

**PREVALENCE AND INTENSITY OF INTESTINAL PARASITE  
INFECTIONS AMONG CHILDREN AT MUKETURI PRIMARY  
SCHOOL, OROMIA REGIONAL STATE, ETHIOPIA**

**A Thesis Submitted to the School of Biological Sciences and Biotechnology, College  
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## **DEDICATION**

I dedicate this thesis manuscript to my family for their continuous love inspiration and support that helped me in accomplishing this thesis.

## **STATEMENT OF AUTHOR**

First, I declare that this is my original work and all sources of material used for this have been duly acknowledged. This thesis has been submitted in partial fulfillment of the requirements of M.Sc. degree at the Haramaya University and deposited at the University Library to be made available to borrowers under rules of the Library. I also declare that this thesis is not submitted to any other institutions anywhere for the award of any academic degree, diploma or certificates.

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## **BIOGRAPICAL SKETCH**

The author was born in 1968 in Ebinat town; South Gonder Zone of Amhara Regional State .He attended his Primary Education at Ebinat Elementary School and his Secondary Education at Ebinat Secondary School, Ebinat.

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## **ACRONYMS AND ABBREVIATIONS**

AIDS	Acquired Immune Deficiency Syndrom
CDC	Centers for Disease Control
DAL	Disability Adjusted of Life Year
ELSA	Enzyme linked Immunosorbent Assay
EPG	Egg per Gram of Feaces
GBD	Global Burden of Disease
GI	Gastro Intestinal
IPIs	Intestinal Parasitic Infections
MDA	Mass Drug Administration
PCR	Polymerase Chain Reaction
SEA	Standard Error of Mean
SPPS	Statistical Package for Social Siense
STH	Soil Transmitted Helminths
WHO	World Health Organization

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**PREVALENCE AND INTENSITY OF INTESTINAL PARASITIC  
INFECTIONS AMONG CHILDREN AT MUKETURI PRIMARY  
SCHOOL, OROMIYA REGIONAL STATE, ETHIOPIA**

**ABSTRACT**

*Intestinal parasitic infections (IPI) are among the major health problems in many countries including Ethiopia. The objective of the present study was to determine the prevalence and intensity of intestinal parasitic infections and to identify the major protozoan and helminthes parasitic species among primary school children in the study area. The design of the study was a cross-sectional school based parasitological survey*

*involving 400 sample student populations drawn from Muketuri Primary School using stratified random sampling method during April –May 2017. The stool samples were examined using direct wet mount, formol ether concentration, modified Ziehl Neelsen method and Kato- Katz methods. From 400 total study participants, 196(49%) were males and 204(51%) were females. The study result showed that 77 (38 % ) males and 100 (49.5% ) females were infected with one or more intestinal protozoan parasites. The overall prevalence of Entamoeba histolytica, Gardia lamblia and Cryptosporidium species, were 28.5%, 12.75% and 3%, respectively. The major intestinal helminth parasite species identified were Ascaris lumbricoides, Hookworms, Trichuri trichuria and Hyminolipes nana with their prevalence of infection 16 %, 5.5%. 1.5% and 4%, respectively. The overall intensity egg burden of A.lumbricoides, Hookworm and T. trichuria were 872.4, 283 and 605.3 eggs per gram of feaces, respectively. Based on these results it can be conclude that intestinal parasitic infections were one of the major problem of the study area.*

*Keywords; Helminths, Intestinal parasitic infection, Muketuri, Oromia, Prevalence, Protozoan, School Childern.*

## 1. INTRODUCTION

Intestinal parasitic infections (IPI) constitute a global health burden causing clinical morbidity and health burden in 450 million people, where it constitute reproductive age and children in developing countries (Quinui *et al.*, 2006). Infection rates with IPIs are highest in children living in Sub-Saharan Africa SSA, followed by Asia and then Latin America and the Caribbean (De Silva *et al.*, 2003; Brooker *et al.*, 2006). In SSA it is estimated that approximately a quarter of the total population is infected with one or more helminths, typically the nematode worms, which are the most prevalent of all gastro intestinal parasites (Brooker *et al.*, 2000). The 2006 propose that of the then 181 million school-aged children in SSA, almost half 89 million were affected by one or more of these parasitic worms (Brooker *et al.*, 2006; Hotez and kamath, 2009). While the whole populations would be geographically at risk, children are observed to disproportionally carry the greatest burden of infection (Hotez *et al.*, 2009). This disproportion has behavioral, biological and environmental bases. Children tend to be more active in the infected environment and rarely employ good sanitary behaviors. Frequently, these potential carriers are crowded together for large periods of time example, schools, orphanages or slums, increasing the likelihood of transmission or environmental contamination with the parasite.

The prevalence of intestinal parasitic infections varies with different geographical regions Poverty therefore, illiteracy, lack of safe drinking water, poor hygiene, and malnutrition is responsible for repeated intestinal parasitic infections which lead to severe morbidity (Pilla, 2003). Additionally environmental factors also play a role in the incidence of IPI as hot and humid tropical climate favor increased parasite prevalence (Bethony *et al.*, 2006). Most of the parasitic infections are spread by faeco-oral route by consumption of contaminated food or water. Many parasitic infections are zoonotics, transmitted from domestic animals including cattle and rodents which act as reservoir (Ramana *et al.*, 2011).

*Giardia lamblia* and *Cryptosporidium* species are intestinal protozoa parasite that are recognized as prevalent and wide spread pathogens of human and many other species of

mammals. Infection with *G.lamblia* and *Cryptosporidium* species are common cause of gastroenteritis known as giardiasis and cryptosporidiosis, respectively. In recent year's massive outbreak of enteritis in people including a high incidence of disease in AIDS patients has increased public awareness of these parasites (Guimier *et al.*, 2004) *G.lamblia* is the most prevalent parasite in developing countries.

Amoebiasis due to infection with *Entamoeba histolytica* is the third leading cause of death from parasitic disease worldwide with its greatest impact on the people of developing countries. The World Health Organization estimates that approximately 50 million people worldwide suffer from invasive amoebic infection each year resulting in 40-100 thousands of death annually (Petri *et al.*, 2000). Spread of this protozoan *parasite* in developing countries mostly occurs through fecal contamination as a result of poor sewage and poor quality of water. Food and water born outbreaks of these protozoan parasites have occurred and the infectious cyst form of the parasite is relatively resistant to chlorine.

The medically important helminths are nematodes roundworms, cestodes tape worms and trematodes (flukes) (WHO, 2000). Parasitic infection particularly intestinal helminths cause hundreds of thousands of death each year and among the world common infectious diseases. The most prevalent and important helminths in developing countries are the soil-transmitted helminths such as *Ascaris lumbricoides*, *Trichuris trichiura*, hookworms, *Hymenolepis nana*. Schistosomiasis is endemic in 74 tropical countries; worldwide affecting over 200 million people, while 500 to 600 million people are at risk of becoming infected (Rosendale, 1997).

School age children are one of the groups at high-risk for intestinal parasitic infections. The adverse effects of intestinal parasites among children are diverse and alarming. Intestinal parasitic infections have detrimental effects on the survival, appetite, growth and physical fitness (Sharma *et al.*, 2004), school attendance, (Nematian *et al.*, 2008) and cognitive performance of school age children (WHO, 2006).

Several studies have been conducted to determine the distribution and prevalence of intestinal parasite infections in Ethiopia. A study among school children by Mengistu and Birhanu, 2004; the study conducted by Tekilu Wogayehu (2009) on the prevalence of *Gardia lambia* and *Cryptosporidium* species and the prevalence of infections with more

than one parasite poly parasitism by Mengistu and Birhanu (2004) a corresponding study conducted in Jiren Elementary and Secondary School by (Girmay *et al.*, 1994). Despite of the study so far conducted, there are still several localities including the study area Muketuri town North Showa Oromia Region for which epidemiological information of intestinal parasitic infections was not available. There is a need to have a clear information of parasitic species and the intestinal affected communities before devising management strategies which can be recognized by the community under the local settings.

Therefore the aim of this study was to assess the magnitude of intestinal parasites among school children at Muketuri primary school. The research finding could provide base line data on the distribution and prevalence of intestinal parasites in the area and assist in proposing strategies to prevent those groups that would be at risk of intestinal parasitic infection.

#### **General Objective;**

The main objective of this research was to determine the prevalence and intensity of intestinal parasitic infections among school children at Muketuri town, Oromia Regional State.

#### **Specific Objectives are;**

1. To determine the prevalence of intestinal protozoan and helminth parasitic infections among school children in the study area.
2. To identify the major intestinal protozoan and helminth parasite species in primary school children in the study area
3. To determine the intensity of intestinal helminth parasitic infection among school children.

## **2. LITERATURE REVIEW**

### **2.1 Human Intestinal Parasite Species**

Intestinal parasitic infections IPI are global health problems causing clinical illness in 450 million inhabitants in developing countries (Quinuine *et al.*, 2006). Parasite found in the

intestine can be categorized into two groups as protozoan and helminthes. The major intestinal parasites of global public health concern are the protozoan species such as *E.histolytica* and *G.lambli*a, soiltransmitted heliminths *A.lumbercoids*, *T.trichura*, hookworms and Schistosomiasis (WHO, 2000). Helminthic infections are enhanced by poor socio economic conditions, lack of sanitary facilities, improper disposal of human feces, and insufficient supply of potable water, poor personal hygiene, poor housing conditions and lack of education (WHO, 1996).

Intestinal parasitic infections and helminthes in particular are associated with increased growth retardation in children, poor increase in body weight in pregnancy, intrauterine growth retardation and low birth weight (Rodrigues *et al.*, 2006). Children infected with soil transmitted heliminths STH have poor education level and performance at school and a higher level of dropout, thus impacting on their future earnings and productivity (Miguel, 2004; Hotez, 2004).

