

1. INTRODUCTION

1.1 BACKGROUND

Health is soundness of body and mind. It is an integral part of life. The state of positive health amplifies “perfect functioning” of the body and mind. It is a process of expanding consciousness that synthesizes disease and non disease and is recognized by patterns of person environment interaction. In ancient times, health and illness were considered as cosmological and anthropological perspective. Thus different communities have their own concept of health, as a part of their culture.

Parasitic infection are widely scattered throughout the world. There are various types of parasites such as ecto-parasites, endo-parasites, facultative parasites, obligatory parasites etc. which can cause disease in humans. They bring serious health problems in humans (CDC, 2014). Intestinal parasitic infection caused by intestinal helminthes and protozoans in human residing in developing countries (Haque, 2007). As per World Health Organization (WHO), globally 3.5 billion people are affected by intestinal parasitic infection and cause clinical morbidity in approximately 450 million, majorities of cases occur among children (WHO, 1998). These parasites are associated with diverse clinical manifestations such as malnutrition, iron-deficiency anemia, malabsorption syndrome, intestinal obstruction, and mental and physical growth retardation (Gyawali, 2009). In context of Nepal, poverty, ignorance and prevalent still that led to several types of disease. The health status is dominated and badly affected by parasitic infections. In Nepal over 70% of mortality and morbidity are associated with infectious disease and is also reflected in top 10 disease of Nepal (Rai *et al.*, 2001). Various studies had been made on intestinal parasites among different group of people. Intestinal parasites are parasites that can infect the gastro-intestinal tract of humans and other animals. They can live throughout the body, but most prefer the intestinal wall. There are several intestinal parasites protozoan parasites and helminthes parasites that dwell in the body. Protozoan parasites include *Entamoeba coli*, *Giardia*, *Cyclospora*, *Cryptosporidium* etc. While helminthes parasites comprises *Ascaris*, *Ancylostoma*, *Trichuris*, *Hymenolepis*, *Strongyloides*, *Taenia* etc. Eggs are passed out of the body through stool and can infect others via contaminations (Birn and Armando, 1999). Means of exposure include ingestion of undercooked meat, drinking infected water, and skin absorption. Millions of people are not getting proper facilities they need because the government is not conscious about their need. Illiteracy, poverty,

malnutrition, high infant mortality rate, inadequate health facilities, poor water supply and unsanitary condition have led the country to a very poor socio-economic condition (Chhetri, 1997).

In Nepal, there are many ethnic group residing in different parts of the country among them are the Chepang. The Chepang are an indigenous Tibeto-Burman people group numbering around sixty eight thousand mainly inhabiting the rugged ridges of the Mahabharata mountain range of central Nepal. They are the most marginalized indigenous ethnic community of Nepal. Over few decades, the Chepang have slowly changed their lifestyle from nomads to a more settled way of life, started relying on cultivation of maize, millet and bananas. The total population of Chepang all around Nepal is about 68,399 (CBS, 2012). They are socially and economically backwarded group. This group relies on nature and farming and has been facing serious health hazards. There have been few researches conducted in the indigenous communities of Nepal (Sharma, 1965). Intestinal parasites are highly prevalent among the population of country. Parasitic helminthes of human are the major health problem around the world where no country can be aloof. Their presence is particularly in developing and under developed countries where transmission of helminthes is accelerated by poor sanitation and hygiene (WHO, 2005; Gordon *et al.*, 2011). Global phenomenon such as climate change, migration, environmental changes, drug resistance and economic factors make things more complicated into public health (Thompson & Conlan, 2011). It is estimated that soil transmitted helminthes have affected more than two billion people worldwide (WHO, 2004). Poverty, poor sanitation, inadequacy of nutrients, lack of health care, dirty water results in helminthes infection. It leads to serious health hazards and may even leads to death due to chronic infection. Intestinal parasitic infection is the major health problem among people of developing countries as the result of food and water contamination (Engels and Savioli, 2006).

1.2 Introduction to Intestinal Parasites

Intestinal parasites are those organisms that dwell in the intestinal parts of the host, obtaining their nutrients from there. The parasites develop and invade intestinal region which is responsible for resulting infections or parasitic infection. The intestinal parasites are basically Protozoan and helminthes (Arora and Arora, 2010).

1.2.1 Intestinal Protozoan Parasites:

Protozoa are single-celled organism usually measuring 1-150 μ . They are generally found in contaminated food and water. They have short life period, high rate of reproduction and a tendency to induce immunity to re-infect the host (Chatterji, 2001).

***Entamoeba* and Amoebiasis:**

Entamoeba histolytica was first described by Losch in 1875 after being isolated in Russia from patient with dysenteric stool. The disease called amoebiasis is the second leading cause of death from parasitic disease worldwide (Stanley, 2003) causing about 450 million infection per year and 50 million incident with around 100000 deaths (Smyth, 1996). *Entamoeba histolytica* lives in the mucous membrane of the intestine, engulfing RBCs. The morphology is divided into three stages as trophozoites, pre-cyst and cystic stages. Trophozoite stage is feeding stage. It has no fixed shape and sizes range 15-30 μ m. Cysts stage is spherical, 10-15 μ m in diameter. Initially cyst is uninuclear and then develops into form two and then four nuclei, binary fission occurs (Ichhpujani and Bhatia, 2002). The amoeba is transmitted by oral-fecal route. Poor hygiene, poverty, ignorance etc facilitates in spreading the disease.

***Giardia* and Giardiasis:**

Giardia was discovered by Leeuwenhoek in 1681 in his own stool and it was described by Lambl in 1859. The highest prevalence of *G. lamblia* occurs in tropics and sub-tropics where sanitation is poor. In developing countries, prevalence rate ranges from 15-20% in children below 10 years old. It dwells in duodenum and the upper part of jejunum in human (Ichhpujani and Bhatia, 2002).

Morphologically, it has two forms: trophozoite and cyst. Trophozoite is pear shaped structure which is a feeding stage and measures 10-20 μ m in length with 5-15 μ m in width.

Trophozoite is bilaterally symmetrical and has a pair of nuclei, one on each side of the midline, one pair of axostyle, one pair of parabasal bodies present on axostyle, four pairs of flagella and probably four pairs of blepharoplasts from which the flagella arise. Mature cyst is oval shaped with two pairs of nuclei. Cyst sometimes converts into trophozoite during unfavorable conditions. Cystic stage is infective stage, transmitted by contaminated water (Arora and Arora, 2010) or through fecal-oral route. Patients may complain of dull epigastric pain, flatulence and chronic diarrhea of steatorrhoea type.

***Cyclospora* and cyclosporiasis**

Cyclospora cayetanensis is a coccidian parasite measuring 8-10µm in diameter. Man acquires infection through contaminated water and food. Unsporulated oocysts are excreted in the feces. During sporulation, the sporant gets divided into two sporocysts and each contains two sporozoites. Sporocysts excyst in the gastro-intestinal tract and invade small intestinal epithelial cells. The life cycle, although not completely understood however cyclosporiasis has been recorded since 1990. It infects small intestine causing self limiting diarrhea, fever, fatigue, cramps lasting for 3-4 days and is associated with poor sanitation (Arora and Arora, 2010).

***Cryptosporidium* and cryptosporidiosis**

Cryptosporidium is a coccidian parasite. It was first reported in 1907 in gastric crypts of a laboratory mouse. Cryptosporidiosis is a zoonosis and is transmitted via fecal-oral route. This infection is now well recognized as causing disease in humans, particularly in those who are in some way immune suppressed or immune deficient. The parasites released in the feces are known as oocyst. Man acquires infection through ingestion of food and water contaminated with feces containing oocysts of the parasites. Oocysts are colourless, spherical or oval and measures 4-5µm in diameter. It has two walls-thin and thick walled which contains 1-6 large dark granules and numerous small granules. In a mature post-sporulation oocyst, 2-4 sausage shaped sporozoites can be seen (Arora and Arora, 2010). The first cases of human cryptosporidiosis were reported in 1976. Cryptosporidiosis is an opportunistic infection and transmitted through fecal-oral route. It appears cholera like diarrheal disease. It may results in respiratory illness. In the immune-competent host, *Cryptosporidium* infection is self limiting and requires only supportive treatment in order to prevent dehydration (Chatterji, 2001).

1.2.2 Intestinal Helminthes Parasites:

Helminthes or parasitic worms are multicellular, bilaterally symmetrical, elongated, flat or round animals. It is classified as Platyhelminthes and Nematelminthes. Platyhelminthes have been classified as cestode and trematode. While Nematelminthes has only one class Nematode. Most helminthes are macroscopic in size and often visible to naked eye. Size of helminthes egg is larger (Chatterjee, 2001).

