3 Location-wise prevalence of gastrointestinal parasites in pigs :

Out of 150 samples examined, 75 samples were taken from Kali-jhoda and 75 samples were taken from Meche gaun of Jalthal VDC. Among the examined samples, highest prevalence of GI parasites was found in Kali-jhoda (93.33%) while Meche gaun (88%) showed the lowest prevalence. Statistically, the difference in the prevalence of GI parasitic infection among the study area was found to be insignificant (χ^2=0.0169, P>0.05).

Table 1 : Location-wise prevalence of GI parasites in pigs :

Location	Number examined	No. of positive samples	Prevalence (%)
Kali-jhoda	75	70	93.33%
Meche gaun	75	66	88%

4.4 Concurrency and intensity of gastrointestinal parasites

4.4.1 Single and mixed infection

During the study different type of parasitic infections were encountered in pigs. Among 136 (90.67%) positive samples, 107 (78.68%) samples were found to have mixed infection with 2 or more species and 29 (21.32%) samples were found to have single infection with only one species in each microscopic field. There was no statistical difference in the prevalence of single and mixed infections (χ^2=29.776, P>0.05).

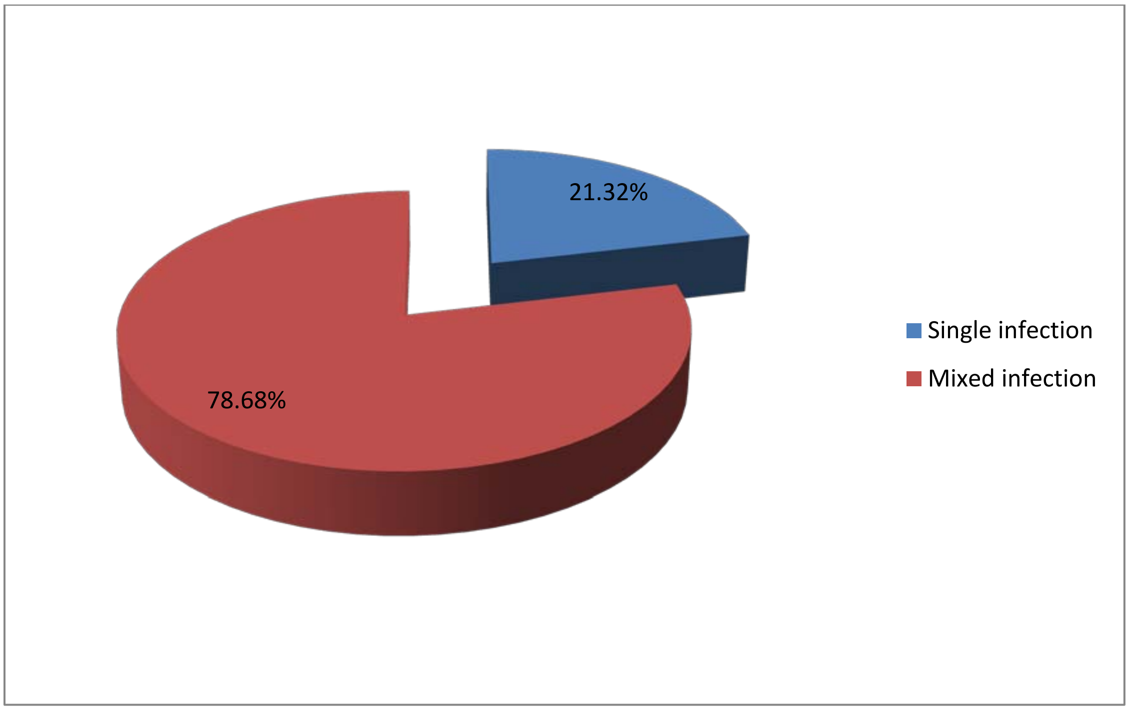

Figure 5 : Single and Mixed infection

4.4.2 Intensity of gastrointestinal parasites in pigs

Intensity of parasitic infection has been calculated based upon the number of eggs/oocyst/cysts and larvae found per microscopic field. Among protozoans, the high intensity of lightly and mildly infected cases were observed in *Eimeria* sp. over *Balantidium coli* and *Isospora* sp. while moderately and heavily infected cases was not found. Similarly, among helminths the high intensity of lightly infected cases was found in *Strongyloides* sp. and that of mildly and moderately were found in *Ascaris* sp. while high intensity of heavily infected cases was found in *Fasciola* sp.

