1. Introduction

1.1 Background

Every animal is equally important in an ecosystem. Some of the species has already been extinct while some are listed in the critically endangered, vulnerable, rare, threaten, protected etc. in global and national status. Animals are generally lost from their natural habitat due to several reasons like; habitat loss, natural disaster, low food quantity etc. It is necessary to protect them in their natural habitat or to control their haphazard use by people, each country has wildlife conservation policy by establishing protected area such as national parks, wildlife reserves, conservation areas etc. In Nepal, there are 10 national parks, three wildlife reserves and six conservation areas. Most of the endangered, vulnerable and rare animals have been protected in the protected areas. Besides the protected areas, in many parts of the country forest has been owned by the community and has been protecting many wild animals.

In Illam, forest is being protected in the form of community forest. People use their community forest to continue their daily life. They mainly use forest for plantation of medicinal plants, grazing of livestock, to cut leaves and branches for firewood or fodder for cattle. Besides protection of medicinal plants and other, the community forest protecting several wildlife species including Red Panda.

Wildlife is getting declined due to several reasons. One of the important reason could be sharing the same pasture areas with the domesticated animals. Sharing the pasture land influence the sharing various dreadful diseases between them. People of Illam had made livestock shed in various inside the community forest which are the important habitat of wild animals including Red Panda as well. Choyatar Community Forest of Jamuna VDC and National Forest of Jamuna and Mabu VDCs of Illam of Nepal are also among them, where Red Panda has been recorded. The forests are also popular for high altitude medicinal plants.

Community peoples practice livestocks grazing inside community forest showed that on the whole day, livestock grazed in forest and bring back to animal shed during night. In summer the livestock sheds were shifted at the high altitude to protect livestocks from high temperature and shifted to the lower altitude during winter because most of the hills remain covered by snow.

1.2 Red Panda (*Ailurus fulgens* Cuvier, 1825)

The Red Panda (*Ailurus fulgens* Cuvier, 1825) is the one living species of family aeluridae (Shrestha 2015) and commonly called Cat bear, Panda bear, Lesser panda etc. In Nepal, it is commonly called "Habre", in local languages "Naututoo" and "Pude Kundo" in different part of communities. There are two sub species of the genera that is *Ailurus fulgens fulgens and Ailurus fulgens styani*. *Ailurus fulgens fulgens* is smaller and lighter than *Ailurus fulgens styani.* Red Panda is an endemic and a flagship species in the Himalayan region (Roberts and Gittleman 1984) which threatened with extinction worldwide (Wang et al. 2008). It inhabits in the temperate zone at low temperatures (Yonzon 1989) in the countries of the Himalayan Mountain Range which includes Nepal, India, Bhutan, Myanmar and China (Roberts and Gittleman 1984). Red Panda is an arboreal, shy, nocturnal and solitary animal however sometime seen in a pair or group and travel a linear distance of 1.57 Km during breeding season and active only in the late in the afternoon and/or early evening hours (Yonzon and Hunter 1991a). The Red Panda resides in evergreen, deciduous, and mixed forests with dense bamboo covered (Roberts and Gittleman 1984, Wei et al. 1999, Choudhury 2001, Pradhan et al. 2001). Despite being a member of the order Carnivora, Red panda is a specialized herbivore with a low nutrient diet. More than 86% of its diet includes ringal bamboo (Pradhan et al. 2001, Yonzon & Hunter 1991), which has forced the animal to inhabit in narrow range of forest types and restricted geographic area (Yonzon 1989, Choudhury 2001, Pradhan et al. 2001b, Sharma and Belant 2009). Because of the specialized diet and narrow range of habitat, Red panda has been considered as an indicator species of ecosystem health in eastern Himalayan broadleaved and conifer forests (Yonzon et al. 2000). Present estimation of the population density and the total area of potential habitat suggest that the global population of Red Pandas are declining and exist currently < 10,000 individuals world wide (Wang et al. 2008). The IUCN classifies the Red Panda as vulnerable status, suggesting a likely extinction globally if conservation measures are not initiated soon (Wang et al. 2008) and by Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES) in its Appendix I (species threatened with extinction which are or may be affected by trade). In Nepal the National Parks and Wildlife

Conservation Act, 1973 has defined this species as an endangered species (Jnawali et al. 2011) and suggested immediate research on identifying existing and potential habitat, and population status for its future conservation.

On the basis of habitat suitability index, Nepal is home to approximately 1.9% of total global population of Red Panda and 38% potential habitat of Red panda covered by protected areas where as remaining 62% by community managed and national forest but the most of the area of Nepal the population conservation of Red Panda is not known (DNPWC 2010). According to Population and Habitat Viability Assessment (PHVA 2010), 317 possible confirmed in 592.39 km^2 habitat and within 3244.52 km^2 including another potential habitat of 2652.13 km^2 the estimated population to be 582 individuals. Recently Red Panda has been reported from Khotang, Jajarkot and Kalikot district (Bista and Paudel 2015) but need to be confirmed scientifically.

Table 1: Population estimation of Red Panda in Nepal

S.N	**Sub populations**	**Area (km2)**		**Population**	
		Confirmed	**Possible**	**Confirmed**	**Possible**
1.	Annapurnna-Manaslu	4.18	84.23	2	20.15
2.	Darchula				
3.	Dhorpatan	89.05	434.92	43	104.05
4.	Gaurishankar	45.17	114.15	22	27.31
5.	Kanchanjungha	111.91	160.76	67	48.13
6.	Khaptad	3.57	211.22	1	36.42
7.	Langtang	47.83	125.7	23	30.07
8.	Rara	55.63	1099.16	19	189.51
9.	Sagarmatha	73.71	150.96	44	45.2
10.	Sakhuwasabha East	101.88	119.01	61	35.63
11.	Sakhuwasabha West	59.46	152.02	36	45.51
	Total	592.	2652.13	317	581.98

Source: Population and Habitat Viability Assessment (PHVA), 2010

Currently, Red Pandas are believed to occur at low densities with a patchy distribution due to habitat fragmentation, loss of foraging habitat, human and livestock disturbances, poaching, and disease (Yonzon and Hunter 1991b, Wei et al. 1999, Choudhury 2001, Patterson-Kane et al. 2009, Sharma and Belant 2009, Dorji et al. 2012, Sharma et al. 2014) while parasitic infection was the one of disease in Panda (Zhang et al. 2007). These are the most serious threats for population decline (Thomas 2002), and thus the loss of genetic diversity (Li et al. 2005, Wei et al. 1999).

1.3. Gastrointestinal parasites in Red panda

Gastrointestinal parasites are known to widespread in vertebrate animals. Parasitic investigation is important for management of animal and animal diseases in domestic and wild animals. Parasitic infection in animals as well as human negatively impact on body weight gain, quality of reproduction due to loss of appetite, nutrient uptake and utilization (Gross et al. 1999) even death in wild animals (Rao and Acharjyo 1984, Hansen and Perry 1994). The variety of infectious and non-infectious diseases in animals are usually due to parasitic origin (Iqbaln et al. 2000, Akhter & Arshad 2006, Siddiki et al. 2010). Parasitism is a universal problem of livestock leading to lower productivity (Hossain et al. 2011) and even death in wild animals (Rao and Acharjyo 1984). By using same grazing site by domestic and wild animals, there are chances of cross- transmission of parasites between wild to domestic or vice-versa (Hoberg and Brooks 2008, Agosta et al. 2010). A regular surveillance and control measures based on correct diagnosis, effective treatment and proper prophylaxis would certainly reverse the situation (Varadharajan and Pythal 1999). The better understanding of cross- transmission help to make management policy for both wild and domestic animals in different geographical area (Morgan et al. 2005). Historically, study of parasites in wild animals have been descriptive account (Thapa 2013, Chaudhary 2014 and Shrestha 2015) rather than clinical effect that the parasites cause i.e. morbidity and mortality. In addition, a few studies have been done regarding cross- transmission of parasites between domestic and wild animals (Boomker et al. 1987, Horak et al. 2004, Van Wyk and Boomker 2011).

Infectious disease theory (Funk et al. 2001) and mathematical models (de Castro and Bolker 2005) predicted that threatened or endangered host populations should harbor fewer parasites. A recent empirical study (Altizer et al. 2007) showed that threatened primates had fewer helminths, protozoa, and viruses than non threatened species. Low parasite richness intuitively seems to be positive for endangered wildlife species (Smith et al. 2009). However, the loss of parasites could be unhealthy for an ecosystem (Hudson et al. 2002) and lead to a loss of genetic variation in immunity and increased susceptibility (Smith et al. 2009).

Currently, Red Panda have been reported to be infected by various intestinal and lung parasites such as: protozoan parasites including*;* *Entamoeba* sp, and *Eimeria* sp (Shrestha 2015) and helminth parasites including: *Baylisascaris, Angiostrongylus, Trichuris* (Lama et al. 2015 and Shrestha 2015), *Strongyloides* sp., Hookworm, *Metastrongylus* sp., *Moniezia* sp. (Shrestha 2015)

1.4 Livestock

Being the Agriculture country (CBS 2001) livestock farming in Nepal is considered as the second major economic activity next to crop Agriculture. It contributes 11% of nation GDP (Gross Domestic Product) and 28% of Agricultural GDP (Shakya 2009). From the multipurpose household survey (Sakya 2009) showed that the total household income, livestock sources contribute 21.2% in the mountain, 19.7% in hill and 9.7% in the terai.

Livestock farming generally provide wool, high quality food (meat and milk), hides, skin, motive power for agricultural operative and transport; the organic manure which is necessary for soil fertility and fuel for farm household are also provide by livestock. Various livestock species, cattle are prominent (47.5%) followed by buffalo (12.3%), goat (15.8%) and sheep (10.4%) (Maharjan 2009). In Nepal consumption of meat, milk, and their products are traditional (Shakya 2009) and annual production are ; milk 1158780 metricton and meat 98895 metricton including buff 12749 metricton, mutton 2823 metricton and goat 18584 metricton (Maharjan 2009).

Due to the gastrointestinal parasite, it affects the production of milk and meat quality (Krecek and Waller 2006, Miller et al. 1998). GIT parasite infection causes direct losses due to drop in production and death of animal and indirect economic losses due to increased cost of control strategies (Soulsby 1982), especially in small ruminants in tropics and subtropics (Perry et al. 2002). In Nepal, parasitic disease is a major problem to livestock and about 24% of deaths in goat were reported to be due to intestinal parasites and total economic losses were 25% (Lohani and Rasai1995). The consequences of GIT parasites infection include reduced feed intake and weight gain, immunity, lower fertility, a reduction in milk production and work capacity and death in critical infection (Fikru et al. 2006).

1.5. Gastrointestinal Parasites of livestocks

All the animals are not equally susceptible to infectious diseases, some has high resistance power and some has low for same infection and cause quick transmission from the animal which has high resistance power to low resistance. Such sort of communicable disease causes serious infection to other (Maharjan 2009). If the transmission of such types of diseases in animals could be reduced it will be good achievement for food safety (Maharjan 2009) and Conservation Area (Chaudhary 2014 and Thapa 2013). To break the transmission cycle, it is necessary to understand, transmission process, effective diagnostic tool, treatment and preventive measure and finally the availability of clear surrounding environment before targeting the disease.

Livestock are more susceptible for various parasitic diseases, currently, livestock are infected with various protozoan parasitic disease such as: *Eimeria* sp., *Entaboeba* sp *Balantidium coli* and *Giardia* sp. (Kanyari et al. 2009, Byanju et al. 2011), nematode: *Trichostrongylus* sp., *Strongyloides* sp., Hook worm, *Ascaris* sp. (Dhakal 2008, Byanju et al. 2011, Neupane 2012), cestode; *Moniezia* sp., *Taenia* sp. *and Dipylidium* sp., (Dhakal 2008, Byanju et al. 2011), trematode*; Paramphiostomum* sp., *Dicrocoelium* sp., *Fasiola* sp., *Schistosoma* sp. and *Amphiostomum* sp. (Dhakal 2008, Kajuria et al. 2013).

1.6 Problem statement and justification

Red Panda is vulnerable species which are indicator of Himalayan mountain ecosystem and global warming (Wang et al. 2008). There is an estimated population of less than 10,000 mature individuals worldwide (Wang et al. 2008) among them about 314 individuals lie in Nepal (Bista and Paudel et al. 2015). The mortality rate of Red Panda is high and one of the causes may be due to parasitic disease (Yonzon 1989). The Red Panda is one of the least studied mammalian species due to the occurrence in high altitude, dwelling in dense forest, arboreal characters, and elusive and nocturnal behavior (Roberts and Gittleman 1984). These factors were also complemented by inefficient government policy, park regulations and political instability (Baral and Heinen 2005). The study on ecology habitat and food analysis has been carried out by different researcher but least research has been carried out about parasitic infection on Red Panda.

1.7 Objectives

1.7.1 General objective

To determine the distribution of gastrointestinal parasites of Red Panda and livestocks in community forest of Illam.