### **2.1.1. Human Intestinal Protozoan Parasites Species**

Intestinal parasitic infections are the most common infections among children in developing countries. The most common intestinal protozoan parasites are *Giardia lamblia*, *Entamoeba histolytica*, *Cyclospora cayetanensis*, and *Cryptosporidium species*. The diseases caused by these intestinal protozoan parasites are known as giardiasis, amoebiasis, cyclosporiasis, and cryptosporidiosis respectively, and they are associated with diarrhoea (Davis *et al.*, 2002). *G. lamblia* is very common in developing countries and the most prevalent parasite that cause diarrhoea. Amoebiasis is the third leading cause of death from parasitic diseases worldwide, with its greatest impact on the people of developing countries (Davis *et al.*, 2002). Cryptosporidiosis is becoming most prevalent in both developed and developing countries among patients with AIDS and among children aged less than five years. Several outbreaks of diarrhoeal disease caused by *C. cayetanensis* have been reported during the last decade (Hrwaldt, 2000). Spread of these protozoan parasites in developing countries mostly occurs through fecal contamination as a result of poor sewage and poor quality of water. Food and water-borne outbreaks of these protozoan parasites have occurred, and the infectious cyst form of the parasites is relatively resistant to chlorine (WHO, 2000; Petri *et al*, 2001). Other species of

protozoan parasites can also be found in the human gut, but they are not pathogenic, except *Microsporidia* species.

In an article published in this issue of the Journal, (Jacobsen *et al.*, 2007 ) looked at the prevalence of intestinal parasites in young Quichua children in the highland or rural Ecuador. They have found a high prevalence of intestinal parasites, especially the intestinal protozoan parasites. They have used the traditional microscopic technique to diagnose intestinal parasitic infections. In total, 203 stool samples were examined from children aged 12-60 months and found that 85.7% of them had at least one parasite. The overall prevalence of intestinal protozoan parasites were *E. histolytica/E. dispar* 57.1%, *Escherichia coli* 34.0%, *G. intestinalis* 21.1%, *C. parvum* 8.9%, and *C. mesnili* 1.7%, while the prevalence of intestinal helminthic parasites in this study were: *A. lumbricoides* 35.5%, *T. trichiura* 0.5 %, *H. diminuta* 1.0%, and *S. stercoralis* 0.7%.

### **2.1.2. Human Intestinal Helminth Parasites Species**

In the vast majority of developing tropical and subtropical regions of the world, helminth infections particularly those caused by soil transmitted helminthes (STHs) and schistosomes constitute major public health and developmental challenges. Infections caused by STHs – including hookworm (*Necator americanus*, *Ancylostoma duodenale*), roundworm (*Ascaris lumbricoides*), whipworm (*Trichuris trichiura*) are associated with poverty and underdevelopment and are most prevalent in the poorest communities of the developing world including almost all countries of the sub- Saharan Africa (Montresor *et al.*, 1998; WHO, 2002).

The burden of these helminth infections has been consistently underestimated in the past, but there is now a general consensus that STH infections represent an important public health problem especially for children (Keiser *et al.*, 2002; WHO, 2002; Tchuente *et al.*, 2003; Bethony *et al.*, 2006). In all this study indicate that an estimated 4.5 billion individuals are at risk of STH infections and the global estimate of number of cases of *A. lumbricoides* is 807 million, *T. trichiura* 604 million, Hookworm (*N. americanus*; *A. duodenale*) 576 million (Bethony *et al.*, 2006; Hotez *et al.*, 2008). Although estimates of

disability-adjusted life years (DALYs) lost due to these helminth infections portray a more

accurate picture of the disease burden caused by the infections, the estimates of DALYs lost differ greatly from one source to another (WHO, 2002; Van der Werf *et al.*, 2003; King *et al.*, 2005; DCCP, 2008). However, total DALYs lost annually may range from 4.7 million to 39 million (DCPP, 2008). Because STHs are transmitted through poor sanitation and hygiene, and schistosomiasis by contact with infected freshwater streams

and lakes. School-aged children are typically at increased risk resulting in high prevalence

and intensity of infection due to high level of exposure (Montresor *et al.*, 1998; WHO, 2002). Although light helminthic infections are often asymptomatic, the adverse health and nutritional impacts of severe worm infections on children are well documented. Helminthic infections often lead to iron deficiency anaemia, protein energy malnutrition, stunting (a measure of chronic under nutrition), wasting (a measure of acute under nutrition), restlessness and abdominal pain (Van der Werf *et al.*, 2003; King *et al.*, 2005; Bethony *et al.*, 2006), and may negatively affect class-attentiveness of schoolchildren (Berhe *et al.*,

*et al.*, 2006), and may negatively affect class-attentiveness of schoolchildren (Berhe *et al.*,

2009).

Hookworm infection in humans is caused by two species, namely *Necator americanus* and *Ancylostoma duodenale* and is transmitted through contact with contaminated soil with third-stage infective larvae, which either penetrate the skin (*N. americanus* and *A. duodenale*) or when they are ingested (*A. duodenale*) (Hawdon and Hotez, 1996).

Hookworm disease occurs when the blood loss exceeds the nutritional reserves of the host, thus resulting in iron-deficiency anemia, zinc-deficiency and protein malnutrition, particularly in pregnant women and children (Curtale *et al.*, 1995). It contributes to anemia by causing blood loss directly through ingestion and mechanical damage of the mucosa, and indirectly, by affecting the supply of nutrients necessary for erythropoiesis (Crompton, 2000).

Hookworm infection is one of the most common chronic infections with an estimated 1.3 billion cases globally and accountable for 65,000 deaths annually (Silva *et al.*, 2003). The presence of more than 40 adult worms in the small intestine is estimated to be sufficient to reduce host haemoglobin concentrations below 11 g/dl, (Lwambo *et al.*, 1992) although the exact number depends on several factors including the species of hookworm. *A. duodenale* causes more blood loss and reduce the host iron reserves than *N. americanus* (Hotez *et al.*, 2004). The clinical manifestations of hookworm disease resemble those of iron-deficiency anemia from other causes. Studies conducted in different parts of Ethiopia showed the presence of both species of hookworm with *N. americanus* accounting for the larger share (92.5%) and (7.5%) prevalence for *N. americanus* and *A. duodenale*, respectively in Gojam (Tedla and Jemaneh, 1985), whereas the same group demonstrated *N. americanus* as an only agent of hookworm infection in Gondar. Chronic hookworm infection in school children contributes to physical and intellectual impairment, learning difficulties and poor school performance (Richared, 2003; Stephenson *et al.*, 1989). More recent evidence suggests that hookworm infection also has subtle yet profound adverse effects on memory, reasoning ability, and reading comprehension in children (Stephenson *et al.*, 1989). These again result in social and economic consequences such as decreased work capacity and productivity in children and adults, increased maternal and fetal morbidity and mortality, premature delivery and low birth weight (Richared, 2003).

*Ascaris lumbricoides*, a soil transmitted round worm, is reported to infect at least one fourth of world's population (Crompton, 1994). In most countries where geo-helminths are endemic, school age children experience the highest prevalence and intensity of infection, particularly with *A. lumbricoides* and *T. trichiura* (Hall *et al.*, 1997). Annual morbidity associated with this parasite has been estimated by WHO at 60,000 with another

250 million people said to be at risk for acquiring the infection (Montresor *et al.*, 1998). Both domestic and wild animals are common reservoirs for *A. lumbricoides*. *A. lumbricoides* is a robust parasite due to the resilient nature of its eggs, which are capable of surviving a wide range of hot and cold temperatures, chemicals, chemical disinfectants and other extreme conditions (Neva and Brow, 1994).

The eggs of ascaris are one of the resilient of the helminth eggs and can remain infective for many years embedded in the soil (Crompton, 1994; Gilgen and Mascie- Taylor, 2000). Ascaris is the largest of the human intestinal parasitic nematodes, in which both mature male and females usually inhabit the jejunum, where they feed on the-semi- digested food present in the host (Neva and Brow, 1994). They also produce or secrete antitrypsin and thus are capable of adequately competing with the host system for ingested proteins and resist the normal peristaltic movement of the gut by assuming an S-shaped configuration, pressing their outer cuticle against the columnar epithelium of the host (Kataz *et al.*, 1989).

Infection is acquired through the ingestion of infective eggs from focally contaminated food or water. Since the eggs are very sticky, they readily adhere to raw fruits and vegetables, which are washed with contaminated water or fertilized with contaminated soil. In highly endemic areas, ascaris eggs may be found on eating utensils, or under the fingernails. They may also be found in household dust and air where they are inhaled or swallowed (OLorcain and Holland, 2000).

The location and burden of worms mostly determine the type and degree of morbidity observed in the host. During the migratory phase, large numbers of larvae may include host sensitization, resulting in asthma, coughing, and shortness of breath, fever, skin rash, and eosinophilia (OLorcain and Holland, 2000; Neva and Brow, 1994). After the worm mature in the intestine, clinical signs and symptoms may include abdominal pain and distension, nausea, vomiting and anorexia. Vitamin A, fat and protein malabsorption also may be present, which lead to anemia. They can also cause lactose intolerance and malabsorption of vitamin and possibly other nutrients which might partly cause the nutritional and growth failure (Taren *et al.*, 1987).

In young children, adult worms can aggregate in the ileum and cause partial obstruction, because the lumen is small. *A. lumbricoides* has been observed to decrease micronutrients and vitamin A absorption, probably by causing a structural abnormality of the mucosa in the small intestine (Curtale *et al.*, 1995; Stacky, 2001).