Table 2: Intensity of parasitic infection in pigs

S.N	Class	Parasites	Light (+)	Mild (++)	Moderate (+++)	Heavy (++++)
1	Protozoan	*Eimeria* sp.	28 (18.67%)	10 (6.67%)		
		Isospora sp.	2 (1.33%)	-	-	-
		Balantidium coli	9 (6%)	5 (3.33%)	-	-
2	Trematode	*Fasciola* sp.	17 (11.33%)	7 (4.67%)	2 (1.33%)	3 (2%)
3	Nematode	*Ascaris* sp.	34 (22.67%)	21 (14%)	12 (8%	2 (1.33%)
		Strongyloides sp.	38 (25.33%)	16 (10.67%)	4 (2.67%)	-
		Ancyclostoma sp.	28 (18.67%)	15 (10%)	5 (3.33%)	1 (0.67%)
		Trichuris sp.	7 (4.67%)	5 (3.33%)	1 (0.67%)	-
		Trichostrongylus sp.	3 (2%)	-	-	-
		Metastrongylus sp.	2 (1.33%	-	-	-

Note - The figure in the column is the numbers of samples

+ = less than 2 egg / cyst / larva (light infection)
++ = 2-4 egg / cyst / larva (mild infection)
+++ = 4-6 egg / cyst / larva (moderate infection)
++++ = 6 or more egg / cyst / larva (heavy infection)

Photographs of identified gastrointestinal parasites

Eggs of Protozoan observed (10x X 40x)

	
Photo 8 : Oocyst of *Eimeria* sp. (25μm× 17μm) (Annex-1)	Photo 9 : Oocyst of *Eimeria* sp. (28μm×20μm) (Annex-1)

	
Photo 10 : Oocyst of *Isospora* sp. (20 μm) (Annex-1)	Photo 11 : Cyst of *Balantidium coli* (40μm) (Annex-1)

Nematodes eggs (10x X 40x)

	
Photo 12 : Egg of *Ascaris* sp. (65 × 50μm) (Annex-1)	Photo 13 : Egg of *Strongyloides* sp.(57×33 μm) (Annex-1)

	
Photo14 : Egg of *Metastrongylus* sp.(50×40μm) (Annex-1)	Photo 15 : Egg of *Trichuris* sp. (58×22μm) (Annex-1)

Photo 16 : Egg of *Trichostrongylus* sp. (85×40μm) (Annex-1)

Photo 17 : Egg of *Ancyclostoma* sp.(70×38μm) (Annex-1)

Trematode eggs (10x X 40x)

Photo 18 : Egg of *Fasciola* sp.(150×80μm) (Annex-1)

Photo 19 : Egg of *Fasciola* sp.(145×78μm) (Annex-1)

5. Discussion

Infection of pigs with gastrointestinal parasites is widely reported from all corners of the world and shown to be influenced by the type of pig management practiced. In context of Nepal different types of protozoan and helminths have been reported in pigs. Gastrointestinal parasite of pigs can result in loss of appetite, poor growth rate, poor feed conversion efficiency and potentiation of other pathogens or even death (Stewart and Hoyt, 2006). These consequently lead to significant economic losses like decreased litter size, poor growth rate, reduced weight gain and their potential to infect humans have recently become a major issue among the public because of recent out breaks of water-borne parasitic diseases such as Giardiasis and cryptosporidiosis (Olson and Guselle, 2000).