***Ascaris* and Ascariasis:**

Ascaris Lumbricoides has worldwide distribution, being especially prevalent in the tropics and sub tropic. It is estimated that 250 million people worldwide are infected by this parasite. The highest prevalence is in malnourished people residing in developing countries. Areas with modern water and water and waste treatment have a low incidence of the infection with parasite. It is found in small intestine, particularly the jejunum of man. Male measures 15-30cm in length and 3-4mm in diameter. The posterior end is curved ventrally to form a hook. Female is longer and stouter than the male worm and measures 25-40 cm in length and 5mm in diameter (Arora and Arora, 2010). The tail is straight and conical. Man acquires infection by ingestion of food, water or raw vegetables contaminated with embryonated eggs. Ascariasis is caused by *Ascaris*. Migrating larva and adult worm can cause symptoms however maximum of infection are symptomless. Fever, cough, dyspnea, rashes, conjunctivitis, hemorrhagic pancreatitis etc are different symptoms observed (Ichhpujani and Bhatia, 2002)..

***Ancylostoma* and Ancylostomiasis:**

Ancylostoma duodenale was described by Dubini in 1843. It is one of the best medically important phasmid nematode, commonly known as “Old world hookworm”. It is cosmopolitan in tropical and subtropical countries. It inhabits in small intestine particularly in the jejunum, less often in duodenum and rarely in ileum. These worms suck blood causing chronic loss of blood producing serious anemia. These worms generally suck about 0.03-0.2 ml blood per day. They are small (male measures 8-12mm by 0.5mm and female 10-13mm by 0.6mm), pinkish and bursiform in shape. The anterior end is slightly bent. Oral cavity is provided with four hooks like teeth on ventral surface and two knobs like teeth on dorsal surface. Copulatory bursa is present in male and females have tapering posterior end and no expanded bursa. Each female can lay about 15000-20000 eggs per

day. Third stage filariform larva penetrates the skin exposed to contaminated soil. The characteristic symptoms of Ancylostomiasis gastrointestinal disturbance and nervous disorder. Iron deficiency anemia, bone marrow defect, heart defect, circulatory failure etc may be due to adult form while larval form may causes ground itching, lesions in the lungs may cause bronchitis, creeping eruptions etc (Arora and Arora, 2010).

***Trichuris* and Trichuriasis:**

Trichuris trichiuria is cosmopolitan in distribution but is more common in the warm, moist regions. It is commonly called whipworm. This worm was first discovered by Linnaeus in 1771. It is medically important parasitic aphasid nematode. These worms are commonly found in the caecum and appendix of man, sometime found in rectum. Sexes are separate. No intermediate host is required. Adult worms are characteristically whip-shaped. The anterior three-fifth is very thin and hair-like and the posterior two-fifth is thick and stout, resembling the handle of the whip. The male measures 30-40mm with tightly coiled posterior end and a single spicule. The female worm is slightly that is 40-50mm and both of them have a narrow anterior portion (Arora and Arora, 2010). Man acquires infection by ingesting embryonated eggs with contaminated food or water. Eggs are barrel shaped. Freshly passed eggs are not infective to human but the eggs take about 3 weeks to develop in moist soil and remain infective for about 2 weeks. Only embryonated eggs when ingested can cause infection, the shell is dissolved by digestive juices and larvae come out with one pole and migrate to caecum ((Ichhpujani and Bhatia, 2002). Infection with *Trichuris trichura* is known as trichiuriasis. The anterior thin portion of the worm penetrates the mucosa and sub mucosa causing trauma. Loss of appetite, anemia, chronic diarrhea, nervousness, loss of weight, bloody stools etc. Appendicitis may occur when the worm enters vermiform appendix (Chatterji, 2001).

***Hymenolepis* and Hymenolepiasis:**

Hymenolepis nana is the smallest tapeworm infecting humans. The name *Hymenolepis* refers to thin membrane covering the eggs (hymen-membrane, lepis-covering) and nana to small size (nanus- dwarf or small). It was first discovered by Bilharz in 1857. It is known as dwarf tapeworm. It measures about 4-5cm in length and 1mm in diameter. Scolex has four cup-shaped suckers and a retractile rostellum armed with a single row of 20-30 hooklets. The neck is long and slender. The strobilus consists of about 200 proglottids.

Eggs are spherical or oval, hyaline, 35-40µm in diameter (Arora and Arora, 2010). Eggs and proglottids are released out in feces of infected humans or rodents. Human get infected by fecal-oral route. The infection is more common in children. Disease caused by it is Hymenolepiasis. Patients develop headache, dizziness, anorexia, weight loss and weakness (Ichhpujani and Bhatia, 2002)..

***Strongyloides* and strongyloidiasis:**

Strongyloides stercoralis was first identified by Normand in 1876, in the feces of French troops who had been suffering from uncontrolled diarrhea in Indochina. It is worldwide in distribution. *Strongyloides stercoralis* causes Strongyloidiasis. Adult worms are cylindrical in shape and is the smallest nematode known to cause infection in man. Females measure about 2-3mm in length and 30-50µm in width where as males are smaller than female measuring 0.7-1µm in length by 40-50µm in width (Arora and Arora, 2010). The cylindrical oesophagus extends through the anterior one third of the body and the intestine extends through the posterior two-third of the body. There are two forms of larva -Rhabditi form and filariform larvae. The females are ovo-viviparous. Rhabditiform larvae are found in the lumen while filariform larva is the infective form. Filariform larva penetrates the skin coming in contact with soil. Once with nine dermis, larvae invade the venous circulation and are carried by blood stream to the right heart and then to the lungs. Then they migrate to the bronchi, trachea, larynx, crawl over the epiglottis to the pharynx and are swallowed. The females burrow way out to the mucous membrane and lay eggs in the tissues. Infection caused is called strongyloidiasis. It is most asymptomatic. In symptomatic cases follows skin lesions, pulmonary lesions, intestinal lesions, etc.

***Taenia* and Taeniasis:**

Taenia saginata has worldwide in distribution in countries where cattle are raised and beef is eaten. *Taenia solium* is not as widely distributed as *Taenia saginata*. It occurs generally where human feces reach pigs and pork is eaten raw or undercooked. It is estimated that as many as 100 million people are infected with *Taenia saginata* and *Taenia solium*. Eggs of both the species are indistinguishable. They are brown, spherical in colour and measures 31-43µm in diameter. *T.saginata* measures about 4-6 meters or more while *T.solium* measures about 2-4 meters or more (Arora and Arora, 2010). Eggs or gravid segments are passed out through fecal matter. The larva hatches out in the small intestine, the scolices

exvaginate and attaches to mucous membrane and develops into adult. Man acquires infection by eating raw or undercooked beef or pork containing encysted larval stage (*Cysticercus bovis*/*Cysticercus cellulosae*). The adult worms live in man's gut causing taeniasis. The clinical features comprises of abdominal discomfort, hunger pain, chronic indigestion etc (Ichhpujani and Bhatia, 2002).

1.3 OBJECTIVES

1.3.1 General objective

To determine the prevalence of intestinal parasites among the Chepang people of Shaktikhor area, Chitwan, Nepal.

1.3.2 Specific objectives

-) To study the age wise and sex wise distribution of parasites among Chepang of Shaktikhor area, Chitwan.
-) To compare protozoan and helminthes parasites.
-) To determine the knowledge, attitude and practices regarding health and sanitation of the studying population.

1.4 Significance of the study

Nepal, a landlocked country has most of the parts rural and undeveloped, contained unhygienic health habit practices among the people that lead to various parasitic infestation. No one remain unaffected by parasitic infection. Chepang people living in marginalized area of Chitwan, Shaktikhor area are still behind in terms of health, education and various other aspects. Due to lack of awareness, they encounter with different types of parasites. Though, several researches have been done in various other ethnic groups but study related to gastro-intestinal parasites among Chepang, is still insufficient.

Parasitic infection has been spreaded throughout the world that has hindered the developmental progress and depicts the exact sanitary status, level of knowledge, and unhygienic way of living. The major serious public health problem in most of the countries occurs as a result contamination of food and water (Engels and Savioli, 2006). The disease like diarrhea, dysentery occurs in most of children and old age people that weakens the

body, and results in low immunity power. This study may be really significant to explore the health status regarding parasitic infection among the Chepang people.

The poor economic condition, low awareness level, sanitary defect, open defecation habit, feeding behavior etc have resulted in the higher prevalence of parasitic infection. Not only has that water pollution resulted in gastro intestinal tract infection (Rai *et al.*, 1994). Chepang community is a marginalized group of people. They have a very low economic status.

The study of intestinal parasites in Chepang of Chitwan has been undertaken to find the prevalence of the infection on the basis of age, sex, sources of drinking, defecation place wise, awareness level, intensity of parasites etc. This study is concerned to find the types of parasites that are prevalent in this community, also promotes in further investigation among them and carries out several researches in the very community.

1.5 Limitation of the study

-) The number of the sample during the study was low. This might be the result of less time, their settlement was very disperse and their shyness.
-) This study was focused on humans only that are not so appropriate for total description of the possible infection.
-) Since only microscopic examination was done using common diagnostic methods due to the financial limitation and constraint.
-) Stool samples of all the individuals of the study area were not observed.

1.6 Hypothesis

The null and alternative hypothesis of this research work is:

Ho=There is no significant differences in the prevalence rate of intestinal parasites among Chepang communities of Shaktikhor area, Chitwan District.

H1=There is significant differences in the prevalence rate of intestinal parasites among Chepang communities of Shaktikhor area, Chitwan District.

