#### **1. INTRODUCTION**

#### 1.1 Background

Nepal is a highly diverse and unique country harbouring an extraordinary variety of landscapes, cultures and wildlife. Despite making up less than 1% of the world's total land mass, its physiographic features range from the highest terrestrial ecosystem in the world, the Himalayas, to the subtropical lowlands of the Terai. This contrast makes Nepal one of the most biodiverse countries in the world, containing within its small area of 141,181 km<sup>2</sup>: 4.2% of all mammals, 8.5% of all birds and 2.2% of all flowering plants on Earth, including threatened flagship species such as the Royal Bengal Tiger (Panthera tigris tigris), Asian Elephant (*Elephas maximus*), Greater One-horned Rhino (*Rhinoceros unicornis*) and South Asian River Dolphin (*Platanista gangetica*) (Shrestha *et al.*, 2007). In addition to the vast faunal diversity, 35 forest types and 118 ecosystems are present in Nepal (GoN, MoFSC 2009). Almost 25% of the country's landmass is designated as protected area, with 10 national parks, three wildlife reserves, six conservation areas and one hunting reserve (DNPWC, 2014). In 2017, the Shuklaphanta and Parsa wildlife Reserve were upgraded to National parks (DNPWC, 2017).

The protected areas of Nepal cover mainly forested land and are located at various altitudes in the Terai, in the Himalayas and in the mountains, thus encompassing a multitude of landscape and preserving a vast biodiversity in the Paleartic and Indomalayan ecozones. Altitudes range from 67 m (220 ft) in the south- eastern Terai to 8,848 m (29,029) at Sagarmatha within a short horizontal span. This extreme altitudinal gradient has resulted in 11- bio-climatic zones ranging from lower tropical below 500m (1,600 ft) to nival zones above 5,000 m (16,000 ft) in the High Himalayas encompassing nine terrestrial eco – regions with 36 vegetation types (Shrestha *et al.*, 2007). Botanist recorded 1,120 species non- flowering and 5,160 species of flowering plants. Nepal rank 10 <sup>th</sup> in terms of richest flowering plant diversity in Asia. Zoologist recorded 181 mammal species 844 bird species, 100 reptile species, 43 amphibian species, 185 freshwater fish species, and 635 butterfly species (DNPWC, 2017).

Chitwan National Park is situated in South Central Nepal, Covering 952.63 sq. km. in the Subtropical lowlands of the inner Terai (DNPWC, 2017). The area comprising the Tikauli forest from Rapti river to the foothills of the Mahabharat - extending over an area of 175 sq. km (Mahendra Deer park) by the late king Mahendra in 1959 AD. In 1963 AD, the area of south of Rapti River was demarcated as a rhinoceros sanctuary. The area was gazetted as the country's first national park in 1973 AD, recognizing its unique ecosystem of the International significance. UNESCO declared RCNP a world Heritage site in 1984 AD. In 1996 AD an area of 750 sq. km surrounding the park was declared a buffer zone, which consists of forests and private lands including cultivated lands. The park consists of a diversity of ecosystem – including the Churia hills, Ox – bow lakes, and the flood plains of the Rapti, Reu and Narayani Rivers. The churia hills rise slowly towards the east from 1500 m to more than 800 m. The western portion of the park is comprised of the lower but

more rugged Someshwor hill. The park shares its eastern boundary with the Parsa Wildlife Reserve. The chitwan valley consists of tropical and subtropical forests. Sal forests cover 70 percent of the park. Grasslands cover 20 percent of the park. There are more than 50 different types of grasses, including the elephant grass (*Saccharum* sp.) renowed for its immense height which can grow upto 8 m in height. The park is home to more than 50 mammals species, over 525 birds and 55 amphibians and reptiles.

#### 1.2 Wild boar (Sus scrofa Linnaeus, 1758)

The wild boar (*Sus scrofa*), also known as the Andamanese pig or Moupin piglet (Lydekker, 1990) and 'Bandel' in Nepali is species of wild boar native to India, Nepal, Burma, Western Thailand and Sri-lanka. Wild boar (*Sus scrofa*) lies on the order Artiodactyla of the Suidae. As of 2005, upto 16 subspecies are recognize, which are divided on four regional grouping based on skull height and lacrimal bone length (Wozeccraft, 2005). Wild boar (*Sus srofa*) are cosmopolitan species. They originated in Europe and Asia, but were widely introduced to North America and are considered an invasive species in the southeastern United States and California. They are common throughout Eurasia, and inhabit every continent except Antartica (Wood and Barrett, 1979; Chapman and Trani, 2007; Oliver and Leus, 2008).

### **1.2.1 Physical description**

Wild boars range from 153 to 240 cm in length and weigh 66 to 272 kg as adults. Females tend to be smaller than males of the same age, with the size difference becoming more apparent as the animal age. Depending on their geographical location they can have speckled or solid pelage color. Their upper canine teeth typically measure 5 to 10 cm and are larger than their lower canines. Their upper canines are usually visible even when their mouth is slosed. Their dental formula is I 3/3, C 1/1, P 4/4, m 3/3 = 44 (Webster *et al.*, 1985; Ickes, 2001; Chapman and Trani, 2007; De Magalhaes and Costa, 2009).

### 1.2.2 Behaviour

Female wild boars are social animals that tend to live in groups. These groups, called sounders, are generally made up of several females and their offsprings. Males tend to be solitary after reaching maturity and join with groups during mating. In cooler condition, the boars may feed during the day, but foraging activities usually increase at the late evening (Graves, 1984; Webster *et al.*, 1985; Boitani *et al.*, 1994).

### 1.2.3 Home range

Their home varies with several factor including the number of individuals in the group, food resource availability, geographic range and predation threats. Females tend to occupy a smaller region and keep to covered areas within a home range to protect themselves and their young. Groups of females accept some overlap between their herd and others, but

sounders remain distinct groups. Males are inclined to occupy a larger area. They tolerate overlap of ranges with other males. On average, wild boars have territorial sizes of 1.1 to 3.9 sq. km (Graves, 1984; Boitani *et al.*, 1994).

### 1.2.4 Food habits

Wild boars are omnivorous. They predominately eat plant matter, particularly crops, fruits, nuts, roots and green plants. They have also been known to consume bird eggs, carrion, small rodents, insects and worms. Wild boars have reportedly preyed on small calves, lambs and other livestock when the opportunity presents itself (Webster *et al.*, 1985; Schley and Roper, 2003; Chapman and Trani, 2007).

## **1.2.5 Distribution in Nepal**

The wild boars lives in grassy or scanty bush jungle of Churia hills of Nepal. This species are widely distributed across Nepal including within all protected areas of lowland Terai and parts of protected areas in the highland Churia to Annapurna ranges and also occurs extensively outside protected areas (Jnawali *et al.*, 2011). It frequents forests after the rains, quite commonly in high crops. Wild boars are wide spread with a surprisingly wide altitudinal range. They are most abundant in Oak and Fir forests.

