PREVALENCE OF GASTRO-INTESTINAL HELMINTH PARASITES OF INDIGENOUS DARAI AND KUMAL COMMUNITIES OF SALYANTAR, DHADING

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Institute of Science and Technology

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DECLARATION

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

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RECOMMENDATION

This is to recommend that the thesis entitled "Prevalence of gastro-intestinal helminth parasites of indigenous Darai and Kumal communities of Salyantar, Dhading" has been carried out by Arjun Thapa for the partial fulfillment of Master's Degree of Science in Zoology with special paper Parasitology. This is his original work and has been carried out under my supervision. To the best of my knowledge, this thesis work has not been submitted for any other degree in any institutions.

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LETTER OF APPROVAL

On the recommendation of supervisor Dr. Mahendra Maharjan, Professor, Central Department of Zoology, Tribhuvan University this thesis submitted by Arjun Thapa entitled "Prevalence of gastro-intestinal helminth parasites of indigenous Darai and Kumal communities of Salyantar, Dhading" is approved for the examination ~~and submitted to the Central Department of Zoology~~ in partial fulfillment of the requirements for Master's Degree of Science in Zoology with special paper Parasitology.

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CERTIFICATE OF ACCEPTANCE

This thesis work submitted by Arjun Thapa entitled "Prevalence of gastro-intestinal helminth parasites of indigenous Darai and Kumal communities of Salyantar, Dhading" has been accepted as a partial fulfillment for the requirements of Master's Degree of Science in Zoology with special paper Parasitology.

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LIST OF ABBREVIATIONS

Abbreviated form	Details of abbreviations
AD	Amino Domino
CBS	Central Bureau of Statistics
CDZ	Central Department of Zoology
χ^2	Chi-square value
et al.	And his associates
GI	Gastro intestinal tract
IFAD	International Fund for Agricultural Development
IJV	Inland Jungle Village
ILO	International Labor Office
IPI	Intestinal Parasitic Infection
KAP	Knowledge Attitude and Practice
NE	North East
NFDIN	National Foundation for Development of Indigenous Nationalities
No.	Number
NTDs	Neglected Tropical Diseases
p-value	Probability value
RPS	Resettlement Plan Scheme
STH	Soil Transmitted Helminth
SDGs	Sustainable Development Goals
T.U	Tribhuvan University
VDC	Village Development Committee

ABSTRACT

In Nepal gastrointestinal helminthiases are endemic in indigenous people living in low land area of Nepal. Their life expectancy is well below in comparison to the other people. Darai and Kumal are marginalized group of people with low socio-economic condition. The present study was carried out on indigenous people (Darai and Kumal) from Salyantar ward no 1 and 2 of Dhading, Nepal from June to September 2018 in order to determine the prevalence of intestinal helminth. A total of 189 (82 from Darai and 107 from Kumal) stool samples from people of two indigenous caste were collected and examined by direct smear methods under microscope along with structured questionnaire related with KAP in relation to intestinal parasitic infection and result were analyzed by "R" version 3.5.1. Among them 25.92% (28.04% of Darai and 24.29% of Kumal) were found positive for one or more intestinal helminths parasites. All together seven species of helminths were encountered. For Darai people, *Trichuris trichiura* (18.29%) was found to be most common than *Ascaris lumbricoides* (4.87%), *Strongyloides stercoralis* and *Hymenolepis nana* (3.65%), *Taenia* spp. (1.21%) without *Enterobius vermicularis* and hookworm infections. As for the Kumal people, *Trichuris trichiura*, *Ascaris lumbricoides* and *Hymenolepis* (7.47%) were most common infection prevailed over *Strongyloides stercoralis* (3.73%), *Enterbius vermicularis* (2.80%), *Taenia* spp. (1.86%) and hookworm infections (0.93%). According to age and sex, in Darai both male and female > 50 years showed high parasitic prevalence 27.27% and 55.55% respectively. But in Kumal male > 50 years showed high parasitic prevalence (27.27%) and female belongs to 36-50 years showed high parasitic prevalence (35.71%). The prevalence of single infection was higher than double and multiple infections with significant difference ($p < 0.05$). Among education attainment, from primary and higher level people showed significant association with prevalence of helminth infection. Similarly, occurrence of intestinal helminth parasites may attributed to lack of knowledge, awareness deficient, food habit, poor sanitation, open defecation, drinking water treatment method, hand washing agent were found to be significantly associated ($p < 0.05$) except occupation, source of drinking water and treatment methods ($p > 0.05$). Hence a remarkable prevalence of helminth infection was indicated by study carried out among Darai and Kumal indigenous people of Salyantar, Dhading. Improvement in personal hygiene, environmental sanitation, periodic MDA, health education, awareness on modes of infection and prevention measures implementation in order to reduce and eradicate these infections.

1. INTRODUCTION

1.1 Background

Indigenous peoples belonging to particular place rather than coming to it from somewhere else or originating naturally in particular place (www.lexico.com). In context to Nepal they are officially described as indigenous nationalities (Adivasi/ Janajati). They make up for 35.8% of the country's total population (CBS, 2011). Despite constituting such a significant proportion of the population, indigenous people have been marginalized in term of language, culture and political as well as economic opportunities throughout the history. Indigenous people in Nepal have distinct culture, language and belief system. They live across the country - the mountain, the hills and the plains. According to Nepal's government 59 indigenous communities have been officially and legally recognized and categoried into 5 caste groups (Endangered, highly marginalized, marginalized, disadvantage and advantage) (NFDIN, 2010). Among them Raute are nomads, forest dwellers and less civilized than rest of the caste group. Most of them rely on agriculture only few indigenous peoples are advanced and better off. However, in terms of ethnic identity, language, religion, culture and backward indigenous peoples have fallen victim to discrimination at the hand of the dominant groups.

In Nepal, there are many indigenous group residing in different parts of the country among them two indigenous ethnic group Darai and Kumal are distributed mostly in Terai region of the country including hilly region, Salyantar of Dhading. Before two three decades they were particularly involved making pot items and brick but most of them are gradually shifting to farming, carpenter and mason. In comparison of Darai, Kumal people maintain well hygiene and sanitation. Salyantar, Dhading is surrounded by four rivers viz. Budhigandaki in west, Netrawoti in south, Hyapinge in east and Kastey in north making it as table like shape. This area is only one plain area of Dhading. Due to its geographical landscape, lack of sufficient good quality of drinking water but possess well facilitated health post, private hospitals, clinic, school and college. Being indigenous community they follow their own traditional culture, behavior, rules and regulation probably that may lead socio-economically backward. This area is one of the biggest and longest Tar of Nepal (www.ddcdhading.gov.np).

According to the WHO, it is estimated that more than 24% of people worldwide get infected with intestinal parasitic infections (IPIs). The majority of whom reside in

developing countries (WHO, 2015). Parasitic infections caused by intestinal helminths and protozoan parasites are among the most prevalent infections in humans in developing countries. In developed countries, protozoan parasites more commonly cause gastrointestinal infections compared to helminths. Intestinal parasites cause a significant morbidity and mortality in endemic countries (Haque, 2007) but it is estimated that almost half of the world's population is infected by helminth parasites at some point of their life (De Silva *et al*., 2003). Similarly, in Nepal indigenous population covers 38.8% of total population. Majority of them live in extreme poverty and deprived socio-economic condition of landlessness. As a result, they are unable to access primary health care, basic education and safe drinking water (ILO, 2005; IFAD, 2010). In Nepal, among top ten infectious diseases gastro-intestinal helminth infection contributed almost 100% in some indigenous communities (Magar *et al*., 2011). Such IPIs are amongst the most common infections worldwide with the most endemic regions being Africa, Southeast Asia, China, South India and South Africa (Ahmed *et al*., 2011; Ng *et al*., 2014).

Intestinal parasitic disease particularly helminths are still neglected disease often physicians neglected them, although worms can cause several clinical manifestations since, they usually have insidious effect on growth and development that rarely cause attendance at health centers. Yet they precisely causes chronic effects, affecting more than two billion people with lifelong infection that have force the public health community to reassess the importance of these infections. And recognition of the simplicity, safety, low cost and efficacy of treatment has now resulted in major global initiatives to achieve control (Awasthi *et al.,* 2003). Soil transmitted helminth (STH) have been documented as causing impairment of growth and nutrition. In areas where helminths are common, deworming activities can be done once or twice per year to the population at risk (Saboya *et al.,* 2011). Infection occurs in contact with parasite's eggs or larva in contaminated soil by direct (without reaching into soil), modified direct (undergoes stages of development in soil) and penetration of the skin (after further development in soil) reach into circulation (Helbig *et al.,* 2012). Infections caused by gastrointestinal helminths are one of the most common health problems for poor people and important cause of anemia and malnutrition which may result in reduced physical and mental development (Ahmed *et al*., 2011; Brooker *et al*., 2008). Yajima *et al.* (2009) hookworm infection is more prevalent in population having frequent contact with soil and presence of latrine alone not sufficient to reduce prevalence of helminths.