2. LITERATURE REVIEW

2.1 History of Parasitology

The knowledge about parasites and parasitology was limited to only few common external parasites such as lice, fleas and few internal parasites like tapeworm, *Ascaris*, pinworms and guinea. They were present by the natural products of human bodies. Even Rudolphi and Bremser also supported this idea (Chandler and Read, 1961). In Linnaeus time, people thought that internal parasites were originated from accidentally swallowed free living organism (Chandler and Read, 1961). Antonie van Leeuwenhoek in 1681 observed *Giardia lamblia* as the first protozoan parasites of human. This was the first protozoan parasites of the humans that he recorded, and the first to be seen under microscope.

It has been found that human act as a host to about 300 species of parasitic worms and more than 70 species of protozoa which may be obtained from our primate ancestors and some of them may be gained from domesticated animals and some may be acquired when they come in contact with the source of infection (Cox, 2003). The first written records of parasitic infection came from Egyptian civilization from 3000 to 400 BC, particularly the Eblers Papyrus of 1500 BC discovered at Thebes. Hippocrates (460 to 375 BC) knew about worms from fishes, domesticated animals as well as human. Paulus Aegineta (AD 625 to 690) described about *Ascaris*, *Enterobius* and *Tapeworms* and gave descriptions of the infections caused by them. Linnaeus described and named six helminth worms, *Ascaris lumbricoides*, *Ascaris vermicularis*, *Gordius medinensis*, *Fasciola hepatica*, *Taenia solium* and *Taenia lata*. Hookworms, in human were found in 1838 by the Italian physician Angelo Dubini, and the link between the worms and disease was established by Wilhelm Griesinger in 1854 (Kean, 1978). Strongyloides infections was well accounted by Grove. The human liver fluke was first discovered by James McConnell in 1875. Tyson was the first person to recognize the “Head” of the tapeworm. Louis Alexis Norman, a physician recognized *S. stercoralis* in 1876 (Cox, 2003). Modern parasitology developed in the 19th century with accurate observations by several researchers and clinicians (Arora and Arora, 2010). Rudolphi classified all the parasites known up to his time. There are good accounts of the history of ascariasis by Grove (1990) and Goodwin (1996).

2.2 Global context

Intestinal parasites are distributed world widely and are a major problem in many developing countries. It can infect the gastro-intestinal passage of human and other animals. Mostly they prefer to live in intestinal wall and even in other various parts of the body. An intestinal parasite can damage its host via an infection which is called helminthiasis in case of helminthes (Loukopoulos *et al.*, 2007). An estimated 807-1221 million people in the world are infected with *Ascaris lumbricoides* which is a soil-transmitted helminthes parasite and is the accounted for a major burden of disease worldwide (CDC, 2016).

Most of the studies showed that *A.lumbricoides* as leading parasite that dwells in the body of human (Goli *et al.*, 2014; Opara *et al.*, 2014). *A.lumbricoides* infection has also been reported in pregnant women (Sehgal *et al.*, 2010).

It was reported that *A.lumbricoides*, *T.trichiura*, *A.duodenale*, *E.histolytica*, *H.nana* were the vital parasites among the children of developing countries (Goli *et al.*, 2014; Abahussain, 2005; Sehgal *et al.*, 2010; Opara *et al.*, 2007; Alamir *et al.*, 2013). *A.lumbricoides* and *E.histolytica* were found as the major parasites found in the HIV patients (Akinbo *et al.*, 2010; Kipyengen *et al.*, 2012).

Similarly, the different researches in the American continent regarding the intestinal parasitic infection have been conducted. Fugita, *et al.* (1993) conducted a survey in five rural communities at Paraguay by different method and study method gave 68.4% positive with 57.4% single infection while 28.1% double 9.6% triple, 4.1% quadruple and 0.4% quintuple infection. The most commonly observed species was *Necator americanus* 23.8%, *Roundworm* 10.6%, *Strongyloides stercoralis* 10.1%, *H.nana* 2.3%, *T. trichiuria* 0.8% and *Taenia* spp 0.8%. Adedayo *et al.* (2004) performed a retrospective study by stool samples at the parasitological unit of the medical laboratory services of Princess Margaret Hospital, Dominica, out of 3752 stool samples (10.47%). The main parasites were *Entamoeba coli* 14%, *Giardia lamblia* 1.4%, *Strongyloides stercoralis* 1.0%, *Ascaris lumbricoides* 0.8%, hookworm 1.5% and *Trichuris trichiura* 0.9%.

In Asian continent, studies have been carried out in tribal people which shows that Bharia tribe of India found out that 30% of children suffer from severe anemia and 50% of children had intestinal parasites with common parasite as *Ancylostoma duodenale* (16.3%)

and *Ascaris Lumbricoides* 18.5% (Chakma *et al.*, 2000). While similar study carried out among people of Bhil tribe of Rajasthan and found that 51.78% were infected with diverse species of intestinal parasites where male were highly infected compared to their counterparts (Choubisa *et al.*, 2012). Pestehchian *et al.* (2015) examined 655 total samples of inhabitant and tribe population of Chelgerd, Iran and revealed that (56%) of patients suffers at least one intestinal parasites where (67.7%) in tribal population and (43%) in inhabitants. *Giardia intestinalis* (28.2%) and *Blastocystic hominis* (27.5%) were detected while Motazedian *et al.* (2015) found that the prevalence was 10.4% where most species observed were protozoan parasites as *E.coli*, *G.lambliia* and *B.hominis* meanwhile the only one infection by *H.nana* (0.1%) was detected with mixed infection. Saifi *et al.* (2001) worked on intestinal parasitic infections in University Campus of Aligarh; there were 3695 samples from the people complaining abdominal discomfort out of which 2152 samples (58.24%) positive for *Entamoeba histolytica*, *Ascaris lumbricoides*, and *Giardia lambia*. There was higher prevalence of *E. histolytica* (37.55%) followed by *G. Lambliia* (14.95%), while *A. lumbricoides* showed (5.71%). Nishiura *et al.* (2002) evaluated the prevalence of *A. lumbricoides* in 492 children from five different rural villages in the Northern Area of Pakistan. It was found that the presence of *A. lumbricoides* was 91%. Its infection was most common in 5-8 years children. Rai *et al.* (2002) studied worm infestation and anemia, a public health problem among tribal preschool children of Madhya Pradesh and the result showed that 48% of them had intestinal infection. High prevalence of Anemia was seen among people with parasitic infection.

Almegrin, (2010) studied among immuno-compromised patients in Saudi Arabia and found no significant difference in intestinal parasites were found between male and female. There was evidence that the prevalence of *Ascaris lumbricoides* and *Trichuris trichiuria* infection statistically greater in periurban areas compared to urban and rural while the prevalence of hookworm was higher in rural areas (Pullan *et al.*, 2014).

From different studies, Intestinal parasites were found more common in females (Akingbade *et al.*, 2013; Marothi *et al.*, 2011; Khanum *et al.*, 2013) and common parasites found were *A.lumbricoides*, Hookworm, *T.trichiuris*, *H.nana* (Opara *et al.*, 2007; Sehgal *et al.*, 2014). Abahussain (2005) reported same common parasites from countries like Nigeria, North India, and Saudi Arabia.

Intestinal parasites on the school-going children has been studied by different researcher (Opara *et al.*, 2007; Alamir *et al.*, 2013) that exposed that single infection of parasites was higher than the double and multiple infection. The overall prevalence was found higher in the age group 12 and the soil-transmitted helminth infections were significantly more common compared to protozoa infections (Nguir *et al.*, 2011). The prevalence and diversity of intestinal parasitic infections is high in the population of central Asia with wide range of nematodes, cestodes and protozoa are found common in Afghanistan (Korzeniewski, 2016).

The three main soil-transmitted infections ascariasis, trichuriasis and hookworm, are common clinical disorder in man (Bethony, 2006). Parasitic infections are endemic world widely and have been described as constituting the greatest single worldwide cause of illness and diseases (Keiser *et al.*, 2008). Parasitic infections are reliant on poverty, miserable personal hygiene, piteous environmental care, inadequate health services and lack of proper and necessary awareness of the transmission mechanism and life cycle pattern of the parasites (Adeyeba *et al.*, 2002).

In case of researches carried out in different African countries revealed that in Zimbabwe it was found that the intensity of the hookworm was found to be higher with 61.7% which was followed by *Ascaris Lumbricoides* and *Trichuris trichiuria* (Chandiwana *et al.*, 1989). In the district of Vhembe of South Africa, it was found that *E.histolytica*, *E.dispar* (34.2%), *Cryptosporidium spp.* (25.5%) were the common parasitic cause of diarrhea was *G.lambliia* (12.8%) among the school children (Samie *et al.*, 2009).

It was found that there is higher prevalence of *Ascaris lumbricoides* (20%) followed by *Trichuris trichiuria* (12.9%), *Enteriobus vermicularis* (17.8%) and *Ancylostoma duodenale* (6.5%) in Nigeria (Akinbo *et al.* 2010).