The present study was carried out to determine the prevalence of gastrointestinal parasites in domestic pigs of Jalthal VDC, Jhapa, Nepal. A total of 150 faecal samples were collected from two different locations (Kali-Jhoda and Meche Gaun) of Jathal VDC of Jhapa, Nepal. The collected faecal samples were microscopically examined by differentiation floatation and sedimentation techniques for isolation of parasitic eggs and oocysts. The study revealed 90.67% found to be infected by protozoan or helminth parasites or both including 10 genera. Among 136 positive samples examined, 38 samples (27.94%), 69 samples (50.74%) and 29 samples (21.32%) were found to be positive for protozoan, nematode and trematode infections respectively. Statistically, the difference in the prevalence of GI parasitic infection among protozoans and helminthes were found to be significant (χ^2=8.1053, P<0.05).

The present study established 10 genera of GIT parasites namely *Eimeria* sp., *Balantidium coli*., *Isospora* sp., *Ascaris* sp., *Strongyloides sp.*, *Ancyclostoma* sp., *Trichostrongylus* sp., *Trichuris* sp., *Metastrongylus* sp. and *Fasciola* sp. This study revealed that *Ascaris* sp. (46%) showed the highest prevalence followed by *Strongyloides* sp. (38.67%) *Ancyclostoma* sp. (32.67%), *Eimeria* sp. (25.33%), *Fasciola* sp. (19.33%), and *Balantidium coli* (9.33%), *Trichuris* sp. (8.67%), *Trichostrongylus* sp. (2%), while *Isospora* sp. (1.33%) and *Metastrongylus sp.* (1.33%) showed the lowest prevalence. Among the intestinal protozoan parasites identified, *Eimeria sp.* (25.33%) showed the highest prevalence while *Isospora* sp.(1.33%) showed the lowest prevalence. Similarly, among nematodes, *Ascaris* sp. (46%) showed the highest prevalence while *Metastrongylus* sp. (1.33%) showed the lowest prevalence. Only one species of trematode i.e. *Fasciola* sp. was identified with the prevalence of 19.33%. The study had also revealed that among 136 (90.67%) positive samples, 107 (78.68%) samples were found to have mixed infection with 2 or more species and 29 (21.32%) samples were found to have single infection with only one species in each microscopic field. Statistically, the differences in the prevalence of single and mixed infections were found to be insignificant. (χ^2=29.776, P>0.05). Location wise, the highest prevalence (93.33%) was found in Kali-jhoda while Meche gaun (88%) showed the lowest prevalence. Statistically, the difference in the prevalence of GI parasitic infection among the study area was found to be insignificant (χ^2=0.0169, P>0.05).

The present study revealed the overall prevalence of gastrointestinal parasites was found to be 90.67%. Similar results were reported by Tamboura *et al.* (2006) in Burkina Faso (91%) Waiswa *et al.* (2007) in South Eastern Uganda (94.8%), Dey *et al.* (2014) in Mymensingh (96.4%). The present study is nearly similar with the findings of Nonga and Paulo (2015) in Arusha city (83%) and Jarvis *et al.* (2007) in Western Estonia (82%). The present result is higher than Ismail *et al.* (2010) in Korea (73.5%), Nganga *et al.* (2008) in Kenya (67.8%), Azam *et al.* (2015) in Bangladesh (65%), Geresu *et al.* (2015) in Ethiopia (61.8%) whereas highly differs from findings of Dadas *et al.* (2016) in Mumbai (51.11%), Laha *et al.* (2014) in India (37.77%), Sowemino *et al.* (2012) in Nigeria (35.8%), Bernard *et al.* (2015) in Pankshin Urban (32%), Padilla and Ducusin (2017) in Philippines (29.11%), Atawalna *et al.* (2016) in Ghana (28%), Tomass *et al.* (2013) in Ethiopia and Jufare *et al.* (2015) in Ethiopia (25%). The differences in the prevalence observed between the present study and aforementioned studies could be due to poor management system and lack of veterinary care. Traditional system of management in which pigs are allowed to roam freely or are kept under poor hygienic conditions such as those obtainable in dirty isolated lots or pens encourages helminthosis (Shima *et al*., 2014).