Helminths infections are endemic in tropical and sub-tropical region of world (Hotez *et al*., 2003; WHO, 2019). Likewise global phenomenon such as climate change, migration, environmental changes, drug resistance and economic factor make things more complicated into public health (Thompson and Conlan, 2011). In lowland of Nepal due to warm and moist climate, parasitic prevalence was found to be high. Socio-economic condition can influence the social behavioral of individual with respect to access to primary health, primary education, improve sanitation and safe water are important contributor of helminths infections (Hotez *et al*., 2011; Rai *et al*., 2002). Helminths infections rate could increases into the individuals with lack of water supply, primary health, house hold hygiene and personal behavior (Soares *et al*., 2011; Karan *et al*., 2012). Similarly, poverty, poor sanitation, inadequacy of nutrient, lack of health care, contaminated food and water result in helminth infection. It leads to serious hazards and may even leads to death due to chronic infection. IPI is the major health problem among people of developing countries (Engels and Savioli, 2006). Use of treated water associated with lower odds of STH infection. Piped water access was associated with lower odds of *Ascaris lumbricoides*, *Trichuris trichiura* but not any STH infection. Access to sanitation was associated with decreased likelihood of infection with any STH *T. trichuris* and *A. lumbricoides* but not with hookworm infection. Wearing shoes was associated with reduced odds of hookworm infection and infection with other STH. Hand washing both before eating and after defecating was associated with lower odds of *A. lumbricoides* infection and soap use or availability was significantly associated with lower infection for any STH (Strunz *et al*., 2014). STH infection is associated with hygiene behavior but not with nutritional or socio-demographic characteristics. Health policy focusing on changing individual hygiene behaviors might be useful in addressing STH infection in Nepal (Parajuli *et al*., 2015). Similarly, among the indigenous community in low land Nepal, who is heavily dependent on natural resources for their livelihood. They are more likely to get geohelminths than other counter parts in Nepal due to their occupation and dependency in nature. They have limited excess to the health service, education and safe drinking water due to higher cost of those services in Nepal (Gyawali *et al*., 2013). Most of the indigenous people are infected with IPIs, often shares their house with domesticated animals such as goat, pig, poultry, cow and buffalo. Access to sanitation is also nonexistent and waste disposal is often done at bank of the forest (Uprety *et al*., 2010; Magar *et al*., 2011).

1.2 Objectives

1.2.1 General objective

- To determine the prevalence of gastro-intestinal helminth parasites of indigenous Darai and Kumal communities of Salyantar, Dhading.

1.2.2 Specific objectives

- To determine the prevalence of intestinal helminth parasites of Darai and Kumal communities.
- To assess the Knowledge, Attitude and Practice (KAP) of communities in relation to helminth infection.

1.3 Significance of the study

Most of the developing country including Nepal have a major health problem particularly with helminth parasites especially socio-economically backward communities people (indigenous). Such type of research will help to meet two Sustainable Development Goals (good health and well beings and clean water and sanitation) among seventeen goals. Although, sufficient information are available regarding helminthiasis of children but information regarding parasitic prevalence among indigenous community people is still not sufficient. The present study was carried to determine the prevalence of helminths parasites in Darai and Kumal people on the basis of (age, sex, occupation, literacy) and to assess the knowledge, attitude and practice in relation to helminths infection. This type of survey is new in this area. Thus the study can help for making plan, prevention, control and treatment of helminthes parasites in Darai and Kumal people.

2. LITERATURE REVIEW

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

During relatively short history on Earth, the parasites on *Homo sapiens* was observed and result revealed that human act as a host to about 300 species of parasitic worms and more than 70 species of protozoa which may be derived from our primate ancestors and some of them may be transmitted from domesticated animals and some may be acquired when they come in contact with the source of infection (Ashford and Crewe, 1998). The first written records of parasitic infection came from Egyptian civilization from 3000-400 BC, particularly the Eblers Papyrus of 1500BC discovered at Thebes, Hippocrates (460-375 BC) knew about worms from fishes, domesticated animal as well as human. Paulus Aegineta (AD 625-690) described about *Ascaris*, *Enterobius* and tapeworms and gave description of the infection caused by them. Linnaeus described and named six helminth worms, *Ascaris lumbricoides*, *Enterobius vermicularis*, *Gordius medinensis*, *Fasciola hepatica*, *Taenia solium* and *Diphyllobothrium latum*. Hookworms in human were found in 1838 by the Italian physician Angelo Dubini and link between the worms and disease was established by Wilhelm Griesinger in 1854 (Kean, 1978). *Strongylodes* infection was well accounted by Grove. The human liver fluke was first discovered by James McConnel in 1875. Tyson was the first person to recognize the Heat of the tapeworm. Louis Alexis Norman, a physician recognized *S. stercoralis* in 1876 (Cox, 2003). Modern parasitology developed in the 19th century with accurate observation by several researcher and clinicians (Arora and Arora, 2010). Rudolphi classified all the parasites known up to his time. There are good account of ascariasis by Grove (1990) and Goodwin (1996).

2.1 Global Scenario

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

Most of the research was done on school going children and result revealed that *A. lumbricoides, T. trichiura, A. duodenale, E. histolytica, H. nana* were the vital parasites among the children of developing countries (Opera *et al*., 2007; Sehgal *et al*., 2010; Goli *et al*., 2014; Wordemann *et al*., 2006; Abahussian, 2005; Almir *et al*., 2013; Akingbade *et al*., 2013; Mohammed, 2010; Mote *et al*., 2005; Orish *et al*., 2017; Ragunathan *et al*., 2010; Mukutmoni and Khanum, 2017; Silver *et al*., 2018). Not only in Children *A. lumbricoides* and *E. histolytica* were the most predominant parasite in HIV positive patients (Akinbo *et al*., 2010). Similarly helminth parasites *A. lumbricoides*, hookworms and *T. trichiura* were also reported from indigenous school children of South America (Scolari *et al*., 2001).

Similarly, different researches have done on indigenous people of different country regarding the intestinal parasites. Chin *et al*. (2016) conducted a survey in two indigenous sub-ethnic groups in Malaysia. In Temuan community trichuriasis (64.2%) was found to be most common infection, preceding hookworm infection (34.0%), ascariasis (7.5%), giardiasis (14.4%) and amoebiasis (3.8%). As for the Mah Meri community trichuriasis (77.5%) prevailed over ascariasis (21.3%), hookworm (15.0%), giardiasis (7.5%) and amoebiasis (3.8%). Significant difference in proportion of trichuriasis, ascariasis and hookworm infection were observed between sub-ethnic groups and poly parasitism was found to be common among two community people. Likewise, in Negritos (indigenous) community people of Malaysia 416 participants were taken from lnland Jungle Village (IJV) and Resettlement Plan Scheme (RPS). Prevalence of STH was found to be significantly higher in IJV (91.3%) vs. RPS (83.1%). But prevalence of *Trichuris trichiura, Ascaris lumbricoides* were found to be high in RPS compared to IJV and

prevalence of hookworm infection, *Entamoeba* sp, *Blastocystis* and flukes were found to be high in IJV than RPS (Muslim *et al*., 2019).

Likewise, 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 methods and have shown 68.4% positive with 57.4% single infection while 28.1% double, 9.6% triple, 4.1% quadruple and 0.4% multiple infection. The most commonly observed species was *Necator americanus* 23.8% but *Taenia* spp and *Trichuris trichiura* were found as least prevalence (0.8%) followed by roundworm, *S. stercoralis*, *H. nana*. 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* followed by *Giardia lamblia*, *Strongyloides stercoralis*, *Ascaris lumbricoides*, hookworm and *Trichuris trichiura*.

In Asian continent, studies have been carried out in tribal people which showed that Bharia tribe of India found out that 30% of children suffer from severe anemia and 50% of children had intestinal parasites with common parasites as *A. duodenale* (16.3%) and *A. 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 (Choubisa *et al*., 2012). Pestehchian *et al*. (2015) examined 655 total samples of inhabitant and tribe population of Chelgerd, Iran and revealed that 56% people suffer at least one intestinal parasites where 67.7% in tribal population and 43% in inhabitants. Mishra *et al.* (2013) in Gujarat and Jaroli *et al*. (2012) in Rajasthan prevalence of parasitic infection due to protozoa were found to be higher than helminth. Similarly, in Uttar Pradesh prevalence of STH among school children were found to be 76.6%. The prevalence was more than 50% in the six of the nine agro- climatic zones. *Ascaris lumbricoides* was found to be most prevalent STH (69.6%) followed by the hookworm (22.6%) and *Trichuris trichiura* (14.6%) (Ganguly *et al*., 2017).

Almegrin, (2010) studied among immune-compromised patient in Saudi Arabia and found no significant differences in intestinal parasites between male and female. There was evidence that prevalence of *Ascaris lumbricoides* and *Trichuris trichiura* infection statistically greater in periurban areas compared to urban and rural while the prevalence of hookworm was higher in rural areas (Pullen *et al*., 2014). But Mohammed, (2010) in

Saudi Arabia prevalence of intestinal parasite was found to be high in rural area than urban.

From different studies, Intestinal parasites were found more common in females (Akingbade *et al*., 2013; Khanum *et al*., 2013; Onyido *et al*., 2016; Odugbemi *et al*., 2015) and common parasites found were *A. lumbricoides*, Hookworms, *T. trichiura*, *H. nana* (Opera *et al*., 2007; Sehgal *et al*., 2010. But in some studies prevalence of parasites were found high in male (Mulambalah and Ruto, 2016; Kirorei *et al*., 2014).