Tapeworms are part of the cestoda class of platyhelminths flat worms that live in human host gastrointestinal tract. The adult *Taenia spp* infection in man is referred to as taeniasis and that due to the larval stage cysticercosis (Hancock, *et al.*, 1989). The distribution of *Taenia saginata* is wider in developing countries where hygienic conditions are poor and where the inhabitants traditionally eat raw or insufficient cooked or sun-cured meat (Florova,1982; Symth,1994; Minozzo, *et al* 2002). In Ethiopia intestinal parasitic infection is a major health problem throughout the country (Degarege *et al.*, 2012).

## **2.2.Life Cycle and Transmission Mechanism of Human Intestinal Parasites**

### **2.2.1. Life Cycle and Transmission of Protozoan Parasites**

Protozoa in their motile, feeding, growing, asexually-multiplying forms are known as trophozoites. These are adapted for existence in the host and generally are unable to survive the rigors of life outside host. Under appropriate conditions, which do not yet understand, some trophozoites of gut protozoa coat themselves in a protective shell and shut down metabolically, to become cysts. This is designed to survive in the outside world so that it may infect another host (Huang *et al.*, 2006). The life cycle of *Giardia lamblia* begins with a non-infective cyst being excreted with the feces of an infected individual. The cyst is hardy, providing protection from various degrees of heat and cold, desiccation, and infection from other organisms (Huang *et al.*, 2006). A distinguishing characteristic of the cyst is four nuclei and a retracted cytoplasm. Once ingested by a host, the trophozoite emerges to an active state of feeding and motility. After the feeding stage, the trophozoite undergoes asexual replication through longitudinal binary fission. The resulting trophozoites and cysts then pass through the digestive system in the faeces. While the trophozoites may be found in the faeces, only the cysts are capable of surviving outside of the host (Huang *et al.*, 2006).

The life cycle of *E. histolytica* consists of two stages. The protected cysts and active trophozoites, Cysts measure 10-15  $\mu\text{m}$  in diameter and typically contain four nuclei. Usually cysts, or occasionally trophozoites are ingested by man, ordinarily in contaminated water or food. Trophozoites ingested will be killed by stomach acids, since they have no protective covering. Cysts are carried to the lower ileum, where the amoebae excyst. The resultant eight trophozoites reach the lumen of the colon, multiply, and may invade and destroy the tissue of the colon wall (by secreting an enzyme) or invade and multiply in other organs (lungs, liver, brain, etc.) by way of the circulatory systems (Gill and Beeching, 2004). Trophozoites may multiply in the lumen of the colon, the colon wall, and other organs they invade. Trophozoites in the lumen may either encysted or pass in faeces trophozoites are most common in dysenteric stools. Most trophozoites disintegrate soon after passage; cysts may be ingested and the life cycle continued (Gill and Beeching, 2004).

The life cycle of *cryptosporidium* is monoxeous its life cycle in short 2 days and the infection may be short lived or may be persistent for months completed within the gastrointestinal tract of a single host (Fayer *et al.*, 2000). Subsequent to oocyst ingestion and activation in the upper GI tract, the organisms excite to release sporozoites. The oocyst is spherical in shape measuring 3-6 $\mu\text{m}$  in diameter and it may be either thick or thin walled (Ramirez *et al.*, 2004; Abhay *et al.*, 2009). The resistant stage that is found usually in the environment is the thick walled oocyst excreted together with feces (Fayer *et al.*, 2000).

The life cycle of *Cyclospora* is unknown; however environmental data suggest that *Cyclospora*, like *Cryptosporidium* species, is a water-borne parasite. The oocysts of *C. cayetanensis* are spherical, measuring 8-10 $\mu\text{m}$  in diameter and the mature oocyst contains 2 sporocysts. Oocysts of *Cyclospora cayetanensis*, are twice as large in comparison with *C. parvum* and are not sporulated (do not contain sporocysts - upon excretion). When freshly passed in stools, the oocyst is not infective thus, direct fecal-oral transmission cannot occur; this differentiates *Cyclospora* from another important coccidian parasite, *Cryptosporidium* (Markell *et al.*, 2000).

In the environment, sporulation occurs after days or weeks at temperatures between 22°C to 32°C, resulting in division of the sporont into two sporocysts, each containing two elongate sporozoites. Fresh produce and water can serve as vehicles for transmission and the sporulated oocysts are ingested (in contaminated food or water). The oocysts excite in the gastrointestinal tract, freeing the sporozoites which invade the epithelial cells of the small intestine. Inside the cells they undergo asexual multiplication and sexual development to mature into oocysts, which will be shed in stools. The potential mechanisms of contamination of food and water are still under investigation (Markell *et al.*, 2000).

### 2.2.2. Lifecycle and Transmission of Helminth Parasites

The life cycles of most helminthes follow the same pattern (Stephenson *et al.*, 2000). Adult hookworms of the genera *Necator* and *Ancylostoma* parasites live the upper part of the human small intestine, whereas *Ascaris* roundworms parasites live the entire small intestine and adult *Trichuris* whipworms live in the large intestine, especially the caecum (Crompton, 2001). The parasites can live for several years in the human gastrointestinal tract. Human beings are regarded as the only major definitive host for these parasites, although in some cases *Ascaris* infection can also be acquired from pigs (Crompton, 2001).

Helminths form three main life-cycle stages: eggs, larvae and adults. Adult worms infect definitive hosts (those in which sexual development occurs) where as larval stages may be free-living or parasitize intermediate hosts. Nematodes produce eggs that embryonate in or outside the host. The emergent larvae undergo 4 metamorphoses (moult) before they mature as adult male or female worms. Helminths can be contracted by humans in various ways. Ingestion of contaminated feces through soil, water or food is a main mode of transmission. Hookworm is most commonly contracted directly through the soles of the feet, usually around open areas of defecation and latrines, when people do not wear shoes or food coverings.

**Trematodes:** trematodes (flukes) are leaf shaped with an outer cover called the tegument which may be smooth or spiny. Most trematodes are hermaphroditic and most of the body consists of reproductive organs and their associated structures. Schistosomiasis is chronic

water related parasitic disease caused by blood flukes of the genus *Schistosoma*. It is the most important disease in terms of its public health and socio economic impact next to malaria, and is still a major helminthes infection at the beginning of the 21<sup>st</sup> century in many developing countries of the tropics. The disease is endemic in 74 tropical developing countries (George and Mohb, 2000). People become infected when coming in contact with water containing schistosome infected snails. Trematodes have more complex life-cycles where 'larval' stages undergo asexual amplification in snail intermediate hosts. Eggs hatch to release free-swimming miracidia which actively infect snails and multiply in sac-like sporocysts to produce numerous rediae. These stages mature to cercariae which are released from the snails and either actively infect new definitive hosts or form encysted metacercariae on aquatic vegetation which is eaten by definitive hosts. Schistosomiasis is contracted by swimming or wading in contaminated water.

**Cestodes** are tapeworm, specialized flatworms, looking very much like a narrow piece of adhesive tape. Tapeworms are the largest, and among the oldest, of the intestinal parasites that have plagued humans and other animals since time began. The most important cestodes affecting humans and animals in Ethiopia are *Taenia saginata*, and *Hymenolepis nana*, the former due to the custom of eating raw meat and the latter due to unhygienic food consumption with contaminated hands and fingers that allow the ingestion of eggs from the faeces of an infected person (Belete and Kloos, 2006). The adults of *Taenia saginata* and *Taenia solium* live in the intestine and are very large worms, i.e. several meters in length. Proglottids as well as eggs appear in faeces. The eggs of the two species are identical; they are round to oval in shape, measuring 35-43  $\mu\text{m}$  in diameter and have a thick, radially-striated shell. The egg contains a 6-hooked embryo called an oncosphere or hexacanth. These eggs must be handled with extreme care because the egg of *Taenia solium* is infective to humans and produces cysticercosis (WHO, 2004). The adult worm of *Hymenolepis nana* found in the intestine. It is very small, only a few centimeters long. The egg is unique in appearance. It is small, measuring 30-47  $\mu\text{m}$  in diameter with a thin, colorless shell. The membrane surrounding the hexacanth embryo has 4-8 filaments arising from each pole that fill much of the space between the embryo and the shell (WHO, 2004).

Helminths egg have tough resistance wall to protect the embryo while it develop mature egg hatch to release larvae either within a host or into external environment. The four main modes of transmission by which the larvae infect new host are faecal-oral ,transe dermal, vector borne and predator-prey transmission.

*Ascaris lumbricoides* (round worms) are dioecious, with male and female organs in separate individuals, and have a direct life cycle (none intermediate hosts). They mate in host's small intestine and the female produce egg. The adult female of ascariasis can produce 2000,000 eggs per day (Satosakar *et al.*,2009). The egg that pass out the adult worm are fertilized, but not embryonated. Once the egg exist the host via feces ,embryonation occurs in the soil and the emryonated eggs are subsequently ingested. Within the embryonated egg ,the first stage larva is develops in to the second stage larva. This second stage larva is stimulated to hatch by the presence of both the alkaline conditions in the small intestine and solubilization of its outer layer by bile salts. The hatched parasite that now resides in the lumen of the intestine penetrates the intestine wall and is carried to the liver through the portal circulation. It then travels via the blood stream to the heart and the lungs by pulmonary circulation. The larva molts twice ,enlarges and breaks in to the alveoli of the lungs. They then pass up through the bronchi and in to the trachea are swallowed and reach the small intestine once again within the small intestine the parasite molt twice more and mature into adult worms (Satoskar *et al.*,2009). Hookworm eggs already hatch in the soil and third stage larvae (L3) penetrate the human skin and reach the blood circulation. Their larvae have the ability to actively penetrate the coetaneous tissue ,most often those of the hands, feet, arms and legs due to exposure and usually through hair follicle or abraded skin, following skin penetration the larvae enter subcutaneous venues and lymphatic's to gain access to the host's afferent circulation. Ultimately, they enter the pulmonary capillaries where they penetrate in to alveolar space , ascend the bronchial tree to the trachea, travels the epiglottis in to the pharynx and are swallowed into the gastro intestinal tract (Satosakar *et al.*,2009). Larvae undergo two molts in the lumen of the intestine before developing into egg laying adult approximately 5 to 9 weeks after skin penetrates.