### **1.2.6 Conservation status**

In conservation status, it was considered as least concern in global and National contest because of its wild distribution range and an abundant size (IUCN, 2011). National population size: Total > 30,000. There is no population estimates available for the sp. (wild boar) in Nepal, however it is frequently observed and speculated to be in excess of 30,000 (Jnawali *et al.*, 2011).

## 1.2.7 Parasitic infection in wild boar

Parasites are cosmopolitan and play a significant role in the morbidity and mortality of humans as well as animals in many parts of the world. These may be transmitted to their host through ingestion, skin penetratioin, by the vectors, direct contact etc. Helminths can cause disruption of the host's nutrient absorbtion by utilizing all nutrients that passes through the intestinal tract. Most prevalent are the intestinal helminthes and infection with these are most often diagnosed by finding and identifying eggs and larval form of different helminthes parasite while examing fecal sample. Parsites play a major role in ecosystems, host population growth and regulation (Chandra and Newberne, 1997) and community biodiversity (Hudson *et al.*, 1992). Parasites can impact host survival and reproduction directly through pathological effects and indirectly through pathological effects and indirectly through pathological effects and Hudson, 1992; Hudson *et al.*, 1992; Coop and Holmes, 1996; Chandra and Newberne, 1997). Severe parasitosis can lead to blood loss, tissue damage, spontaneous abortion, congental malformation and death

(Despommier *et al.*, 1995; Chandra and Newberne, 1997). Changes of human habitation to sub areas, increased use of lands for agriculture process, increased human activities and consumption of wild boar meat have increased the chances of exposure of wild boars to domestic animal and humans. Wild boar can act as a reservoirs for many important infectious diseases such as classical swine fever, brucellosis and tricchinellosis and in human diseases such as hepatitis E, tuberculosis, leptospirosis and tricchinellosis (Meng *et al.*, 2009).

Wild boars are estimated to host at least 20 different parasitic worms, with maximum infection occurring in summer. Wild boars also carry parasite worms, known to infect humans including Gastrodiscoides, Trichinella spiralis, Taenia solium and Balatidium coli (V.G. H. et al., 1998). Wild boars host a variety of parasites including Trichinella species, Toxoplasm gondii, Gongylonema species, Lungworms (Metastrongylus elongatus), Kidneyworms (Stephanurus dentatus), Stomachworms (Physocephalus sexalatus), Ascarids (Ascaris lumbricoides), Whipworms (Trichuris suis) (Henry and Comley, 1970; Ickes, 2001; Ickes et al., 2005; Chapman and Trani, 2007; Meng et al., 2009). Parasites in wild boar are Metastrongylus apri, Dicrocoelium found dendriticum, Macracanthorhynchus hirudnaceus, Gongylnema pulchrum, Physocephalus sexalatus, Taenia hydatigena larva (Yagoob et al., 2014). Sato et al. (2008) recorded eighteen helminth parasites, including seventeen nematode sp. (Metastrongylus elongatus, Metastrongylus salmi, Metastrongylus asymmetricus, Metastrongylus pudendotectus, Stephanurus dentatus, Gnathostoma doloresi, Physocephalus sexulata, Ascarops strongylina, Capillaria suis, Ascaris summ, Globocephallus somoensis, Globocephalus longimucronatus, Strongyloides ransomi, Trichuris suis, Bourgelatia diducta, Oesophagostomum watanabei), and one cestoda sp (Pseudanoplocephala nipponensis). Disease monitoring in wild animals has recently a necessary componenet for preventing further infections and conservation. The presence of parasite in an animal, particularly in young animals resulted into reduced body weight gains and reproductive disorders. In addition to that the parasite affect the quality of animals products (meat, skin, antlers) and ultimately death (Fox, 2000).

### **1.3 Objectives**

### **1.3.1 General Objective**

To study the gastro-intestinal parasites of wild boar (Sus scrofa) in Chitwan National Park.

### **1.3.2 Specific objectives**

- ) To determine the prevalence of parasites in wild boar.
- ) To analyze the intensity of parasitic infection in wild boar

#### 1.4 Significance of the study

In global contest, various researches have been carried out regarding intestinal parasites of wild boar. CNP was one of the ideal habitat for many natural study sites due to its high potentials of biodiversity. It provides ideal habitat for rare and endangered species like Tiger, Elephant, One horned rhino, Hispid hare, Wild boar, Monkey, Spotted deer etc. For the study of wild boar this National Park was one of the most important places in the world. On the basis of literature survey, very few listed article have been found in relation to Gastro-intestinal parasites of domesticated pig but not any article in wild boar was found in contest of Nepal. This study will be the first attempt on the prevalence of Gastro-intestinal parasites infection on wild boar and also help in understanding the prevalence of Gastro-intestinal parasites as well as help to cover the research gap on this species and also help in finding the zoonotically associated diseases with reference to wild boars. So, it was realized that the research study should be launched to investigate prevalence of gastro-intestinal parasites by fecal examination. This study will provide first overview on parasites in wild boar in the vicinity of CNP.

# 2. LITERATURE REVIEW

The most common source of new emerging infectious diseases that put a risk, the health of human beings and livestock are wild animals. The wild and domestic animals most commonly interact through direct competition for food, predation, pathogen exchange or hybridization (Foufopoulos *et al.*, 2003). Parasites are the living organisms which depend on the host for their food, shelter and metabolic activities. Parasites can affect host survival and reproduction directly through pathological effects (blood loss, tissue damage, spontaneous abortion, congenital malformation and death) and also reduce the host's immunity by affecting on the physical condition (Thawait *et al.*, 2014). In case of wild boar, a very little research work has been carried out regarding parasitic infection. Wild boars can be infected by different parasites including protozoans, trematodes, cestodes and nematodes. Here, some of the important published work related with the present work has been reviewed.

#### 2.1 In global contest

Endoparsites are those organisms which inhabit in the gut, body cavity, liver, lungs and blood or within the internal cavities and tissue or cell of their host causing parasitic infection. Parasites usually include gastro-intestinal helminthiosis, coccidiosis, fasciolosis and mange (Meng *et al.*, 2009). The suitable temperature and humidity play an important role for the development of endoparasites. Some of the protozoan and helminthes parasites have been reported in wild boar from various part of the world. The wild boars were susceptible to internal parasites because these animals seek rivers, polls or swamps for wallowing and searching for food where the higher risk of infection with snail born helminthes.