Intestinal parasites on the school going children has been studied by different researcher (Opera *et al*., 2007; Ragunanthan *et al*., 2010; Almir *et al*., 2013; Kirorei *et al*., 2014) that exposed that single infection of parasites was higher than double and multiple infections. The overall prevalence was found higher in age group below 12 years and STH infection were significantly more common compared to protozoa infections (Ngui *et al*., 2011). The prevalence and diversity of intestinal parasitic infection is high in population of central Asia with wide range of nematodes, cestodes and protozoa are found common in Afghanistan (Korzeniewski, 2016).

In case of researches carried out in different African countries revealed that in northwestern Amazonia it was found that intensity of *Ascaris lumbricoides* was found to be high followed by *Entamoeba* spp. and *Giardia* spp. however, the frequency of helminth such as round worm and hookworm was found to be high in adult (Oliveira, 2016). Similarly, in western Kenya more prevalence was recorded in 9-12 years age than 5-8 years age school going children and more male people were infested than female (Kirorei *et al*., 2014). Similarly in different region of Africa researchers were found to be high prevalence of helminth parasites (Mirisho *et al*., 2017; Mulambalah and Ruto, 2016; Odugbemi *et al*., 2016; Onyido *et al*., 2016; Orish *et al*., 2017).

Parasitic helminthes and protozoan infection are also common among the Inuit in Canada and Greenland and among Alaskan natives where the major helminthiasis among Arctic population is trichinellosis, diphylobothriasis and echinococcosis but toxoplasmosis infection was considered very high with giardiasis and cryptosporidiosis (Hotez, 2010).

Similarly in eastern Europe, the soil transmitted helminthes infections, giardiasis and toxoplasmosis remain endemic whilst in southern Europe, vector-borne zoonoses have emerged like leishmaniasis and Chagas diseases (Hotez, 2011). Besides, intestinal parasitic disease such as enterobiasis, giardiasis and ascariasis are detected more

frequently in Romania but their importance is definitely surpassed by trychinellosis, cystic echinococcosis and toxoplasmosis (Neghina, 2011). When a study 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 females. Common parasites were *G. intestinalis, E. histolytica*, *C. cayetanensis*, *A. lumbricoides, H. nana*, *T.* spp, *S. stercoralis, E. vermicularis* and *T. trichiura* (Masucci, 2011). Similarly, in Columbia the prevalence of protozoa and helminth from children belongs to six indigenous communities were found to be 98% and 16.7% respectively (Florez *et al*., 2012).

In case of Australian continent, the prevalence of *Blastocystic* 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).

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 personnel hygiene, piteous environmental care, inadequate health service and lack of proper and necessary awareness of the transmission mechanism and life cycle pattern of the parasites (Mote *et al*., 2005; Ubhavawardana *et al*., 2018; Odugbemi *et al*., 2016; Chin *et al*., 2016; Muslim *et al*., 2019).

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

2.2 National Scenario

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 disease (Rai *et al*., 2005). Sharma (1965) reported that the round worm infestation is very common in some parts of our country. He studied that 976 stool samples and found 40% round worm infestation in Bhaktapur area.

In Nepal, the common intestinal parasitic infection among people is ascariasis, amoebiasis, giardiasis and taeniasis (Acharya, 1997). Similarly, prevalence of STH infection from difference places of Nepal was found to be 3.3%-51.5% (Parajuli *et al*., 2015). In eastern region of Nepal, prevalence of intestinal protozoa and helminthes parasites were found to be 5.72% and 2.45% respectively. Among them *Giardia intestinalis* was found to be most prevalent infection (3.34%), followed by *Entamoeba histolytica* (1.96%) and Hookworm (0.97%). Parasitic infection also observed high among 20-50 years age and lowest in the below 10 years (Baral *et al*., 2017). Pokharel *et al*. (2004); Agrawal *et al*. (2012); Uga *et al*. (2004) reported that protozoan infection rate was higher in school going children of Nepal than helminth infection.

Different researches were done on indigenous people of Nepal. Among them in Kumal people of Gaidakot, hookworm was found to be common parasite with 30.87% followed by *Ascaris lumbricoides*, *Hymenolepis nana*, *Trichuris trichiura*, *Strongyloides stercoralis* and *Taenia* spp. with 16.10%, 6.04%, 3.35%, 2.68% and 2.01% respectively. People belong to over forty year showed high prevalence (Gyawali, 2012). Similarly in indigenous group of chitwan the prevalence of helminth infection was found to be 54% (Gyawali *et al*., 2013).

Among different indigenous groups of Nepal, prevalence of parasites were found to be high in female than male (Thapa, 2019; Dhakal, 2018; Budhathoki, 2015; Yadav, 2014; Karki, 2003). But in some indigenous groups of Nepal prevalence of intestinal parasites were found to be high in male than female (Oli, 2016; Yadav, 2017; Adhikari, 2018).

Tharu, (2006) surveyed among Chepang adults at Taklung VDCs of Gorkha and examined 410 stool samples of different age group and sexes, found that stool 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 female 93.22%. Prevalence of *Ascaris lumbricoides* was found as common among helminth followed by hookworm, *T. trichiura*, *E. vermicularis*, *S. stercoralis* in helminthes while *E. histolytica* showed high prevalence (24%) than *G. lamblia*, *Cryptosporidium* sp, *C. cayetanensis* in protozoan parasites.

Khadka *et al*. (2020) prevalence of intestinal parasites in Nawalparasi among Chepang and Musahar prevalence of intestinal parasites were found to be 39.8% and 33.3%

respectively. The most predominant helminth was *Ascaris lumbricoides* (15.6%) while *Entamoeba histolytica* was found to be most prevalent protozoan (5.4%).

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. Similarly, when studied among the school children revealed that 19.8% were infected with common protozoan parasite and highest positive rate was in Dalit (20.3%) and least in Indo-Aryan (19.6%). Higher prevalence of *Ascaris lumbricoides* was found followed by *Trichuris trichiura*, *Strongyloides stercoralis*, hookworm, *Taenia solium*, *Hymenolepis nana* and *Enterobius vermicularis* reported by Shrestha and Maharjan (2013).

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 one month to 60 months old, were admitted and measured height and weight, among them 167 were checked for intestinal permeability and 173 for parasitic infection. Subedi *et al*., (2012) studied on feeding practices in Chepang communities which revealed that literate mother were more conscious and early initiatives at breastfeeding than illiterate mother. The meal feeding practice and diverse food for children were found lower.

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

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

Different researches were carried on school going children from different locality, their result revealed that among helminth *Ascaris lumbricoides* was found to be high (Sah *et al*., 2016; Tiwari, 2012; Yadav and Mahato, 2017). Similarly *Taenia* spp also showed as high by Shrestha *et al*., 2016 and Shah *et al*., 2013 and in few research prevalence of *H. nana*, hookworm and *T. trichiura* was showed high by Pradhan *et al*., 2014; Kunwar *et al*., 2016 and Khanal *et al*., 2011 respectively. Similarly, among protozoa prevalence of

Giardia intestinalis was found to be common by Chandrashekhar *et al*., 2005; Tandukar *et al*., 2013; Tiwari, 2012; Shrestha *et al*., 2016. Besides that *Entamoeba histolytica* also found as common parasites by Khanal *et al*., 2011; Kunwar *et al*., 2016.

In indigenous groups, *Ascaris lumbricoides* was reported as most prevalent helminth by Karki, 2003; Dhakal, 2018; Adhikari, 2018; Yadav, 2017; Budhathoki, 2015; Yadav, 2014. In Kisan community of Palpa prevalence of *Trichuris trichiura* was found to be high (Thapa, 2019). Likewise among protozoa prevalence of *Entamoeba histolytica* was reported high by Yadav, 2014; Budhathoki, 2015; Adhikari, 2018. Besides that *Entamoeba coli* was reported as high prevalence by Dhakal, 2018 and Thapa, 2019. Yadav, (2017) reported that *Giardia lamblia* was found to be high in Musahar community of Siraha.

Low socio-economic status, poor hygienic condition (Khanal *et al*., 2011; Gyawali, 2012), lack of pure drinking water, lack of proper sanitary disposal and lack of education (Gwawali *et al*., 2013; Khadka *et al*., 2020) are to be the root cause of parasitic infection.

It was found that non-vegetarian was highly infected with parasitic infection than vegetarian (Pandey *et al*., 2015; Oli, 2016; Dhakal, 2018; Yadav, 2017). Besides, intestinal parasites are associated with hand washing behavior (Gyawali *et al*., 2013; Khadka *et al*., 2020), labourer and farming professional Gyawali *et al*., 2013 and Tandukar *et al*., 2013 respectively, unawareness (Pandey *et al*., 2015), lack of toilet (Gwawali *et al*., 2013; Khadka *et al*., 2020).

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

3. MATERIALS AND METHODS

3.1 Study Area

Dhading is a part of province of no. 3, is one of the seventy seven district of Nepal. It lies in the Himalayan north to the Kathmandu, occupying 1926 sq. km area. It contains 11 village municipality and 2 municipalities. Among them present study area lies in Tripura Sundari Gaunpalika. Two indigenous groups of people, Darai and Kumal of Salyantar 1 and 2 wards were selected for this study as the study population. This place was famous for production of Mas, Dahl and Ghayeaa. It is also one of the longest and biggest Tar of Nepal. The major cast living in study area was Darai and Kumal (www.ddcdhading.gov.np). Indigenous people of the study area have limited access of sanitation, education, occupation and safe drinking water. Health practices of the communities depended on native plants and traditional healers known as Dhami and Jhakri who perform ancient right to protection, blessing and healing. In this area 7292 peoples were living among them 807 (368 male and 439 female) Darai and 2242 (1013 male and 1229 female) Kumal. Similarly among 1791 household, 555 household were without toilet, 1029 household with flush toilet, 200 with ordinary toilet and 7 household still unknown (CBS, 2011). It lies in LAT 27.98765/LON 84.81209, 29^0 NE and 554 m altitude.