1.7.2 Specific objectives

1. To determine the prevalence of intestinal parasites of Red Panda.
2. To determine the prevalence of intestinal parasites of livestocks.
3. To determine the probable sharing of intestinal parasites in between Red Panda and livestocks**.**

2. LITERATURE REVIEW

Parasites are cosmopolitan in distribution infecting human, domestic and wild animals. Large number of researches on parasitic infection have been carried out in human and domestic animals while studies on parasitic infection in wild animals are scanty. Red Panda is a wild carnivore but adapted to herbivore food habit. Due to this reason, possibility of parasitic infection in Red Panda belonging to carnivore as well as herbivore animals.

2.1 Global scenario of gastrointestinal parasites of Red Panda and other wild carnivore.

Taxonomically Red Pandas belong to carnivora and phylogenetically related to other carnivores including; Giant panda, Raccoons, Bear, Jackals etc. Hence probability of parasites of Red Panda are similar with those carnivore. Few studies have been carried out on parasitic infection in Red Panda. Red Panda has been found to be infected with gastrointestinal as well as parasites of lungs and heart.

Lungs worm have been reported to be dreatful parasites causing pneumonia in Red Panda. A group of lung worms infecting Red Panda includes metastrongyloid group which are: *Angiostrongylus* sp., *Crenosoma* sp.. The metastrongyloid nematode *Angiostrongylus vasorum* has been reported in Red Pandas (*Ailurus fulgens fulgens*) from European zoos (Jensen et al. 2005, Grondahl et al. 2005, Bertelsen et al. 2010). In the similar study, this species was also reported in a 9 year old male, captive Red Panda from urban Zoo in the United Kingdom (Janet et al. 2009). Similarly, in a coprological survey of Red Pandas from different zoos in Europe, 2.6% Red Pandas were found to be infected with *A. vasorum*, 4.3% with *Crenosoma* and 27.8% Red Pandas with previously unidentified metastrongyloid species but morphologically distinct from *A. vasorum* as the most prevalent (Bertelsen et al. 2010). From Aalborg Zoo, Denmark Willesen et al. (2012) examined four captive-breed Red Pandas and found infected with a newly discovered metastrongyloid nematode. Other cases of *Angiostrongylus* sp. and Crenosomatidae had been reported from 52 Red Pandas from the National Zoos of America (Montali et al. 1984).

Out of 115 faecal samples of Red Panda examined in Padmaja Naidu Himalayan Zoological Park (PNHZP), India, seven Pandas has been reported to be infected with endoparasites (Pradhan et al. 2011) and identified genera includes protozoan parasites; *Trichomonas* sp., trematode; *Schistosoma* sp. and nematode; *Ascaris* sp. Another trematode *Ogmocotyle ailuri* was previously reported from Red Panda (*A. fulgens*) of National Zoological Park in the United States (Price 1954).

In USA two neonatal Red Pandas have been reports to be infected with *Sarcocystic* spp. infection and concluded vertical transmission of this parasites by histopathological, immunohistochemical, molecular and ultrastructural diagnosis (Zoll et al. 2015).

Chowdhary (2001) has reported *Toxocara transfuga* and hookworm species from Red Pandas from Indian Subcontinent. Similarly Neiffer et al. (2002) confirmed heartworm *Dirofilaria* sp. infection, presumably with *D. immitis* in Red Panda (*A. fulgens fulgens*) from North America and China (Lan et al. 2012). *Toxoplasma gondii* has also been reported in Red Panda (*A. fulgens fulgens*) from America (Sikarskie et al. 1991).

Baylisascaris spp. are widely distributed in the Giant Panda (*Ailuropoda melanoleuca*), Red Panda (*Ailurus fulgens*), raccoon (*Procyon lotor*), Ursid species (*Ursus maritimus*, *Ursus arctos pruinosus*, *Selenartos thibetanus mupinensis* and *Ursus arctos lasiotus*) and other mammals including humans (Sato et al. 2004, Zhang et al. 2011, Xie et al. 2011, Xie et al. 2013).The nematode *Baylisascaris shroederi* is the intestinal parasite of giant pandas, and can cause intestinal obstruction, inflammation, and death (Zhang and Wei, 2006, Hu 2001) while transmission of parasites depends on direct contact between the host and ingestion of free-living infective stages or penetration of free-living stages present in the environment (Karamon et al. 2013). Many food or water resources can become polluted because pandas defecate while feeding (Zhang and Wei 2006). Zhou et al. (2013) and Zhang et al. (2011) investigated the infection of *B. schroederi* in faecal samples of Giant panda (*Ailuropoda melanoleuca*) in China using PCR.

B. procyonis was reported from North America (Logiudice 2003), New York (Kidder et al.1989), Portland(Yeitz et al. 2009), North Carolina (Hernandez et al. 2012) Western Maryland (Ainsley et al. 2012), Illinois (Birch et al. 1994), Germany (Baeur and Gey 2002, Kazacos 2001, Miyashita 1993, Sato et al. 2001), Canada (Sexsmith et al. 2009), West Virginia (Kazacos 2001, Souza et al. 2009) and Poland (Karamon et al. 2013) from

Raccoons, in giant panda (Yang 1998, Zhang 2008, He 2009, Xie et al. 2011, Yang and Zhang 2013), in human (Huff et al. 1984, Park et al. 2000) and in Red Panda (Xie et al. 2011and Pradhan et al. 2011).

A study carried out in Illinois by Adams et al. (1981) and they showed that reccoons was infected with Coccidian parasites *Eimeria nuttalli* oocysts, *E. procyonis* oocysts and sporocysts of *Sarcocystis* sp., Similarly, Dubey (1982) surveyed in Columbus and he found *Baylisascaris procyonis*, trichurid, capillarid, trichostrongyloid and *Eimeria procyonis* oocysts in raccoons. In the similar study, *Eimeria procyonis*-like oocysts were reported in 15 of 15 captive raccoons and endogenous coccidian stages (large and small schizonts) in 6 of 6 juvenile wild-caught raccoons (Dubey et al. 2000). Foster et al. (2004) reported *Eimeria procyonis* (84%), *Eimeria nuttalli* (10%), 1 unidentified species of *Eimeria* (3%) and an unidentified species of *Sarcosystis* (3%) from adult raccoons (*Procyon lotor*) in U.S.A. One nematode worm; *Baylisacaris procyonis* and one unidentified tapeworm has been reported in road-killed Raccoons from West- Maryland (Ainsley et al. 2012).

Kelley (2006) reported 72.2% prevalence of cestode in raccoons (*Procyon lotor*) from Texas including *Atriotaenia procyonis*, *Mesocestoides* spp., and the immature species of Taeniidae. In the similar study, Kelley et al. (2008) reported cestodes from 35 raccoons (*Procyon lotor*) in North Central Texas, including *Atriotaenia procyonis*, *Mesocestoides* sp. and immature *Taenia pisiformis*. Another study carried out in North Central Texas, showed, other species of trematode parasites in raccoons (*Procyon lotor*), the mammalian Schistosome *Heterobilharzia americana* with prevalence rate of 47.2% (Kelley 2010). Among 19 raccoons captured, 8 species of helminths including five nematodes (*Physaloptera rara*, *Placoconus lotoris*, *Molineus barbatus*, *Baylisascaris procyonis, Toxoascaris procyonis*); one acanthocephalan (*Macrocanthorhynchus ingens*), and two cestodes (*Atriotaenia procyonis* and *Mesocestoides lineatus*) were reported in raccoon (*Procyon lotor*) from Duval County, Texas.

Kresta et al. 2009 examined 590 faecal samples of Raccoons in Texas and identified 20 different helminthes. Among them; 13 nematodes (*Physaloptera rara, Gnathostoma procyonis, Gongylonema sp. Placoconus lotoris, Molineus barbatus, Toxascaris leonine, Baylisascaris procyonis, Lagochilascaris sp., Synhimanthus sp, Aonchotheca putorii,* *Pharyngodon* sp.,*Syphacia* sp.,and *Cruzia americana*), two Cestoda (*Atriotaenia procyonis, Mesocestoides lineatus*), two Acanthocephala (*Macracanthorhynchui ingens* ,*Moniliformis* sp.) and three Trematoda (*Pharyngostomoides* sp., *Heterobilharzia* sp.,*Grysoma singularis*). Birch et al. (1994) were found Six species of helminths: four species of nematodes (*Arthrocephalus lotoris, Physaloptera rara, Gnathostoma procyonis,* and *Baylisascaris procyonis*); one species of cestode (*Mesocestoides variabilis*); and one species of acanthocephalan (*Macracanthorhynchus ingens*). Michael et al. (1999) collected samples from April 1997 through April 1998 of 128 raccoons (*Procyon lotor*) from 7 sites representing 4 physiographic areas in South Carolina were examined for gastrointestinal helminth parasites and they found four species of nematodes (*Gnathostoma procyonis*, *Physaloptera rara*, *Arthrocephahis lotoris* and *Molineus barhatus*) and 2 species of acanthocephalans (*Macracanthorhynchus ingens* and *Centrorhynchus conspectus*).

2.2 National scenario of gastrointestinal parasites of Red Panda and other wild ruminents.

In Nepal, most of the investigations have been performed in ecological basis like ecology, behaviour, and population status of Red Panda in different protected area like National Parks, Conservation Areas and Hunting Reserves (Yonzon 1989, Yonzon and Hunter 1991, Sharma and Belant 2009 and Sharma and Belant 2010). Less numbers of research have been carried out in out of protected area. In the national context, only two studies have been done in gastro-intestinal parasites of Red Panda (Shrestha 2015 and Lama et al. 2015) from RNP and KBCA respectively.

Lama et al. (2015) studied the intestinal parasitic infections in free-ranging Red Panda (*Ailurus fulgens*) from Kothi Bhir Community Area (KBCA), Rolpa, Nepal and showed 100% prevalence of protozoan parasites including; Coccidian parasites and some unidentified species while nematodes includes; *Trichuris* sp., *Baylisascaris* sp., *Angiostrongylus* sp. and one unidentified species and in case of cestode and trematode eggs were present but species has been identified. Shrestha (2015) carried out study on gastrointestinal parasites in Red Panda (Ailurus fulgens) in RNP, Nepal and found 93% Red Panda were infected with either protozoan or helminth or both. Altogether 12 parasitic genera were identified in Red panda and these were; *Entamoeba* and *Eimeria* as protozoan; Hook Worm, *Trichuris, Strongyloides, Baylisascaris, Toxoasacaris, Oxyuris, Crenosoma, Angiostrongylus, Metastrongylus* as nematodes and *Moniezia* as cestode but no trematode were recorded.

Being the group carnivore, Red Panda is herbivores in food habitat and parasite of Red Panda may be similar with ruminants because most of the parasites transmitted through food. In Nepal, few investigations have been carried out in wild ruminants.

Thapa (2012) carried out a survey of gastrointestinal infection of parasites in Himalayan Tahr and Barking Deer in Rara National Park and reported altogether 10 genera of intestinal parasites which includes: *Eimeria* sp., *Oxyuris* sp., *Strongyloides* sp., *Ascaris* sp., *Trichostrongylus* sp., *Dictyocalus* sp., *Mullerius* sp., *Haemonchus* sp., and *Moniezia* sp., in Himalayan Tahr while in Barking Deer, *Eimeria* sp. *Oxyuris* sp. *Ascaris* sp. *Trichuris* sp. *Dictyocalus* sp., *Haemonchus* sp. and *Moniezia* sp. have been reported.
Chaudhary (2014) carried out a survey of the gastrointestinal parasites in Krisnasar (*Antilope cervicapra*) of Blackbuck conservation area, Bardia and Shuklaphanta wildlife reserve, Kanchanpur, Nepal and recorded *Entamoeba* sp. and *Eimeria* sp. as a protozoan parasites and among helminth parasites *Oxyuris* sp., *Strongyloides* spp., *Ascaris* sp., *Trichostrongylus* sp, *Haemonchus* sp., *Trichuris* sp. and *Bunostomum* sp. as a nematodes; *Paramphistomum* sp., *Schistosoma* sp. and *Fasiola* sp. as a trematode, and *Moniezia* sp. as a cestode.

2.3 Gastrointestinal parasites of livestocks

Livestock are important animals for economic point of view but generally infected with several types of gastrointestinal parasites eg; protozoan and helminth parasites. GI parasites are the major problems in livestock management due to high mortality, weak body condition, low productivity, high cost of drugs (Rajakaruna and Warnakulasooriya 2011). Due to unmanaged farming system and sharing pasture land parasites of one animal can easily transmit to other.

Cattles are important domesticated large ruminants in terms of agricultural and economic aspects. Cattle are found to be infected with various intestinal parasites. Common intestinal parasites of cattle includes; protozoan parasite: *Entamoeba* sp., *Balantidium* sp., *Giardia* sp., *Isospora* sp., *Emeria* sp. (Sathaporn et al. 2011, Biu et al. 2009, Paul et al.2010, Pfukenyi et al. 2007, Ntonifor 2013). Similarly the cattle were reported to be infected with helminth parasites. Among helminth, nematodes species are more common and reported species are; *Strongyle* sp., *Strongyloides* sp., *Trichostrongylus* sp., *Oesophagostomum* sp., *Haemonchus* sp., *Bunostomum* sp., *Ascaris* sp., *Capilaria* sp., *Ostertagia* sp., *Trichuris* sp., *Cooperia* sp., *Toxocara* sp., *Chabertia* sp., *Avitellina* sp.,*Nematodirus* sp., *Dictyocaulus* sp., while cestode; *Taenia* sp., *Moniezia* sp., and among trematode; *Fasiola* sp., *Paramphiostom* sp., *Schistosoma* sp., *Amphiostomum* sp. (Hook et al. 2004, Frooq et al. 2012, Chowdhury et al. 1993, Masood and Majid 1989, Bilal et al. 2009, Mersha 2012, Rajakaruna and Warnakulasooriya 2011, Jeon et al. 2010).