Ineson (1953) examined twenty two wild pigs from Newzealand to determine the presence and intensity of parasitic infestation and finally recorded the presence of fifteen species of parasites namely, *Balantidium* (61.9%), *Eimeria debliecki* (4.7%), *Anaplasma* (6.6%), *Cysticercus tenuicollis* (4.7%), *Hyostrongylus rubidus* (28.5%), *Ascaris suum* (42.8%), *Metastrongylus elongatus* (66.6%), *Choerostrongylus pudendodectus* (38%), *Haematopinus suis* (68.1%), *Sarcoptes scabie* var *suis* (4.5%), *Oesophagostomum dentatum* (14.2%), *Echinococcus granulosus* (10%), *Trichuris suis* (20%), *Fasciola hepatica* (2%) and *Globocephalus urosubulatus* (4%). Similarly, in Aransas National Wildlife Refuge (Coombs, 1974) examined ten feral pigs (*Sus srofa domesticus*) x European Wild boars (*Sus srofa cristatus*) and finally reported, *Gongylonema pulchrum,Ascaris suum*, *Globocephalus urosubulatus*, *Stephanurus dentatus*, *Sarcocystis* sp. and three sp. of *Metastrongylus*.

Similarly, in France, Humbert and Henry (1989) identified five species of nematode (*Metastrongylus asymetricus*, *Metastrongylus confuses*, *Metastrongylus elongatus*, *Metastrongylus pudendodectus*, *Metastrongylus salmi*) and two species of stomach

nematodes (*Ascarops strongylina*, *Physocephalus sexalatus*). Likewise, in Iran, Eslami and Farsad-Hamdi (1992) examined fifty seven wild boars (*Sus srofa*) from protected regions of Iran for helminthes and reported sixteen species of helminthes, out of which ten are nematodes, one acanthocephalon, two trenatodes and three larval cestodes. Correspondingly, from Finland, Roepstorffa *et al.* (1998) examined 516 swine herds and recorded the presence of *Ascaris suum*, *Oesophagostomum* sp., *Isospora suis, Eimeria* sp., *Trichuris* sp. and *Strongyloides ransomi*. Likewise, from North –west Poland, Balicka-Ramisz *et al.* (2000) examine fecal samples from 63 wild boars and reported the prevalence of 82.5% in wild boar. Four coccidian sps were (*Eimeria debliecki, Eimeria suis, Eimeria splita* and *Eimeria scraba*) and one *Isospora suis* are reported.

Fiere et al. (2001) examined forty – seven wild boars from eastern Spain and reported Taenia hydatigena cysticercus (19%), Ascarops strongylina (87%), Physocephalus sexulatus (6%), Ascaris suum (21%) and Macracanthorhynchus hirudinaceus (21%). No trichinella species were found. Correspondingly, from Eastern Stavonia, Republic of Croatia, Rimac et al. (2002) investigated the fecal samples of 47 wild boars (Sus srofa) and finally revealed the presence of 14 helminth species. The predominant nematodes include, *Metastrongylus* apri, Metastrongylus pudendotectus, Globocephalus urosubulatus, Ascarops strongylina, Physocephalus sexalatus, Gnathostoma hispidum, *Oesophagostomum* Trichinella the acanthocephalon sp., sp., species Macracanthorhynchus hirudinaceus, the intestinal fluke Echinochasmus perfoliatus (Trematode) and larval stage (Cysticercus tenuicollis, Echinococcus hidatidosus) of two tapeworm species (Taenia hydatigena, Echinococcus granulosus) cestode were recorded.

Soloyamani-Mohammadi et al. (2003) examined twelve wild boars (sus scrofa) during a survey from 2000-2001 from western Iran and recorded two cestode larva, Cysticercus tenuicollis (25%), Cysticercus cellulosae (8.3%), four nematode species, Metastrongylus apri (41.6%), Metastrongylus pudendotectus (16.6%), Metastrongylus salmi (8.3%), Trichuris suis (8.3%) and the acanthocephalon Macrecanthorhynchus hirudinaceus. Correspondingly, from central Spain, Isabel et al. (2003) compaired the helminth population in agroup of wild boars and finally identified eleven helminth species, including ten nematodes (Ascaris suum, Gongylonema pulchrum, Oesophagostomum dentatum, Trichuris suis, Globocephalus urosubulatus, Metastrongylus sp, Physocephalus sexalatus, Simnondsia parasoxa, Capillaria garfiai and Ascarops strongylina) and one acanthocephalon Macracanthorhynchus hirudinaceus. Mudim et al. (2004) examined fecal samples of 79 wild boars (Sus srofa) finally reported the prevalence of 97.5% of the samples for intestinal parasites namely, Strongilides (70.9%), Ascaris suum (46.9%), Trichuris suis (29.1%), Metastrongylus sp. (12.6%), Strongyloides ransomi (3.8%), Balantidium coli (38.0%), Entamoeba sp. (15.2%), Giardia sp. (1.3%), Blastocystis sp. (12.6%). Coccidian oocyst were observed in 59.5% and five species of *Eimeria* and one Isospora were recovered. Foata et al. (2005) collected and examined 160 (stomach and intestine) and 58 livers of wild boars from Corsica and reported six speces of helminth, namely one trematode Dicrocoelium dendriticum, the only larval stage of cestode, Echinococcus granulosus, three nematode species, Ascaris suum, Globocephalus *urosubulatus* and *Metastrongylus* species and one acanthocephalon *Macracanthorhynchus hirudinaceus*. Likewise, Eijck and Borgsteede (2005) examined the prevalence of gastrointestinal parasites in suking piglets, weners, fattening and sows and finally reported the infection with *Ascaris suum* (50%), *Oesophagostomum* sp. (25%) and *Trichuris suis* (37.5%). Similarly, from Estonia, Jarvis and Magi (2007) examined one hundred wild boars (*Sus srofa*) and finally reported seven helminth species, *Metastrongylus salmi*, *Metastrongylus elongatus*, Ascaris suum, *Trichuris suis*, *Dicrocoelium dendriticum* and *Taenia hydatigena* larva.

Among twenty nine Japanese wild boar (Sus srofa leucomystax) during hunting season 2005-2006 were examined and finally recorded the presence of eighteen helminth parasite, including 17 nematodes species. (Metastrongylus elongatus, Metastrongylus salmi, Metastrongylus assymetricus, Metastrongylus pudendotectus, Stephanurus dentatus, Gnathostoma doloresi, Physocephalus sexulata, Ascarops strongylina, Capillaria suis, Globocephalus longimucronatus, Strongyloides ransomi, Trichuris suis, Bourgelatia diducta, Oesophagostomum wantabei) and one cestode species. (Pseudanoplocephala nipponensis) (Sato et al., 2008). Similarly, from Turkey, Boral et al. (2009) examined 238 fecal samples. Out of 238 pigs specimen 105 were yonger than 6 months and rest are older than 6 months and finally revealed the presence of *Giardia* sp. 9 (3.7%), *Balantidium coli* cysts 4 (1.6%), Ascaris suum 9 (4.1%) of pigs yonger than 6 month and Cryptosporidium sp. 9 (6.7%), Balantidium coli cyst in 2(1.5%) and Ascaris suum in 9 (6.7%) in above 6 months. Similarly, in Italy, Moretta et al. (2010) examined the fecal samples of 123 wild boars and finally recorded the presence of (73.98%) strongyles and (33.33%) coccidian and other paraites including lungworms (8.94%), Acanthocephalon (6.55%), Trichuris suis (4.88%), Capillaria sp. (2.44%), Spiruridae sp. (1.63%), Ascaris suum (0.81%) and Strongyloides ransomi (0.81%).