This study revealed that *Ascaris* sp. (46%) was the most prevalent parasite. This is similar with the findings of past studies of Tamboura *et al.* (2006) in Burkina Faso (40%), Dey *et al.* (2014) in Mymensingh (50.9%), Laha *et al.* (2014) in India (65.46%), Dadas *et al.* (2016) in Mumbai (32.59%) whereas Tomass *et al.* (2013) in Northern Ethiopia (25.9%) and Junhui *et al.* (2013) in China (10.23%) are slightly lower than present findings. The highest prevalence of *Ascaris* sp. in the present study might be associated with farm management systems and access of free roaming of pig in the environment which facilitates ingestion of thick shelled eggs of *Ascaris* sp. These thick-shelled eggs are resistant to adverse environmental factors as well as chemicals and can maintain infectivity for long periods of time (Roepstorff and Nansen, 1998).

The prevalence of 38.67% recorded for *strongyloides* sp. in this study was the second most prevalent parasite after *Ascaris* sp. which highly differs from findings of Salifu *et al.* (1990) in Nigeria (87.7%). The present study is nearly similar with the findings of past studies of Dey *et al.* (2014) in Mymensingh (29.1%), Tamboura *et al.* (2006) in Burkina Faso (21%), Azam *et al.* (2015) in Bangladesh (20%), Nonga and Paulo (2015) in Arusha city (15.0%) while higher than than the findings of past studies of Geresu *et al.* (2015) in Ethiopia (16.2%), Laha *et al.* (2014) in India (13.06%), Junhui *et al.* (2013) in China (6.49%), Nganga *et al.* (2008) in Kenya (4.3%), Kaur *et al.* (2017) in India (4.5%) and Dadas *et al.* (2016) in India (0.74%). The differences in the prevalence could be due to the differences in climatic conditions, management systems and local circulating parasites in the locality. The survival of *strongyloides* larvae depends on the environmental temperature and moisture. The larvae of these species are susceptible to desiccation with the dry areas providing unfavourable environment for survival of *strongyloides* larvae (Marufu *et al*., 2008).

Previous studies by Tamboura *et al.* (2006) in Burkina Faso and Dey *et al.* (2014) in Mymensingh reported *Ancyclostoma* sp. with prevalence of 10% and 3.6% respectively which is lower than the report of the present study (32.67%). Contrary to this, from findings of Salifu *et al.* (1990) in Nigeria reported the prevalence of 83.2% which is higher than the result of this study. The aforementioned differences in the prevalence could be due to poor management system, differences in climatic conditions as this parasite is endemic in warm climate and poor sanitary condition.

The prevalence recorded for *Eimeria* sp. (25.33%) in the present study is lower than the findings of past studies of Matsubayashi *et al.* (2009) in Japan (40.3%), Dey *et al.* (2014) in Mymensingh (56.4%) and Laha *et al.* (2014) in India (34%) while highly differs from findings of Junhui *et al.* (2013) in China (6.35%), Karamon *et al.* (2007) in Poland (2.6%) and Tomass *et al.* (2013) in Northern Ethiopia (1.7%). The differences in the prevalence could be due to differences in the ecological factors between the origin of pigs, differences in management system and the ability of the cysts/oocysts to survive for long in the environment (kagira *et al*., 2002).

Fasciola sp. is the only trematode observed in this study with a prevalence of 19.33%, which is nearly similar with the past studies of Geresu *et al.* (2015) in Ethiopia (10.5%) and Dey *et al.* (2014) in Mymensingh (14.6%). However, lower than the report of Nwoha and Ekwurike (2011) in Abia state (50%) and higher than the report of Tomass *et al.* (2013) in Northern Ethiopia (1.8%). Factors such as presence of reservoir hosts, presence of snail intermediate hosts and ability of *Fasciola* sp. to colonise and adapt new hosts contribute for its spread in livestock in an area (Mas-coma *et al*., 2005).

The prevalence of *Balantidium coli* observed in this study was found to be 9.33%. The present study is nearly similar to the findings of past studies of Ananda *et al.* (2014) in Shimoga (6.6%). However, lower than the report of Dadas *et al.* (2016) in India (31.85%), Dey *et al.* (2014) in Mymensingh (40%) and highly differs from the findings of Weng *et al.* (2005) in China (47.2%), Edmund *et al.* (2005) in Malaysia (53.3%) and Ismail *et al.* (2010) in Korea (64.7%). Overcrowded pens or barns and poor sanitation practices are some of the factors that can lead to the spread of *Balantidium.* Therefore, differences in the prevalence could be due in the differences in management system, husbandry practices and sanitation measures and also due to differences in climatic conditions as this parasite is fairly rare in temperate areas.