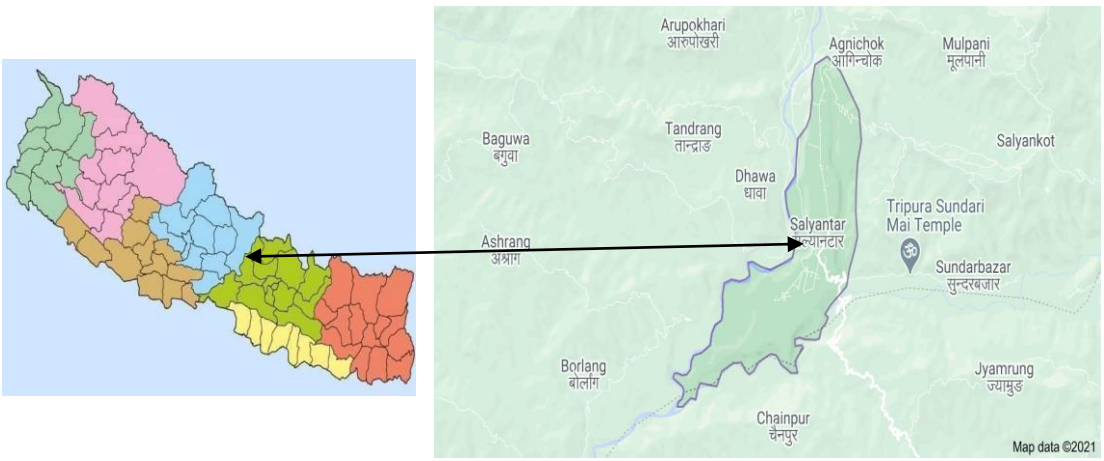

Figure 1: Map showing study area

3.2 Materials Required

i. Compound microscope

ii. Collecting vials

iii. Wooden spoon

iv. Glass slides

v. Coverslips

vi. Cotton

vii. Ocular and stage micrometer

viii. Bamboo toothpicks

ix. Stickers

xi. Dustbin

xii. Test tubes

xiii. Filter paper and Beaker

3.3 Chemicals Required

i. Potassium dichromate

ii. Normal saline

iii. Lugol's iodine solution

iv. 70% alcohol

v. Soap

vi. Formalin

3.4 Study Design

The present study was designed to determine the parasitic helminth prevalence among two indigenous groups of people by applying faecal sample collection and examination method along with the structured questionnaire survey to correlate them with the knowledge, attitude and practices as shown in the flow chart (Fig 2).

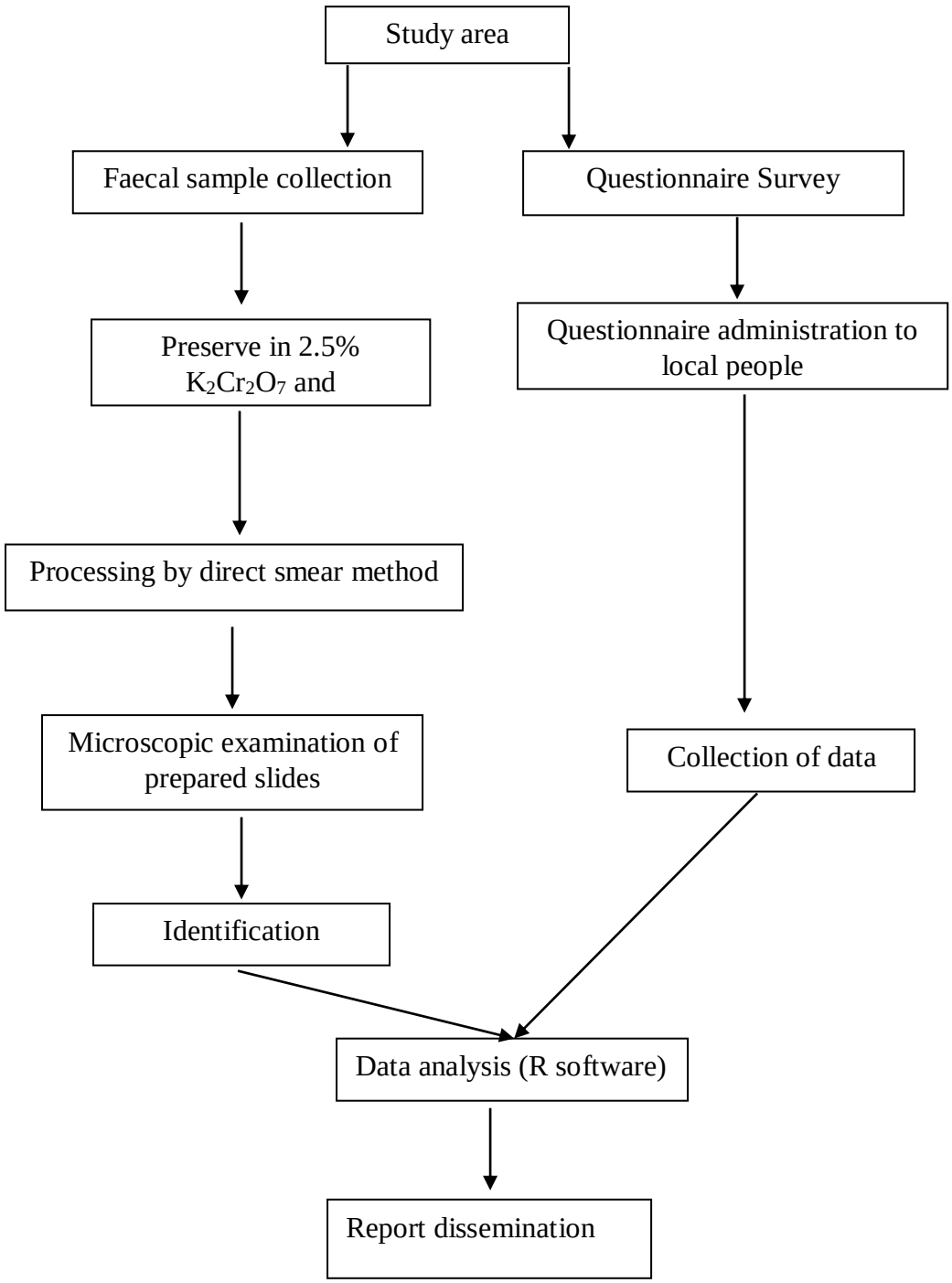

Figure 2: Flowchart showing study design

3.5 Sample Size

On the basis of objectives, purposive sampling technique was done for collection of stool sample. Total sample size of the study was 189 (82 from Darai and 107 from Kumal) from two indigenous (Darai and Kumal) peoples of Salyantar, Dhading. Stool samples were collected irrespective of age, sex, occupation and literacy from the study area.

3.6 Sample Collection and Preservation

The stool samples were collected during a month period (June, 2018). The verbal consent was taken from the study population. The participants were provided labeled sterile vials and application sticks with clear instruction on how to collect the samples. From each targeted person about two gm of fresh stool sample was collected. Each of the vial was checked for its labeling and proper amount of sample. The collected stool samples were preserved in potassium dichromate (2.5%) and transported to the Parasitology Laboratory of Central Department of Zoology, Kathmandu, Nepal for further investigation of eggs and larva of intestinal parasites.

3.7 Preparation of Chemicals

3.7.1 Potassium Dichromate ($K_2Cr_2O_7$)

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

3.7.2 Normal Saline

Normal saline was used for observing the characteristics movement of parasites. This solution was prepared by dissolving 8.5 gm of sodium chloride in 1000 ml of distilled water. It was used in unstain preparation (Soulsby, 2012).

3.7.3 Lugol's Iodine

Iodine solution was used for studying the internal characters for identification of the species of protozoan parasites as well as helminths eggs a stained preparation was required. Iodine solution was prepared by dissolving 10 gm of potassium iodide in 100 ml of distilled water and slowly 5 gm of iodine crystals were added in it. The solution was filtered and then kept in bottle (Soulsby, 2012).

3.8 Methods of Observation

Both stained and unstained smear preparation were first examined under the low power (100X) of microscope. Observation was starting from one end of the slide to another. When the parasite's eggs were seen then objects were centered and focused under high power (400X) when necessary for detail diagnosis.

3.9 Examination of Stool Samples

Laboratory diagnosis of helminth parasites in the collected stool samples includes

3.9.1 Macroscopic examination

Immediately after collection of vials the stool samples but before addition of potassium dichromate ($K_2Cr_2O_7$) as preservatives, the samples were examined directly through naked eyes. The samples were observed for color, odour, blood content, mucus content as well as visual parasites and their segments.

3.9.2 Microscopic examination

Direct smear or wet films, both unstained and stained were prepared for this purpose. For accurate identification of the parasites, measurements of the size of the parasite's eggs were done with ocular micrometer.