From Turkey, Senlik et al. (2010) investigated the status of helminth infection in wild boars and finally twelve species of helminthes were detected, Metastrongylus apri (59%), Metastrongylus salmi (52%), Metastrongylus pudendotectus (52%), Dicrocoelium dendriticum (33%), Globocephalus urosubulatus (22%), Macracanthorhynchus hirudinaceus (19%), Gongylonema pulchrum (11%), Physocephalus sexalatus (7%), Trichuris suis (7%), Ascarops strongylina (4%), Hyostrongylus rubidus (4%) and Taenia hydatigena larva (4%). Tomass et al. (2012) examined seven hundred fourteen pigs from Ethopia and finally recorded the presence of Ascaris suum (25.9%), Fasciola hepatica (1.8%), *Eimeria* sp. (1.7%) and *Trichuris* sp. (0.3%). Correspondingly, from Brasil, Muller and Silva (2013) examined forty gastro - intestinal tracts of wild boars and finally reported the presence of Ascaris suum, Trichostrongylus colubriformis, Oesophagostomum dentatum, Trichuris suis. Similarly, from Iran, Yagoob et al. (2014) necropsied and examined the hunted wild boar and finally recorded the the presence of seven helminth *Metastrongylus* apri (34%),Dicrocoelium dendriticum (22%), species, Macracanthorhynchus hirudinaceus (19%), Gongylonema pulchrum (9%), Physocephalus sexalatus (7%), Trichuris suis (6%) and Taenia hydatigena larva (3%). Likewise, from India, Allwin *et al.* (2015) examined 90 fecal samples in total (n = 30 of wild pigs, n = 30

desi pigs and n = 30 cross breed pigs) and reported the presence of four species of nematode, *Ascaris suum*, *Trichuris suis*, *Strongylus* sp., *Strongyloides* sp. Begum *et al.* (2014) examined 110 fecal samples and 20 vicera from Bangladesh and finally reported the presence of 12 types of endoparasites namely *Ascaris suum* (50.9%), *Strongyloides* sp. (29.1%), *Oesophagostomum* sp. (12.7%), *Trichuris suis* (9.1%), *Ancylostoma* sp. (3.6%), *Hyostrongylus rubidus* (1.8%), *Fasciola* sp. (12%), *Dicrocoelium* sp.(8.2%), *Schistosoma suis* (7.3%), *Eimeria* sp.(56.4%), *Balantidium coli* (40%) and *Isospora suis* (9.1%).

Sen et al. (2015) examined 100 fecal sample of wild boars from Bangladesh and finally identified the presence of six species of nematode, Ascaris suum (38%), Macracanthorhynchus hirudinaceus (22%), Strongyloides ransomi (20%), Trichuris suis (5%), Hyostrongylus rubidus (5.17%) and Oesophagostomum sp (6.25%). Recently, from spain, Mansouri et al. (2016) examined 11 males and 14 female wild boar and finally recorded the presence of cysticercus tenucollis 1(4%), the larval stage of Taenia hydatigena, 13 (52%) Macracanthorhynchus hirudinaceus, 17 (68%) Metastrongylus sp. and 20 (80%) with Ascarops sp. No trichinella sp. were recorded. Likewise, From India, Dadas et al. (2016) conducted a survey to determine the gastro- intestinal parasites and finally reported the presence of six parasites species, namely Ascaris suum (32%), Balantidium coli (31.85%), Trichuris suis (11.11%), Isospora suis (1.48%), Strongyloides ransomi (0.74%) and Globocephalus urosubulatus (0.74%). Correspondingly in the recent time, from Serbia, Stojanov et al. (2017) examined 52 fecal samples and reported the presence of Metestrongylus sp., Ascaris suum, Trichuris sp., Hyostrongylus sp., *Physocephalus* sexalatus, **Strongyloides** Gnathostoma hispidum, ransomi, Oesophagostomum sp., Globocephalus sp., Hyostrongylus rubidus., Eimeria deblecki and Eimeria suis.

### 2.2 In National Context

On the basis of literature survey, very poorly listed article has been found in relation to gastro-intestinal parasites of wild boar in Nepal. However, some related work on the basis of domesticated pig has been reviewed. Joshi (1991) observed the presence of *Taenia* cysts in pig in Kathmandu. Similarly Joshi *et al.* (2001) demonstrated the presence of *Cysticercosis* in domestic pig in Dharan and Kathmandu. Poudyal (1998) identified presence of *Cysticercosis* in domesticated pig in Dharan and Kathmandu. Shakya (2009) revealed the presence of *Taenia* cysts in domesticated pig in Kirtipur Municipality. Recently, from Chandragiri Municipality, Kathmandu, Khanel (2017) has reported the overall prevalence of 88.57% and presence of three genera of protozoa, *Eimeria* sp. 45 (42.8%), *Isospora* sp. 8 (7.61%), *Balantidium* sp. 25 (23.80%), two genera of trematode, *Fasciolopsis* sp.10 (9.52%), *Schistosoma* sp. 5 (4.76%), and three genera of nematode, *Ascaris* sp. 40 (38.09%), *Strongyloides* sp.15 (14.28%) and *Trichuris* sp. 8 (7.61%).

# **3. MATERIALS AND METHODS**

#### 3.1 Study Area

#### **3.1.1 Chitwan National Park**

The study area, Chitwan National Park is situated in South Central Nepal, Covering 952.63 sq km. in the subtropical lowlands of the inner terai (DNPWC, 2017). Its geographic location co-ordinates 27°30'0"N 84°20'o"E. In altitude it ranges from about 100m (300ft) in the river valleys to 815 m (2674ft) in the churia hills (Shrestha et al., 2007). The area comprising the Tikauli forest – from Rapti river to the foothills of the Mahabharat – extending over an area of 175 sq. km (Mahendra Deer Park) by the late king Mahendra in 1959 AD. In 1963 AD the area of south of Rapti River was demarcated as a rhinoceros sanctuary. The area was gazette as the country's first national park in 1973 AD, recognizing its unique ecosystem of the International Significance. UNESCO declared CNP a world heritage in 1984 AD. In 1996 AD an area of 750 sq. km surrounding the park was declared as buffer zone, which consists of forest and private lands and cultivated lands. The Chitwan valley consists of Tropical and subtropical forests. Sal forests covers 70 percent of the park. Grasslands cover 20 percent of the park. There are more than 50 different types of grasses, including the elephant grass (Saccharum sp.), renowed for its immense height. The park is home to more than 50 mammals sp., over 525 sp. birds and 55 amphibians and reptiles. It provides habitat for wide range of vertebrates which include endangered species such as wild elephant (Elephas maximus), Bengal Tiger (Panthera tigris), Spotted Deer (Rusa alfredi), Blue Bull (Boselephus trogocumelius), Barking Deer (Muntiacus muntjak), Hog Deer (Hyelophus porcinus), Wild Boar (Sus srofa), Jackal (Canis mesomelus), Crocodile (Crocodylus porusus), Cobra (Naja naja) etc.