The major neglected infections include helminthes infection toxocoriosis, strongyloidiasis, ascariasis and cysticercosis, in case of protozoan infection trichomoniasis, bacterial infections and vector borne infection including Chagas disease in United States of America (Hotez, 2008).

Parasitic helminthes and protozoan infections are also common among the Inuit in Canada and Greenland and among Alaskan natives where the major helminthiasis among Arctic population is trichinellosis, diphyllbothriasis and echinococcosis. *Toxoplasma gondii*

infection was considered very high along with Giardiasis and Cryptosporidiosis (Hotez, 2010).

Similarly, in the Eastern Europe, the soil transmitted helminthes infections, giardiasis and toxoplasmosis remain endemic whilst in southern Europe, and vector-borne zoonoses have emerged like leishmaniasis and Chagas disease (Hotez, 2011). Besides, intestinal parasitic disease such as enterobiasis, giardiasis and ascariasis are detected most frequently in Romania but their importance is definitely surpassed by trichinellosis, cystic, echinococcosis and toxoplasmosis (Neghina, 2011). When a study was carried out in a teaching hospital in Italy it was found that 11.1% of total surveyed were contaminated with intestinal parasites and the prevalence was higher in males than in females. Common parasites were *Giardia intestinalis*, *Entamoeba histolytica*, *Cyclospora cayetanensis*, *Ascaris lumbricoides*, *Hymenolepis nana*, *Taenia* spp, *Strongyloides stercoralis*, *Entamoeba vermicularis* and *Trichuris trichiura* (Masucci, 2011).

In case of Australian continent, the prevalence of *Blastocystis* spp (57%), *Giardia intestinalis* (27%) and *Dientamoeba fragilis* (12%) was found around the Sydney city business district, while pockets of giardiasis were identified in the rural areas .(Fletcher, 2012). Some researches carried out shows that the water- borne disease transmitted through either drinking water or recreational water exposure (Gibney *et al.* 2017).

Hence, parasitic infections are widely spreaded in different continents. However, Asia and Africa have the highest infection rate than other continents even the developed continents possess varieties of infections.

In National Context

Like in other developing countries, Nepal is also untouched from infections caused by intestinal parasites. Several studies have been carried out and reports have been published that showed the prevalence of different intestinal parasites. 70% of morbidity and mortality are associated with infectious diseases (Rai *et al.*, 2005). Sharma (1965) reported that the round worm infestation is very common in some parts of our country. He studied 976 stool samples and found 40% roundworm infestation in Bhaktapur area.

In Nepal, the common intestinal parasitic infection among people is ascariasis, amoebiasis, giardiasis and taeniasis (Acharya, 1997). Khetan (1980) examined 2073 stool samples in

Narayani Zonal Hospital and found 1522 stool samples had worm infection of which 458 samples had *Ascaries*, 591 had hookworm, 203 had *Trichuris trichiura*, 175 had *Giardia lamblia*, 83 had *Entamoeba histolytica* and 11 had other infection. Sherchand and Cross (1996) studied on intestinal parasites from Kathmandu area and found out that 28.1% parasitic infection was among healthy children, 38.8% among healthy adults whereas 62.7% among children with abdominal discomfort. Pokheral *et al.* (2004); Agrawal *et al.* (2002); Uga *et al.* (2004) reported that the infection of protozoan parasites was found to be higher than helminthic parasites in school going children. Investigation in two rural villages in Chitwan was carried out where the total intestinal parasitic infection was 44% and the cases were higher in female than in male (Yong, 2000). In case of soil transmitted parasites, Shrestha *et al.* (2014) has reported that the contamination of soil with different parasitic form was found to be 28.5%. Rai *et al.* (2001) studied the intestinal parasitic infection in rural hilly area of Western Nepal, Achham district and revealed 76.4% prevalence of intestinal parasites in the children of that district. Shrestha (2001) reported the prevalence of parasites in rural 73.45% and in urban was 71.66% and the common parasite found was *A. lumbricoides*.

Goto *et al.* (2002) conducted a survey on Nepali children to correlate the weaning practices and *Giardia lamblia* infection where 210 poor urban Nepali children, less than 1 month to 60 months old, were admitted and measured height or length and weight, among them 167 were checked for intestinal permeability and 173 for parasitic infections. Subedi *et al.* (2012) studied on feeding practices in Chepang communities which revealed that literate mother were more conscious and early initiative at breastfeeding than illiterate mother. The meal feeding practice and diverse food for children were found lower.

Tharu, (2006) surveyed at Taklung V.D.C of Gorkha and examined 410 stool samples of different age group and sexes and found that stools of 225 people (91.11%) were infected by intestinal parasites where positive samples were collected and the prevalence rate in male was 88.78% and in females 93.22%. Prevalence of *Ascaris lumbricoides* (6.22%), *S. stercoralis* (2.66%) in helminthes while *E. histolytica* (24%), *G. lamblia* (11.11%), *Cryptosporidium* sp (4%), *C. cayetanensis* (3.11%) in protozoan parasites. Kunwar *et al.* (2006) revealed that the pattern of occurrence was 53%, 20% and 2.7% for Hookworm, *Ascaris lumbricoides* and *Trichuris trichiuria*. *Ascaris* and hookworm prevalence rate was noticeably increased with increasing age and group. Similarly, Shakya *et al.* 2006 carried

out another study among elderly people in Kathmandu valley found out that 41.7% were infected in total. Out of which 30.6% had multiple parasitism. The infection rate with protozoa (25.8%), helminth (27%), *Trichuris trichiuria* (39.4%) and *Entamoeba histolytica* (19.7%) were the commonest. Raghav *et al.* (2008) investigate intestinal parasites of Siraha district and discovered the prevalence rate was 63.20% with *A.lumbricoides* had higher prevalence (41.23%). Bhandari (2011) found that the rate of prevalence was 40% among which the positive case was higher in female. Parajuli *et al.* 2008 studied the behavioral and nutritional factors as well as geohelminthic infection among two ethnic groups in the terai region where prevalence was 42.1%.

Thapa *et al.* (2011) investigated parasitic infection among young children of rural community of Nepal which showed 45.5%. About 60% of total population is victimized by different intestinal parasites and the distribution of parasites were found higher in rural than Urban area. When a study was carried out among the Kumal community of Chitwan, Nepal, it was found that half of the population of studied population was infected with at least one of the intestinal parasite where the highest prevalence was shown by hookworms (Gyawali, 2012). Similarly, when studied among the school children revealed that 19.8% were infected with common protozoan parasite was *E.coli* and highest positive rate was in Dalits (20.3%) and least in Indo-Aryan (19.6%). Higher prevalence of *Ascaris lumbricoides* (22.63%) was found followed by *Trichuris trichiuria* (6.06%), *Strongyloides stercoralis* (1.82%), hookworm (1.6%), *Taenia solium* (1.01%), *Hymenolepis nana* (0.81%) and *Enteriobus vermicularis* (0.40%) when reported by Shrestha and Maharjan (2013).

Singh *et al.* (2014) found the higher prevalence of parasites in males which is similar reports from different authors (Mohammed *et al.*, 2014; Dash *et al.*, 2010; Yesmeen *et al.*, 2015) which show that males were infected higher.

Intestinal parasitic infection has been a significant problem in HIV patients worldwide, the prevalence of IP was 22.4%. Age, sex, marital status and being under Tuberculosis treatment were significantly associated with increased odds of intestinal parasite infection and are common in HIV infected people (Tiwari *et al.*, 2013).

Another study showed that *Ascaris lumbricoides*' propensity to produce large number of eggs that are resistant to extreme of environmental conditions that have made them one of the highly prevalent and geographically well distributed (Kanchan, 2015).

Oli, (2016) studied 200 stool samples of Tharu people of Pawanpur VDCs of Dang district and found that 29.9% of the total samples were found to be infected with parasites. *E.histolytica* was most predominant as protozoan parasite and *Ascaris lumbricoides* was detected as major helminthes parasites (45.76%).

Low socio-economic status, poor hygienic condition (Khanal *et al.*, 2011; Ojurongbe *et al.*, 2014; Rosino *et al.*, 2008), Lack of pure drinking water, lack of proper sanitary disposal (Rayapu *et al.*, 2012), Lack of health education are to be the root cause of parasitic infection (Rashid *et al.*, 2011). Soil transmitted helminthes infections are distributed widely in tropical and subtropic (Dada *et al.*, 2015).

It was found that non-vegetarian was highly infected with parasitic infection than vegetarian (Pandey *et al.*, 2015). Besides, intestinal parasitic is associated with hand washing behavior (Karunaithas *et al.*, 2011, Sah *et al.*, 2016), farming professions(Tandulkar *et al.*, 2013), unawareness (Pandey *et al.*, 2015), lack of toilet (Karunaithas *et al.*, 2011).

Most of the researcher showed that the parasitic infection is high among school going children (Ragunathan *et al.*, 2010; Sharma *et al.*, 2004). Not only that, the infection of parasites was found to be present in HIV patients (Kipyengen *et al.*, 2012, Tiwari *et al.*, 2013).