3.9.2.1 Unstained smear preparation of stool/saline wet mount

A small quantity of fecal sample was taken with the help of wooden applicator and emulsified with normal saline on a clean glass slide. Then cover slip was placed over it and excess of fluid was removed with cotton or filter paper. Unstained smear made chromatid bodies more visible. The smear was observed under microscope so as to demonstrate helminths eggs and larvae.

3.9.2.2 Stained smear preparation of stool/Iodine wet mount

A portion of stool samples was put on glass slide to which a drop of Lugol's iodine was added and mixed. Then cover slip was placed over it and excess of fluid was removed with cotton/filter paper. The smear was observed under microscope. Iodine wet mount was necessary for the identification and study of the nuclear character.

3.10 Questionnaire Survey to Assess the KAP

In order to assess the KAP analysis and to co-relate with parasitic prevalence, a set of structured questionnaire was prepared. The questionnaire was pretested among colleagues

(friends) and tested among community people who are not involved in the present study. The final set of questionnaire was prepared and administered in the study population to assess knowledge, attitude and practices of Darai and Kumal people in relation with helminth parasites and its infection. In brief the questionnaire includes (a) Socio-demographic: address, age, gender and socio-economic status (b) Knowledge: knowledge on IP, modes of transmission, prevention method and awareness on IP (c) Attitude (d) Behavioural data: drinking water treatment method, food habit, hands washing agent, source of drinking water drinking water and latrine facilities (e) Participant's medical history: any complaints of abdominal pain/discomfort, nausea and vomiting. Peoples were interviewed in their own wise and for below ten years their survey form were fill up by respective guardians. After all, the entire questionnaires were checked for accuracy and completeness.

3.11 Data Analysis and Interpretation

Existence of helminths eggs in stool was used as the outcomes variable thus all the subjects were categorized into infected and not infected based on the presence of at least one parasitic helminth egg. After that, all data as well as laboratory findings were analyzed according to their cast, species-wise, age - sex, occupation and literacy infection rate. Thus analyzed data was interpreted by representing with table and bar-diagram. Besides that prevalence of intestinal parasites with KAP were calculated. With the help of "R" version 3.5.1 software package, p-value and Pearson's Chi-square value were calculated, which helps to interpretation for significance association between parasitic prevalence and different variables (if p-value < 0.05 it showed significant relation but p-value > 0.05 it showed insignificant relation).

4. RESULTS

4.1 Result of stool examination

The Darai and Kumal community people are considered as backward indigenous people of Salyantar Dhading. Helminth parasitic infection causes significant impact in the public health. In order to assess the helminths parasitic infection 189 community people including Darai and Kumal were surveyed with structured questionnaire and feacal examination. The result revealed as a whole 25.92% helminthic parasitic prevalence in Salyantar, Dhading.

4.1.1 Prevalence of helminthic parasitic infection

Both Darai and Kumal community people live together sharing same environment. The parasitological result revealed almost similar prevalence rate between two indigenous community people without insignificant statistical association (χ^2 = 0.26873, p = 0.6042) (Fig 3).

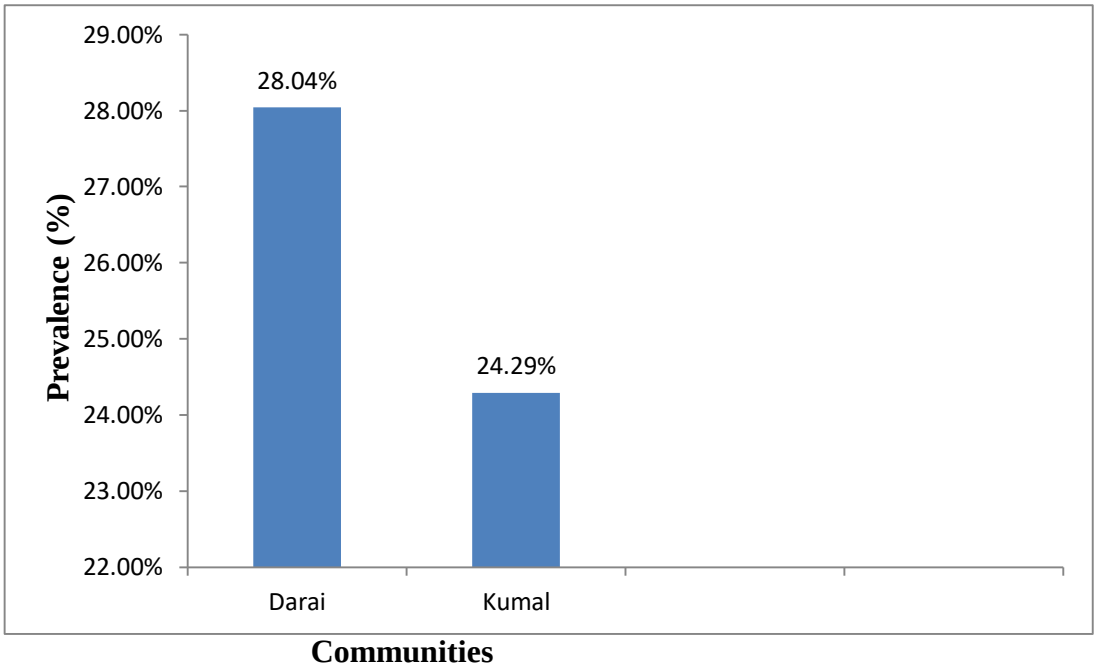

Figure 3. General prevalence of helminth parasites (n=189)

4.1.2 Species-wise Prevalence of Intestinal Helminth Parasite

In order to determine the species wise prevalence of helminths parasitic infection all together seven species were recorded that belongs to two classes. Among them parasites belongs to nematodes showed maximum species than cestodes. Besides that prevalence of

Taenia spp found to be low but prevalence of *Trichuris trichiura* was found to be high with statistically significant ($\chi^2 = 4.5447$, $p = 0.0302$) (Table 1).

Table 1. Species-wise prevalence of helminth parasite

Class	Parasites	Darai (N=82)	Kumal (N=107)	Total (N=189)	Chi-value (χ^2)	p-value
Nematode	*Ascaris lumbricoides*	4.87%	7.47%	6.34%		0.4592
	Enterobius vermicularis	-	2.80%	1.58%		
	Hookworm	-	0.93%	0.52%		
	Strongyloides stercoralis	3.65%	3.73%	3.70%		0.9765
	Trichuris trichiura	18.29%	7.47%	12.16%	4.5447	0.03302
Cestode	*Hymenolepis nana*	3.65%	7.47%	5.82%		0.252
	Taenia spp.	1.2%	1.86%	1.58%		0.706

4.1.3 Age and Sex-wise Prevalence of Helminth Parasite

In indigenous surveyed communities, respondents were categories into four age groups among them high prevalence of helminth infection was found to be in >50 year and low prevalence was found in 36-50 age group in Darai people but in Kumal people high prevalence was found in 36-50 age group and low in >50 year age. Similarly according to sex high prevalence of helminth was recorded in male of >50 year and low in 21-35 in Darai and prevalence of female was high in >50 year and low in 36-50 year was recorded. Similarly in Darai 5-20 year age group male showed low prevalence and >50 year showed high prevalence but 21-35 year age male showed no any prevalence but age of 36-50 year female showed high prevalence and >50 year showed low prevalence. Thus age and sex

wise prevalence of helminth infection showed statistically insignificant (χ^2 = 6.5458, p = 0.08787) (Table 2).

Table 2. Age and Sex-wise prevalence of helminth parasite

Age Groups (In Years)	Darai			Kumal		
	Male (N=37)	Female (N=45)	Total (N=82)	Male (N=49)	Female (N=58)	Total (N=107)
5-20 (N=61)	25%	23.07%	24%	25%	25%	25%
21-35 (N=57)	20%	33.33%	28%	-	30.43%	21.87%
36-50 (N=35)	25%	12.5%	16.66%	11.11%	35.71%	26.08%
>50 (N=36)	27.27%	55.55%	40%	27.27%	20%	25%
Total (N=189)	24.32%	31.11%		18.36%	29.31%	

4.1.4 Occupation-wise Prevalence of Helminth Parasite

Occupationally, most of the respondents were farmer. The result revealed that maximum housewives of both community showed high parasitic prevalence but students showed low prevalence in Darai and other occupational groups including teacher, tailor and businessman showed low prevalence in Kumal. Statistically there was insignificant difference in prevalence of helminth parasite according to occupation of people (χ^2 = 2.570, p = 0.4617) (Table 3).

Table 3. Occupation-wise prevalence of helminth parasite

Occupation	No of Darai	Prevalence %	No of Kumal	Prevalence %	Chi-value (χ^2)	p-value
Students	24	20.83	30	23.33	0.14153	0.7068
Farmers	37	27.02	40	22.5	0.41257	0.5207
Housewives	10	40.00	14	35.71	0.24309	0.622
Others	11	36.36	23	21.73	3.6846	0.05492

4.1.5 Literacy Status-wise Prevalence of Helminth Parasite

Although in surveyed area most of the respondents were illiterate and showed the high prevalence of helminth infection in Darai but in Kumal higher education attained people showed high prevalence rate. Similarly low parasitic prevalence was also showed just reverse in between two community. Primary level and higher level education attainment people showed statistically parasitic significant difference (p-value < 0.05) (Table 4).