Features: The park consists of a diversity of a diversity of ecosystem – including the Churia hills, Ox – bow lakes and the flood plains of the Rapti, Reu and Narayani Rivers. The churia hills rise slowly towards the east from 1500 m to more than 800 m. The western portion of the park is comprised of the lower but more rugged Someshwor hill. The park shares its eastern boundary with the Parsa Wildlife Reserve (DNPWC, 2017).

Climate: The park has arrange of climatic seasons each offering a unique experience. October through February with average temperature of 25°c offer an enjoyable climate. From March to June temperatures can reach as high as 43°c, the hot humid days give way to the monsoon season that typically lasts from late June until September (DNPWC, 2017).



Figure 1: Map showing study area in Chitwan National Park (www.google.com).

## **3.2 Instruments and Materials**

Following instruments, materials and chemicals were used to carry out the research.

## **3.2.1 Materials and Chemicals**

The materials used during the research were centrifuge machine, measuring cylinder, Volumetric flask, electric microscope, glass rod, stage micrometer, ocular micrometer, refregerator, tea strainer, motor and pestle. The chemicals required were 10% formalin, distilled water, saturated Sodium chloride solution, Methylene blue.

## 3.3 Study Design



Figure 2: Research flow chart

# Glimpse of photograph during field and lab work



Photo 1: Juvenile of wild boar



Photo 3: Fecal sample collection



Photo 5: Examination under microscope



Photo 2: Wild boar with juvenile



Photo 4: Juvenile at Kasara



Photo 6: Measurement of size of parasite

### **3.3.1 Sample collection method**

A sum total of 100 fecal samples were collected from wild boars at CNP for a time span of two months ranging from April 2017 to May 2017. Fresh fecal samples were collected from the dropping ground. Proper care were taken when collecting the fecal samples from the ground to prevent contamination.

# **3.3.2 Preservation of samples**

After collection of fecal samples about 20-30 gm of feces were placed in clean 50ml vials and mixed with 10% formalin and stored at  $4^{\circ}$ c in laboratory facilities provided by Department of Zoology, Kirtipur, Kathmandu.

## **3.3.3 Examination of fecal sample**

All the fecal samples so collected were processed using standard sedimentation, floatation and direct smear technique as described by Soulsby (1982) followed by microscopic examination for the presence of helminth ova and protozoan cyst/oocysts.

## **3.3.3.1 Direct smear method**

Two mg of fecal samples were mixed in a drop of saline on a slide and coverslip was applied. The entire field of coverslip was examined under low microscope and eggs were visualized and counted (Soulsby, 1982).

## 3.3.3.2 Differential floatation (D.F.) Technique

The D.F. technique is widely used for the detection of nematode and cestode egg. Three gm of fecal sample was taken in a beaker and 42 ml of water was added. With the help of mortar and pestle, the sample was grinded lightly and filtered with a tea strainer. The filtered sample was poured into a centrifuging tube of 15 ml and centrifuge at 1000 rpm for 5 minutes. The tube was taken out and the upper part of water was removed with the help of pipette. The tube was noted then filled with the saturated sodium chloride (Nacl) solution and again centrifuge at 1000 rpm for 5 minutes. More saturated sodium chloride solution was added to develop convex surface at the top of the tube and one drop of methylene blue was added. A cover slip was placed over the top of the tube so that the Nacl touches the cover the slip for a few minutes and then the cover slip was placed on a slide and examined under microscope ( $10x \times 40x$ ) (Soulsby, 1982).

#### 3.3.3.3 Sedimentation technique

Sedimentation was used to isolate of flukes, some other tapeworms and nematodes whose eggs didnot float readily in common floatation solution. Three gm of fecal was weighted. Sample was grinned with water (45-50 ml) using mortar and pestle. The Sample was filtered with a tea strainer, filterate was allowed to stand for 5 minutes in a beaker. The supernant was removed and sediment was resuspended with water (till the suspension becomes clear). The suspension was then allowed to sediment for next 5 minutes. The suspenant was drawn off. A drop of sediment was placed on clear slide, and few drop of methylene blue was added into it. The fecal smear was covered with cover slip and examined using a microscope (Soulsby, 1982).

### 3.4 Identification of oocysts, eggs and larvae of parasites

Oocyst, eggs and larvae were identified on the basis of morphological characters (shape and size) as described by Yamaguti (1961), Soulsby (1982), Sloss *et al.* (1994) and other published and unpublished articles and also from internet sources. Caliberation obtained using Ocular and Stage micrometer was used to measure length and breadth of eggs, Oocysts and larvae.

### 3.5 Intensity calculation of parasite

Intensity of parasites was calculated depending on the number of eggs /oocysts and larvae found per field.

Light infection= < 2 eggs/oocysts /larvae per field Mild infection= 2 - 4 eggs /oocysts/larvae per field Moderate infection= 4 - 6 eggs/oocysts/larvae per field Heavy infection= > 6 eggs /oocysts/larvae per field

### 3.6 Data analysis

Since the study was mainly focused on identification of different parasites, the data were analyzed by using MS-EXCEL 2007 and statistical analysis was performed using "R" version 3.3.1 software packages. Chi-square test was used for statistical analysis of data. In all cases 95% confidence interval (CI) and P < 0.05 was considered for statistically significant difference.

# 4. RESULTS

### 4.1 Overall Prevalence of Gastro - Intestinal Parasites in wild boar

Out of 100 fecal samples of wild boar (*Sus scrofa*), 95 (95%) samples were found to be positive for parasitic infection in wild boar of CNP. There is significant difference in the general prevalence of parasites.



Fig 3: General prevalence of intestinal parasites

## 4.2 Distribution of GI parasites in wild boar.

Out of 100 sample examined, nine genus of parasites were identified with Protozoan, *Eimeria* sp. with micropyle 40 (40%) and *Eimeria* sp. without micropyle 70 (70%). Similarly, in Trematode, *Fasciola* sp. 12 (12%) and in Nematode, *Ascaris* sp. 7 (7%), *Stephanurus* sp. 44 (44%), *Strongyloides* sp. 56 (56%), *Strongyle* sp. 49(49%), *Metastrongylus* sp. 12 (12%), *Trichuris* sp. 6 (6%) and *Globocephalus* sp. 38 (38%). There is highly significant difference between the genus wise prevalence of parasites on wild boar ( $^2 = 208.34$ , df = 9, P < 0.05).



Fig 4: Genus wise parasitic prevalence of wild boar

#### 4.2.1 Prevalence of Protozoan parasites

Out of total 100 samples, protozoan parasites with the genus *Eimeria* sp. without micropyle showed the highest prevalence 70% followed by *Eimeria* sp. with micropyle 40%. There is high significant difference between *Eimeria* sp. without micropyle and with micropyle sp. ( $^2 = 21.662$ , P < 0.05).