In Nepal, several species of helminthes and protozoan parasites have been recorded by different researchers. Among helminthes parasites, *Ascaris lumbricoides* is found in highest prevalence and *Entamoeba* spp is found higher among different species of protozoan parasites while *Giardia lamblia* is highest in diarrhoel patients.

3. MATERIALS AND METHODS

3.1 STUDY AREA

Chitwan is one of the 75 districts of Nepal, and is located in the southwestern part with an area of 2,238.39 km² with a population of 5,79,984 comprising 2,79,087 male and 3,00,897 female (CBS, 2011). Shaktikhor, 22 km away from the main highway (27.73⁰N 84.59'E) is inhabited by 500-600 population of Chepang. It lies in Chitwan district in the Narayani Zone of Southern Nepal. Elevation of Shaktikhor varies from approximately 250-1200 meter above sea level.



Figure: 1 map of Nepal showing Shaktikhor area (Source- www.google.com)

This area is the home of several castes and the main castes are Brahmin, Chettri, Magar, and Chepang. Chepang are marginalized group of people. Most of the people of this area are engaged in agricultural activities. People residing in this area have piteous health condition and lower level of awareness. They defecate in open places though they have latrines at their home. They lead a nomadic life with their primary lifestyles, hunting, foraging for wild roots, fishing as well as traditional farming near jungle.

3.2 Study design

The study design is based on the Laboratory examination.

3.3 Sample Size

A total 125 stool samples were examined from randomly selected houses were oriented about the methods to collect the stool. Orientation was focused on methodology of collection of stool and avoidance of urine in stool.

3.4 Stool Sampling

One hundred twenty five people were randomly selected for the study purpose. After proper instructions, vials were given to the children regarding collection of the stool sample, they were sterile labeled vials and application sticks. From each Chepang, about 2 gm. of fresh stool were collected. Each of the specimens was checked properly. Potassium dichromate (2.5%) was used to preserve the collected stool samples and it was transported to the Parasitological Laboratory of Central Department of Zoology for further investigation of eggs, adult of intestinal parasites.

3.5 Apparatus

3.5.1 Equipments

-) Compound microscope
-) Centrifuge
-) Ocular and stage micrometer
-) Filter paper
-) Sampling vials
-) Forceps
-) Needles and toothpicks

3.5.2 Chemicals

-) 2.5% Potassium dichromate
-) 70% alcohol
-) Glycerin
-) Distilled water
-) Salt solution Iodine solution

3.5.3 Preparation of Potassium Dichromate

2.5 gm of potassium dichromate was weighted accurately with the help of electric balance and dissolved in 100ml of distilled water. This solution was used for the preservation of stool samples.

3.5.4 Preparation of Normal saline

Normal saline was used for observing the characteristics of movement of parasites and this solution was prepared by dissolving 8.5 gm of Sodium chloride in 1000ml of distilled water, which was used in unstained preparation (Zajac and Conboy, 2012).

3.5.5 Preparation of Iodine solution

Iodine solution was prepared by dissolving 10 gm of potassium iodine in 100 ml of distilled water and slowly adding 5gm of iodine crystals in it. This solution was filtered and then kept in bottle (Zajac and Conboy, 2012).

3.6 Laboratory process

Lab process was conducted at Central Department of Zoology, Tribhuvan University, Kirtipur, Kathmandu.

3.6.1 Microscopic examination

Microscopic examination is required for the detection and identification of protozoal cysts, oocysts and helminthes eggs or larva. Further examination of stool samples were done in various ways before microscopic examination.

3.6.2 Unstained smear preparation of stool:

A portion of stool sample was picked up with a wooden application and emulsified with freshly prepared normal saline on a clean glass slide. The resulting mixture was made thin, and its consistency was made clear. A clean cover slip was placed over it and the excess fluid if any was removed with the help of blotting paper (Chatterji, 2001).

3.6.3 Stained smear preparation of stool:

Stained preparation was required for identification and the study of nuclear character of protozoan cysts or dead specimens of trophozoites. The iodine stained preparation was used for this purpose which was diluted in the ratio of 1:5 with distilled water (Chatterji, 2001).

3.6.4 Differential Floatation (D.F.) technique:

About 15 ml of water was added in the beaker and fecal matter was stirred properly and the mixture was filtered. The filtrate was poured into the centrifugal tube of 15ml and centrifuged at 1000 rpm for 5 minutes. The supernatant water was replaced with sodium chloride solution and again centrifuged. Again more chlorine was added to develop convex surface at the top of the tube and then cover slip was placed at the top. It was left for 10 minutes. Then cover slip was transferred slowly to slide. The slide was observed in an electric light microscope thoroughly for the eggs and cysts presence (Chatterji, 2001).

3.6.5 Sedimentation technique :

About 1 gm of stool sample was emulsified in about 4 ml of 10% formal saline solution and shaken well and the suspension was allowed to stand for 30 minutes for enough fixations. Addition of further 3-4 ml of 10% formal saline solution and was shaken well. The suspension was sieved through cotton gauge in a funnel into a 15 ml centrifuge tube. Addition of 3-4 ml of ether was followed by vigorous shaking for 5 minutes. Immediate centrifuging of the tube containing solution at 2000 rpm for 2 minutes and was allowed to settle. After this, four layer suspensions were obtained. The sediment lying at the bottom was examined for the parasites by unstained and stained smear. All the preparation was first examined under the low power (10X) objective and ocular starting from one corner of the cover slip to another corner then it was examined under the high power (40X) (Chatterji, 2001).

3.7 Data analysis and Interpretation

According to the primary data collected, statistical analysis was done with the help of Microsoft Excel 2007. All data as well as laboratory findings were analyzed according to their age, gender, feeding, habit and infection rate. Chi-square tests were done for the analysis and the total observed value was assumed as positive sample for the significance differences.

4. RESULTS

This study was carried out in Shaktikhor area to find out the prevalence of intestinal parasites among the Chepang people. A total of 125 stool samples were collected from them and examined from May to October, 2017 at parasitological lab, Central Department of Zoology. The results of the study are divided under following headings:

4.1 Results of stool examination

4.1.1 General prevalence of intestinal parasites:

Out of 125 samples, 52% were found to be infected with single or double parasites. The major groups parasites found are protozoan and helminthes.

Table 1: General prevalence of intestinal parasites of the study area

Name of area	Total no. of examined samples	Positive cases no.	Positive% Cases	Negative cases no.	Negative% cases
Shaktikhor	125	65	52	60	48

Distribution of protozoan and helminthic infection:

Out of 65 infected samples, the helminthic infection was found to be higher that is 86.15% and protozoan infection 13.85% among Chepang people.

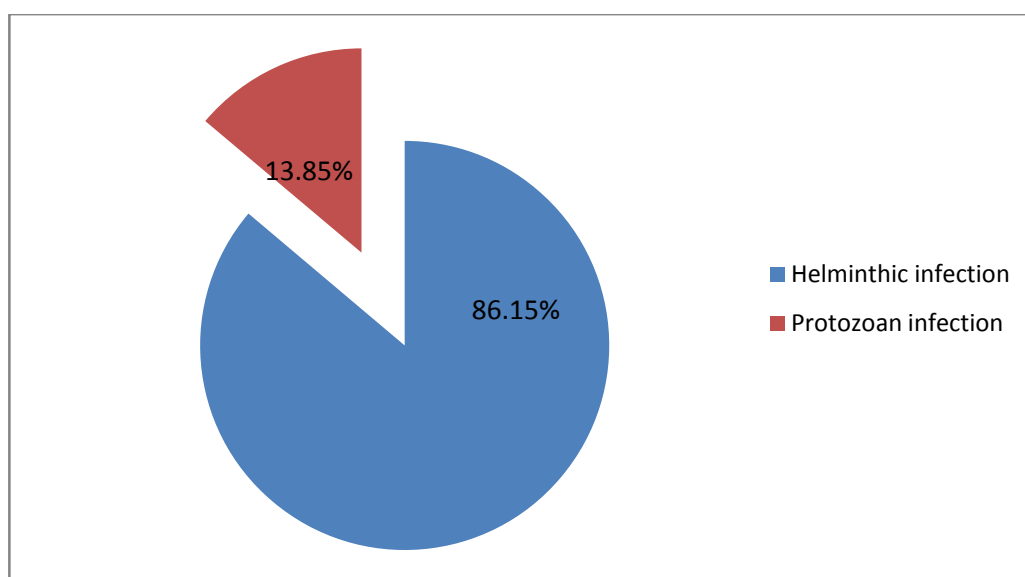


Figure 2: Distribution of protozoan and helminthes infection

4.1.2 Sex-wise prevalence of intestinal parasites:

Out of 125 stool samples examined, 63 were male, where 53.96% were found to be infected while among 62 female, 50% were found to be positive for intestinal parasites. Hence, the infection among male was found to be higher than female in this study. Sex-wise prevalence of parasites of the people was found to be insignificant between male and female of Chepang ($\chi^2=0.0721$, $df=2$, $p=0.791$).