Goat and sheep are considered as a small ruminants and rear in mixed farm for wool, milk, meat by farmer. Due to mixed farming system cross transmission of parasites between goat and sheep are possible. The most common parasites reported in goat and sheep includes: *Entamoeba* sp., *Balantidium* sp., *Giardia* sp., *Isospora* sp., *Emeria* sp. (Diaz et al. 2011, Ntonifor et al. 2013, Bui et al. 2009, Kajuria et al. 2013) among protozoan parasites; *Strongyle* sp., *Strongyloides* sp. *Trichostrongylus* sp., *Oesophagostomum* sp., *Haemonchus* sp., *Bunostomum* sp., *Ascaris* sp., *Capilaria* sp., *Ostertagia* sp., *Trichuris* sp., *Cooperia* sp., *Toxocara* sp., *Necator* sp., *Chabertia* sp., *Avitellina* sp.,*Nematodirus* sp., *Dictyocaulus* sp., among nematodes; *Taenia* sp., *Moniezia* sp., among cestodes; *Fasiola* sp., *Paramphiostom* sp., *Schistosoma* sp., *Dicrocoelium* sp., *Amphiostomum* sp., and among trematodes (Kanyari et al. 2011, Basir 2009, Neupane 2012, Tefera et al. 2009, Ikpeze and Nzemaka 2009, Nwake et al. 2015, Bandyopadhyay et al. 2010, Yadav and Tandon 1989).

Yak (*Poephagus gruenniens)* is a multipurpose animal which provides meat, milk, hair, wool and hair as well as play vital role in agricultural, transportation and rural economies of the people. People practice free ranging and migratory grazing system of Yak and during winter they share grazing land with cattle as well as parasites too (Goswami et al. 2013) while infection of parasites causes degradation of health and reduce production of milk, meat, hair (Waller 2002). Some reported GI protozoan and helminth parasites in Yak from different countries includes; *Emeria* sp., *Coccyx* sp , *Haemonchus* sp., *Nematodirus* sp., *Cooperia* spp., *Strongyle* sp. *Moniezia* spp., *Nematodirus* sp., *Trichuris* sp. *Fasiola* sp., *Paramphistomum* sp., *Amphiostomum* sp., *Ascaris* sp., *Trichuris* sp., *Dicrocoelium* spp., (Byanju et al 2011, Bandyopadhyay et al. 2010, Hui Dong et al. 2012, Shrestha and Bindari 2013, Kuchai et al. 2010).

Horses are important animal for transportation in the resources poor communities where road facilities are not available. Beside that horses are used in agriculture for secondary and tertiary cultivation land preparation (Abaynen et al. 2002) and recently they are used for sporting. Horses are prone to number of infectious parasitic diseases and reported parasites of horse includes: *Parascaris* sp., *Strongyloides* sp., *Fasciola* sp., *Oxyuris* sp., *Trichuris* sp., *Dicrocoelium* sp. *Eimeria* sp. *Trichonema* sp., *Triodontrophorus* sp., *Grastrodiscus* sp., *Habronema* sp., *Strongyloides* sp., *Dictyocaulus* sp., *Trichostrongylus* sp., *Cyathostome* sp., *Triodontophorus* sp., *Strongylus* sp cestode ; *Anoplocephala magna* (Uslu and Guclu 2007, Lem et al. 2012, Pam et al. 2013, Berhanu et al. 2014, Tesfu et al. 2014).

2.4 Parasite sharing between wild and domestic animals

Transmission of infectious diseases between wild and domestic animals are becoming major issaue in global (Gortazar et al. 2007). Being the same group, same food habitat or sharing same pasture land domestic and wild animals may be sharing same types of parasites. Sharing of the similar types of parasites by different animals are the neglected subject in society due to low economic condition and lack of knowedge (Pedesen et al. 2005). Transmission of the infectious disease is due to the rising interaction between the different species (Gortazar et al. 2007). Non host specific or multihost parasites are sharing between the wild and domestic animal as a host due to sharing same ecological factors (Abal and Torger 2013) and intermediate arthopode host (Otranto et al. 2015). The investigation conducted by Pence et al. (2003) has been showed that the transmission of parasites between Cat and Ocetot (*Leopardus pardalis*) was possible because similar types of cyst, eggs and larva were recorded despite different in size. In another study by using PCR method, it has been showed that the transmission of protozoan parasite (Neospora caninum) between livestock and wild animals were possible (Gondium et al. 2004).

3. MATERIALS AND METHODS

3.1 Study Area

The study area Illam district is a hilly district (26°54'0" north and 87°56'0"east) of Mechi zone of Nepal’s eastern development region. It lies in the western slop of the Singhalila ridge, adjacent to Singhalila National Park of Darjeeling; India and South of Kanchanjunga conservation Area; Nepal. This area forms one of the five Prioritized landscape of the eastern Himalayas, the eastern Himalayan broadleaf and conifer Global 200 eco region (Olson and Dinerstein 1998) and Himalayan Bio diversity hot sport (Myers et al. 2000). This Area is important for Red Panda mainly because this provides a biological linkage between two protected Area, Kanchanjunga Conservation Area in Nepal and Singalila National Park in India. The elevation of Illam is 300m to 4000m. Red panda has been recorded from Jogmai, Jamuna, Mabu, Puwamajhuwa, Maipokhari, Maimajhuwa and Pyang VDCs of Illam. The present study area covers Chhintapu and Dhanepa community forest of Maimajhuwa VDC, National forest of Mabu and Jamuna VDC and Kalikhop and Dadeli Community forest of Jogmai VDCs.

Illam is rich in bio-diversity. Different varieties of vegetation have been identified in the area, including forests of *Pinus roxburghii, Schima-Castanopsis, Alnus, Pinus wallichiana*, *Pinus patula, Rhododendron, Quercus lanata* and Temperate mountain oak forest (W.W.F and ICIMOD 2001). Different fauna found in this area includes: Common leopard (*Panthera pardus*), Clouded leopard (*Neofelis nebulosa*), Leopard cat (*Prionaulurus benghalensis*), Wild boar (*Sus scrofa*), Yellow throated martin (*Mattes flavigula*), Assamese macaque (*Macaca assamensis*), Barkimg deer (*Muntiacus muntijak*) and Red Panda (*Ailurus flulgens*) (Kandel 2009). The major attraction Illam for the researches includes abundant availability of rare birds and Mammals including Red panda.

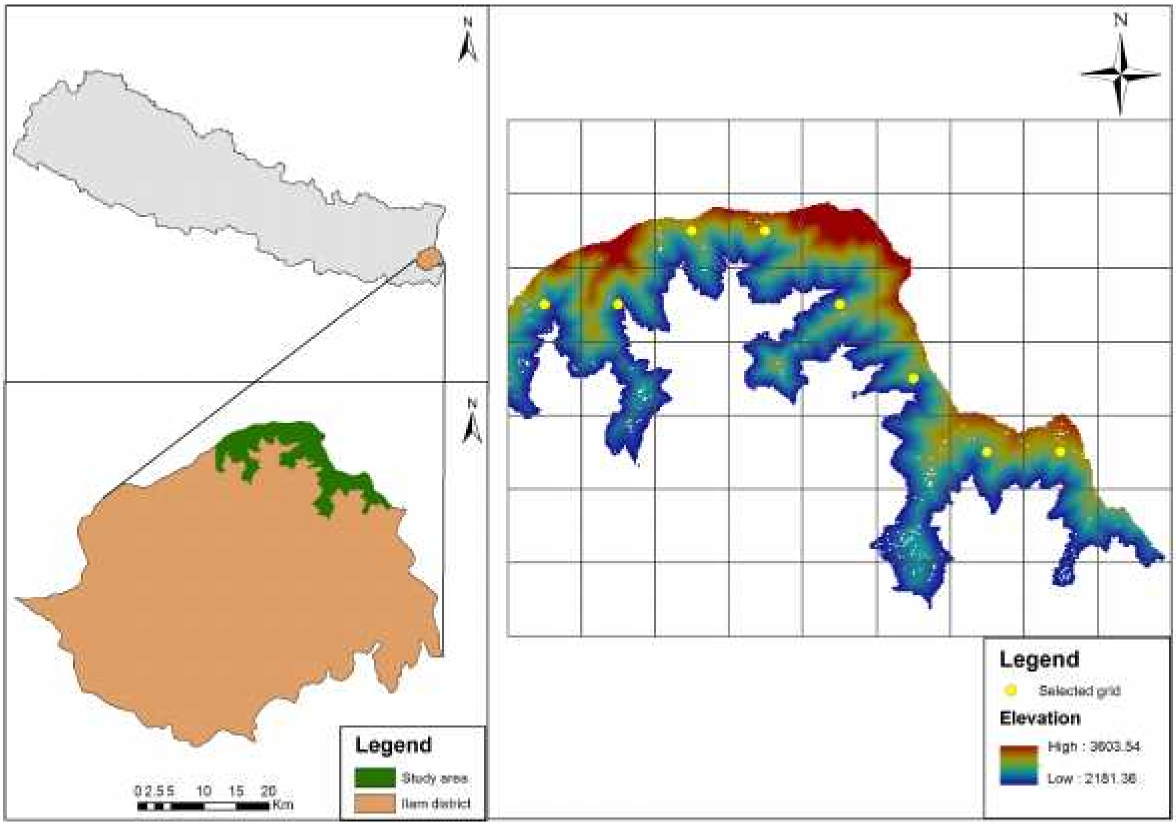


Fig 1: Map of Nepal including Illam district showing study area

3.2 Materials

During the research the materials used have been listed below:

3.2.1 Materials for field

i. Sterile vials ii. GPS iii. Binacular iv. Camera

v. Verner caliber vi. Globes

3.2.2 Materials for Laboratory

i. Beaker ii. Motor/Pistle iii. Glass rod iv.Globes

v. Slides vi. Cover slips vii. Volumetric flask

viii. Droppers ix. Tea strainer x. Measuring cylinder xi. Toothpicks

xii. Niddle xiii. Centrifuge Machine xiv. Centrifuge tube

xv. Electric balance xvi. Cotton xvii. Electric microscope

xviii. Gloves xix. Stage micrometer xx. Mask

xxi. Ocular-micrometer xxii. Refrigerator

3.3 Chemicals

i. Potassium dichromate ($K_2Cr_2O_7$)
ii. Distilled water (D/W)
iii. Saturated NaCl solution
iv. Methylene blue
v. Lugol's Iodine solution

3.4 Study Design

The present study was designed to assess the intestinal parasitic infection in Red Panda and livestock in Illam community forest. The study includes;

a) Selection of animal habitat by GIS system.
b) Collection of fresh pellet samples in sterile vials from transects.
c) Preservation of faecal samples in 2.5% Potassium dichromate solution.
d) Examination of faecal samples by using flotation, sedimentation and Stoll's counting technique.
e) Identification and measurement of eggs/cysts of different parasites.

3.4.1 Sample collection method

Using GIS system possible habitat of Red Panda was selected, for this elevation was selected from 2200m to 4800m because Red Panda was recorded from 2200 to 4800m altitude (Roberts and Gittleman 2009) and prepared grids of size 1.7×1.7 (2.89) km^2, because the average home range of Red Panda is 2.89 Km^2 (Yonzon 1989, Yonzon et al. 1991) to reduce the chances of duplicate samples from the same individuals . From the selected area eight grids were randomly selected and in each grid two vertical transects were made. First transect was 500m apart from starting point of grid and second transect was 700m apart from first transect. Samples were collected from the established transects (Burnham et al. 1980) and to increase sample size, we repeated collection of samples from same transect or grid and opportunistically while walking from one transect line to another. Fresh faecal samples encountered during study period were measured using verner caliber, different pellets are likely from different individuals. Livestock samples were collected immediately after they were defecated, and each sample was visually confirmed to be from different individuals. Approximately 20 gm of faecal samples of both Red Panda and livestock were collected on either side of 2 m apart of each transect and also the opportunistically from the trees branches.

3.4.2 Preservation of samples

Collected faecal samples of Red Panda and livestock were preserved in 2.5% Potassium dichromate that help in maintaining the morphology of protozoan parasites and preventing further development of some helminthic eggs and larvae.

3.4.3 Sample size

From the study area 55 samples were collected. Out of them:

Table 2: Proportion of faecal samples collected from Illam district.