Table 1: Prevalence of	protozoan	parasites
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S.N	Name of parasites	Prevalence
1	<i>Eimeria</i> sp. without micropyle	70%
2	Eimeria sp. with micropyle	40%

#### **4.2.2 Prevalence of helminth parasites**

Out of 100 samples, 80 samples were found to be positive for helminth parasites. Eight genus of helminth were identified.Out of which trematode (1) *Fasciola* sp. 12 (12%) and nematode (7) with genus *Strongyloides* sp. 56% followed by *Strongyle* sp. 49%, *Stephanurus* sp. 44%, *Globocephalus* sp. 38%, *Metastrongylus* sp. 12%, *Ascaris* sp.7% and *Trichuris* sp. 6%. Helminth parasitic infection in wild boar were found to be highly significant ( $^2$  =149.7, P < P0.05).

Class	Name of parasites	Prevalence
Trematode	Fasciola sp.	12%
Nematode	Strongyloides sp.	56%
	Strongyle sp.	49%
	Stephanurus sp.	44%
	Globocephalus sp.	38%
	Metastrongylus sp.	12%
	Ascaris sp.	7%
	Trichuris sp.	6%

Table 2: Prevalence of helminth parasites

### 4.2.3 Trematodes

Out of 100 samples, prevalence of trematodes was observed only in 12 samples i.e presence of *Fasciola* sp. only.



Fig 5: Prevalence of Trematode parasite in wild boar of CNP.

### 4.2.4 Nematode

Among 100 samples, 80 samples were found to be positive for nematodes. Seven genus of nematodes were identified. Among them *Strongyloides* sp. 56 (56%) accounts highest prevalent in wild boar at CNP followed by *Strongyle* sp. 49 (49%), *Stephanurus* sp. 44 (44%), *Globocephalus* sp. 38 (38%), *Metastrongylus* sp. 12 (12%), *Ascaris* sp. 7 (7%) and *Trichuris* sp. 6 (6%). Nematode parasitic infection in wild boar were found to be statistically significant (2=129.08, P < 0.05).



Fig 6: Prevalence of nematode parasites in wild boar of CNP.

### 4.3 Mixed infection

Out of 100 samples, 95 samples were found to be infected with different intestinal parasites of wild boar. Double infection showed the highest rate 48 (50.52%) followed by multiple infection 19 (20%), triple infection 18 (18.94%) and single infection 10 (10.52%). The result revealed the significant difference in mixed infection (2=46.75, P < 0.05).

S.N.	Types of infection	Total 95 (100%)
1	Single	10 (10.52%)
2	Double	48 (50.52%)
3	Triple	18 (18.94%)
4	Multiple	19 (20%)

### 4.4 Intensity of Parasites in wild boar of Chitwan National Park

Intensity of parasitic infection has been assessed based upon the number of eggs/oocyst/cysts and larva found per microscopic field. Among protozoans, maximum number of positive samples were found with light intensity followed by mild, moderate and heavy intensity. In heavy intensity of protozoan, *Eimeria* sp. without micropyle showed higher intensity over *Eimeria* sp. with micropyle. Similarly, in case of trematodes light intensity is followed by mild intensity. In case of nematodes, *Stephanurus* sp. possesses the highest intensity followed by *Strongyloides* sp., *Strongyle* sp., *Metastrongylus* sp. and *Globocephalus* sp. No heavy intensity was found between *Ascaris* sp. and *Trichuris* sp. Maximum number of nematode samples revealed the light intensity followed by mild and moderate.

Table 4: Intensity	of pa	rasites in	wild boar
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Class	Parasites	Light	Mild	Moderate	Heavy
Sporozoa	<i>Eimeria</i> sp.	18 (45%)	10 (25%)	7 (17.75%)	5 (12.5%)
	with micropyle				
	<i>Eimeria</i> without	34 (48.57%)	17 (24.28%)	12 (17.14%)	7 (10%)
	micropyle sp.				
Trematode	Fasciola sp.	8 (66.66%)	4 (33.33%)	-	-
	Ascaris sp	4 (57.14%)	3 (42.85%		
				-	-
	Stephanurus sp.	10 (22.72%)	13 (29.54%)	6 (13.63%)	15 (34.09%)
Namatada	Trichuris sp.	4 (66.66%)	2 (33.33%)	-	-
Nematode	Metastrongylus	2 (16.66%)	7 (58.33%)		3 (25%)
	sp.				
	Globocephalus	8 (21.05%)	12 (31.57%)	10 (26.31%)	8 (21.05%)
	sp.				
	Strongyloides	18 (32.14%)	12 (21.42%)	8 (14.28%)	18 (32.14%)
	sp.				
	Strongyle sp.	18 (36.73%)	7 (14.28%)	10 (20.40%)	14 (28.57%)
1				1	

### Photographs of identified Gastro-intestinal parasites



Photo 7: Ascaris sp. (50um x48um)



Photo 9: *Eimeria* sp. without micropyle (24um x 16um )



Photo 8: Trichuris sp. (55um x26um)



Photo 10: *Eimeria* sp. with micropyle (26um x16um)



Photo11: *Eimeria* sp. showing (with or without) micropyle



Photo 12: Stephanurus sp. (98um x60um)



Photo 13 : Globocephalus sp.(55um x 45um)



Photo 15: Strongyloide sp. (55um x 36um)



Photo 14: Fasciola sp. (110um x 60um)



Photo 16: Strongyle sp. (80um x 60um)



Photo 17: *Metastrongylus* sp.(57um x 34um)



Photo18: Larva of Strongyle sp.

# Photographs of unidentified gastro-intestinal parasites



Photo 19: unindentified (91um x 60um)



Photo 21: unidentified (90um x 60um)



Photo 23: unidentified (122um x 84um)

![](_page_22_Picture_7.jpeg)

Photo 20: unidentified (100um x 36um)

![](_page_22_Picture_9.jpeg)

Photo 22: unidentified (122um x 84um)

![](_page_22_Picture_11.jpeg)

Photo24: unidentified (88um x86um)

### 5. DISCUSSION

National park is an area of land of unusual ecological and scenic interest set aside by government, where flora and fauna are protected as far as possible in their wild state for their scientific, educational and recreational value and for the benefit of the nation and mankind as a whole (DNPWC, 1973). Wild animals are concurrently infected with multiple parasites and interactions among these parasites may influence both diseases dynamics and host fitness. However, the sublethal cost of parasite infection are difficult to measure and the effects of concomitant infections with multiple parasite species on individual physology and fitness are poorly described for wild host. Both microparasites (eg. Bacteria, viruses) and macroparsites (eg, helminths, arthopods) can have far reaching effects on the fitness of their hosts, ranging from reducing body condition below levels critical for reproduction to causing castration or direct mortality (Stien, 2011). Wild boars are inhabits a diverse array of habitats including heavily brushed areas providing shelter from predators, water for drinking and bathing purpose and swampy areas (Henry and Conley, 1970; Ickes, 2001; Ickes *et al.*, 2005; Chapman and trani, 2007; Meng *et al.*, 2009).