The present result shows 8.67% of *Trichuris* sp. infection in faecal sample which was found to be similar with previous findings of Dey *et al.* (2014) in Mymensingh (9.1%), Dadas *et al.* (2016) in India (11.11%), Sowemimo *et al.* (2012) in Nigeria (12.2%), Borkotoky *et al.* (2014) in

Nagaland (6.25%), Laha *et al.* (2014) in India (16.66%), Edmund *et al.* (2005) in Malaysia (17.5%) and Geresu *et al.* (2015) in Ethiopia (12.6%). The present status of *Trichuris suis* infection is little bit higher than the previous findings of Weng *et al.* (2005) in China (5.2%), Azam *et al.* (2015) in Bangladesh (5%), Padilla and Ducusin (2017) in Phillipines (3.1%), Jufare *et al.* (2015) in Ethiopia (2.9%), Tamboura *et al.* (2006) in Burkina Faso (1%) and Atawalna *et al.* (2016) in Ghana (0.5%) whereas highly differs from previous findings of salifu *et al.* (1990) in Nigeria (47.2%) and Nganga *et al.* (2008) in Kenya (32.2%). The differences in the prevalence of *Trichuris* sp. could be due to the ability of the eggs to survive for long in the environment (Pittman *et al*., 2010).

The prevalence of *Trichostrongylus* sp. observed in this study was found to be 2% which is lower than the findings of previous studies of Nganga *et al.* (2008) in Kenya (10.4%). The prevalence of *Metastrongylus* sp. was found to be 1.33% which is little bit higher than the findings of previous studies of Salifu *et al.* (1990) in Nigeria (3.7%) and Nonga and Paulo (2015) in Arusha city (4%). The differences in the prevalence could be due to differences in the ecological factors and husbandry practices.

Isospora sp. is the coccidian parasite which was found at the rate of 1.33% in the present study which shows similar result with findings of Dadas *et al.* (2016) in India (1.48%). However, lower than findings of Sowemimo *et al.* (2012) in Nigeria (6.3%) and Dey *et al.* (2014) in Mymensingh (9.1%) while highly differs from Karamon *et al.* (2007) in Poland and Laha *et al.* (2014) in India (52.34%). The aforementioned differences in the prevalence could be due to differences in the ecological factors between the origin of pigs, differences in management system and the ability of the cysts/oocysts to survive for long in the environment (Kagira *et al*., 2002).

The present study revealed higher prevalence of GIT parasites in pigs originated from Kali-jhoda (93.33%) when compared to Meche gaun (88%). The reasons for the variations in GIT parasite prevalence among the origin of the pigs might be related to difference in management practice performed. There was insignificant association ($\chi^2 = 0.0169$; $P > 0.05$) in origin groups of Jalthal VDC which is similar with the results obtained by Geresu *et al.* (2015) with location.

The present study also revealed that there was higher mixed infection (78.68%) compared to single infection (21.32%). Similar result was found by Waiswa *et al.* (2007) in Uganda 80.3% in mixed infection and 19.7% in single infection. The present study is just opposite of the results obtained by Nganga *et al.* (2008) in Kenya (single 67.8%, mixed 31.3%) and Geresu *et al.* (2015) in Ethiopia (single 37.69%, mixed 11.28%).