Common Name	**Scientific Name**	**Collected Sample (%)**
Red panda	*Ailurus fulgens*	25.45 %
Cow	*Bos* sp.	36.36 %
Goat	*Capra hircus*	16.36 %
Horse	*Equus caballus*	9.1 %
Sheep	*Ovis aries*	7.27 %
Yak	*Bos grunniens*	3.63 %
Buffalo	*Bubalus bubalis*	1.82 %

These samples were transported to a laboratory of the Central Department of Zoology, Tribhuvan University, Kathmandu in preservatives and refrigerated and was conducted following techniques for the identification of eggs and oocysts present in feces.

Photos of Field activities:

Photo 1: Questioning with local people

Photo 2: Scat of Red Panda

Photo 3: Measuring the Red Panda scat

Photo 4: Collecting of scat of Red Panda

Photo 5: Collection of pellet of Sheep

Photo 6: Collection of dung of Cow

Photo 7: Fresh and dry scat of Red Panda

Photo 8: Yak in the habitat of Red Panda

Photo 9: Defecating pellet by Goats

Photo 10 : Cattle in the habitat of Red Panda

Lab work Photos:

Photo 11: Running the centrifuge mechain

Photo 12: Preparation of Slide

Photo 13: Microscopic observation of slide

3.5 Microscopic Examination

All the samples were examined at the laboratory of Central Department of Zoology, Tribhuwan University, Kirtipur. The samples were processed for microscopic examination. The ova/oocysts/cyst and larvae of different parasites were identified according to the morphology and quantitative estimation by using saline and iodine wet mount and concentration method (flotation and sedimentation) and Stoll's counting technique to determined mix infection and intensity of parasites(Soulsby 1982).

3.5.1 Saline wet mount

Small quantity (about 2mg) of faeces was mixed in a drop of saline placed on a clean slide. Any grass fiber or particles were removed and covered with coverslip. The smear was examined under microscope at 10X and 40X (Soulsby 1982).

3.5.2 Iodine wet mount

It was done by emulsifing the facal samples in a drop of Lugol's Iodine solution on a slide covered with a clean coverslip and examined under microscope as above.

3.5.3 Concentration method

Eggs/cysts are often low number in faeces that they are difficult to be detected in direct smears or mounts. Therefore, this procedures were performed which includes flotation and sedimentation techniques (Soulsby 1982).

a. Differential Floatation Technique

Nematode and cestode eggs present in Red Pandas and livestock feces is detected through this technique. This technique ensures the egg float in the floatation liquid, which helpes to identify the eggs.

Approximately 3 gm of faecal sample was taken in a beaker and added 20 ml of water then the sample was grinded lightly with the help of motor and pistle and filter the solution by tea strainer. The filtrate solution was poured into a centrifuge tube of 15 ml and centrifuged at 1000 rpm for 5 minutes. The tube's water was replaced with saturated sodium chloride solution and again centrifuged.

After centrifuge more saturated sodium chloride solution was added to develop convex surface at the top of the tube and one drop of methylene blue (to stained) where a cover slip can be placed for a few minutes and then cover slip was removed and placed on a 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.

b. Sedimentation

This technique is used for the detection of trematode eggs. It provides good results as the eggs of the trematode are bit heaver than the other, where sediments of centrifuged contents is taken for eggs detection (Veterinary Lab. Tecniques 2003).

Saturated salt solution was removed gently from the test tube after examined the flotation portion and pure the sediment content into the watch glass and stirred the content gently to mixed it. One drop from the mixture was taken to prepared a second slid. The specimen was stained with iodine wetmounts solution.

In this way two slides were prepared from one sample (one from flotation and one from sedimentation) were examined under 10X and 40X of microscope to detect eggs of helminthes and trophozoites or cyst of gastrointestinal protozoans.

c. Egg cyst and larva size measurement

- by using ocular and stage micrometer

- length and breath measured by calibration

d. Egg, cyst and larva identification

- on the basis of shape and size along with published literatures (Barutzki and Schaper 2009, Bhir 1998, Villeneure 2013, Brianti et al. 2012).

3.6 Data Analysis

Feacal flotation and sedimentation procedures yielded similar results to parasite identified. Therefore presence/absence information reflects combined results from both method. So to analyzed the data MS Excel 2007 and SPSS 19 were used.

4. Results

4.1. Gastro-intestinal parasites of Red Panda

A total of 14 fresh faecal samples of Red Panda were collected by using line transect technique from the community forest of Illam and examined by using direct smear and concentration methods for gastro-intestinal parasites detection. All the samples were found to be positive for intestinal parasites.

4.1.1. Protozoan parasites

Red Panda were found to be infected with protozoan parasites belonging to three classes (Table: 3). Among them Coccidian parasite showed the highest prevalence (64.28%) followed by *Entamoeba* sp. (57.14%) and *Balantidium* sp. (14.28%). Study revealed the existence of several species of Coccidian parasites, which were grouped into two broad groups*; Eimeria* with micropyle and without micropyle. The Red Panda were found to be almost equally infected with both groups of this parasites.

Table 3: Prevalence of Protozoan parasite of Red Panda from Illam community forest

S.N	Class	Name of Parasite	Prevalence Rate (%)
1.	**Sarcodina**	*Entamoeba* sp.	8(57.14%)
2.	**Sporozoa**	*Eimeria* with micropyle	9(64.28%)
		Eimeria without micropyle	8(57.14%)
3.	**Litostomatea**	*Balantidium* sp.	2(14.28%)

4.1.2. Helminth Parasites

Among the helminth parasites, Trematode and Cestode parasites were not observed from Red Panda. But they were found to be highly infected with nematode parasites belonging to seven different genera. Among them *Oxyuris* sp. showed highest prevalence rate (100%) followed by *Ascaris* sp. (57.14%), *Trichostrongylus* sp. (50%), *Strongyloides* sp. (50%), *Crenosoma* sp. (42.85%), *Trichuris* sp. (42.85%) and Hook worm (35.71%).

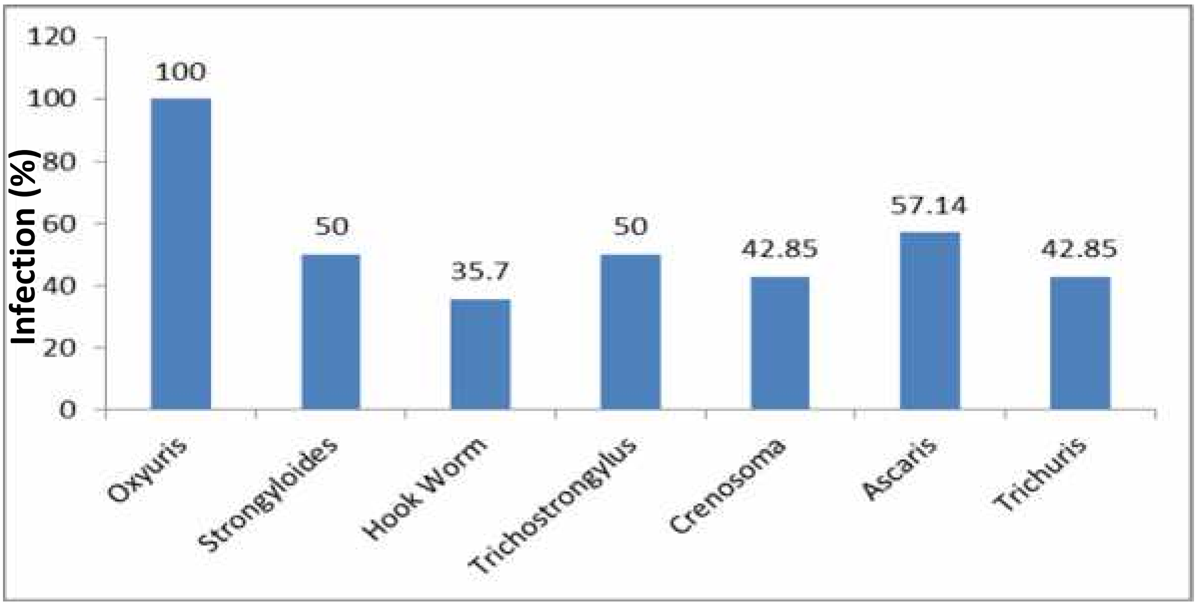


Fig 2: Prevalence of nematode in Red Panda

4.1.3. Mixed infection in Red Panda

Red Panda were found to be infected with different gastro-intestinal parasites. All 14 samples of Red Panda revealed mixed infection with 3-5 different genera in each sample. Among the 14 samples examined, multiple and triple infection found to be (78.57%) and (21.42%) respectively. No single and double infection were observed in this study (Figure 2).

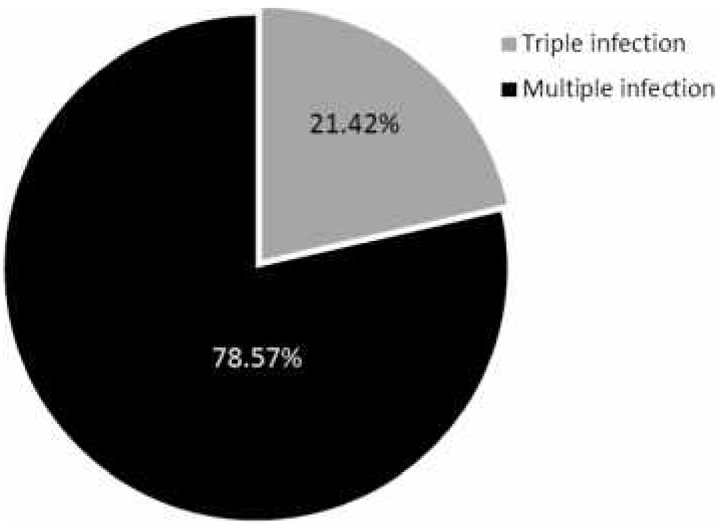


Fig 3: Mixed infection on Red panda

4.1.4. Intensity of parasitic infection

Heavy parasitic infection was considered in those samples which has six or more ova or oocyst observed per field. Maximum five samples of Red Panda showed heavily infected with Coccidian parasite; *Eimeria*. While two each samples showed high intensity of *Oxyuris*, *Trichostrongylus* and *Ascaris* and four samples with *Oxyuris,* three samples with Coccidian and two each samples showed moderate intensity of *Crenosoma* and *Trichostrongyloid* which considered 4-6 ova/oocyst per field. Maximum six samples showed mild infection with Oxyuris followed by four each samples with *Entamoeba* and *Eimeria* without micropyle, which considered 2-4 ova/oocyst per field and <2 ova/oocyst observed per field considered light infection; maximum four sample each showed light intensity of *Strongyloides* and *Trichuris,* three samples each with *Entamoeba, Cenosoma and Ascaris*.

Table 4: Intensity of infection of intestinal parasite in Red Panda

S.N	Class	Name of Parasite	+	++	+++	++++
1.	**Sarcodina**	*Entamoeba* sp.	3	4	1	-
2.	**Sporozoa**	*Eimeria* with micropyle	-	1	3	5
		Eimeria without micropyle	1	4	2	-
3.	**Litostomatea**	*Balantidium* sp.	2	-	-	-
4.	**Nematoda**	*Oxyuris* sp.	2	6	4	2
5.		*Strongyloides* sp.	4	2	1	-
6.		Hook worm	2	2	1	-
7.		*Crenosoma* sp.	3	1	2	-
8.		*Trichostrongylus* sp.	2	1	2	2
9.		*Ascaris* sp.	3	2	1	2
10.		*Trichuris* sp.	4	1	1	-

4.2 Gastro-intestinal parasite of livestock

Fresh faecal samples of domestic animals were collected from the line transect technique from community forest of Illam. By this technique total 41 samples of domestic animals were collected including 20, 9, 5, 4, 2, 1 samples of cow, goat, horse, sheep, yak, buffalo respectively. All the samples tested were found to be positive for either protozoan or helminth parasites.

4.2.1 Protozoan parasite

The result revealed that cows and horse were infected maximum with Coccidian parasite followed by Sarcodina and Litostomatea. Similar result was observed in case of goat, sheep and yak but Sarcodina and Litostomatea were absent. In case of buffalo, prevalence of litostomatea was absent but other three genera were present out of four protozoan genera (Table: 5).

Table 5: Prevalence of Protozoan parasites of Livestock from Illam community forest

Protozoan parasites	**Cow (n=20)**	**Goat (n=9)**	**Horse (n=5)**	**Sheep (n=4)**	**Yak (n=2)**	**Buffalo (n=1)**	**Total (n=41)**
Sarcodina *Entamoeba* sp.	6(30%)	0	2(40%)	0	0	1	9(21.95%)
Sporozoa *Eimeria* with micropyle	9(45%)	4(44.44%)	2(40%)	3(75%)	1	1	20(48.78 %)
Eimeria without micropyle	12(60%)	5(55.55%)	3(60%)	3(75%)	1	1	25(60.97 %)
Litostomatea Balantidium sp.	2(10%)	0	1(20%)	0	0	0	3(7.31%)

4.2.2 Helminth Parasites

Domestic animals of Illam which share the same grazing area of Red Panda habitat were found to be infected with all three classes of helminth parasites; nematode, cestode and trematodes.