Wild boars are estimated to host at least 20 different parasitic worms, with maximum occuring in summer (Heptner *et al.*, 1998).Wild boars host a variety of parasites including *Trichinella* species, *Toxoplasm gondii*, *Gongylonema* species, lungworms (*Metastrongylus elongatus*), kidneyworms (*Stephanurus dentatus*), stomachworms (*Physocephalus sexalatus*), Ascarids (*Ascaris lumbricoides*), whipworms (*Trichuris suis*) (Henry and Conley, 1970; Ickes, 2001; Ickes *et al.*, 2005; Chapman and trani, 2007; Meng *et al.*, 2009). The current epoch of ecological time is driven by human interference. Multiple anthropogenic stressors – including climate change, pollution, ocean acidification, habitat loss and fragmentation, urbanization, agriculture expansion and intensification, together with other changes in the use of water and land resources are directly or indirectly impacting all species on earth (Faye *et al.*, 2006). These changes may be lead to the crossing or corrosion of critical thresholds or planetary boundaries that include physological stress or complete system dysfunction, with negative consequences for individuals, populations and species. Such processes will have significant impacts on parasite natura history and infectious disease risk (Harvel *et al.*, 2002).

The current study aimed to evaluate the prevalence of gastro-intestinal parasite of wild boars in CNP. Out of 100 fecal sample examination for gastro-intestinal parasites, 95 samples were found to be infected with an overall prevalence of 95%. On parasitological examination wild boars were found to be harboring different parasites which include one genus protozoan, one genus of trematode and seven genus of nematodes.With respect to species wise prevalence, *Eimeria* sp without micropyle (70%) was found to be the most prevalent parasite followed by *Strongyloides* sp. (56%), *Strongyle* sp. (49%), *Stephanurus* sp. (44%), *Eimeria* sp. with micropyle (40%), *Globocephalus* sp. (38%), *Metastrongylus* sp. (12%), *Fasciola* sp. (12%) and least prevalence of about 7% and 6% was noted for

Ascaris sp. and Trichuris sp. Overall Eimeria sp. was found to be most prevalent parasite as earlier reported by researcher (Balicka-Ramisz et al., 2000) of 82.5% prevalence. But the researcher, (Ineson, 1953; Moretta et al., 2010; Tomass et al., 2012) reported the low prevalence of *Eimeria* sp. following 4.75%, 33.33% and 1.7% respectively. High prevalence of Eimeria species in the study area may be due to the overcrowding of other animals and habitat because oocyst require moist condition to undergo sporulation. The variation might be due to the difference in sample size, selection of samples, techniques of sample collection, period and place of the study, environment factors etc. The prevalence of trematode i.e Fasciola sp. was 12% which was similar to the report of (Begum et al., 2014). However, the report from Northen Ethopia, Tomass et al. (2012) indicates low prevalence just only 1.8%. Factors such as presence of reservoir hosts, presence of snail intermediate host, and ability of *Fasciola hepatica* to colonise and to adopt new hosts contribute for its spread in livestock in an area. No any cestodes were recorded in the present study in wild boar which supported the findings of the result of (Roepstorffa et al., 1998; Tomass et al., 2012; Allwin et al., 2015). Most of the studies on gastro-intestinal nematode ecology in wild boar have conducted that temperature and humidity play an important role in the survival and transmisson of parasites, eggs and larvae (Kutz et al., 2004). The nematode species identified in the fecal examination of wild boar of CNP includes Strongyloides sp. (56%) followed by Strongyle sp. (49%), Stephanurus sp. (44%), Globocephalus sp. (38%), Metastrongylus sp. (12%), Ascaris sp. (7%) and *Trichuris* sp. (6%).

*Ascaris* sp. (large roundworm) is the most important gastro-intestinal worm of pigs. It is more common in growing pigs than in adult pigs (Lee, 2012). Heavily infested pigs may have upto twenty five worms blocking the small intestines and bile duct causing loss of appetite, vomiting, icterus and death if the small intestine is ruptured. Growth rate and feed efficency can be depressed by upto 10% (Lee, 2012). The prevalence rate of *Ascaris* sp. in the present study was 7 (7%) which was nearly similar to the report of (Boral *et al.*, 2009). However, the researcher, (Ineson, 1953; Eijck *et al.*, 2005; Begum *et al.*, 2014 Mundim *et al.*, 2015; Sen *et al.*, 2015; Dadas *et al.*, 2016) obtained the higher prevalence rate following 42.8%, 50%, 50.9%, 46.9%, 38% and 32.9%. On the cotrary, the report from (Fiere *et al.*, 2001; Moretta *et al.* 2010) indicates low prevalence just only 2% and 0.81%.

Pigs are considered the natural hosts of *Trichuris suis* (whipworm) although primates and humans may be infected. *Trichuris* infection is a fairly common problem in pigs (Lee, 2012). This parasite have direct life cycle and animals become infected after ingestion of infective eggs. The present prevalence rate of *Trichuris* sp. was 6% which was nearly similar to the report of (Senlik *et al.*, 2010; Yagoob *et al.*, 2014; Sen *et al.*, 2015) following the prevalence of 7%, 6%, and 5% respectively. But, the report from (Ineson, 1953; Soloyamani-Mohammadi *et al.*, 2003; Mudim *et al.*, 2004; Eijck and Borgsteede, 2005; Begum *et al.*, 2014; Dadas *et al.*, 2016) indicates the higher prevalence rate of 20%, 8.3%, 29.1%, 37.5%, 9.1% and 11.11%. On contrary, the researcher, (Moretta *et al.*, 2010; Tomass *et al.*, 2012) reports the lower prevalence of *Trichuris* sp. just only

4.88% and 0.3%. Similarly, *Globocephalus* is a genus of hookworms that affects swine and wild boars worldwide. The most relevant species of veterinary importance is *Globocephalus urosubulatus*. The life cycle is not completely elucidated (Junguera, 2017). Most probably they have a direct life cycle without intermediate host. Pigs get infected after ingesting the infective larva released from eggs to the environment. The present prevalence rate of *Globocephalus* sp. was 38% which was higher than the earlier reported by (Senlik *et al.*, 2010; Dadas *et al.*, 2016) following 22% and 0.74% prevalence respectively.

*Strongyloides* sp. (Intestinal threadworm) causes strongyloidiasis. *Strongyloides* worms may be present in a host as a parasitic and free living form in the soil. They have direct life cycle and causes infection by ingestion of contaminated vegetation and drinks with the larva of this species (Staphen and Gareth, 2003). The present prevalence rate of *Strongyloides* sp. was 56% which was higher than the earlier reported by (Mudim *et al.*, 2004; Moretta *et al.*, 2010; Begum *et al.*, 2014; Dadas *et al.*, 2016) following 3.8%, 0.81%, 29.1% and 0.74%, respectively. Since wild boars were associated with feeding of earthworms, beetles, bugs and numerous larvae which functioned intermediate or paratenic hosts for various helminthic fauna (Coobs and Spinger, 1974) and also habitat overlap and competition for food also contribute the higher prevalence of *Strongyloides* in animals (Ezenwa, 2002).