Table 4. Literacy status-wise prevalence of helminth parasite

Education attainment	No of Darai	Prevalence %	No of Kumal	Prevalence %	Chi-value (χ^2)	p-value
Illiterate	30	40	38	26.31	2.8264	0.09273
Primary level	19	10.52	35	22.85	4.5559	0.03281
Secondary level	27	29.62	28	21.42	1.3174	0.2511
Higher level	6	16.66	6	33.33	5.5589	0.01839

4.1.6 Helminth Parasitic Infection among Darai and Kumal Community

Infection with single helminth parasite in both community people were found more common than double and multiple infection. Regarding double infection in Darai co-

infection of *Trichuris* with *Strongyloides* was found higher than *Trichuris* with *Ascaris* but lack of multiple infections. Similarly in Kumal people, double infections were showed by *H. nana* with *Trichuris*, *Ascaris* with *Strongylooides* and hookworm with *Strongyloides*. In multiple infection co-infection of *Ascaris*, *Trichuris* and *Taenia* spp and co-infection of *Ascaris*, *Trichuris* and *Enterobius* were observed. That showed prevalence of helminth parasitic infection was found to be statistically significant (χ^2 = 106.55, p = 2.2e-16) (Fig 4).

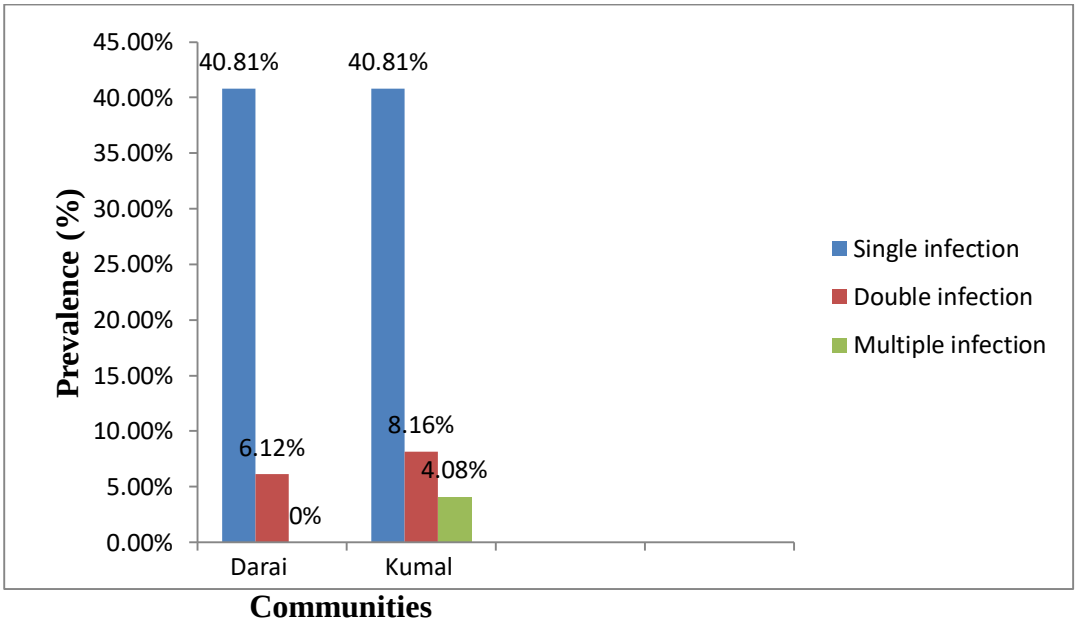

Figure 4. Helminth parasitic infection among Darai and Kumal

4.2 Assessment of knowledge, attitude and practice of people in relation to helminth infection

Among the 189 study population structural questionnaire survey was carried out from each individual except the children below 10 years. On behalf of children survey form was filled up from their respective guardians.

4.2.1 People's knowledge and parasitic prevalence

The survey was conducted to cover the knowledge of Darai and Kumal community people with respect to intestinal parasitic infection, transmission, preventive measure as well as awareness on intestinal parasites.

Result indicated that half of the Darai people don't have knowledge of intestinal parasites. Most of them had heard about *Ascaris lumbricoides* commonly called Juka. The

parasitological result showed that Darai people who don't have knowledge of intestinal parasite were found to be infected comparatively high than people having knowledge of intestinal parasites. Similarly in Kumal community people most of the people were unaware regarding intestinal parasite although they were comparatively less infected. Thus knowledge on intestinal parasite was found to be statistically significant (χ^2 = 9.1675, p = 0.002464) (Table 5). Similarly in both community people very few had knowledge as contaminated water and food are major sources of intestinal parasite infection. In both indigenous community people lack of knowledge about modes of transmission showed high infection but statistically significant association with modes of transmission of intestinal parasite (χ^2 = 7.7715, p = 0.005308). In these communities maximum prevalence was observed among people who don't know about any preventive methods. It showed statistically significant association (χ^2 = 32.562, p = 8.498e-08). Most of the respondents were found to be unaware on intestinal parasite that corresponded to high prevalence and statistically significant (χ^2 = 5.5692, p = 0.01828).

Table 5. Assessment of Knowledge of people in relation to helminth infection

Knowledge	Number of Darai people	Prevalence %	Number of Kumal people	Prevalence %	Chi-value (χ^2)	p-value
Knowledge on IP						
Yes	37	10.81	43	16.27	9.1675	0.0024
No	45	42.22	64	29.68		
Mode of Transmission of IP						
Know	13	15.38	29	10.34	7.7715	0.0053
Don't know	69	30.43	78	29.48		
Prevention methods of IP						
Don't know	13	53.84	14	64.28		

Partially know	49	26.53	60	18.33	32.562	8.5e-08
Know	20	15.0	33	18.88		
Awareness on IP						
Aware	24	16.66	36	13.88	5.5692	0.0188
Unaware	58	32.75	71	29.57		

4.2.2 People's attitude and parasitic prevalence

Both Darai and Kumal community people believes that diarrhoea and dysentry are regular and common phenomenon so the disease are not fatal and these disease don't kill the people. Having this attitude of the community people indicated high parasitic prevalence with statistically significant association (χ^2 = 6.9003, p = 0.008618) (Table 6). In case of barefoot most of the respondent's opinion were found to be misunderstanding type that is due to barefoot can cause maximum chances of foot injury rather than intestinal parasitic infection. Their attitude towards possibility of intestinal parasitic infection particularly hookworm infection by walking bare foot showed statistically significant association with parasitic infection (χ^2 = 19.833, p = 4.935e-05). Most of the respondents were agree with IPS is due to contaminated food and water and those who are agree showed low prevalence than disagree respondents. Statistically it was found to be significantly associated (χ^2 = 42.766, p = 6.171e-11). Similarly most of the respondents know that domestic pets like hens, cats, duck, dog and pig can transmit and causes zoonotically important diseases. Their attitude towards transmission of intestinal parasitic infection due to playing with domestic pets showed significant association with parasitic infection (χ^2 = 13.971, p = 0.0009253). In surveyed community, knowledge about *Ascaris* was found to be common so most of the respondents agreed that *Ascaris* can causes the vomiting and some respondents have ever seen *Ascaris* in their vomit. Their attitude towards particular symptom of *Ascaris* cause vomiting showed statistically significant association with parasitic infection (χ^2 = 10.727, p = 0.001056).

Table 6. Assessment of Attitude of people in relation to helminth infection

Attitude	No of Darai	Prevalence %	No of Kumal	Prevalence %	Chi-value (χ^2)	p-value
Diarrhoea and dysentery can kill the people						
Agree	17	11.76	23	13.04	6.9003	0.008618
Disagree	65	32.30	84	27.38		
Walking with bare foot cause IPI						
Yes	6	16.66	13	7.69		
No	57	33.33	61	36.06	19.833	4.935e-05
Don't know	19	15.78	33	9.09		
Most of IPI is due to contaminated food and water						
Agree	63	14.28	89	13.48	42.766	6.171e-11
Disagree	19	73.68	18	77.77		
Playing with domestic pets can cause IPI						
Agree	47	17.02	57	12.28		
Disagree	22	50	31	32.25	13.971	0.0009253
Don't know	13	30.76	19	47.36		
Ascaris can causes the vomiting						
Agree	69	23.18	87	19.54	10.727	0.001056
Disagree	13	53.84	20	45.0		

4.2.3 People's practice and parasitic prevalence

Due to its geographical distribution in this area most of the household consume tap water than from direct resource and river. Common sharing of source of drinking water was not found to be statistically significant (χ^2 = 2.0678, p = 0.3556) (Table 7). People using

drinking water directly without any treatment acquired higher infection as compared to filter and whereas people using boiling water was found to be less infected. Statistically, drinking water consumption had significant association with parasitic infection (χ^2 = 25.703, p = 2.622e-06). Most of the people use closed toilet and very few of them defecate on open place. The result revealed that defecation was significantly associated with parasitic infection (χ^2 = 6.2987, p = 0.04288). According to food habit most of them were non-vegetarian. Non-vegetarian consumed all kind of meat item available in that area such as buffalo, chicken, goat, pig and fish. Prevalence of non-vegetarian was found to be slightly higher than vegetarian. Statistically, there was no significant difference between food habit and parasitic infection (χ^2 = 15.582, p = 7.899e-05). Most of the people use water and soap as cleaning agent which showed less parasitic prevalence and high prevalence was showed by the people who were using soil and water as cleaning agent. Statistically, there was significant difference in the prevalence of intestinal parasitic infection with cleaning agent used (χ^2 = 44.192, p = 1.374e-09). Similarly in that community most of the people were found to be believed in consulting with doctor for treatment than direct taking medicine and traditional (Dhami and Guruwa) method. Among them who believed on traditional method showed slightly higher parasitic prevalence than rest methods. Statistically, there was no significant difference in the prevalence of intestinal parasitic infection of people on the basis of treatment methods (χ^2 = 1.7751, p = 0.4117).