Adult whip female worms shed between 3,000 to 20,000 eggs per day, which passed with the stool. In the soil the egg develop in to a 2-cell stage and an advanced cleavage stage and then embryonate . It is the emryonated egg that is actually infectious. Environmental factors such as high humidity and warm temperature quicken the development of the embryo .This helps explain the geographic predelication for tropical environments .under optimal conditions, embryonic development occur between 15-30 days .Infections begins when these embryonated eggs are ingested (Satoskar *et al.*,2009).

### **2.3 Pathogenesis and Clinical Manifestation Intestinal Parasitic Infection**

The main clinical manifestation of infections with IPIs is diarrhea, with abdominal cramping, vomiting flatulence and weight loss being common symptoms. The symptoms can be several in younger children, as well in under nourished and immune compromised patients. Besides protozoan parasites intestinal helminthic infections are also a huge burden in developing countries, including Ethiopia (Abera and Nibret, 2014; Getaneh *et al.*, 2010). Intestinal schistosomiasis and soil transmitted helminthes (STHs) are common among school children and preschool children in the country (Alemu *et al.*, 2011; Dana *et al.*, 2015).

The wide spectrum of intestinal infection ranges from asymptomatic to temporary intestinal infection. In up to 90% of *E.histolytica/dispar* infections, the symptoms are absent or very mild (Reed, 2000). Although people can be asymptotically colonized with *E.histolytica/dispar* they should be treated. Otherwise, the cyst carries may be dangerous environmentally or may develop amoebic colitis (dysentery ) after a period of month (Reed,2000). It was thought that signs and symptoms of invasive amebiasis develop in approximately 10% the infected population. Symptom commonly attributed to *E .histolytica* colitis or dysentery is abdominal pain or tenderness and diarrhea (watery ,bloody ,mucous).

As in any parasitic infections host parasite interaction is the initial steps in the pathogenesis of gardiasis. In this interaction first the Gardia trophozoites attach to the cell surface of villi by means of a disk on their posterior or ventral surface. Lactin a protein on the trophozoite lining recognize specific receptors on the intestinal cell and may be partly responsible for the tight attachment between the parasite and the villi ( Wiser, 2006). Following

attachment of trophozoites, there will be major structural and functional abnormalities in small intestine. Some of these abnormalities include mucosal damage as a result of mechanical obstruction or blockage of the intestine by large number of parasites, the release of cytopathic substances such as thiol proteinases and lactins from *Gardia thropozoites* , the stimulation of a host immune response with release of cytokines and mucosal inflammation and deconjugation of bile salts (Wiser, 2006).

Symptom associated with giardiasis range from a symptomatic to acute gastro intestinal manifestations. Generally, the symptoms are sever the first time a person experiences giardiasis and children are at the greatest risk of contracting clinical giardiasis (Buret *et al.*,2002). In the majority of un treated patients the infection resolves spontaneously but it can become chronic and last for several months or even years in rare cases. The acute symptoms are a sudden explosive watery, foul smelling diarrhea the stool are generally describes as loose bulky, frothy and greasy with no blood or mucous. This is sometimes a companied by nausea colicky epigastric pain, vomiting, and prolonged bleaching (Wit Mas *et al.*,2001).

Soil transmitted helminthes (STH) such as hookworms (*Necator americanus* and *Ancylostoma duodenale*) and whipworms (*Trichuris trichuria* ) contribute to iron deficiency, anemia by ingesting blood and by damaging the intestinal mucosa during feeding (Brooker,2010). The manifestation of several disease include fatal intestinal obstruction or Chronic and intestinal STH infections can contribute to malnutrition and iron deficiency anemia and also can adversely affect physical and mental growth in children (Drake *et al.*, 2000, Stephenson *et al.* ,2000; Hotez *et al.* ,2004). Much of the pathophysiology of these parasites is nutritional in nature ,and their geographic distributions overlap with those of the four most common forms of malnutrition, iron deficiency and anemia's, vitamin A deficiency, and iron deficiency disorders affect hundreds of millions of people, especially children and women and girls of children bearing age (Stephenson ,*et al.* ,2000b). Deficiencies of vitamin B12 and other nutrients are also important in a number of areas.

*A.lumericoides* is a well known cause of malnutrition ,intestinal obstruction biliari colic and pancreatitis estimated to infect a quarter of the world 's population (Hall,2000). The

most severe manifestations include chronic dysentery, rectal prolapsed anemia, and growth stunting (Stephenson *et al.*, 2000). Intellectual and cognitive impairments and delays are also associated with chronic heavy infections causing TDS (Partovi *et al.*, 2007). STH is most often associated with malnutrition status as they induce intestinal bleeding, competition for nutrients, frequent anemia and diarrhea (Steinmann, 2012).

## **2.4. Epidemiology of Human Intestinal Parasitic Infections**

### **2.4.1. Global Distribution of Human Parasitic Infection**

Gastrointestinal (GI) parasites are infectious diseases of poverty. Thus, while still found in North America and Europe, their prevalence is highest in areas of intense poverty in low- and middle-income countries in the tropical and subtropical regions of SSA, Asia and LAC (Garg *et al.*, 2005; Hotez, 2007; Hotez *et al.*, 2008). In North America and Europe, these infections are most prevalent within immigrant and refugee community's (Barnet, 2004; Garg *et al.*, 2005; Stauffer and Weinberg, 2009). Globally, in immunocompetent individuals ascaris stands first with a prevalence of 600 million per year followed by schistosomiasis (300 million/year) and amebiasis (40–50 million per year).

Intestinal protozoan infections are endemic worldwide. In developed countries the prevalence of human intestinal parasitic protozoan infections is estimated to be between 1-7%, but it may be as high as 50% in developing countries. All age groups are equally affected during epidemics, but both subclinical infection and clinical disease are more common in children in endemic areas. Outbreaks occur regularly in childcare facilities. Immuno-compromised individuals are also more commonly affected than members of the general population. *Giardia lamblia* is also a cause of “travellers’ diarrhea, in which the disease is sometimes also called sewer fever. *E. histolytica* infection is common in most developing countries. It is reported to be responsible for approximately 50 million cases of invasive Amoebiasis and upwards of 100,000 deaths each year. Thus, it is second only to malaria as the cause of mortality due to protozoan infection (WHO, 2006). In immunodeficient individuals, on the other hand, cryptosporidiosis is the commonest intestinal parasite known to occur.

Human intestinal Parasitic helminthes are known causes of morbidities such as nutritional deficiency, impaired physical development and learning ability, and socioeconomic

deprivations in populations living in the tropics where poor hygiene conditions provide an optimal environment for the development and transmission of the parasite. Because the most significant physical and intellectual growth disturbances occur as a consequence of moderate and heavy worm burdens, the age-associated epidemiology of *A. lumbricoides* and *T. trichiura* infections has focused attention on infected school children in developing countries (Bradley *et al.*, 2004). It is estimated that more than one billion of the world's population is chronically infected with soil-transmitted helminthes. A revised estimate of the probable number of Ascaris infections in the world with the morbidity indicated that 59 million of the 1.2 billion people infected (including 51 million children less than 15 years of age) are at risk of faltering growth, decreased physical fitness, or both as a result of infections (Bradley *et al.*, 2004).

#### **2.4.2. Human Intestinal Parasitic Infection in Ethiopia**

In Ethiopia, intestinal parasitic diseases are among the ten top causes of morbidity nationwide. Most of the intestinal parasites are more and their manifestation is more severe in children than adults. Infection in children is also associated with malnutrition, growth retardation and school performance. In Ethiopia, intestinal parasitic infections are highly prevalent being the first or second most predominant cause of outpatient morbidity in the country (Ali *et al.*, 1999; Leggese and Erko, 2004; Taddese, 2005). Although the prevalence rates of individual parasite infections vary considerably in different parts of the country, most studies showed a higher prevalence of *A. lumbricoides* followed by *T. trichuria* and hook worm.

In the epidemiological study of STHs, South Gonder, Ethiopia, 49% of the examined children had one or more types of helminth infection, of which 32.5%, 13.3% and 2.4% were single, double and triple infections respectively. About 28.4%, 8.3% and 12.1% of the children had moderate infections of ascariasis, trichuriasis and hookworms, respectively (Leykun, 2000).

Reports from different parts of Ethiopia showed different prevalence rates of the intestinal protozoan parasites. The prevalence of *Cryptosporidium* infection in children with diarrhea ranged from 3.3% in Jimma, 5.6% in Addis Ababa to 9% in Northwestern Ethiopia (Mengistu *et al.*, 2007). In a study conducted in South Western Ethiopia, the

prevalence of Giardiasis was 13.7% though the rate is much lower than *A.lumbricoides* (Mulat *et al.*, 2013). Intestinal protozoan parasites including Giardia, Cryptosporidium and amoeba are widely distributed in the country (Karanis *et al.*, 2007).

Based on a countrywide survey of giardiasis, the overall prevalence among school children and residents were 8.9% and 3.1%, respectively and that of the non- school children were 4.4 % (Merid *et al.*, 2001). In a countrywide survey of amoebiasis in 97 communities, the overall prevalence of *E. histolytica* infections, as measured by rate of cyst-passers, in school children and non-school communities were 15.0% and 3.5%, respectively (Girum, 2005).