4.2.2.1 Nematode

Domestic animals of Illam community forest were infected with six genera of nematode parasites. Among them *Oxyuris* sp. (90%) showed the highest prevalence in cows followed by *Ascaris* sp. (55%), each of 50% prevalence by *Strongyloides* sp. and *Trichostrongylus* sp., Hook Worm (45%) and Trichuris sp. (40%). In case of goat, *Oxyuris* sp. (88.88%) also showed highest prevalence followed by 66.66% prevalence by *Strongyloid* sp. and *Ascaris* sp. and almost similar by *Trichuris* sp., Hook Worm and *Trichostrongylus* sp. Equal prevalence (80%) were showed by *Oxyuris* sp., *Strongyloides* sp., *Ascaris* sp. but *Trichuris* sp. and *Trichostrongylus* sp. were absent in case of horse while Hook Worm showed 20% prevalence rate. The rate of prevalence of *Oxyuris* sp. and *Trichostrongylus* sp. were high in case of sheep but *Strongyloides* sp., *Trichuris* sp., Hook Worm and *Ascaris* sp. showed almost similar prevalence (Fig: 3).

Table 6: Prevalence of Nematode parasites in livestock

Nematode Parasites	Cow(n=20)	Goat(n=9)	Horse(n=5)	Sheep(n=4)
***Oxyuris* sp.**	18(90%)	8(88.88%)	4(80%)	3(75%)
***Strongyloid* sp.**	10(50%)	6(66.66%)	4(80%)	2(50%)
***Trichuris* sp.**	8(40%)	4(44.44%)	0	1(25%)
Hook worm	9(45%)	3(33.33%)	1(20%)	2(50%)
***Trichostrongylus* sp.**	10(50%)	3(33.33%)	0	3(75%)
***Ascaris* sp.**	11(55%)	6(66.66%)	4(80%)	1(25%)

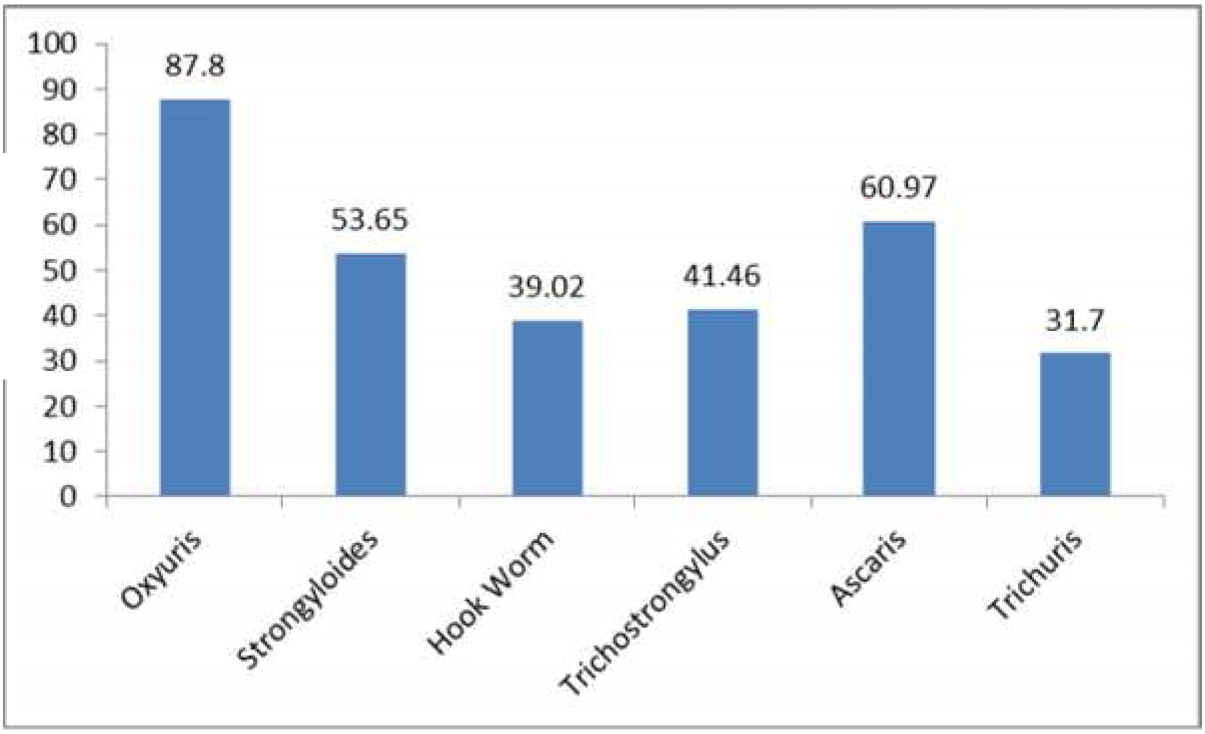


Fig 4: Nematode parasitic Infection in Livestock

In general, 87.8% domestic animals weree infected with *Oxyuris* sp. which was the highest prevalence than any other nematode followed by *Ascaris* sp. (60.97%), *Strongyloid* sp. (53.65%), *Trichostrongylus* sp. (41.46%), Hook Worm (39.02%) and *Trichuris* sp. (31.70%) (Fig: 3).

4.2.2.2 Cestode and Trematode

Only one genera of Cestode *i.e Moniezia* and one genera of trematode *i.e Paramphiostome* sp. were observed during the study and prevalence of *Monizia* sp. and *Paramphiostomum* sp. was found to be 14.63% and 2.43% respectively.

Table 7: Infection of Cestode and trematodes on Livestock

Class	**Parasite**	**Prevalence (%)**
Cestode	*Moniezia* sp.	6(14.63%)
Trematode	*Paramphiostomum* sp.	1(2.43%)

4.2.3 Mixed infection

During the study, different types of parasitic infections were encountered in livestock. Multiple infection (73.17%) was found to be highest followed by triple (19.51%) and double (7.31%) infection but no single infection was encountered.

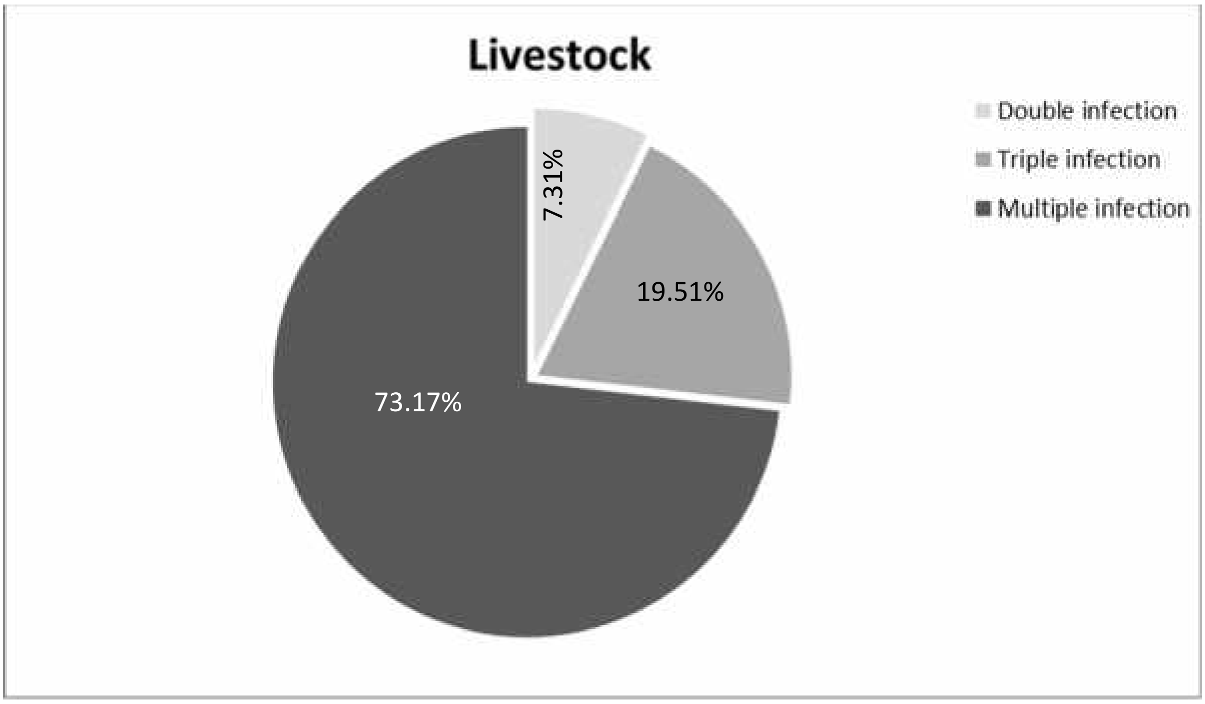


Fig 5: Mixed infection in Livestock

4.3 Comparison of GI parasites in between Red Panda and Livestock.

As a comparison of GIT parasites of Red Panda and livestock, protozoan and nematode showed 100% prevalence rate in Red Panda while 87.8% and 82.92% in case of Livestock. Cestode and trematode are absent in Red Panda but 2.43% and 14.63% prevalence rate in case of livestock. So prevalence of nematode and protozoan were equal in Red Panda while nematode was slightly greater than protozoan in case of livestock.

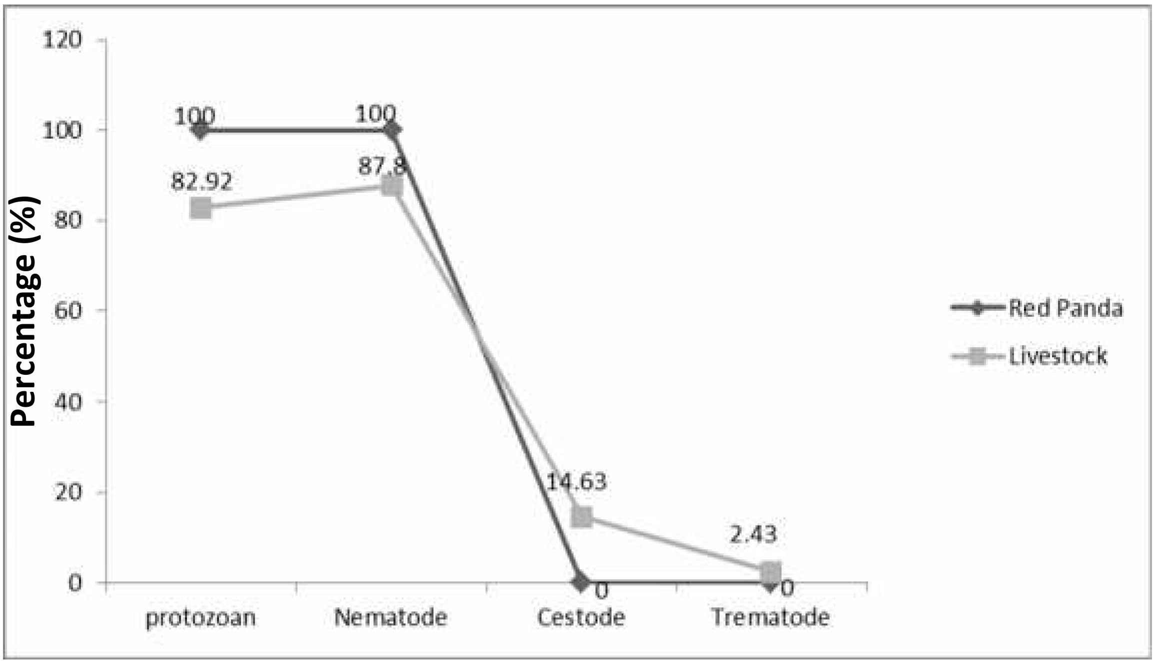


Fig 6: Comparison of GI parasites in between Red Panda and Livestock

4.3.1 Prevalence of protozoan parasites in between Red Panda and Livestock

The protozoan parasites in Red Panda and Livestock were compared. Among four protozoan parasites encountered, *Entamoeba* sp. and *Balantidimum* sp. were found to be most prevalence in Red Panda than livestock. While with regard to *Eimeria* sp., *Eimeria* with micropyle was comparatively higher in Red panda but *Eimeria* without micropyle was almost similar in livestock and Red Panda. Statistically there was not significant difference in protozoan infection between Red Panda and livestock *i.e.* (P= 0.709, X2= 0.625 at d.f.=1). Species wise association showed that there is statistically significant difference in prevalence of Entamoiba (X^2= 6.052, P= 0.014 at d.f.= 1). While association of *Eimeria* spp. and *Balantidium* sp. in Red Panda and Livestock were not significant difference (Table: 8).

4.3.2 Nematode parasites in between Red Panda Livestock

During the study seven different nematode parasitic infection were reported. *Oxyuris, Trichostrongylus* and *Trichuris* showed highest prevalence in Red Panda than Livestock and the case was just reverse for *Strongyloides,* Hook Worm and *Ascaris.*While *Crenosoma* was only reported in Red Panda. Nematode parasitic infection was statistically not significant difference in between Red Panda and Livestock i.e. (P=0.314, X^2=1.878 at d.f. =1). Species wise association between Red Panda and livestock of Illam showed that, although the prevalence rate of *Oxyuris* sp. was high in Red Panda, statistically there was not significant difference observed (Table: 8) but in case of Hook Worm, *Trichuris* sp., *Ascaris* sp. and *Trichostrongylus* sp. were almost equally infected in both groups except *Crenosoma* sp. which is dreadful long parasite.