Table 7. Assessment of Practice of people in relation to helminth infection

Variables	Practices	No of Darai	Prevalence %	No of Kumal	Prevalence %	Chi-value (χ^2)	p-value
Drinking water treatment method	Boiling	19	5.26	27	3.70		
	Filter	24	29.16	37	22.85	25.703	2.6e-06
	Not treatment	39	38.46	45	37.77		
Defecation	Open	10	80	17	64.70		
	Closed	54	5.35	63	9.32	6.2987	0.04288
	Dug	18	66.66	27	33.33		
Food habit	Vegetarian	2	-	5	20	15.582	7.8e-05
	Non-veg	80	28.75	102	24.50		

Hand washing agent	Only with water	18	27.77	27	25.92	44.192	1.3e-09
	Ash and water	24	37.5	32	34.375		
	Soil and water	11	63.63	9	55.55		
	Soap and water	29	6.89	39	7.692		
Source of drinking water	Tap	49	30.61	69	24.63	2.0678	0.3556
	Direct resource	17	29.41	27	25.92		
	River	16	18.75	11	18.18		
Treatment methods	Direct taking medicine	17	29.41	29	17.24	1.7751	0.4117
	Traditional	19	33.33	15	26.66		
	Consulting doctor	56	26.78	63	26.98		

5. DISCUSSION

Intestinal parasitic infection directly relates with personal hygiene and sanitation as well as social, demographic and economic standards. Since most of the indigenous people in the country are far backward, health status along with parasitic diseases seemed to be very high compared to others. Most of the earlier studies carried out among the indigenous people from different part of the country showed high parasitic prevalence. Among them Chepang as well as Musahar indigenous people which are considered to be most backward showed high parasitic prevalence 91.11% among the Chepang of Gorkha (Tharu, 2006), 48.8% Kumal in Chitwan (Gyawali, 2012). Similarly among Magar of Palpa revealed 66.87% parasitic prevalence (Karki, 2003). Besides these other indigenous people from Chitwan also shown to be high (Adhikari, 2018, Gyawali *et al*., 2013). While some of the indigenous people, the prevalence rate had shown to be less such as Chepang and Musahar of Makwanpur and Nawalparasi 36.6% (Khadka *et al*., 2020), Kisan of Baglung 31.32% (Thapa, 2019), Meche of Jhapa 27.33% (Dhakal, 2018), Tamang of Kathmandu Sangla (Budhathoki, 2015) 31.20% and Musahar of Siraha 13.65% (Yadav, 2017). Not only in Nepal, the parasitic disease prevalence scenario of other countries are also seems to be same. The parasitic prevalence among indigenous people of Malayasia (Muslim *et al*., 2019, Chin *et al*., 2016), Philippines (Delaluna *et al*., 2020) and Amazonian southern border (Sandoval *et al*., 2017) had also shown to be high. The parasitic disease status of Darai and Kumal (28.04% and 24.29%) of Salyantar also was as earlier studies.

On the basis of the published and unpublished previous research, that suggest the risk factor for STH is related to personal hygiene, socio-demographic and socio-economic factor. Mostly helminth parasites are identified into three classes among them prevalence of parasitic infection belong to class nematode was found to be high with common helminth parasites *Trichiris trichiura* and *Ascaris lumbricoides.* In most of the research done on helminth in national and foreign country, result revealed that 5-6 parasites were reported but in this study altogether seven species of helminthes were detected. Among them prevalence of *Trichuris trichiura* (12.16%) was found to be high. Such similar finding was showed by other researchers also (Thapa, 2019, Chin *et al*., 2016 and Muslim *et al*., 2019). Similarly hookworm also reported as a least prevalence helminth parasite (0.52%) harmonized by the research done on Musahar community of Ithari (Yadav, 2014). Besides that *Taenia* spp. was not found as common cestode in various researches.

But in Darai and Kumal, prevalence of *Taenia* spp was found to be 1.58%. That suggests the Darai and Kumal people of that community consume various meat products from pork as well as beaf and buff. In different region of indigenous community of Nepal also showed prevalence of *Taenia* spp. Besides that indigenous people of Magar of Palpa and Chepang of Chitwan showed high prevalence. While some of the researchers showed low prevalence of *Taenia* spp. (Thapa, 2019, Dhakal, 2018, Yadav, 2017, and Yadav, 2014) in some of the indigenous people.

Most of the study done among indigenous people had shown that female were infected comparatively high than male. That also supported the present result. In both indigenous groups male were less infected than female. But there was no signification variation between sex groups in relation with parasitic prevalence. This might due to sharing of similar type of environment, feeding habit and due to male dominant community, female are most disadvantage group member they are force to leave school, marriage in early age and more female were involved in house work such as cooking, cleaning, washing as well as agricultural labor in spare time. Which is similar to the finding on prevalence of parasitic infection on indigenous people of Nepal and other country (Karki, 2003; Tharu, 2006; Gyawali, 2012; Chin *et al*., 2016; and Muslim *et al*., 2019). Similarly according to age group, the prevalence of parasitic infection was found to be insignificant. But in most of the finding based on age group below 20 years showed high parasitic prevalence (Thapa, 2019; Yadav; 2017; Karki 2003) which is antagonistic to present findings due to totally under care of parents, they are not involved in field work and government also provided deworming time to time.

Due to low socio-economic standard in indigenous group most of the people involved in agriculture as farmers but housewives showed high parasitic prevalence in both Darai and Kumal (40% and 35.71%) respectively. Which is similar to the result shown by indigenous sub-ethnic groups of Peninsular Malaysia (Chin *et al*., 2016). That reflect those housewives not only engage in housework and also involved in farming. According to government plan based on to prevent from STH infection different education learning package health education and MDA plan were lunched and result found to be effective to minimize parasitic prevalence in school age children. In overall students of both communities showed low parasitic prevalence (22.08%). Similar finding was showed by other researchers also (Tharu, 2006; Gyawali *et al*., 2013 and Oli, 2016).

Parasitic prevalence in between Darai and Kumal were compared according to their educational attainment. Surprisingly, the result indicated significantly high parasitic prevalence among Kumal people attending higher level education compared to Darai community people. Rest of the other categories seemed to be similar.

Indigenous population with poor knowledge, attitude and practices further aggravating the situation. Due to different risk factor in-relation to parasitic prevalence (sanitation, personal hygiene, food habit) it might be slightly different. Various research done on indigenous peoples of Nepal showed high parasitic prevalence by single helminth parasites followed by double and multiple infections. Similar observation was previously reported by Gyawali, 2012 and Gyawali *et al.*, 2013. The same case was observed on other country (Chin *et al.*, 2016; Delaluna *et al.*, 2020). Polymorphism is not merely a marker for poor sanitation, poverty but its prevalence rate are crucial to know individual with multiple infection may suffer from multiple morbidity and increased susceptibility to other infections.

The prevalence of parasitic infection depends not only on socio-economic, socio-demographic and geographical location, other factors also play vital role for the parasitic prevalence like knowledge, attitude and practices regarding parasitic infection. Among them knowledge and practices are considered as indicator for prevalence of parasitic diseases. All the parameter belongs to knowledge (knowledge on IP, modes of transmission, prevention methods and awareness) were found to be highly significant association with parasitic prevalence in Darai and Kumal indigenous people. Such results were showed in Gaidakot by Gyawali *et al*. (2013). In other indigenous groups from Siraha, Saptari and Chitwan also reported high parasitic prevalence on unawareness than aware people. Regarding knowledge, not only reported high parasitic prevalence from Nepal it also reported from indigenous people of Malaysia by other researchers (Chin *et al*., 2016; Muslin *et al*., 2019).

In present result, people's perceptions and prevalence of parasite infection was found to be highly associated in indigenous community. Among different attitude related variables mostly walking with bare foot, contaminated food and water and playing with domestic pets found to be correlated with parasitic prevalence. Such result showed by the other researchers also (Khadka *et al*., 2020; Gyawali, 2012) among the indigenous people of Nepal. Besides that in Malaysia parasitic disease prevalence and people's attitudes on

above three parameters also showed same result as in indigenous people of Nepal (Chin *et al*., 2016; Muslim *et al*., 2019).

In most of the research prevalence of parasitic infections were found to be intimately related on people's practices. The study revealed that the drinking water treatment methods used by people had played an important role in helminth infections. Such result might be harmonized with the result showed by Tharu, 2006 and Khadka *et al*., 2020. Similar findings also reported by Oli, 2016; Dhakal, 2018; Chin *et al*., 2016 and Muslim *et al*., 2019. Similarly, those people who used open place for defecation showed high helminth prevalence in comparison to facilitated latrine. That might be due to contaminating the surrounding environment throughout night soil. It is harmonized with the result showed by Gyawali *et al*., 2013; Adhikari, 2018; Chin *et al*., 2016 and Muslim *et al*., 2019.