In Ethiopia high prevalence of intestinal parasite transmission is attributable to factors associated with low socio economic status, such factors include poor personal hygiene and environmental sanitation and lack of clean water supplies for most parts of developing countries for instance, Ethiopia has one of the lowest quality of drinking water supply and latrine coverage in the world. Besides that as comparison by 2000, Ethiopia had only 12% latrine coverage while Kenya had 87% (Kumie and Ali, 2005). More over a number of surveys had show that protozoa, and helminthes parasite infections were prevalent in varying magnitude (Eriko and Medhin, 2003).

## **2.5. Diagnosis of Human Intestinal Parasitic Infection**

In the diagnosis of intestinal parasites a wide variety of laboratory methods can be employed (Ahmedi *et al.*, 2007). Stool microscopy using direct wet mounts, formol ether concentration and the Kato –katz technique offers many relative advantages over other diagnostic methods for detecting intestinal parasites (Parija and Srinivasa, 1999; Bogoch *et al.*, 2006). Direct saline wet mount provides economical and rapid diagnosis for intestinal parasite when they are present insufficient density in the fecal sample (Ukaga *et al.*, 2002).

The formol ether concentration method described by (Allen and Ridely, 1970) increases the sensitivity and specificity of stool microscopy to allow the detection of low number of organism recovers most ova, cyst and larvae and retains their morphology, there by facilitating identification (Ahmadi *et al.*, 2007). It is well established that the direct wet mount technique lacks sensitivity but most hospital laboratories in developing countries rely on this method for routine stool examinations as a result of its affordability and

simplicity (Ukaga *et al.*, 2002; Ahmadi *et al.*, 2007; Wirkom *et al.*, 2007). The ether concentration method was often used for the diagnosis of helminths infections, particularly in specialized laboratories (Marti and Escher, 1990; Utzinger *et al.* 2010).

The most prominent tool for detection of helminth egg in stool samples was the kato –katz thick smear test Katz *et al.*, (1972). Probably due to its simplicity, low material and infrastructure requirements, and cheap price. The Kato-katz technique Katz *et al.*, (1972) was useful for the qualitative estimation of worm burden (Marikell *et al.*, 1999). It was especially useful for field surveys for helminths infections since it provides estimate of intensity of infection (Martin and Beaver 1968). The technique entails the examination of a standard sample (determined by the size of the template) of fresh faeces pressed between a microscope slide and a strip of cellophane that has been soaked in glycerin (Ukaga *et al.*, 2002).

Others recommend screening of at least three stool samples for correct diagnosis (CDC, 2006). Though serological tests including ELISA for the detection of antigen in stool, as well as antibody in blood per serum are available, their usefulness has been found to be limited except in case of extra intestinal manifestations. Usefulness of PCR in the diagnosis of parasitic infections is limited due to cost affordability in low income countries. (Katz and Tayler, 2001)

## **2.6. Control and Prevention and Human Parasitic Infection**

Health education is aimed at reducing transmission and reinfection by encouraging healthy behaviors. For STH infections and schistosomiasis, the aim is to reduce contamination of soil and water by promoting the use of latrines and hygienic behavior. Without a change in defecation habits, periodic deworming cannot attain a stable reduction transmission. Health education can be provided simply and economically and presents no contain indications or risks. Furthermore, its benefits go beyond the control of helminth infections. In this perspective, it is reasonable to include this component in all helminth control programs. (Hotez *et al.*, 2004).

Improved sanitation is aimed at controlling transmission by reducing soil and water contaminates. Sanitation is the only definitive intervention to eliminate STH infections, but to be effective it should cover a high percentage of the population. Therefore, because of

the high costs involved, implementing this strategy is difficult where resources are limited (Asaolu and Ofozie 2003). Moreover, when used as the primary means of control, it can take years or even decades for sanitation to be effective (Brooker *et al.*, 2004).

Typical public-health interventions (such as the provision of clean water, community health education, observation of food hygiene, and maintenance of functioning sanitation systems) are essential to long-term control in a community. However, the implementation and sustainability of interventions is complex, and variable between local contexts. Slum living is an increasingly common phenomenon that is creating a new urban parasitological where polyparasitism is magnified, and this will require a new way of thinking for the development and implementation of community control endeavors. Large-scale epidemiological surveys with concomitant cartographic modeling using morbidity questionnaires are increasingly used to guide targeted, specially explicit and cost-effective sanitation and pharmaceutical interventions (Brooker and Utzinger, 2007; Simoonga *et al.*, 2009; Stensgaard *et al.*, 2009), such as Mass Drug Administration (MDA). MDA entails the large scale, often nationwide, deployment of drugs based on local needs gathered from these parasitological surveys and geospatial modeling of infected populations Hotez (2009). Examples of other geospatial parasitological surveys are available from East Africa (Brooker and Clements, 2009, Brooker, Kabatereine, Smith, *et al.*, 2009).

Anthelmintic Drug Treatment (“deworming”) is aimed at reducing morbidity by decreasing the worm burden. Repeated chemotherapy at regular intervals (periodic deworming) in high-risk groups can ensure that the levels of infection are kept below those associated with morbidity and will frequently result in immediate improvement in child health and development. For ascariasis and trichuriasis, for which intensity peaks among school-age children, frequent and periodic deworming may reduce transmission over time. Obstacles that diminish the effectiveness of periodic deworming are the low efficiency of single-dose mebendazole and albendazole for the treatment of hookworm and trichuriasis, respectively (Adams *et al.*, 2004; Albonico *et al.*, 1994).

### **3. MATERIAL AND METHODS**

#### **3.1. Description of the Study Area**

The present study was conducted from April up to May 2017 in Muketuri town north showa, Oromia region to determine the prevalence and intensity of intestinal parasitic infection among school children. Muketuri is located at about 80 km north of Addis Ababa. It has one Kebele with total population of 21,480 of this 10,000 were male and 11,480 were females according to Muketuri Administrative Office's report (2008). The inhabitant of Muketuri town use agriculture, trade and some employer to lead their livelihood. Agriculture is the source of income in the area, where the farming system is characterized by small scale production of dairy cattle and trade is the main source of income.

There are three primary schools and one secondary school. The study was carried out in the Muketuri Primary School Number One which consists of grade 1-8 and the other two Primary Schools consist of grade 1-4. There is one health center in the town and three rural drug houses. Muketuri Primary School Number One has a Pipeline water supply but poor toilet service and absence of health education. In general the town has poor sanitation, poor toilet facilities, sewage system, insufficient pipeline water supply and many of the households have individual hand dug water well. Although there was no published report on the relative water born disease the woreda, health office report (2008/2009) indicates that intestinal parasite infections together with high malnutrition rate among preschool and school age children were major health problem in the study area.

#### **3.2. Research Design**

A cross sectional study was conducted among children in Muketuri Primary School to determine the prevalence and intensity of intestinal parasitic infections. A pre survey contact was done during which permission was obtained from the woreda education department and health bureau of local government. Verbal consent was also seeked from

the parents of participating students. Before collecting the stool samples, the aim of the study was explained to the parents, the lab assistants, the participants by using local language. Then after a clean piece of plastic sheet and stick applicator were distributed to the study participants and they were instructed to provide 3 g of fresh stool sample, after that it was taken for lab diagnosis at medical laboratory of Muketuri Health Center. Laboratory examinations of stool samples were carried out using a direct wet mount ,formol-ether concentration ,modified Ziehl-Neelsen and Kato-Katz methods .

### 3.3. Study Population

The entire grade 1-8 students in the selected study were considered as the total population of the present study. According to the data obtained from Muketuri Primary School, the total number of students from grade 1-8 that were registered in 2016/17 academic year were 1463 of these, 738 and 725 were male and female , respectively. These constituted the study population of the present study.

### 3.4. Sample Size Determination and Sampling Techniques

The sample size was determined by using the formula of sample size determination for single population proportion of participant who was infected with intestinal parasite (Naing et al., 2007). Sample size determination for study was by using the formula indicated below. The sampling technique was random sampling and the students from each grade levels were selected randomly from their class attendance.

$$n = Z^2 P (1 - P) / d^2$$

Where n = number of participant

$$Z = 1.96 \text{ (Statistic for level of confidence)}$$

$$P = 0.05 \text{ (Expected prevalence or proportion)}$$

$$d = 0.05 \text{ ( Precision )}$$

In determining the sample size, the P value was taken as 50% (maximum value ) because of absence of recent data on prevalence of intestinal parasite in the study area. Thus, the sample size used in this study, as determined by the above formula was 400. In this formula, P (1-P) is maximized when P=0.05, which ensures a sample size to be large

enough to satisfy the precision and confidence level. The study samples were represent three age groups,(6-9, 10 -14, 15 -18 } and both sexes.

Random sampling method was utilized to select the sample population. The students were first group according to their grade level (grade 1 to 8). A proportional quota was allocated for each grade and each section. A random sampling technique was employed to select students from each grade and classroom by using class rosters as sampling frame.

### **3.5. Collection of Stool Samples**

The students were trained on how to collect the fresh stool samples using plastic container provided to them. Stool sample was collected from each student as they arrive at school premises. Since the school was far from the health center approximately 400m the fresh stool samples were taken to the health center of medical laboratory room with in 1hr in order to ensure proper identification of helminths eggs, cysts and oocysts of protozoa. The appearance of each fecal sample was carefully examined for consistence, presence or absence of blood and mucus .The fecal samples were then examined microscopically for parasites by direct smear saline method as recommended by {WHO 2003}. Negative samples were later subjected to concentration method. Based on WHO {2003} criterion for quantifying the number of eggs per gram {EPG} of faces, positive stool samples were further reexamined.