Table 8: Comparative analysis of GI parasites in between Red Panda and livestock

S.N	**Class**	**Parasites**	**Prevalence in Red Panda**	**Prevalence in livestock**	**X^2-Value**	**P-Value**
1.	**Sarcodina**	*Entamoeba* sp.	8(57.14%)	9(21.95%)	6.052	0.014
2.	**Sporozoa**	*Eimeria* with micropyle	9(64.28%)	20(48.28%)	1.007	0.316
		Eimeria without micropyle	8(57.14%)	25(60.97%)	0.064	0.800
3.	**Litostomatea**	*Balantidium* sp.	2(14.28%)	3(7.31%)	0.613	0.592*
4.	**Nematoda**	*Oxyuris* sp.	14(100%)	36(87.8%)	1.878	0.314*
5.		*Strongyloides* sp.	7(50%)	22(53.65%)	0.056	0.813
6.		Hook worm	5(35.7%)	16(39.02%)	0.048	0.826
7.		*Trichostrongylus* sp.	7(50%)	17(41.46%)	0.309	0.578
8.		*Crenosoma* sp.	6(42.85%)	0	19.723	0.000*
9.		*Ascaris* sp.	8(57.14%)	25(60.975)	0.064	0.800
10.		*Trichuris* sp.	6(42.85%)	7(31.70%)	0.574	0.449
11.	**Cestode**	*Moniezia* sp.	0(0%)	6(14.63%)	_	-
12.	**Trematode**	*Paramphiostomum* sp.	0(0%)	1(2.43%)	-	-

*= Fisher exact test accepted due to less than five expected value

From the analysis it showed that the transmission of parasites between Red Panda and livestock were possible. To know the parasites sharing groups with Red Panda, livestocks were categories into cattle and goat/sheep. Morphologically similar parasitic egg/ cyst and larvae were compared.

4.3.3 Comparison of GI parasites in between Red Panda and Cattle

Prevalence of GIT parasites in between Red Panda and Cattle were also compared. Incase of protozoan parasites, highest prevalence rate showed by *Eimeria* with micropyle (64.28%) and (47.61%) in Red Panda and Cattle respectively while *Entamoeba* sp., *Eimeria* without micropyle and *Balantidium* sp. showed almost equal prevalence rate in Red Panda and cattle. Statistically, there was not significant difference in prevalence of all intestinal protozoan parasites between Red Panda and Cattle (Table: 9).

Among the seven genera of nematode, 100% and 90.47% prevalence rate showed by *Oxyuris* sp. in Red Panda and cattle respectively. *Ascaris* sp. showed equal and *Strongyloides* sp., Hook worm, *Trichostrongylus* sp. and *Trichuris* sp. Showed almost equal prevalence rate in Red Panda and cattle. Statistically, there was not significant difference in prevalence of intestinal nematode in between Red Panda and Cattle (Table: 9).

Table 9: Prevalence of gastro-intestinal parasites in Red Panda and cattle

S.N	**Class**	**Parasites**	**Prevalence in Red Panda**	**Prevalence in cattle**	**X^2-Value**	**P-value**
1.	**Sarcodina**	*Entamoeba* sp.	8(57.14%)	7(53.84%)	1.944	0.163
2.	**Sporozoa**	*Eimeria* with micropyle	9(64.28%)	10(47.61%)	0.940	0.332
		Eimeria without micropyle	8(57.14%)	13(61.53%)	0.079	0.778
3	**Litostimatidea**	*Balantidium* sp.	2(14.28%)	2(9.52%)	0.188	1.00*
4	**Nematoda**	*Oxyuris* sp.	14(100%)	19(90.47%)	1.414	0.506*
5		*Strongyloides* sp.	7(50%)	10(47.61%)	0.019	0.890
6.		Hook worm	5(35.7%)	9(42.85%)	0.179	0.673
7		*Trichostrongylus*	7(50%)	11(52.38%)	0.19	0.778

		sp.				
8.		*Ascaris* sp.	8(57.14%)	12(57.14%)	0.00	1.00
9.		*Trichuris* sp.	7(42.85%)	8(38.09%)	0.079	0.778
10.		*Crenosoma* sp.	6(42.85%)	0	1.414	0.506*

*= Fisher exact test accepted due to less than five expected value

4.3.4 Comparison of GI parasites in between Red Panda and Goat/Sheep

In the comparison of prevalence of protozoan parasites in between Red Panda and Goat and Sheep; among the four protozoan parasites, *Eimeria* with micropyle (64.28%) showed the highest prevalence rate followed by *Eimeria* without micropyle and *Entamoeba* sp. showed equal rate and *Balantidium* sp. in case of Red panda and like Red Panda *Eimeria* without micropyle (61.53%) also dominant in Goat and Sheep followed by *Eimeria* with micropyle but *Entamoeba* sp. and *Balantidium* sp. was absent. Statistically, there was no significant difference in prevalence of GIT protozoan parasites (*Eimeria* with micropyle without micropyle and *Balantidium* sp.) where as significant difference in prevalence of *Entamoeb*a sp. (Table: 10).

Incase of nematode genera, prevalence rate of Red Panda was same that mentioned previously and incase of Goat and Sheep, *Oxyuris* (84.62%) also showed the highest prevalence rate and followed by *Strongyloides*, *Ascaris*, *Trichostrongylus*, Hook worm and *Trichuris* where as *Crenosoma* sp. was absent. Statistically, there was no significant difference in prevalence of nematode parasites except *Crenosoma* which was significant difference (Table: 10).

Table 10: Prevalence of gastro-intestinal parasites in Red Panda and goat/sheep

S.N	Class	Parasites	Prevalence in Red Panda	Prevalence in Goat and Sheep	X^2-Value	P-value
1.	**Sporozoa**	*Eimeria* with micropyle	8(57.14%)	7(53.84%)	0.304	0.581
		Eimeria without micropyle	9(64.28%)	8(61.53%	0.054	0.581
2	**Sarcodina**	*Entamoeba* sp.	8(57.14%)	0	10.556	0.002*
3	**Litostomatea**	*Balantidium* sp.	2(14.28%)	0	2.006	0.481*
4	**Nematoda**	*Oxyuris* sp.	14(100%)	11(84.61%)	2.326	0.222*
5.		*Strongyloid* sp.	7(50%)	8(61.53%)	0.363	0.547
6.		Hook worm	5(35.71%)	5(38.46%)	0.22	0.883
7.		*Trichostrongylus* sp.	7(50%)	6(46.15%)	0.40	0.842
8.		*Ascaris* sp.	8(57.14%)	7(53.84%)	0.30	0.863
9.		*Trichuris* sp.	6(42.85%)	5(38.46%)	0.054	0.816
10.		*Crenosoma* sp.	6(42.85%)	0	7.163	0.016*

*= Fisher exact test accepted due to less than five expected value

4.3.5 Parasitic Mixed infection in between Red Panda and Livestock

Multiple infection was most prevalent in both Red Panda and Livestock but slightly high in Red Panda than Livestock and followed by triple infection which also slightly increase in Red Panda than Livestock. Single and double infection were not observed in Red panda while double infection was encountered in livestock (Fig:6).

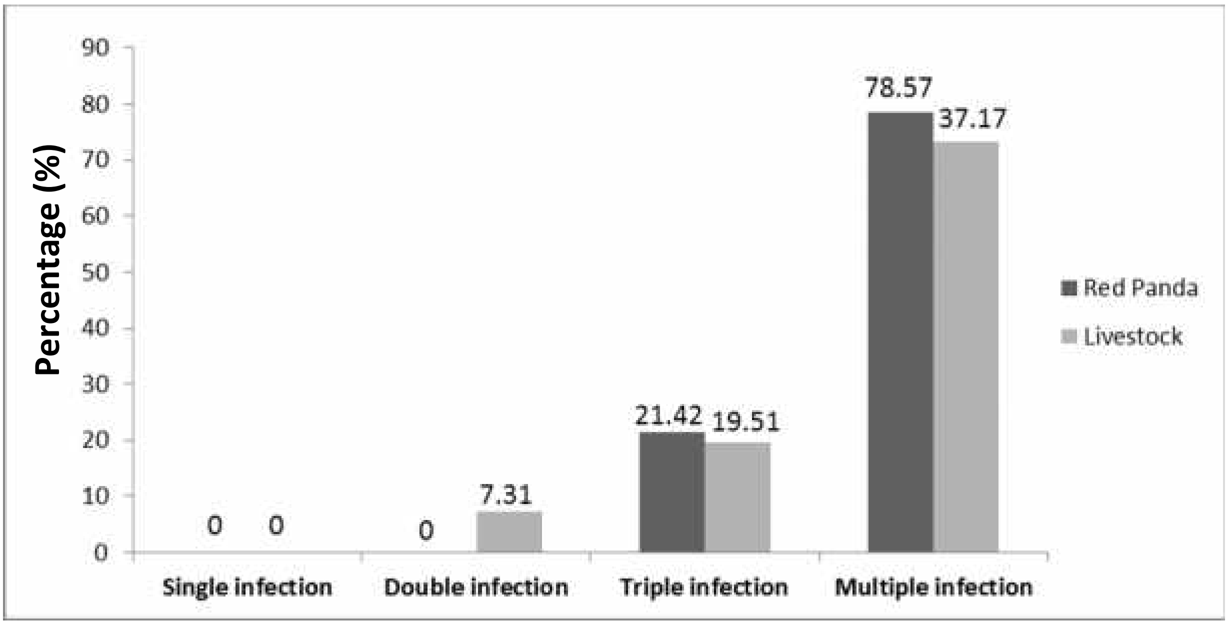


Fig: 7 Comparative analysis of mixed infection in between Red Panda and livestock

The prevalence of trematode and cestode were not statistically compared due to low rate and absence in Red Panda.

The common parasitic genera of Red Panda and livestock showed higher prevalence compared to host-specific genera, which suggesting that cross-transmission of parasites between the livestock and Red Panda was possible.

4.4 Diameter of Eggs/Cysts of Different GI Parasites of Red Panda and Livestock

In the present study, the diameter (length by with) of eggs/cysts of different gastrointestinal parasites were measured which is given below;

- ***Eimeria* sp.**

  Diameter of oocyst of *Eimeria* sp. was 35.5 ± 19.5 µm according to without micropyle while 27 ± µm with micropyle. Eggs are smsll in size, pink in colour and contain morula which is located centrally or sub-centrally filled up. Micropyle occurs in one side.

- ***Entamoeba* sp.**

  Cysts are small, rounded or spherical in shapes having four nucleus and measure 20 ± 6 µm in diameter.

- ***Balantidium* sp.**

  The trophozoid is oval shape, present of cilia, kidney shaped macronucleus and measures 30 ± 10µm × 20 ± 5µm while cyst are circular and measures 25 ± 7µm.

- **Hook Worm**

  Ova have a thin hyaline shell bluntly rounded at the ends. They usually are in the 4-8 cells stages and developing ovum fill the shell and measures 65 ± 10 µm ×30 ± 5µm.

- ***Trichostrongylus* sp.**

  An egge are 90 ± 10 × 48 ± 12 µm in size, oval or kidney bean shaped with thine and transparent outer shell and wrinkled inner membrane, bilaterally symentrical, colourless, central mass usually in 8- 12 cell stages or multisegmented and varies from 16-32 in numbers. The space between the egg shell and embryonic mass is relatively conspicuous. One side is more rounded than other or somewhat both sides rounded.

- ***Ascaris* sp.**

  Eggs are 31.5 ± 7.5 µm in size, nearly spherical, yellowish brown, granular contents and unsegmented, thick aleveolated albuminous shell.

- ***Strongyloides* sp.**

  Eggs are small, measure 130 ± 30 × 52.5 ± 7.5 µm in size, oval with rounded edges or ellipsoidal, thin shelled and contain fully developed larvae that can be seen under low power.

- ***Trichuris* sp.**

  An eggs are in 55 × 30 µm size, contains unsegmented embryo, brown in colour, barrel shaped with a transparent plug at either pole.

- ***Oxyuris* sp.**

  Eggs are bean and oval shaped having segmented embryonic mass surrounded by stiky fluide and measure 112.5 ± 7.3 × 55.5 ± 19.5 µm in size.

- ***Moniezia* sp.**

  Eggs are triangular or quadriangular in shaped, somewhat irregular having a circular or pear shaped (pyriform) apparatus at one end and measure 45 ± 4.5 µm in size.

- ***Paramphiostomum* sp.**

  The eggs of it is 96 ±5 × 52 ± 7.5 µm in size, operculum in one pole, pale grey or greenish in colour, contains five blastomeres sirrounded by about 50 yolk cells, morula located centrally or somewhat subcentrally.