Likewise, according to food habit, most of the report revealed that non-vegetarian showed high prevalence of STH infection. This might be due to consumption of different source of meat without properly cooked. Such similar finding was showed by other researcher Oli, 2016; Dhakal, 2018 and Yadav, 2017 too on their earlier studies. Most of the study reflected helminth infection and use of cleaning agent had reciprocally related. Thus people who used soap and water showed low parasitic prevalence. It is an agreement on report published by Chin *et al*., 2016; Gyawali *et al*., 2013 and Muslim *et al*., 2019 but those used soil and water as hand washing agent showed high STH infection. Because of lack of awareness, believe in traditional customs, low socio-economic status and careless about personal hygiene. Such result was showed by Yadav, 2014; Oli, 2016 and Dhakal, 2018.

Similarly, on the basis of drinking water no significant difference was found but high parasitic prevalence was found on people who directly used water from resources that might be polluted to open place defecation practice of people near water resources and mixing of night soli. Such similar finding was shown by other researchers Budhathoki, 2015; Gyawali, 2012; Gyawali *et al*., 2013; Chin *et al*., 2016 and Muslim *et al*., 2019. Likewise the finding of the study revealed that those people believed in traditional method such as Dhami and Guruwa showed high parasitic prevalence. It is due to lack of knowledge, attitude, cultural and behavior variation. Such result was harmonized with other researchers also (Oli, 2016; Dhakal, 2018; Adhikari, 2018, Yadav, 2014 and Yadav, 2017).

6. CONCLUSION AND RECOMMENDATIONS

6.1 Conclusion

The findings suggest that among helminthes infection, particularly *Trichuris trichiura*, *Ascaris lumbricoides* and *Hymenolepis nana* are endemic in this indigenous population of Dhading. Likewise, the current study showed significant difference of single, double and multiple helminth infections between Darai and Kumal. Future study should include other different indigenous people of Nepal are heterogenous in their levels of education, lifestyle, hygiene practices. Additionally, the findings that both the Darai and Kumal indigenous people harbored high levels of overall parasitic infections highlights the urgent need for diseases control measures, such as periodic MDA, provision of safe water and improved sanitation. To consolidate and ascertain long term sustainability of disease control greater efforts are required to instil understanding of personal hygiene and health education among indigenous people in order to enhance their knowledge and awareness about the transmission and prevention of these infections.

6.2 Recommendations

The followings are the recommendation for the prevention and control of helminths parasites.

- ✓ Public health education should be provided to every member of tribal community and it should be included in school curriculum.
- ✓ Mass Drug Administration (MDA) program against helminth parasites should be implemented every six months, especially before peak season of parasitic infection.
- ✓ Habit of defecating on open field and near water resource should be prohibited.
- ✓ Pure and safe drinking water facility should be made easily accessible for the tribal communities.
- ✓ Proper hand washing practices should be demonstrated in community as well as school.

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ANNEX-1

PHOTOGRAPHS

Photo No. 1

Photo No. 2

Photo No. 1 & 2: Observation of stool samples under microscope in CDZ's Laboratory.

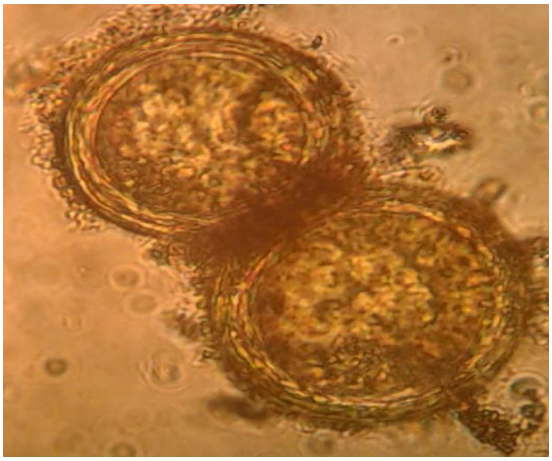

Photo No. 3: Egg of *Ascaris lumbricoides* at 400X

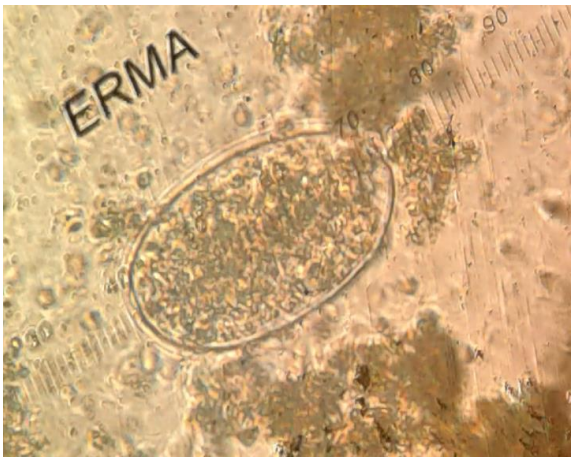

Photo No. 4: Egg of Hookworm (291 µm x 18 µm) at 400X

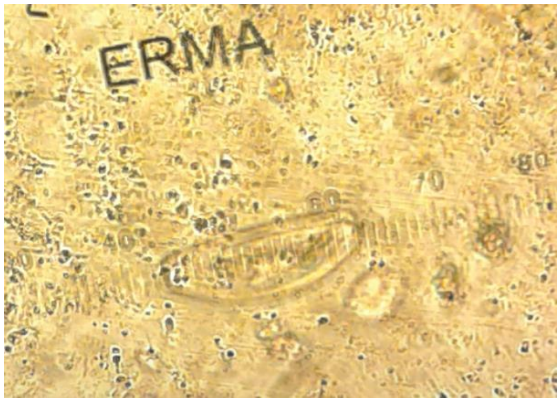

Photo No.5: Egg of *Enterobius vermicularis* (50 µm x 25 µm) at 400X

Photo No.6: Egg of *Hymenolepis nana* (40 µm) at 400X

Photo No. 7: Egg of *Strongyloides stercoralis*

(65 µm x 32 µm) at 400X

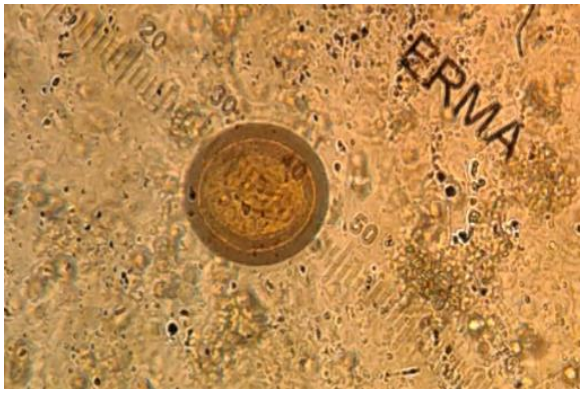

Photo No. 8: Egg of *Taenia* spp.

(33 µm) at 400X

Photo No. 9: Egg of *Trichuris trichiura*

(52 µm x 22 µm) at 400X

ANNEX-2

QUESTIONNAIRE

1. Name:

Age: ……. Sex: ……. Locality: ……. Education: …….

2. What is your main occupation?

a. Farmer b. Housewives c. Students d. Others

3. Have you any knowledge about IPI

a. Yes b. No

If yes, What are they

4. Disease prevalent in the community

a. Bacterial b. Viral c. Parasitic d. Others

5. Have you ever seen worms in your/family member faeces

a. Yes b. No

If yes, When?

1. Within year 2. Between five years 3. Many years ago

What you called them? If you know give the Common name

6. Do you know the cause of worm infection?

a. Yes b. No

If yes, What are they? ……

7. Do you know the prevention of worm infection?

a. Yes b. No

If yes, What are they? ……

8. How do you treat incase of infection?

a. Traditional methods b. By taking medicine c. By consult doctor

9. Do you know any disease transmitted by

a. Water, Yes/No, If yes ……

b. Food, Yes/No, If yes ……..

c. Meat, Yes/No, If yes ……..

10. Do you know the cause of diarrhoea?

a. Yes b. No

If yes, What are they? ……

11. Diarrhoea and dysentery can kill the people

a. Agree b. Disagree c. Others

12. Diarrhoea and dysentry transmitted by water

a. Agree b. Disagree c. Others

13. *Ascaris* can causes the vomiting

a. Agree b. Disagree c. Others

14. Playing with domestic pets can cause IPI

a. Agree b. Disagree c. Don't know

15. Walking with bare foot cause IPI

a. Yes b. No c. Don't know

16. From where do you get drinking water

a. Tap b. River c. Direct resource

17. How do you use water for family?

a. Filter b. Boil c. Not treatment

18. Where do you defecate?

a. Open field b. Toilet c. Dug toilet

19. Do you wash your hands before eating and after defecate?

a. Yes b. No

If yes, by which agent

1. Soap and water 2.Water only 3. Soil and water 4. Ash and water

20. What type of food habit you have?

a. Vegetarian b. Non-vegetarian

- If non-vegetarian, which meat frequently you eat?

Chicken Buffalo Mutton Pork/Pig Fish

- How do you prepare your meat to eat?

Boiled Well cooked Half boiled Sekuwa masu