Table 2: Sex-wise prevalence of intestinal parasites

S.N	Sex	Total examined samples	Positive cases no.	Positive cases%	Negative cases no.	Negative cases %
1.	Male	63	34	53.96	29	46.03
2.	Female	62	31	50	31	50
	Total	125	65	52	60	48

4.1.3 Age group-wise prevalence of intestinal parasites:

This study was categorized into three age groups which were 2-10 years, 11-20 years and above 21 years. The highest prevalence of parasite in this study was found to be among age group above 21 and minimum was found to be in age group 2-10 years. Statistically, there is significant difference regarding parasitic infection among different age group ($\chi^2=6.19$, $df=3$, $p=0.045$).

Table 3: Age group-wise prevalence of intestinal parasites

S.N.	Age(years)	Total samples	Positive no.	Positive %
1.	2-10	14	6	42.85
2.	11-20	75	34	45.85
3.	Above 21	36	25	69.44
	Total	125	65	52.0

4.1.4 Prevalence of specific intestinal parasites:

Out of 65 positive samples, 47 were infected with *Ascaris lumbricoides* followed by *Entamoeba coli* 9, Hookworm 5, *Trichuris trichuria* 4, *Taenia* spp 4, *Hymenolepis nana* 3 and *Strongyloides stercoralis* 2.

Table 4: Infection rate of specific intestinal parasites

S.N.	Parasites	Numbers	Infected %
1.	<i>Ascaris lumbricoides</i>	47	72.30
2.	Hookworm	5	7.69
3.	<i>Strongyloides stercoralis</i>	2	3.07
4.	<i>Entamoeba coli</i>	9	13.85
5.	<i>Trichuris trichiuria</i>	4	6.15
6.	<i>Hymenolepis nana</i>	3	4.61
7.	<i>Taenia</i> spp	4	6.15

4.1.5 Types of infections

:

The intensity of single infection was found to be more out of total infective samples 125. The single infection was found to be 83.07% while that of double infection was 10.76% and that of triple infection was 1.53% in Chepang people.

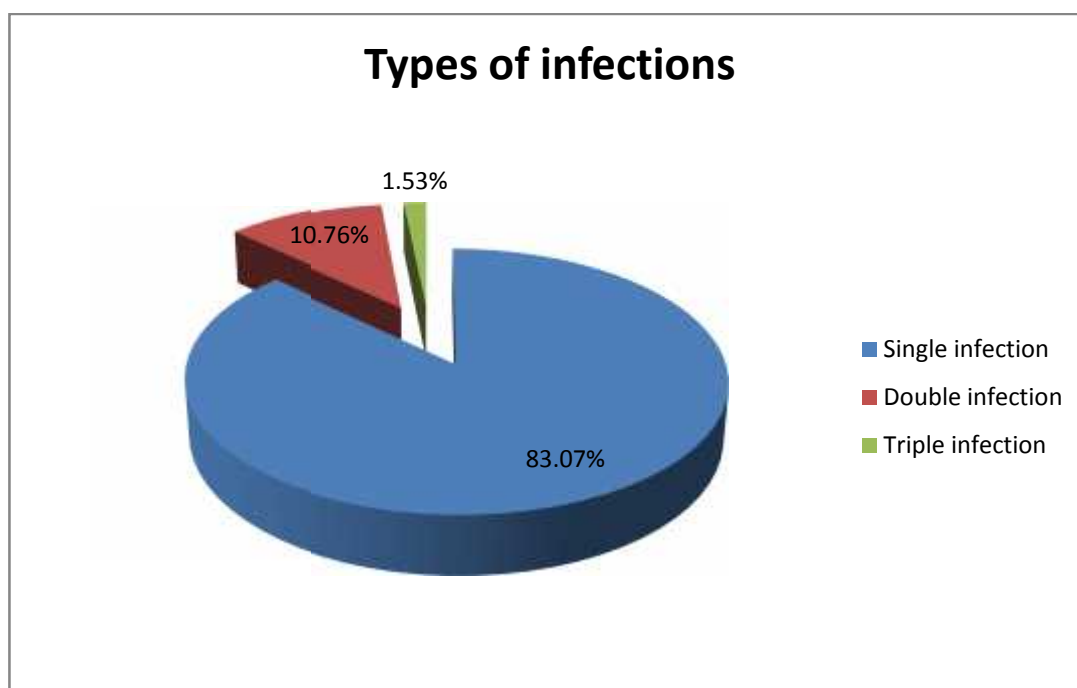


Figure 3: Types of infection.

❖ **Single infection:**

Among 65 total infections, 54 were affected with single infection. Out of which the highest intensity of single infection was of *Ascaris lumbricoides* 72.22%, *Taenia* spp 7.40%, Hookworm, *Entamoeba coli*, *Hymenolepis nana*, *Trichuris trichiuria* all have same intensity 5.55%.

Table 5: Single infection

S.N.	Parasites	Number	% of positive cases	No. of infected male	No. of infected female
1.	<i>Ascaris lumbricoides</i>	39	72.22	25	14
2.	<i>Taenia</i> spp	4	7.40	3	1
3.	Hookworm	3	5.55	1	2
4.	<i>Entamoeba coli</i>	3	5.55	1	2
5.	<i>Hymenolepis nana</i>	3	5.55	2	1
6.	<i>Trichuris trichiuria</i>	3	5.55	1	2
	Total	57		34	23

Double infection

Altogether 10 samples were found to be infected with double infection. Out of which *E.coli* and *A.lumbricoides* have the highest intensity 40% and all other have 10%.

Table 6: Double infection

S.N.	Parasites	No.	% of positive cases	No. of infected male	No. of infected female
1.	<i>A.lumbricoides</i> + <i>T.trichuria</i>	1	10	-	1
2.	<i>A.lumbricoides</i> + <i>H.nana</i>	1	10	-	1
4.	Hookworm+ <i>E.coli</i>	1	10	-	1
5.	<i>E.coli</i> + <i>A.lumbricoides</i>	4	40	1	3
6.	<i>T.trichuris</i> + <i>S.stercoralis</i>	1	10		1
7.	Hookworm + <i>E.coli</i>	1	10		1
8.	<i>S.stercoralis</i> + <i>E.coli</i>	1	10		1
	Total	10			

❖ Multiple infection

Among 65 positive samples the intensity of multiple infections was 1.

Table 7: Multiple infection

S.N.	Parasites	No.	Positive case %	No. of infected male	No. of infected female
1.	<i>T.trichuria+S.stercoralis+A.lumbricoides</i>	1	1	-	1
	Total	1	1.53	-	1

4.2 Results of survey analysis

Chepang people were interviewed during sample collection time through a set of questionnaire prepared earlier so as to find out their feeding habit, knowledge about parasites, health practices, economic status, occupation and other several health habits. The questions were prepared in such a way that covers all these queries. The result prepared after the questionnaire survey analysis is as given below:

4.2.1 Hand -washing wise prevalence of intestinal parasites:

The prevalence of parasitic infection was found higher among the person who used only water as hand washing material (75%) followed by soil and water (73.3%). However use of ash and water was 52% and that of soap and water was found to be 42.02%. Statically, there was highly significant difference in the prevalence of intestinal parasites in case of hand washing methods ($\chi^2 = 8.87$, $df=4$, $p=0.03$).

Table 8: Hand washing wise prevalence of parasites

S.N	Agent	Observation no.	Positive cases	positive %	Infected male	%	Infected female	%
1.	Soap and water	69	29	42.02	12	41.37	17	58.62
2.	Ash and water	25	13	52	8	61.54	5	38.46
3.	Soil and water	15	11	73.33	6	54.54	5	45.45
4.	Only water	16	12	75	8	66.67	4	33.33
	Total	125	65	52	34	52.31	31	47.69

4.2.2 Prevalence on the basis of knowledge:

Many people were interviewed so as to find out their knowledge towards the different factors like intestinal parasites, knowledge regarding mode of transmission, ways to control different parasitic infections. Most of the people were unaware and the only few of them know about the parasites and their consequences. Statically, there was significant difference in parasitic infection rate between male and female in case of awareness level ($\chi^2 = 5.41$, $df = 2$, $p = 0.03$).

Table 9: Knowledge of interviewed people (aware or unaware)

S. N	Gender	No of interviewed	Aware				Unaware			
			No.	%	Infected	%	No	%	Infected	%
1.	Female	62	10	16.12	5	50	52	83.87	24	46.15
2.	Male	63	7	11.11	4	57.14	56	88.88	32	57.14
	Total	125	17	13.6	9	52.94	108	86.40	56	51.85

4.2.3 Food- habit wise prevalence of intestinal parasites

It was found that out of 125 respondents, non vegetarian were found to be infected more than vegetarian. 52.46% male non vegetarian were infected and 52.54% were infective female were non vegetarian. People use pork, chicken, buff etc as meat sources. There was no significant difference in the prevalence of parasites in food habit wise prevalence ($\chi^2 = 0.434$, $p = 0.933$).