- ***Crenosoma* sp.**

  Eggs are 0.070 - 0.076 mm long and 0.050 - 0.052 mm wide First-stage larvae are 310 ± 40µm long, and 16 ± 5µm wide These larvae have a slightiy blunt, rounded anterior end, whereas the tail tapers noticeably into a point, with a slight deflection just before the tip.

Comparative photos of Parasites of Red Panda and Livestock

Protozoan parasites:

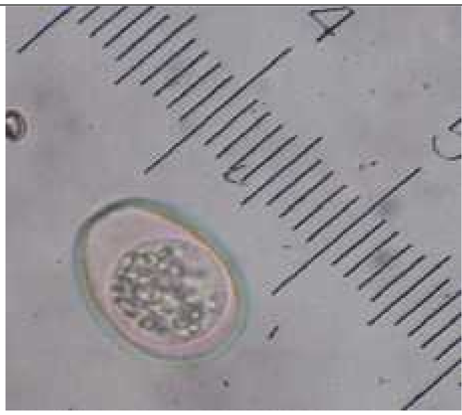
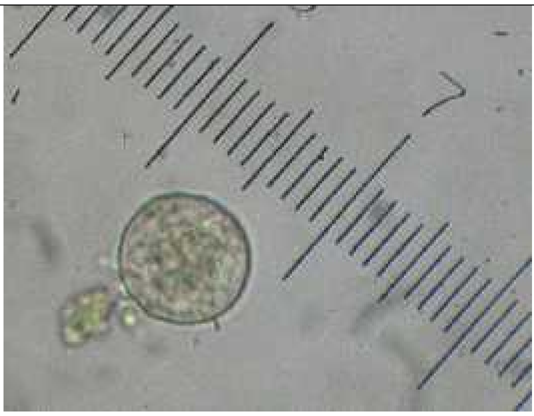
Parasites of Red Panda		Parasites of Livestocks
 Photo 14: *Entamoeba* sp. (18µm)		 Photo 15: *Entamoeba* sp. (18µm)
 Photo 16: *Eimeria* with Micropyle (21µm)		 Photo 17: *Eimeria* with Micropyle (31µm)
 Photo 18: *Eimeria* with out Micropile (21µm)		 Photo 19: *Eimeria* without Micropile (33µm)

 Photo 20: *Balantidium* sp. (30µm)	 Photo 21: *Balantidium* sp. (33µm)

Nematode parasites:

Parasites of Red Panda	Parasites of Livestocks
 Photo 22: *Oxyuris* sp. (105µm)	 Photo 23: *Oxyuris* sp. (69µm)
 Photo 24: *Strongyloides* sp. (155µm)	 Photo 25: *Strongyloides* sp. (135µm)

Photo 26: Larva of *Strongyloides* (245µm)

Photo 27: Larva of *Strongyloides* (270µm)

Photo 28: *Crenosoma* sp. (360µm)

Absent

Photo 29: Hook Worm (45µm)

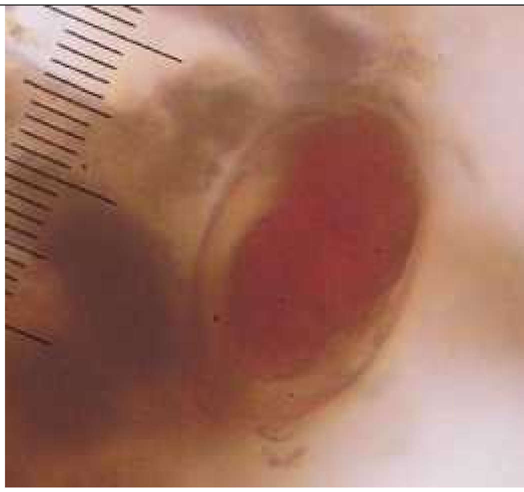


Photo 30: Hook Worm (54µm)

Photo 31: *Trichostrongylus* sp. (96µm)

Photo 32: *Trichostrongylus* sp. (103µm)

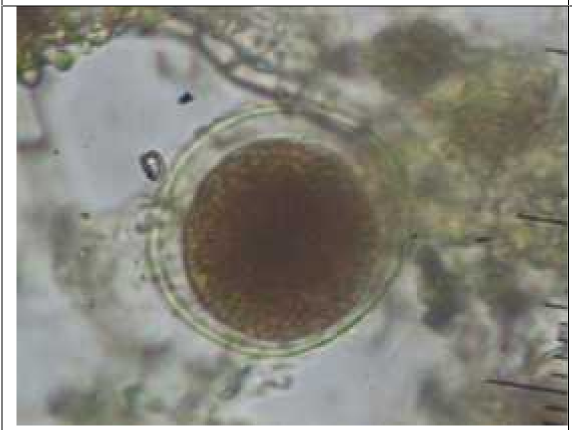


Photo 35: Corticated egg of *Ascaris* (38µm)

Photo 36: Corticated egg of *Ascaris* (31µm)

Photo 37: Decorticated egg of *Ascaris* (72µm)

Photo 38: Decorticated egg of *Ascaris* (91µm)

 Photo 39: *Trichuris* sp. (48µm)		 Photo 40: *Trichuris* sp. (63µm)

Cestode:

Absent		 Photo 41: *Moniezia* sp. (47µm)

Trematodes:

Absent		 Photo 42: *Paramphistomum* sp. (92µm)

5. Discussion

Red Panda inhabits temperate broadleaved forest with bamboo in the understory with an altitudinal range preference of 2200-4000 m with less numbers and nearly extinction (Roberts and Gittleman 2009). Forest fire, livestocks grazing, slash and burn cultivation, timber and fire wood collection, predation by dogs, natural dying of ringal bamboo species, drought, landslide, lack of awareness and diseases are identified as the major conservation threats for Red Panda. The origin of any types of diseases are the parasitic infection. Parasites are transmitted through direct contact or indirect methods (food, water, air etc). Sharing the same pasture mean the sharing the same food, water and air. So sharing the same pasture by wild and domestic animals means may be the sharing the same types of parasites. Livestocks and wild animals are suffering from different types of parasite and they shed parasites by defecation, urination etc. and when another animals come in contact with that area they become infected. Most of the protozoans are not imported and causes poor body condition but helminth parasites can causes death of the animals. Parasites are the neglected topic but imported in conservation aspect.

The present study revealed that high prevalence of gastrointestinal tract (GIT) parasites in both Red Panda and livestock. Both Red Panda and livestock share the same parasite species with different levels of infection. The prevalence of GIT parasites were highest in Red Panda compared to livestock (Farooq et al. 2012).While livestock had shown high parasitic prevalence in compared to Red Panda.

A total of 14 faecal samples of Red Panda and 41 samples of livestock were collected from the community forest of Illam and examined by concentration methods. All the samples of both Red Panda and livestock were found to be positive either for protozoan or helminths. This prevalence rate of Red Panda was almost similar as compared to 93.02% reported in Red panda from Rara National Park (RNP) (Shrestha et al. 2015) and 100% in Kothi Bhir community area (KBCA), Rolpa (Lama et al. 2015). But higher than the reports of Bartelsen et al. (2010) and Pradhan et al. (2011) which showed 35% and 46.66% parasitic infection from European zoo and Darjeeling, India respectively. The prevalence of gastrointestinal parasites of livestock in this study was similar with report of Bandyopadhyay et al. (2010) who reveal 92.4% prevalence rate in India and the prevalence was higher as compared to 81.82%, 28.25%, 66.29% observed by Byanju et al. (2011), Laha et al. (2012) and shirale et al. (2009) respectively. High prevalence of parasites in Red panda and livestock clearly indicates that the parasites shared between them probably due to sharing the same pasture area.

From the economic and sanitary point of view, coccidian parasites are the most prevalent among protozoa. Coccidian parasite infects large number of wild animals including Red Panda and Raccoons. Coccidia are considered to have direct one-host fecal-oral-intestinal life cycles of little biological and public health importance. *Eimeria* is the most common Coccidian prasites among wildlife and livestock. The high prevalence rate of *Eimeria* infection has been reported from various researcher of America from Raccoons which is more closely related to Red Panda. There are two distinct group of *Eimeria* infecting Red Panda with micropyle and without micropyle on the basis of morphological structure. According to previous study, *Eimeria* can't be differentiating into species level since it required culture to identify the species of this parasite. The prevalence of *Eimeria* with micropyle and without micropyle in Illam almost similar with *Eimeria* reported from RNP (Shrestha et al. 2015). High prevalence of *Eimeria* infection has been also reported from Raccoons of America (Dubey et al. 2000, Wright and Gompper 2005, Dubey 1982, Foster et al. 2004, and Adams et al. 1981). Large number of livestock were also showen to be infected (Byanju et al. 2006, Ntonifor et al. 2013, Kanyari et al. 2009, Swai et al. 2006, Bui et al. 2009). In the present study, *Eimeria* was found to be 64.28% which was higher than 35% and 48% observed by Kanyari et al. (2009) in sheep and goat respectively, 17.8% by Laha et al. (2012) and 20.9% by Ntonifor et al. (2013) and lower than 85.7% (Apio et al. 2013) and 76.5% (Matsubayashi et al. 2009).

Almost similar prevalence of *Eimeria* in Red Panda and livestock may be due to the herbivorous food habitat of Red Panda sharing the same grazing area with livestock.

Other coccidian parasites such as *Isospora* sp., *Cryptosporidium* sp. and *Cyclospora* sp. have been reported from various other wild mammals including Raccoons (Taylor et al. 2007) and Red Panda (Lama et al. 2015). *Isospora* sp. and *Cryptosporidium* sp. are also absent in livestock.

Besides the coccidian parasites, the Red Panda were found to be infected with two other protozoan parasites, *Entamoeba* sp. and *Balantidium* sp. Amoebic dysentery, an intestinal disease caused by infection with the protozoan parasite *Entamoeba* sp. is an important disease of man and animals throughout the world. *Entamoeba* sp. had also been reported from Red Panda of RNP (Shrestha et al. 2015). In both RNP and Illam, Red Panda were infected with more than 50% by *Entamoeba* species. *Entamoeba* is causative agent of amoebiosis among livestock it has been reported from cattles of Kenya (Kanyari et al. 2009). In the present study, this parasite is also reported in cattle with prevalence rate 21.95% which was found to be less than 87% and 77% in Sheep and Goat by Kanyari et al. (2009) and 83% by Paul et al. (2010) in cattle. The prevalence of *Entamoeba* in Red Panda was higher than livestock it was may be due to behavioral and genetic factors associated in Red Panda and livestock (Gillespie et al. 2005).

Balantidium coli is the ciliate zoonotics protozoan parasites. Non-human primates have been considered the most important reservoirs for human infection (Walzer and Healy 1982, Nakauchi 1999). *Balantidium* sp. has been reported from White-handed gibbon (*Hylobates lar*), squirrel monkey (*Saimiri sciurea*), Japanese macaque (*Macaca fuscata*), wild boar (*Sus scrofa*) and chimpanzee (*Pan troglodytes*) from Japan (Nakauchi 1999). This is the first case to report the *Balantidium* sp. in Red Panda in the global context with prevalence rate 14.28%. The prevalence of *Balantidium* sp. in livestock during the present study was lowest (7.31%) than other protozoans which is similar to Kanyari et al. (2009) who reported 2% and 3% prevalence in sheep and goat, 1.6% and 6.6% in cattle by Uysal et al . (2009) and Paul et al. (2010) respectively. *B. coli* was reported in pig by Ismail et al. (2010), Weng et al. (2005) and Carneri 1972 observed 64.7% , 47% and 58% prevalence which was greater than prevalence of this study. The greater prevalence may be due to many food or water resources can become polluted because pandas defecate while feeding (Zhang and Wei 2006).

The first report on isolation and maintenance of *B. coli* was done by Barrett and Yarbroug (1921) in animals. *B. coli* are a ciliated and a normal inhabitant of intestine of wild and domestic animals, probably capable of becoming somewhat pathogenic under favorable condition. It has been identified by Varadharajan and Kandasamy (2000) from India, Hossain (2012) from Bangladesh.The infection of *B. coli* may be due to the contamination of water or food with cyst in the grazing area (Schuster and Ramirez 2008).

Generally wild animals become infected with nematode, cestode and trematode helminth parasites. To compared the life cycle of cestode and trematode, a suitable intermediate host is required but not for most of the nematode parasites. Interestingly, Red Panda of Illam were found infected with only nematode parasites but livestock were infected by cestode and trematode too.

However the trematode, *Ogmocotyle ailuri* was previously described from Red Panda at a zoo in the America (Price 1954 and 1960). *O. ailuri* also isolated from the small intestine of Taiwanese monkey (*Macaca cyclopis*) (Yoshimura et al.1996) and Japanises monkey (*Macaca fasaculata*) (Iwaki et al. 2012).

Another trematode, *Heterobiharzia americana* also recorded in Archer and Wichita countries of north contra Texa and overall prevalence was 47.2% (Kelley 2010) and other trematodes *Alaria* sp., *Digenea* sp. and *Eurytrema procyonis* were observed in Raccoon (Wright and Gompper 2005). Prevalence of trematode were found 13% in Red Panda from KBCA (Lama et al. 2015) but genera was unidentified. Absent of trematodes in present study might be due to absent of suitable intermediate host in Illam. Trematode infection was most common among livestock (Bandyopadhaya et al. 2010, Yadav and Tando 1989, Byanju et al. 2011, Kanyari et al. 2009, Chouwdhary et al.1993, Pathak 2011).