6. CONCLUSION AND RECOMMENDATIONS

6.1 Conclusion

The present study was conducted in order to determine the prevalence of gastrointestinal parasites in pigs of Jalthal VDC of Jhapa. A total of 150 samples were collected from two different locations (Kali-jhoda and Meche gaun) of Jalthal VDC and microscopically examined by differentiation floatation and sedimentation techniques for isolation of parasitic eggs or oocysts. This study had revealed an overall prevalence of 90.67% in the study area which includes ten genus of gastrointestinal parasites namely *Eimeria* sp., *Balantidium coli*, *Isospora* sp., *Ascaris* sp., *Strongyloides* sp., *Ancyclostoma* sp., *Trichostrongylus* sp., *Trichuris* sp., *Metastrongylus* sp. and *Fasciola* sp. Among 136 positive samples, 38 samples (27.94%), 69 samples (50.74%) and 29 samples (21.32%) were found to be positive for protozoan, nematode and trematode infections respectively. Among the identified parasites, *Ascaris* sp. (46%) was the most prevalent parasite followed by *Strongyloides* sp. (38.67%) *Ancyclostoma* sp. (32.67%), *Eimeria* sp. (25.33%) *Fasciola* sp. (19.33%), and *Balantidium coli* (9.33%), *Trichuris* sp. (8.67%), *Trichostrongylus* sp. (2%), while *Isospora* sp. (1.33%) and *Metastrongylus sp.* (1.33%) showed the lowest prevalence. Single infection was found in 29 (21.32%) samples while multiple infections were observed in 107 (78.68%) samples. Location wise, the highest prevalence (93.33%) was found in Kali-jhoda while Meche gaun (88%) showed the lowest prevalence. Intensity of parasitic infection was found to be different in different faecal samples. Some samples were highly infected and some are lightly infected with parasites. Among protozoans, *Eimeria* sp. was found to be heavily infected while among helminthes, *Ascaris* sp., *Strongyloides* sp. and *Ancyclostoma* sp. respectively were found to be highly infected. From the study, it has been concluded that the gastrointestinal parasites in pigs in the study area are highly prevalent and infection of intestinal parasites is still an important factor that hinders the development of the pig farming industry. Therefore, the findings here would have important implications to control and prevent intestinal parasites infection in pigs in the study area.

6.2 Recommendations

- Identification of parasites was done on the basis of their morphological character. To know the exact parasites upto species level molecular identification is necessary.
- Formulation of appropriate and cost effective strategies to control GI parasitic infections in pigs is very much needed.
- Strategic anthelmintics drug treatment for helminthes should be provided under supervision of expert veterinarian.

- Further studies are required to study the possible impact of parasitic infestations of pigs on public health.

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ANNEX-1

Identified eggs and cysts of parasites found in pigs:

In the present study, the diameter of eggs/cysts of different gastrointestinal parasites were measured which is given below:

Identified eggs and cysts of parasites found in pigs :

Name of parasites	Range of diameter of eggs and cysts (μm)		Morphological Characters	Reference values (Soulsby, 1982)
	Length	Width		
Eimeria sp.	25	17	Oocysts are oval to ellipsoidal, sub-spherical and oocyst wall thin.	11-35μm ×13-25μm
Isospora sp.	20	-	Oocysts are sub-spherical, light yellow in colour and micropyle absent.	20-24 μm×18-21 μm
Balantidium coli	-	40	Cysts are ovoid, to spherical, faintly yellowish green in colour.	40-60 μm
Ascaris sp.	65	50	Oval, thick shells, brownish-yellow in colour, albuminous layer bears prominent projections.	50-75 μm×40-50 μm
Ancyclostoma sp.	70	38	Ovoid, thin membrane and contain 4-8 blastomeres.	56-75 μm×34-47 μm
Strongyloides sp.	57	33	Blunt ends and contain fully developed embryos or larva.	40-60 μm×26-40 μm
Trichuris sp.	58	22	Brown, barrel-shaped with transparent plug at either pole.	50-60 μm×21-25 μm
Trichostrongylus sp.	85	40	Irregular, ellipse dissimilar, kidney-shaped not very wide poles, one of which has more rounded than the other, dissimilar side-walls.	79-118 μm×31-52 μm
Metastrongylus sp.	50	40	Thick, rough shells and contain a fully developed embryo.	45-60 μm×38-42 μm
Fasciola sp.	145	78	Thin shells, operculated at one pole, regular ellipse, granular yellowish-brown contents filling whole egg.	125-150 μm×63-90 μm