*Metastrongylus* is a genus of nematodes of the family Metastrongyloidae, usually found as lungworms in pigs and sometimes causing parasitic bronchitis. The life cycle is indirect. Infection only occurs where pigs have access to earthworms. The present prevalence was 12%, which was similar to the report of (Mudim *et al.*, 2004) who had revealed the prevalence of 12.6%. However, the report from (Ineson, 1953; Yagoob *et al.*, 2014; Mansouri *et al.*, 2016)) indicates the higher prevalence rate of 66.6%, 34% and 68%, respectively. The difference in prevalence rate might be due to the improper geographical distribution of different earthworm species which form part of diet of wild boars and act as intermediate hosts for these parasites. *Stephanurus* sp. (kidneyworm) is a *strongyloide* nematode of pigs. The prevalence of *Stephanurus* sp. was 44%. The high prevalence rate might be due to the high persistant rainfall and high humidity.

In the study, the eggs were doubted either as *Hyostrongylus* sp. or *Oesophagostomum* sp. Therfore, those eggs were considered as *Strongyle* group. Both of these worms have direct life cycle. The prevalence of *Strongyle* sp. in the present study was 49% which was higher to the earlier reported by (Ineson, 1953; Eijck and Borgsteede, 2005; Senlik *et al.*, 2010; Sen *et al.*, 2015). Habitat overlaping, competition for food and overcrowding have a significant effect on *strongyle* abundance (Ezenwa, 2002). In the present study the mixed infection was higher on double infection (50.52%) followed by multiple infection (20%), triple infection (18.94%) and single infection (10.52%). The mixed parasitic infection were more common in wild boar due to common overlaping habitat, its feeding pattern, climatic condition, competition for food and parasite exchange between between domestic and wild boars. The intensity of different parasites in wild boars of CNP was

observed in the study. According to the results, maximum numbers of wild boars were found to be infected with light infection which is asymptomatic condition and cannot cause disease in the animals while less number of wild boars were infected with moderate and heavy infection revealed by *Eimeria* sp., *Stephanurus* sp., *Metastrongylus* sp., *Globocephalus* sp., *Strongyloides* sp., *Strongyle* sp. The heavy and moderate infection is symptomatic and can cause serious diseases in animals.

### 6. CONCLUSION AND RECOMMENDATIONS

#### 6.1 Conclusion

The present study showed that overall prevalence of gastro-intestinal parasites of wild boar (Sus scrofa) was 95% with higher prevalence rates of protozoa (85%) followed by nematodes (80%) and trematode (12%) but no any cestode were recorded during the study period. Nine different parasitic species were revealed in wild boar of CNP such as *Eimeria* sp. (with or without) micropyle among protozoan; *Fasciola* sp. among trematode and Strongyloides sp., Stephanurus sp., Metastrongylus sp., Ascaris sp., Trichuris sp., Globocephaus sp. and Strongyle sp. among nematodes. Out of all these identified gastrointestinal parasites, *Eimeria* sp. without micropyle (70%) showed higher prevalence followed by Strongyloides sp. (56%), Strongyle sp.(49%), Stephanurus sp. (44%), *Eimeria* sp. with micropyle (40%), *Globocephalus* sp.(38%), *Metastrongylus* sp. (12%), Fasciola sp. (12%), Ascaris sp. (7%) and Trichuris sp. (6%). In mixed infection, double infection showed the highest rate 48 (50.52%) followed by triple infection 18 (18.94%) and single infection 10 (10.52%). Similarly, in intensity of parasitic load maximum number of samples revealed the light intensity followed by mild, moderate and heavy intensity. The results indicated the highly significant difference in the prevalence of parasites on examination samples of wild boar. Damages associated with the presence of parasites in wild boars are difficult to estimate. This problem requires a systematic and continuous investigation in the future. Damages resulting from reduced weight, low fertility, Burdening of the immune system and increased susceptibility to other infectious diseases can be assessed directly or indirectly. In that respect, development of an appropriate methodology adjusted to our conditions is necessary for determining the actual damage in the segment of the ecosystem and improving the wild life welfare. Though there are some studied of gastrointestinal parasites that have been done in wild and captive but this is the first attempt of study of gastrointestinal parasites of wild boar in wild condition in CNP.

#### 6.2 Recommendations

On the basis of conclusion the following recommendations are made to reduce the risk of Gastro-intestinal parasites to wild boar.

- ) Molecular techniques should be applied for accurate identification of parasites upto species level.
- ) National parks should establish veterinary laboratory and gastrointestinal parasites should not be neglected by the conservation biologist because they are one of the main threats for wild animals.
- ) Seasonal study of parsitic prevalence in wild boar should be conducted to know the prevalence of parasites in a season-wise pattern.
- ) Gastro-intestinal parasites have shown intense infection in wild boar but these parasites have a high possibility of infecting other wild animals as well. Therefore, similar studies should be carried out in other wild animals also.
- ) National parks should be strictly prohibited for domestic animals since they are the major source of infection to wild animals.

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# ANNEX - 1

Table 5: Size of eggs/ cysts/oocysts of different Gastro - intestinal parasites of wild boar.

Name of Parasites	Size in um		Remarks
i ui usites	Length	Breadth	
<i>Eimeria</i> with micropyle sp.	24um-30um	18um-22um	Oval in shape
<i>Eimeria</i> without micropyle sp.	22um - 28um	16um - 20um	Sporulated oocysts have four sporocysts with two sporozoites each.
Ascaris sp.	50 - 70 um	40 - 60um	Thick walled, golden brown, thin inner yolk membrane, uneven lumpy aliminous outer wall unsegmented, granular contents.
Fasciola sp.	130-150 um	63 - 90 um	Oval, has operculum at one end, unsegmented, bile stained
Trichuris sp.	50 - 68 um	21 - 31 um	Thick shell, two transparent polar plugs unsegmented, brownish granular contents.
<i>Metastrongylus</i> sp.	51 - 63 um	32 - 42 um	Wall thick, deep grey with slightly wrinkled surface.
<i>Globocephalus</i> sp.	55 - 63 um	38 - 45 um	Ovoid, thin membrane 4-8 cells when shed with feces.
Strongyle sp.	50 - 85 um	26 - 47 um	Wall thin, smooth colorless, slightly to strongly barrel shaped, blastomeres present.
Strongyloides sp.	40 - 55 um	20 - 30 um	Single cell, grayish green short, thick L1 larva present.
<i>Stephanurus</i> sp.	90 - 114 um	53 - 70 um	Wall thin, transparent, blastomeres present.