Cestode infection in Red Panda (Lama et al. 2015) had shown from KBCA similarly the Red Panda of RNP have been reported to be infected with *Moniezia*, a common herbivore cestode parasite but none of the Red Panda samples collected from Illam were positive. However three genera of cestode had been reported from Raccoon in Archer and Wichita countries of North Central Taxas including *Atrioenia procyonis, Mesocestoides spp., Taenia pisiformis* (Kelley and Horner 2008). The cestode spp. described from cattle includes *Moniezia* (Horak et al. 2004, Kanyari et al. 2009, Laha et al. 2012, Rafiullah et.al. 2011), and *Taenia* (Bui et al. 2009). In the present study, only one cestode, *Moniezia* was recorded in cattle with prevalence rata 14% which was almost similar with 21% and

16% observed by Kanyari et al. (2009) in sheep and goat 10% and 11% infection reported by Laha et al., (2012) and Farooq et al. (2012) in cattle and higher than 0.65% and 0.48% revealed by Rafiullah et al. (2011) in male and female cattle respectively.

Altogether seven genera of nematodes in Red Panda and six genera of nematodes in livestock were observed from Illam community forest. Among them *Oxyuris* sp. showed the 100% prevalence rate in Red Panda which was highest than 58.14% recorded by Shrestha et al. (2015). The highest prevalence rate of *Oxyuris* may be due to the cool climate of the area. Cool climate is suitable for the development of *Oxyuris* larva. The parasite was observed equally high prevalence (87.8%) in livestock too.

Generally round worm of Red Panda are considered as *Balyascaris* but due to the indistinguishable shape and size, they are simply consider here as *Ascaris*. *Balyascaris* is an important intestinal nematode of Red Panda as well as Raccoon. This parasite had been recorded from Spain in white-headed lemurs (*Eulemur albifrons*) (Martinez et al., 2015), North America (Kazacos, 2001) Germany (Bauer et al. 2011). In this study the prevalence of *Ascaris* was found 57.14% which was higher than 38.88% and 13.04% prevalence rate of *Balyascaris* reported in Red Panda from Rara National Park, Mugu, Nepal (Shrestha et al.2015) and Kothi Bhir Community area, Rolpa, Nepal (Lama et al.2015) respectively. *Balyascaris* is found in Red Panda, Gient Panda, Raccoon, Cat, Dog etc. Nematodes like, *B. procyonis*, *Capillaris acrophili*, *C. plica*, *C. procyonis, C. putorii and Placoconus lotoris* had been reported in Raccoons from Southern New york (Wright and Gompper 2005). Similarly, *B. procyonis* was also reported in Raccoons from Western North Carolina (Hernandez et al. 2012). The highest prevalence rate were recorded from North-Eastern, mid-western ,mid-Atlantic, some western states (California, Washington, Oregon and Coloradol and some region of Taxas (Kazacos 2001, Long et at. 2006, Chavez et al. 2012). In more than 85% of cases infections have no symptoms, especially if the number of worms is small. During the study, the prevalence of *Ascaris* in livestock was 57.14% which agree with 57% recorded by Awraris et al. (2012) in Ethiopia and greater than 1.5%, 25.9%, 17.6%, 14.7% and 3.7% recorded by Chowudhury et al. (1993), Tomass et al. (2013), Ismail et al. (2010), Matsubayashi et al. (2009) and Uysal et al. (2009). Almost equal prevalence rate was observed in between Red Panda and livestock but it is indistinguishable between *Ascaris* found in Red Panda and livestock.

During the study, *Trichostrongylus* sp. was recorded for first time from Red Panda in the global context. The prevalence of *Trichostrongylus* sp. and *Strongyloides* sp. in Red Panda was found 41.46% and 50% respectively. *Strongyloides* sp. had been recorded in Red Panda by Shrestha (2015) and prevalence was low compared to present study. The paraste was already recorded in American Raccoons from New York (Wright and Gompper 2005). In case of livestock, the parasite has been reported in Kenya (Kanyari et al. 2009), India (Laha et al. 2012), Pakistan (Rafiullah et al. 2011), Tanzania (swai et al., 2006) and Nigeria (Bui et al. 2009) with prevalence rate 13%, 25.13%, 4.21%, 20% and 11% respectively. The prevalence rate of *Strongyloides* sp. was higher (53.65%) as compare to all previous finding rate. The prevalence of *Trichostrongylus* sp. was found 41.46% which was higher than 13.83% and 16.24% infection recorded by Rafiullah et al. (2011) in male and female cattle respectively and lower than 55.8% recorded in goat and higher than 28.8% and 9.7% observed in sheep and cattle by Ntonifor et al. (2013). This result showed that Illam community forest was highly contaminated with *Strongyloides* and *Trichostongylus* eggs both from Red Panda and livestock.

Trichuris is another common nematode parasite prevalent in both wild and domestic animals. The parasite has been reported from RNP (Shrestha 2015) and KBCA (lama et al. 2015). The prevalence of *Trichuris* was 42.85% which was higher than 4.65% and 26.08% recorded by Shrestha et al. (2015) and Lama et al. (2015) respectively. High worm load may cause growth retardation, anemia and hemorrhagic diarrhea (Hale and Stewart 1979). *Trichuris* sp. have been infecting large number of livestock (Horak et al. 2004, Yadav and Tendon 1989, Byanju et al. 2011, Kanyari et al. 2009, Bui et al. 2009, Laha et al. 2012, Rafiullah et al. 2011). In the present study, *Trichuris* sp. (31.70%) was isolated from cattle. The prevalence of *Tichuris* sp. was comparatively higher than 2%, 13.08%, 5.27%, 2.6% recorded by Kanyari et al. (2009), Laha et al. (2012), Rafiullah et al. (2011) and Bui et al. (2009) respectively.

Crenosoma spp. is the Metastrongylus lungworm infecting wild and domesticated canids in Europe (Morgan et al. 2005, Traversa et al. 2010). Recently, emergence of this parasite was observed in several European countries (Traversa et al. 2010) due to population increase and urbanization of Red foxes (*Vulpes vulpes*) (Deplazes et al. 2004) which is the major reservoir hosts of this parasite in Europe. The prevalence of *Crenosoma* was found 42.85% in Red Panda which was almost similar 34.88% by Shrestha et al. 2015

Rara National Park, Mugu, Nepal and higher than 4.3% from European zoos (Bertelsen et al. 2010). Hook Worm infection has been reported from Red Panda of RNP (Shrestha et al. 2015) with prevalence rate 44.19% which was almost similar with present study which revealed 35.7%. Hookworms are cosmopolitan in distribution (Bowman et al. 2003) and can be transmitted orally but also by cutaneous penetration and cause high mortality in animals and human (Hotez et al. 2004). *Crenosoma* spp. was not observed in cattle because it is the parasites of carnivore.

Angiostrongylus vasorum was recorded in Red Panda from different countries. It is a most important lungworm which causes pathogenic pneumonia to Red Panda. *A. vasorum* was recorded from Denmark, U.K and European zoos by Bolt et al.1992, Janet et al. 2009, Bertelsen et al. 2010) respectively. *Angiostrongylus* sp. was also reported in Nepal (Lama et al. 2015 and Shrestha 2015) but during this study *A. vasorum* was not recorded. Several nematodes like, *Dirofilaria* spp., *Toxocara* spp., *Trichinella* spp. are important nematode parasites to carnivores and omnivorous which was reported in Red Panda from different parts of the World (Chowdhary 2001, Lan et al. 2012, Neiffer et al. 2002). This parasite was negative in Red Panda and domestic animals of Illam community forest.

Aelurostrongyloid spp. have been reported in carnivora by different researchers in global and national context. In the present study, none of these nematodes were isolated in the faecal matter of Red Panda of Illam community forest, Nepal. Some reports have been indicated the presence of *Toxocara* spp., *Bunostomum* spp., *Haemonchus* spp., *Oesophagostomum* spp., *Cooperia* spp., *Ostertagia* spp. etc from cattle (Hoorak et al. 2004, Bandyopadhyay et al. 2010, Bilal et al. 2009).

Among 14 samples, multiple infection was found to be highest (78.57%) in Red Panda followed by double (21.42%) which was similar with Shrestha et al. (2015). No single infection was observed during study. Mixed infection 52.78% and single infection 47.23% were recorded by Byanju et al. (2011). Bui et al. (2009) observed double infection (9.7%) and triple infection (1.3%) which was lower than the present study.

The intensity of different parasites in Red Panda of Illam community forest were observed in this study. According to result maximum number of Red Panda were found to be infected with light infection. Some of the faecal samples of Red Panda found to be positive for heavy infection by *Eimeria, Oxyuris, Trichostrongylus and Ascaris*. The heavy infection indicates symptomatic condition causing serious diseases in Red Panda. The result of present study was similar to the report of Shrestha (2012).

6. Conclusion and Recommendations

6.1 Conclusion

Overall result revealed that all the Red Panda of Illam community forest were found infected with protozoan and helminth parasites. Three genera of protozoan parasites were identified infecting Red Panda among them *Eimeria* sp. showed the highest prevalence followed by *Entamoeba* sp. and *Balantidium* sp. In case of helminth parasites only nematodes were observed. *Oxyuris* sp. showed 100% prevalent followed by *Ascaris* sp. (57.14%), *Strongtyloides* sp. and *Trichostrongylus* sp. (50%), *Crenosoma* sp. and *Trichuris* sp. (42.85%) and Hook worm (35.7%). Multiple infection (78.57%) was more common in Red panda followed by triple (21.42%) while single infection was absent. In case of intensity rate, maximum number of Red Pandas were found to be infected with light infection followed by mild, modred and heavy.

Almost similar rate of protozoan and helminth parasites were recovered from livestock which share the same pasture land as Red Panda. Cow, Goat, Horse, Sheep, Yak and Buffalo were found to be infected by *Eimeria* sp. while *Entamoeba* sp. was found in Cow, Horse and Buffalo but prevalence of *Balantidium* sp. was reported in Cow and Horse only. Among the helminth, nematode parasites showed the highest prevalence while cestode and tematode infection were relatively low. Eight different parasitic helminth were identified in livestock such as *Oxyuris* sp., *Ascaris* sp., *Strongyloides* sp., *Trichostrongylus* sp., *Trichuris* sp., Hook worm among nematodes, *Moniezia* sp. among cestoded and *Paramphistomum* sp. among trematode. Out of all these parasites, *Oxyuris* sp. showed the highest prevalence in all the domestic animals; 90%, 88.88%, 80% and 75% in cow, goat, horse and sheep respectively. Infection of *Strongyloides* sp. revealed heavy in horse (80%) followed by goat (66.66%) and 50% in each of cow and sheep. *Trichuris* sp. was absent in horse and in other animals prevalent almost equal, 40% and 44.44% in cow and goat while 25% in sheep. Prevalence of Hook worm was highest (50%) in sheep and lowest (20%) in horse which revealed similar result for *Trichostrongylus*. *Ascaris* showed dominant parasites for horse followed by goat, cow and sheep. Prevalence of *Moniezia* and *Paramphistomum* in cattle was 14.63% and 2.43% respectively. Multiple infection was dominant (73.77%) in livestocks followed by triple (19.51%) and double infection (7.31%).

Present stusdy revealed the prevalence of some protozoan and helminth parasites common to Red Panda and livestock , where helminths were more prevalence compared to protozoa. Among the protozoan parasites *Eimeria* sp., *Entamoeba* sp., and *Balantidium* sp., shared a higher prevalence in Red panda and cattle as well as Red Panda and goat/ sheep. Similarly, *Oxyuris* sp., Hook worm, *Strongyloides* sp., *Trichuris* sp., *Ascaris* sp., and *Trichostrongylus* sp., were found morphologically similar in both animals.

Statistical analysis showed that all these helminth parasites can be shared between Red Panda and livestock particularly from cattle and sheep /goat to Red Panda and vice-versa. *Crenosoma* sp. seen to be Red Panda specific which was not shared in livestocks. Similarly, cestode and trematode parasites found in livestocks were not found in Red Panda.

6.2 Recommendations

On the basis of conclusion following recommendations have been made to reduce the risk of gastrointestinal parasitic transmission between Red Panda and livestock.

- Morphologically similar parasites were compared in between Red Panda and livestocks but verification or identification should be done in molecular basis.
- Considering the detection of several zoonotic parasites, investigation of the role of human being in the transmission of zoonotic parasites in between Red Panda, livestocks and other wildlifes has been recommended.
- It is recommended to establish veterinary laboratory in the study area in order to carry out continuous diagnosis,surveillance and to prevent loss of samples during transpotation as well as for prompt laboratory work without loss of samples.
- Red panda habitat should be strictly prohibited for grazing of domestic animals.
- Red Panda conservation plans, specific for community forest must be develop for long term conservation of Red Panda.
- Livestocks grazing area should be well manage and separated from Red Panda habitat.

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