### **3.6. Laboratory Examination of Stool Samples**

#### **3.6.1 Direct Wet Mount Method**

A direct wet mount with normal saline {0.85% NaCl solution} was prepared and to examine stool samples for the presence of motile intestinal parasites and trophozoite under light microscope at 10× and 40× magnification. Lugoal's iodine staining was used to observe cyst of intestinal parasite{WHO, 1999}.

#### **3.6.2 .Formol-Ether Concentration Method**

A portion of each stool sample was used for detection of parasitic ova and protozoa cysts using the formol-ether sedimentation technique. One gram {1 g} of each stool sample was first emulsified with 3-4 ml of 10% formol saline. An additional 3-4 ml of 10 % formol saline was added this was mixed thoroughly and passed through gauze. Three to four

(3-4) ml of diethyl ether was added and mixed by inverting and intermittent shaking for 1 minute, and centrifuged at 3,000 rpm for 5 minute. After centrifugation, the supernatant (layer of ether, debris and formol saline ) was discarded and the sediment (containing the parasites at the bottom of the test tube ) was re-suspended in formol saline. The sediment was examined microscopically under 10× and 40× magnifications for the presence of any parasitic organisms (WHO, 2000c).

### 3.6.3 Modified Ziehl –Neelsen Method

A thin faecal smear was made, leaved to air dry and fixed in methanol for 2-3 minutes. Further fixation in formalin vapor should be performed if possible, to reduce infectivity. For detection *cryptosporidium*. This was then stained with cold carbol-fuchsin for 5-10 minutes which was then washed off boiled water. The smear then decolorized using 1% hydrochloric-ethanol for 1 minute and were counter –stained with 1% methyl blue for another 1 minute. The stain was again washed off with boiled water and the smear would be microscopically examined by using 100× magnification (Henriksen and pholenz, 1981).

### 3.6.4 Kato-Katz Method

Kato –katz technique was applied to determine the intensity of STH infections briefly portion of fecal specimen was taken by clean wood /plastic spatula and forced through the nylon screen to separate fecal materials from the large debris. The screened fecal material was transferred to the template which was laid flat centrally on microscope slide. The template hole was completely filled with screened fecal material and leveled to the surface of the template. Cellophane square which was soaked in malachite green glycerin solution was placed over the specimen. The specimen was made to spread evenly under the cellophane tape by pressing it with glass slide prepared for this purpose ( Bogoch *et al .* , 2006).

The prepared kato katz slide was examined under the microscope for STH ova by principal investigator with assistance of experienced laboratory technician at Muketuri Health Center. The number of eggs of each species were recorded in to the number egg per gram of stool (EPG) in order to analyze intensity of STH infections. The eggs per gram of stool (EPG) of each STH species was obtained by multiplying the number of eggs

by 24 (kato slides delivering 41.7 mg of stool plug } (Brooker *et al .* ,2007}. Based on WHO (2003) criterion for quantifying the number of eggs per gram (epg) of faces, positive stool samples were further reexamined by single thick smear technique using 41.7 mg Kato-Katz template and the number of helminths eggs counted were multiplied by 24 in order to quantify the number of eggs per gram of faces. To ensure consistency of the result and as a form of quality control, 20 % of the slide randomly selected and read again (Andrage *et al.*, 2001).

### **3.7. Data Analysis**

Collected data was coded, checked and analyzed using SPSS version 20 Software. Descriptive statistics such as frequency, percentage, mean standard deviation and range were determined for each intestinal parasite. In order to analyze intensity of infection for intestinal parasites, the number of eggs were converted into the number of Eggs Per Gram of stool and transformed to log scales for analysis of geometric mean. The class of intensity of infection were expressed as light, moderate and heavy based on the egg count per gram of stool. The 95% CI was used to show the accuracy of data analysis. Probabilities less than 5% ( $P < 0.05$ ) for null hypothesis testing were considered statistically significant.

### **3.8. Data Quality Control**

To ensure quality control, all the laboratory procedures including collection and handling of specimens were carried out in accordance with standard protocols. All the reagents were checked for contamination time. To ensure general safety, disposable gloves were worn and universal bio safety precaution (NCCLS,2000} were followed at all time for quality control of the concentration method preserved stool specimen known to contain parasite ova and larvae were included in each batch of sample to be concentrated to ensure that the procedure were precise.

### **3.9. Ethical Consideration**

Before conducting the investigation, the investigator made discussion with concerned bodies in the study area. Their agreement was obtained after the objectives and purpose

were explained. Ethical clearance was obtained from Wuchale Woreda Health Office and Educational Office. Additionally, after explaining the importance, purpose, and procedure of the study briefly, a written consent were obtained from the study participant. Any one not willing to take part in the study had full right to do so and confidentiality of the study participants were also maintained. Any study participant who was positive for intestinal parasite was referred to physicians for treatment.

## 4. RESULTS AND DISCUSSIONS

### 4.1. The Age and Sex Distribution of the Study Participants

The age and sex distribution of school children are summarized and presented in Table 1. A total of 400 school children were selected by stratified random sampling technique from grade 1 to 8 to participate in the study for prevalence and intensity of intestinal parasite. Sex distribution showed that, 198 (49.5%) were male and 202 (50.5 %) were females. The age range of the study population ranged between 6-18 years. More than 52.8% of the students were in the age groups of 10-14 years old. The age category of the students were 149 (37.3%) in 6-9 years, 210 (52.8%) in 10-14 years, and 40(10%) in 15-18 years. Table (1).

Table 1. The Age and Sex Distribution of Study Participants in the Study Area

Variables	Category	Frequency	Percent (%)
Age	6-9	150	37.5
	10-14	210	52.5

	15-18	40	10.0
Sex	Male	198	49.5
	Female	202	50.5

#### 4.2. Prevalence of Intestinal Protozoan Parasitic Infections among School Children

The overall prevalence of protozoan parasitic infection among all age groups of the pupil was 44.25 %. Of these the prevalence of protozoan parasitic infection for males and females was 38% and 49.5%, respectively (Table 2). The prevalence of intestinal protozoan parasitic infection in males and females of age group 6-9 years old was 47.1% and 46.3% respectively, while for the age group 10-14 was 37% for male and 49% for female. The prevalence of intestinal protozoan parasitic infections for the age groups of 15-19 years was 22.1% for males and 66.66% for females, respectively (Table 2).

Although the difference in the prevalence of intestinal protozoan parasitic infection among all age group in male and females were not statically significant ( $\chi^2 = 0.136$ ,  $p=2.656$ ), the prevalence was somewhat higher in age group 6-9 compared to the other groups in both males and females. In the present study, the observed prevalence of protozoan parasitic infection in age group 6-9 was 46.66%, which was higher than the other age groups. This could be due to low immunity in children, less awareness of washing hands and other personal hygiene measure in this age group.

Higher prevalence of parasitic infection among school children may be due to the poor sanitary condition in the schools (Oguntibeju, 2006). Children usually do not take care of their personal hygiene for instance they play in contaminated outdoor environment, in and around the area which was contaminated with intestinal protozoan parasitic infections that can certainly cause series health problems to them, they also face difficulties in proper use of latrine due to lack of basic hygiene practice, such as washing hand before and after meal, and after use of latrine (Abu Mourad, 2004). In this study, male children (47.1%) were infected more than female children (46.6%) with the same age (6-9 years old). This result is in line with a study conducted in Kenya, which demonstrated that there were more males than females who contracted amebiasis (Chabalala and Mamo 2000) but it was

contradictory with observation made by Odikamnoro and Ikeh (2004), females children were found to have higher prevalence than male children.

In current study, females had higher prevalence rate than male in age group 10-14 and 15-18 years old, which was 49% and 66.66% respectively (Table 2). However the difference was not statistically significant ( $\chi^2 = 0.039$   $P = 0.0852$ ,  $\chi^2 = 0.06$   $P = 0.291$ ). This may be due to the fact that females were more often involved in food processing and handling activities than male and hence water and food contamination is one of the most common modes of transmission (WHO, 1997a) would account for this gender associated infection difference.

**Table 2.** . Prevalence of intestinal protozoa parasitic infections by age and sex among school children Muketuri Primary school from April-May 2017.

Age group (in years)	Male		Female		Both sex		$\chi^2$	p-value
	No Exam	No Pos (%)	No Exam	No Pos (%)	No Exam	No Pos (%)		
6-9	68	32(47.1)	82	38(46.3)	150	70(46.6)	0.136	2.656
10-14	108	40(37)	102	50(49)	210	90(42.8)	0.039	0.852
15-18	22	5(22.1)	18	12(66.66)	40	17(42.5)	0.06	0.291
Total	198	77(38)	202	100(49.5)	400	177(44.2)	0.016	0.975

No Exam=Number of examined No Pos=Number of positive

#### 4.3. Intestinal Protozoan Parasite Species Identified from Examined School Children

In the present study microscopic stool examinations were done. The result showed that infection with various intestinal protozoan parasites species were common among the study school children. As shown in table 3, three major intestinal protozoan parasites species were identified in examined stool of school children. The parasite species in their order of dominance were *E.histolytica* (28.5%), *G.lamblia* (12.75%) and *Cryptosporidium specie* (3%). The higher prevalence of *E.histolytica* infection in current

study might be attributed to the fact that high degree of food and water contamination with human excreta and lack of awareness in simple health promotion practice such as personal hygiene and food hygiene (Endeshaow 2000).

High prevalence of intestinal protozoan parasite infections of school children were commonly reported in different part of Ethiopia. For example Ayalew (2006) reported 38% prevalence of amebiasis among children in Dire Dawa, Eastern Ethiopia. According to the finding done by Yenenh (1994), the prevalence of intestinal protozoan parasitic infections among resident of four villages in South Western Ethiopia was 82.7%.