Table 10: Food habit wise prevalence of intestinal parasites

S.N.	Gender	Respondent	Vegetarian				Non-vegetarian			
			No	%	Infected no	%	No.	%	Infected no	%
1.	Male	63	2	3.17	1	50	61	96.82	32	52.46
2.	Female	62	3	4.83	1	33	59	95.16	31	52.54
	Total	125	5	4.0	2	40	120	96	63	52.5

4.2.4 Occupation-wise prevalence of intestinal parasites

According to this survey, it was found that the highest prevalence of parasites were found to be among drivers (66.67%) followed by students (61.29%), followed by housewives (52.63%) and least was among farmers (37.5%). Statically, there was no significant difference in the existence of parasites according to occupation wise ($\chi^2=5.7704$, $p=0.123$).

Table 11: Occupation-wise prevalence of intestinal parasites

S.N.	Occupation	Respondents No.	No. of positive samples	%
1.	Farmer	40	15	37.5
2.	Students	62	38	61.29
3.	Housewives	19	10	52.63
4.	Driver	3	2	66.67
5.	Service holder	1	-	-
	Total	125	65	52

4.2.5. Prevalence of intestinal parasites on the basis of sources of drinking water

Sources of drinking water are different among the Chepang people. Most of them rely on river water for various purposes. However, many NGOs/INGOs have established taps for their convenience but still they depend on rivers for their water supplement. It was found that most of the people depending on river water were found to be infected greater than (61.17%) the people using tap water (32.5%). In case of sources of water, there was significant difference shown ($\chi^2=7.85$, $df=2$, p value=0.005).

Table 12: Prevalence of parasites on the basis of sources of drinking water

S.N.	Water sources	Observation no	Positive cases	%
1.	River	85	52	61.17
2.	Tap water	40	13	32.5
	Total	125	65	52

4.2.6 Prevalence of intestinal parasites on the basis of livestock and domestic animals

Out of 125 people interviewed, most of the respondents have domestic animals like pigs, buffaloes, cows, hen, dogs, ducks and goats. The prevalence of intestinal parasites was found to be higher among those having domestic animals (52.99%) than those not having domestic animals (37.5%). There was no significant difference in the prevalence of intestinal parasites on the basis of livestock and domestic animals ($\chi^2=0.23$, $df=2$, $p=0.63$).

Table 13: Prevalence of intestinal parasites on the basis of livestock and domestic animals

S.N.	Animal husbandry	Observation	Positive cases	Positive %
1.	Having	117	62	52.99
2.	Not having	8	3	37.5
	Total	125	65	52

4.2.7 Defecation place wise prevalence of intestinal parasites:

From the analysis of questionnaire, it was found that 83.2% respondents were using toilet where as 16.8% were not using toilet. Among 83.2% toilet user 80.95% were male and 85.48% were female while 19.40% male and 1.45% female were not using toilet, they go to open areas for defecation. In toilet user, 49.01% males and 45.28% females were infected while in non-toilet user, 75% males and 77.77% females were infected with intestinal parasites. Statically, In case of defecation place wise, there was no significant difference observed ($\chi^2=6.07$, $p=0.12$).

Table 14: Defecation place wise prevalence of intestinal parasites

S.N.	Gender	No Of responders	Toilet User				Toilet Non User			
			No.	%	Infected no.	%	No.	%	Infected no.	%
1.	Male	63	51	80.95	25	49.01	12	19.04	9	75
2.	Female	62	53	85.48	24	45.28	9	1.45	7	77.77
	Total	125	104	83.2	49	47.11	21	16.80	16	76.19

4.2.8 Prevalence of intestinal parasites on the basis of medication method:

Based on the survey, people still believe in traditional methods for curing diseases hence the prevalence rate is higher among the people following traditional methods for the treatment. Maximum infection was among traditional methods 49.25% and least was direct taking medicine 29.41%. There was no significant difference in the occurrence of parasites on the basis of treatment ways ($\chi^2=3.59$, $df=3$, $p=0.16$).

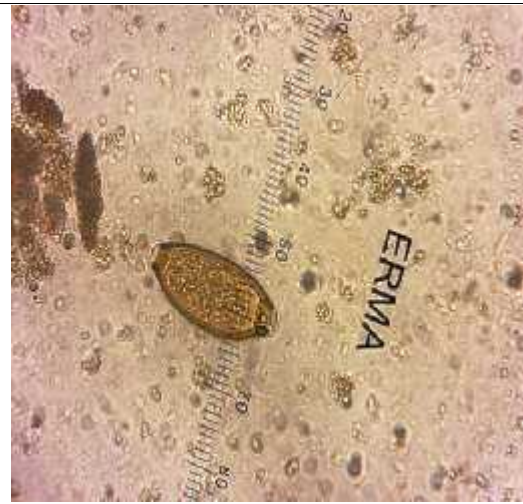
Table 15: Treatment method wise prevalence of intestinal parasites

S.N.	Methods	No. of Respondent	Positive cases	%
1.	Direct taking medicine	17	5	29.41
2.	Consulting doctors	41	14	34.14
3.	Traditional methods	67	33	49.25
	Total	125	65	52

PHOTOGRAPHS



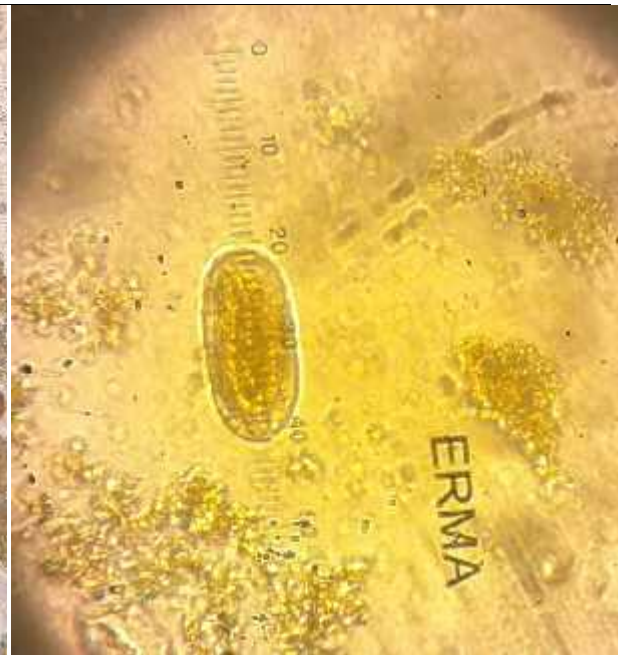
Picture 1: Egg of *Taenia solium*
Size- 25 μm



Picture 2: Egg of *Trichuris trichuria*
Size- (55 \times 20) μm



Picture 3: Egg of *Hymenolepis nana*
Size - 50 μm



Picture 4: Egg of *Strongyloides stercoralis*
Size-(52 \times 30) μm



Picture 5: Egg of *Ascaris lumbricoides*
Size- 60 μ m



Picture 6: Egg of *Entamoeba coli*
Size- 25 μ m



Picture 7: Egg of hookworm
Size- (60 \times 34) μ m



Picture 8: Storage of samples



Picture 9: Using centrifuge machine



Picture 10: Microscopic Examination



Picture 10: Chepang lady with her son on right.



Picture 11: Chepang students at chepang school.

5. DISCUSSION

Parasitic infection continue to be the present as a major challenge to the health and well being of several people around the world and mostly prevalent in the poor countries where the level of health habit status is low. Many types of helminthes and protozoal intestinal parasites affect Human that provokes a wide range of symptoms that is associated with gastrointestinal tract. Out of 60 million deaths in the world, more than 25% are accounted for parasites (Arora and Arora, 2010). In Nepal, about 4.8% people died of cholera or diarrhea (CBS, 2002).

From the calculation of data, the prevalence of intestinal parasitic infection was found to be 52%. The total samples were 125 which were collected from Chepang of Shaktikhor area, out of which 65 samples were found to be infected with either single infection or multiple infection. Helminthes infection (86.15%) was found to be higher than protozoan infection that is 13.84% among Chepang people. Hookworm infection was detected among population which might be as a result of poor farmers residing in the locality who usually work bare foot in farm and might have contaminated with infective stage of hookworm.

Sex wise parasitic infection rate was slightly higher in males (53.96%) which is similar to findings with other studies like Tandulkar *et al.* (2013); Khanal *et al.* (2011); Singh *et al.* (2014); Pradhan *et al.* (2014); Pandey *et al.* (2015); Mohammed and Hussain (2014); Wordmann *et al.* (2006); Chandrashekhar *et al.* (2005); Regmi *et al.* (2014). So this present study shows gender is not so related for parasitic infection.

As per helminthic parasites, several studies show that *Ascaris lumbricoide* was the most common infection which was 72.30% followed by *Ancylostoma duodenale* 7.69%, *Trichuris trichiuria* and *Taenia solium* had the same prevalence 6.15% and *Hymenolepis nana* had 4.16%. The least prevalent parasite was *Strongyloides stercoralis* 3.07%. This study showed similarity with other studies as well Nishiura *et al.* (2002); Oli (2016); Ojurongbe *et al.* (2014), Thapa *et al.* (2011) etc. The contamination of soil and water was found to be the main reason for the result obtained. Besides, improper defecation, unhygienic health habits, less awareness about the parasites is also other reasons. In Chepang community, the life style is below average thus they are more prone to several intestinal parasites and that may easily get transmitted among them. In case of protozoan parasite, *Entamoeba coli* were the highest prevalence (13.85%).