In this study the most prevalent intestinal protozoa parasite identified in Muketuri children was *E.histolytica* infections in all age group. The prevalence of *E. histolytica* infection in male school children of age group 6-9, 10-14 and 15-18 years old was 4.8%, 5.3% and 22.7%. This show as age increase the prevalence of *E.histolytica* infection also increase of similar pattern was observed in female school children. That means the prevalence of *E. histolytica* infection in female children of age groups 6-9, 10-14 and 15-18 years old 7%, 7.5% and 66.1%respectively, (Table 3). The reason for this as age increase children engaging themselves in different playing games, cleaning cattle homes and eating contaminated foods without washing their hands.

The difference in findings among the various studies can be explained by variation in geography, environmental sanitation, inadequate medical care, socio economic condition of the study subjects and health care as well as prevailing climatic and environmental conditions under consideration (WHO, 2006)

Intestinal protozoan parasite including *E.histolytica* ,*G.lamblia* and *Cryptosporidium species* are widely distributed in Ethiopia. Report indicated that the prevalence of *Cryptosporodium species* and *G.lamblia* among diarrheic patients referred to Ethiopian Health and Nutrition Research Institute were 20.8% and 8.6% respectively, (Endeshew *et al.*, 2004). A study conducted by Legesse and Erko (2004) among school children around Lake Langano showed that the prevalence of intestinal protozoan parasite infections was 83.8% which was higher than the overall prevalence of intestinal protozoan parasitic infection of school children in the present study ( 44.25%).

**Table 3.** Major intestinal protozoa parasite species identified from examined stool samples of school children.

Age group years and sex	No pos	Eh	Gl	C.species	Double infection	X <sup>2</sup>	P-value
		No Pos%	No pos%	No pos%	No Pos%		
6-9 Male	32	19(4.8)	7(1.8)	6(1.5)	2(2.94)	2.28	0.240
Femal	38	28(7)	6(1.5)	4(1.0)	5(6.09)		
10-14 Male	40	21(5.3)	17(4.3)	2(0.5)	3(2.77)	0.93	0.363
Female	50	30(7.5)	20(5.0)	0(0)	5(4.9)		
15-18 Male	5	5(22.7)	0	0	1(4.54)	0.25	0.160
Female	12	11(66.1)	1(0.3)	0	0		
Total	177	114(28.5)	51(12.75)	12(3)	16(4)		

Key *Eh*=*Entamoeba histolytica*, *Gl* =*Gardia lamblia*, *Cryptosporidium species* No pos =Number of positive individual.

#### 4.4 Prevalence of Intestinal Helminths Parasitic Infection among School Children

Microscopic stool sample examination showed that infections with various intestinal helminths were common in school children. Out of the total 400 children examined, 24.75% (99/400) children were found positive for one or more of intestinal helminth parasite infection (Table 4). In this current study the prevalence of helminthes infections among young children (6-9 years) old 36.66% which was higher than the other age group,

and statically significant difference was observed between male and female children within this age group (Table 4). This indicated that younger children were more exposed to intestinal helminthes parasite infections which could be happen due to their higher exposure to contamination of the environment, especially the soil where the children usually play and eat food without washing their hands. This could also be explained by the fact that in this age their immunity to parasitic infections has not been fully developed (Stephenson *et al.*, 2000). Eventhough gender is not a significant risk factor for prevalence of helminths infections (Wani *et al* 2008), in this study male children (29.79%) were more infected with intestinal parasitic helmiths than female (19.8%) (Table 4). This was in agreement with previous studies in Lake Zway Health Center (Tesfamichale and Tekelemariam, 1983; Tekelemariam and Tesfamichale 1987).

The overall prevalence of intestinal parasitic helmints infection among children in the present study (24.75%) was lower to the result of studies conducted in school aged children in Babile town, 27.2% by Girum(2008), the study around Lake Zeway island by Tesfamichale and Tekelemariam (1983), which showed an overall prevalence of 56% and study reported from South Gonder 49% by leykun (2000). This difference in prevalence might be a reflection of the difference in local sanitary standard, environmental condition, time and seasonal differences in the design of the survey work and personal hygiene (Albonico *et al.*, 1999). Regarding gender this study had shown that, male had higher risk for helminthes infections than that of female children, however, the difference was not statically significant (  $P > 0.05$  ).

**Table 4.** Prevalence of intestinal helminths parasitic infections by age and sex among school children.

Age group ( in year)	Male № Exam	№ Pos(%)	Female № Exam	№ Pos (%)	Both Sex № Exam	№ Pos (%)	X <sup>2</sup>	p-value
6-9	68	32 (47.5)	82	23 (28.04)	150	55(36.66)	10.96	0.010
10-14	108	27 (25)	102	17 (16.66)	210	44(20.95)	0.217	1.032
15-18	22	-	18	-	40	-		
Total	198	59 (29.79)	202	40 (19.80)	400	99 (24.75)	12.809	0.071

№ Exam=Number of examined № Pos=Number of positive

#### 4.5 Intestinal Parasitic Helminths Species Identified among examined School Children

The overall prevalence of intestinal helminthes parasite infections was 24.75% in the school children. Among the intestinal parasite species *Ascaries lumbricoides* 16%, Hookworm 5.5%, *Trichuries trichuria* 1.5% and *Hymenolipes nana* 4% were predominant parasite in the school children. The higher prevalence of *Ascaries lumbricoides* observed in this study might be the result of inadequate sanitary condition and poor hygiene practice.

In the present study, the prevalence of *Trichuris trichuria* infection (1.5 %) was less than the result of similar study reported in Babile (3.6%) by Girum (2005). However, it was much lower than the study conducted in Lake Lanngano (14.7) by Mengistu and Birhanu, Chilga District (14.8%) by Leykun (2001) and different part of Ethiopia (30%) by Gezahegn, (2008). The observed difference in the prevalence of intestinal helminths parasite infections in the present study from prevalence of helminths parasite species reported in other part of Ethiopia might be due to difference in diverse environmental

conditions of the study site as epidemiology of soil transmitted helminths is highly affected by surface temperature, attitude, soil type and rain fall (Brooker *et al.*,2003).The observed difference could also be explained by the fact that the prevalence and distribution of helminths parasite infections varies by place and with age in Ethiopia as reported by Yesnamole *et al.*, (2010). In the present study the higher prevalence of *H.nana* infection was found to be the most prevalent (4%) parasite compared to the report from the study conducted in Babile town ,eastern Ethiopia (Girum,2005), but it was higher than that of the study in Mexico (Espinoza *et al.* 2003). The high prevalence rate of *H.nana* infection observed in this study indicates that hygienec practice of the school children in the study area were poor and it was an important factor for auto infection and transmission to other ( Grang and Zumla,2003)

Age specific prevalence of *A. lumbricoides*, Hookworms, *Trichuris trichuria* and *H.nana* infection in the present study showed similar pattern of infection with the highest prevalence seen among children in the age group between 6-9 years and then a further decline in older age group (Table 5).This was in agreement with previous studies (Girum,2005, Erko *et al.*, 2003) .WHO reported higher prevalence of parasitic infections among younger children .This phenomena probably reflect age related change in exposure to intestinal parasite infection.

A similar study in Brazil was also reported high prevalence of *Ascaris lumbricoides* in the 5-10 years age group (Fleming *et al.*,2006).This could be because of high level of soil contact activity and low personal hygiene in the young age group. In addition, this indicated that younger children were more exposed to infections which could be transmitted due to the contamination of soil where the children usually play and eat food without of washing their hands, Thus as age increase, the likely hood acquiring intestinal nematode infection decrease possibly due to important personal hygiene and basic life skill as reported by Alemayehu *et al.*, (1998).

**Table 5.** Intestinal helminths parasite species identified from examined stool sample of school children of Muketuri Primary School, from April-May 2017.

Age Group	Sex	No of ps	Al No Pos%	Hw No Pos%	Tt No Pos%	Hn No Po%	$\chi^2$	P-value
6-9	Male	32	16 (4)	6 (1.5)	2 (0.5)	8 (2)	0.23	0.15
	Female	24	17 (4.3)	4 (1.0)	1 (0.3)	2 (0.5)		
10-14	Male	27	18 (4.5)	2 (0.5)	1 (0.3)	6 (1.5)	2.18	0.20
	Female	18	13 (3.3)	2 (0.5)	2 (0.5)	0 (0)		
15-18	Male	0	0 (0)	0 (0)	0 (0)	0 (0)	0.8	0.74
	Female	0	0 (0)	0 (0)	0 (0)	0 (0)		
Total		101	64 (16)	10 (5.05)	6 (1.5)	16 (4)		

Key; Al=*Ascaris lumbricoides*, Hw=Hookworms, Tt=*Trichurias trichuria*, Hn =*Hayminolepis nana*. No pos=number of positives

#### 4.6 Intensity of Helminths Parasitic Infections among School Children in the study Area.

Kato-katz thick smear were prepared as described by WHO (1991). The wilcoxon signed rank test was used to test for differences in fecal egg count between the methods. Furthermore, samples were sub divided in to low, moderate and high egg excretion intensities to thresholds proposed by WHO. *Ascaris lumbricoides* low 1-4999 EGP, moderate 5000-49999 EGP and high > 49999 EGP *Tricuris tricuria* low 1-999 EGP, moderate 1000-9999 EGP and high >9999 EGP and hookworm low 1-1999 EPG, moderate 2000-3999 and high >3999.

In the present study area more of positive cases for helminthes parasitic infections were within the range of light and few moderate infections, and there were no cases with heavy infection of helminth. The total egg count of *A.lumbricoides*, Hookworms, and

*T.trichuria* range 264-5160, 144-528, and 192-1032, respectively. The overall average eggs burden for *A.lumbericoides* Hookworm and *T.trichuria* infections were 872.4, 283.6 and 605.3, respectively. The mean egg count of *A.lumbericoides*, Hookworms and *T.trichuria* students in male students was 892.8, 284.6, and 642.56 respectively and the total egg burden were 726.62 (Table 6) in the same way the mean eggs of *A.lumbericoide*, Hookworms and *T.trichuria* in female students were 841.5, 281.2 and 562.2, respectively and the total egg load were 723.9 (Table 6).

Much epidemiological research has shown variation in the intensity of helminths infections by age (Chan *et al.*, 1996). Changes in the average intensity of infection with age implies that there is rising in child hood and declining in adult hood. Children aged 5-15 years are most affected by *A.lumbericoides* and *T.trichuria*, however, the intensity and reinfection decline in adult hood (Gilles 1996), in contrast hookworm frequently exhibits a steady rise in intensity of infection with age, peaking in adult hood (Bethony *et al.*, 2002)

This trend was also observed in other studies (Brooker *et al.*, 2004). This could be because of high level of soil contact activity and low personal hygiene in the youngest age group. Similarly, lower intensity of hookworm infection was observed in age group 15-18 with an increase in 6-9 years age group with mean egg count 153.73 and 236.5, respectively. In the case of *A.lumbericoides* the mean egg count (1018.5 and 694) was recorded in 7-10 and 11-14 years age group, respectively (Brooker *et al.*, 2004) on the other hand no mean egg count was observed in 15-18 years age group. This could be because of high level of soil contact activity and low personal hygiene in the youngest age group. As the result shown in (Table 6) the intensity of *A.lumbericoides* and *T.trichuria* infection were found to be higher in male children than female

In this study the intensity of helminthes infection as measured by egg per gram of faeces is generally low in egg range. The overall intensity of helminths infection among the present study subject for *A.lumbericoides*, Hookworms, and *T.trichuria* were 872.4, 283.6 and 605.3, respectively (Table 6). It is comparable with report from Abosa around Lake Zway, South Ethiopia (Gezahegn, 2008). The low intensity level of helminths (STH) infection in the present study might be due to low humidity, unfavorable soil formation and chance difference in exposure to infection.

**Table 6.** Mean  $\pm$  SEM and Range of egg count of intestinal helminthic parasite egg per gram of faces (epg)

Age group	Sex	N	Al	Hw	Tt	Total egg load
			Mean $\pm$ Sem	Mean $\pm$ Sem	Mean $\pm$ Sem	Mean $\pm$ Sem
6-9 e	Mal	34	1036.6 $\pm$ 348.6	328 $\pm$ 56.3	612 $\pm$ 240	845 $\pm$ 252.6
	Fem	24	147.3 $\pm$ 367.5	211.2 $\pm$ 31.5	552 $\pm$ 0	878.1 $\pm$ 213.4
10-14 e	Mal	27	1102 $\pm$ 175.4	186.6 $\pm$ 26.6	840 $\pm$ 0	824.9 $\pm$ 119
	Fem	18	1088 $\pm$ 618	153 $\pm$ 36.4	576 $\pm$ 144	714 $\pm$ 316.5
15-18 e	Mal	-	-	-	-	-
	Fem	-	-	-	-	-
All age	Mal	59	892.8 $\pm$ 243.7	284.6 $\pm$ 33.2	642.5 $\pm$ 237	726.6 $\pm$ 170
	Fem	42	841.5 $\pm$ 244.3	281.2 $\pm$ 72.6	565.2 $\pm$ 85	723.9 $\pm$ 169
Total		101	872.4 $\pm$ 172.7	283.6 $\pm$ 24.1	605.3 $\pm$ 118	692.8 $\pm$ 116.1

Key; Al=*Ascaris lumbricoides*, Hw=Hookworms, Tt=*Tricuris trichuria* , Sem =standard error mean, No pos=number of positives

## 5. SUMMARY, CONCLUSION AND RECOMENDATION

### 5.1 Summary

The objective of the present study was to identify the prevalence and intensity of intestinal parasite species and to determine the major protozoan species and the major helminths species that cause infection among school children in the study area.

The design of the study was a cross sectional parasitological survey involving a sample population of 6-18 aged school children in Muketuri Primary School number one at Muketuri town during April - May 2017. A total of 400 stool samples were collected and examined by Direct Wet Mount, Formol Ether Concentration, Ziehl-Neelsen and Kato-Katz Method and showed that the prevalence of infections with protozoan and helminths, parasites were common in the study area. The overall prevalence of protozoan transmitted parasite infection was 177 (44.25% ), and the overall helminthes transmitted parasite infection was 99 (24.7).

The prevalence of protozoa parasite *E.histolytica*, *G.lamblia* and *Cryptosporidium species* was 144(28.5%), 51 (12.7%) and 12 (3%) respectively. The prevalence of protozoan infection among school children both male and female in age group 6-9, 10-14 and 15-18 were 70 (46.6), 90 (42.85), and 17(42.5) repectively.

The prevalence of helminths parasite *A.lumbericoides*, *Hookworms*, *T.trichuria* and *H. nana* was 64 (16%), 10 (5.5), 6 (1.5) and 16 (4%) respectively. The prevalence of helminths infection among school children both male and female in age group 6-9,10-14,and 15-18 were 55 (36.6 %), 44 (20.95%) and 0 (0%) respectively.

The intensity of helminths were also analysed and the mean egg count and range of *A.lumbericoides*, *Hookworm* and *T.trichuria* were 872.4(range 264-5160),283.6(range 144-528 ) and 605.3( range 192-1032 ) egg per gram of faces respectively.

In the present study area more of positive cases for helminthes parasitic infections were within the range of light and few moderate infections. There were no cases with heavy infection of the helminths.

## **5.2. Conclusion**

The major intestinal parasite species diagnosed in the school children of Muketuri town were *E.histolytica*, *G.lambliia*, *Cryptosporidiumspecies*, *A.lumbericoides*, Hookworm, *T.trichuria* and *H.nana*. The findings in the present showed that intestinal parasite protozoan infections were the major public health problems in the study area (Muketuri town). *E.histolytica* and *G.lambliia* infections were common for the school children. *A.lumbericoides*, Hookworms and *T.trichria* were found as a dominant species of intestinal helminths parasite diagnosed in the stool samples of school children.

The intensity of intestinal helminths parasite infections was analyzed and the result showed that the burden of intestinal helminths was within the range of light and moderate infections and there was no case with heavy infection of intestinal helminths.

Intestinal parasitic infections represent a series threat because of their water born and soil transmission. An increase prevalence of intestinal parasitic infection in the study area relate with factor such as poor drinking quality water for consumption, indiscriminate defecation by the inhabitants and their animals absence of toilet poor personal hygiene and environmental sanitation poor handling practice of water source and less awareness to intestinal parasitic infection.

### 5.3. Recommendation

Based on the finding of the present study about the prevalence of intestinal parasitic infections, the following recommendation can be suggested.

- Local health sector and any concerned bodies should collaborate with school health program for delivering health education to increase the knowledge, attitude and practice of school children as to how transmission of intestinal helminths infection is prevented, such as improvement of personal hygiene and environmental sanitation, shoe wearing habit.
- Moreover in depth studies should be made on socio economic factors like latrine usage hand washing practice to better evaluate the epidemiology of intestinal parasite infection in the area.
- Improving the pipe line water supply and its sanitation.
- School deworming program should be implemented.

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## 7. APPENDICES

### Appendix 1

#### 7.1.Consent Form(English Version) For Participation As Volunteer In Research Under Taking

I am a post graduate student from haramaya university . I am here to study the prevalence and intensity of parasitic infection among children in your school .The aim of this study is to determine the prevalence and intensity of parasitic infection in children to fulfill the requirement for M Sc thesis research . I am requesting you and /or your child’s genuine response to collection of stool .There is no any health related risk in participating. When your children are found positive for intestinal parasitic infections, you will receive standard drug free of charge .The information that you/your child provide me during the results of the laboratory investigation are strictly confidential. The participation of your child in this study completely voluntary and he/she can refuse to participate or free to withdraw him/her self from the study at any time .If you understood the explanation well enough. I am requesting you to your signature as illustrated below.

I the undersigned have been informed and understood that the purpose of this particular research project is to find out the prevalence and intensity of intestinal parasitic infection of school children. I have been told that I can refuse my child from participating in the study at any time. Hence with this understanding, I am here by giving my agreement to participate my child in this research voluntarily.

Name of Parent/Guardian ----- Sign----- Date----

## Appendix 2

### 7.2. Laboratory Data Collection Format

C o d e	S e x	A g e	Dat e/m onth	Pres ence /Abs ence	Parasitic Identification in the Stool							S i n f	D i n f
					Examination								
					E h	G l	C p	A l	H w	T t	Hn		

--	--	--	--	--	--	--	--	--	--	--	--	--	--

Key ;- 1. E.h = *Entamoeba histolytica*    5. Al = *Ascaris lumbricoides*  
 2. G.l = *Giardia lamblia*                6. Hw = Hook worms  
 3. CP = Cryptosporidium                 7. Tt = *Tricuris tricurria*  
 8. Hn = *Hymenolipes nana*

### Appendix 3

**7.3. Table 7. The Age and Sex Distribution of Study Participants in the Study Area**

Variables	Category	Frequency	Percent (%)
Age	6-9	150	37.5
	10-14	210	52.5
	15-18	40	10.0

Sex	Male	198	49.5
	Female	202	50.5

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## Appendix 4

### 7.4 .Cut of Values for Classification of Intensity of Infection

**Table 2 Egg Per Gram Of Stool, STH Infection Status (Light, Moderate and Heavy) Based**

	Light	Moderate	Heavy
<i>A.lumbricoides</i>	1-4999	5000-49999	>50000
<i>T.tricurria</i>	1-1999	1000-9999	>10,000
Hookworm	1-1999	2000-3999	>4000

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