Regarding the age groups, the highest prevalence was found to be among age above 21 years. This might be as a result of less awareness towards the factors responsible for transmission of eggs and their parasites. They had very low economic conditions; they worked in field and walked on bare foot. Defecation usually occurred at open places which increased the risk factor. Thus adults didn't usually take any anti-helminthic and anti-protozoan drugs. However, students were sometimes accessible to these drugs at drug distribution programmes launched.

Various studies showed that the intensity of single parasitic infection was higher than that of double and multiple infections. Shrestha and Maharjan (2013); Agrawal *et al.* (2012); Opara *et al.* (2007); Uga *et al.* (2004); these studies resemble single infection was highly prevalent 81.53% followed by double infection (16.92%) and the least was triple infection (1.53%). It also shows that *Ascaris lumbricoide* infection was highly prevalent followed by *Taenia solium* 7.40% while *Ancylostoma duodenale*, *Entamoeba coli*, *Hymenolepis nana* had the similar prevalence as 5.55% of positive cases and the least was of *T.trichiuris* 3.70%.

The prevalence of parasitic infection was found to be higher among the person who used only water as hand sanitizer which showed similarity with Sah *et al.* (2016). Besides that they also use soil and water (73.3%). The use of soap and water and ash and water was 42.02% and 52% respectively. The reason behind may be lack of health education and awareness.

It was also found that among Chepang people, non-vegetarian (52.5%) was found to be infected more than vegetarian (40%). They consumed meat of different animals like goat, chicken, pigs, buff etc. Sometime they consume meat of dead animals and fowls as well. Eating of raw meat uncooked or undercooked meat had higher risk factors for transmitting the disease. Even several studies have given the same result as higher prevalence among non-vegetarian (Pandey *et al.*, 2015).

The prevalence was found to be higher among the people with livestock and domestic animals (52.99%) for transmitting the different parasites. Lack of proper sanitation of sheds and farm, sheds near or attached to homes might be cause of this results. People without domestic animals (37.5%) had lesser chances of prevalence.

In the present study, *T.solium* was found in 4 cases (7.34%) which were higher in comparisons with Parajuli (2004) in Mushar community in Chitwan and Raghav and Houston (2008) in Siraha in which prevalence rate of *Taenia* sp. is 1.63% and 2.3% respectively. Most of the households of the community were pig keeper and this may be due to the consumption of improperly cooked pork meat. It has been found that *Taenia solium* is excreted in the excreta of the patient but its proglottid was sneezed out by asymptomatic child (Pant, 2016).

Here in this study, it was found that still people believed in traditional methods of treatment hence prevalence rate was also higher among them (49.25%). People also took medicine without concerning and the rate is 29.41% and that of consulting with doctor was 34.14%.

People use river water for several purposes drinking, washing, cleaning etc. Several Chepang were depending on this source of water. Among the river water user 61.17% were infected. While 32.5% of total tap water user were infected. This might be as a result of contamination of sources of water with sewage and garbage. The use of consumption of unboiled water resulted in 95% of cases parasitic infection (Bhandari, 2011; Shan L.V. *et.al*, 2013; Gibney 2017).

Pre-school and school-age children as well as women of childbearing age, including adolescent girls, tend to have the higher proportion of worm infections. Although intestinal worms can infect all members of populations, these specific groups are at greater of heavy infections than others and are more vulnerable to the harmful effects of chronic infections.

6. CONCLUSION AND RECOMMENDATIONS

This study was carried out to observe the prevalence of gastro intestinal parasites in Chepang communities of Shaktikhor, Chitwan. Out of 125 samples, 65 samples were positive which 52% of the total was and the infection rate was found to be higher among the males than females with a slight difference (male -34 infective (53.96%) and female-31(50%). The age group above 21 years was found to be highly infected than other age group which was 69.44% followed by 10-20 years (45.33%) and the age group 2-10 years were 42.85% infected. It was found that the *Ascaris lumbricoides* infestation was higher among them. Altogether seven types of parasites were detected in the lab where helminthes have the wide variants namely *Ascaris lumbricoides* (72.22%), *Strongyloides stercoralis* (3.70%), *Trichuria trichiuris* (3.70%), *Hymenolepis nana* (5.55%), *Taenia* (7.40%), and Hookworm (5.55%) and *E.coli* (5.55%). The single infection was higher than double and multiple infections. The double infection of (*E.coli* and *A.lumbricoides*) was higher that is 40%. Multiple infections were of *Trichuris trichuria*, *Strongyloides stercoralis* and hookworm which covers 1.53% of total. Helminthic infection was found more than protozoan infection among Chepang people that might be the outcome of consumption of dirty and contaminated food and water that degrades the health of them. The lack of knowledge, poor sanitation and unhygienic practices were the major causes of prevalence of intestinal parasites among Chepang people. This study analyzed that Chepang people were living nomadic life and in terms of health and hygiene, their level of standard was low. In present study, it was found that people of that community are uneducated and hence was unaware about different intestinal parasites and follow witch doctors for the treatment of the diseases. 86.40% of people were unaware and 13.60% were aware about parasitic infection. Similarly, the use of river water was higher among them. River gets polluted easily. Epidemiological surveys is necessary in Chepang community so that helped to explore the sanitary condition, level of knowledge, socio-economic conditions etc. that can be fruitful for producing data for the reduction of parasites in near future. Further, this study reflected that the poverty, low awareness level, consumption of raw and uncooked food, poor environmental hygiene, contaminated water etc. were the major factors responsible for parasites' multiplication and distribution. Thus, it was very vital to change their concept and knowledge level, improve their sanitary condition so as to uplift their behavioral prospectus.

RECOMMENDATIONS

Following recommendations are extracted after this research work which is listed below:

-) Since the knowledge regarding health and hygiene is low, basic health education programmed should be launched among them from time to time. Public health education in the school curriculum must be made compulsory.
-) Feeding habit should be changed in the way like use of boiled water, avoiding consumption of raw and uncooked vegetables etc. Approach of Pure and safe drinking water facility should be made easier among the tribal community.
-) People should avoid defecation in the open areas and use of toilet should be promoted.
-) Every house must contain proper latrine facility so as to minimize the infection that passes on through fecal-oral route.
-) People should not walk on bare foot and farmers should be more careful while walking on field and animal sheds.
-) Research work should give emphasis to the study of intestinal parasites of different tribes.

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ANNEX-1

Table showing measurement of size of various parasitic eggs:

S.N	Parasites	Size (μm)	Content of egg	Morphological characteristics of egg
1.	<i>Ascaris lumbricoides</i>	(60-75 \times 40-50) μm	Embryonated	Oval or round
2.	<i>Hymenolepis nana</i>	35-40 μm	Embryonated	Smooth, thin outer shell with hexacanth embryo
3.	<i>Taenia</i> spp	31-43 μm	Embryonated	Spherical and brown in colour
4.	<i>Trichuris trichiura</i>	(50-55 \times 22-24) μm	Embryonated	Barrel shaped with mucus plugs at each pole.
5.	Hookworm	(60-75 \times 36-40) μm	Embryonated	Thin transparent hyaline shell, colourless
6.	<i>Strongyloides stercoralis</i>	(50-58 \times 30-34) μm	Embryonated	Oval, transparent, thin-shelled

ANNEX-2
QUESTIONNAIRE

Questionnaire for baseline health survey among Chepang people of Shaktikhor of Chitwan District.

1. a. S No/ Household No b. Date

2. Name of the respondent

3. a. Age b. Sex c. Locality
d. Occupation

4. Are you literate Yes No?

If literate, Primary level Secondary level Higher secondary level

General read and write

5. Where do you get drinking water for family?

a. Tap b. Kholsa c. Well d. Others

6. How do you use water for family?

a. Direct water b. Boiled c. Using germicides d. Others

7. What do you use to clean hands?

a. Water only b. Water soap c. Water and Ashes d. All of above

8. When do you wash hands?

a. Before meal b. After meal c. After defecation d. After working in field

9. Do you cut nail regularly? Yes No

If yes when?

a. Once a week b. Once a month c. Randomly

10. Which type of domestic animals do you keep?

- a. Hens
- b. Chicken
- c. Buffalo
- d. Goats
- e. Cow
- f. Buffalo
- g. Pigs

11. What type of food habit you have?

- a. Vegetarian
- b. Non vegetarian

12. If Non vegetarian, which meat frequently you take?

- a. Pork
- b. Chicken
- c. Buffalo
- d. Fish
- e. Mutton

13. How do you prepare your meat to eat?

- a. Raw meat preparation
- b. Sekuwa
- c. Half cooked
- d. Boiled
- e. Well cooked

14. How do you clean the vegetables and fruits?

- a. Rubbing on clothes
- b. Tap or well water
- c. Without water

15. Have you suffered by diarrhea / dysentery worms?

Yes No

16. Have you taken de-worming tablet before?

Yes No

17. Do you know the methods of prevention of worm infection?

Yes No

If yes, what are they?