Prevalence of intestinal parasitic infection among Libbefana Kindergarten Children in Kirkos Sub city, Addis Ababa, Ethiopia

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Acronyms

Al	*Ascaris lumbricoides*
CDC	Center for Disease Control and prevention
CI	Confidence Interval
CSA	Central Statistical Authority
DQC	Data Quality Control
Eh/d	*Entamoeba histolytica/dispar*
Gl	*Giardia lamblia*
Hn	*Hymenolepis nana*
IHPI	Intestinal Helminthes Parasitic Infection
IPI	Intestinal Protozoan Parasitic Infection
IPPI	Intestinal Parasitic Infection
NCCLS	National Committee on Clinical Laboratory Standard
RPM	Revolutions Per Minute.
SPSS	Statistical Package for Social Science
STH	Soil Transmitted Helminthes
Tt	*Trichuris trichuria*
MOH	Ministry of Health
WHO	World Health Organization

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ABSTRACT

Background*: Intestinal protozoan and helminthic parasitic infections are still one of the major health concerns in developing countries. Monitoring of intestinal protozoan and helminthic infection and associated risk factors are essential for intervention strategies. Therefore, the aim of this study was to assess the prevalence of intestinal parasitic infection among and associated risk factors among Libbefana Kindergarten children at Kirkos sub city, Addis Abeba, Ethiopia*

Objectives*: to assess the major protozoan and helminthic parasite species and determine their prevalence rate of protozoan and helminthic parasite in preschool children in Libbefana kindergarten School, Addis Ababa during Oct. 2017- June. 2018.*

Methods: *facility based cross-sectional study was conducted among preschool children students at Libbefana kindergarten children Kirkos sub city from October 2017 to June 2018. Three hundred twenty three participants were involved in the study by providing stool specimens and detailed personal information. Fresh stool samples were collected from each preschool child and processed by wet mount and formal-ether fecal concentration technique. Data were analyzed using SPSS version 20.0 statistical software and p value <0.05 were used as statistically significant.*

Results*: Among the 323 students participated in the study, (21.4%) were positive for one or more intestinal parasites. The rates of protozoan and helminthic parasites among preschool children were 15.8% and 5.6%, respectively. The most prevalent parasite detected in this study was G. lamblia (8.9%) followed by E. histolytica /dispar/ Moshkoviskii (6.8%). Among the different variables assessed in the study, parents' educational level, hand washing habits, water consumption, personal hygiene, parents' awareness to parasitic infection were statistically significance with intestinal parasitic infection (p<0.05)*

Conclusion and recommendations: *The major protozoan and helminthic parasite species diagnosed in the preschool children of Kirkos sub city were G.lamblia, E. histolytica, A.lumbricoides, T. trichiura and H. nana in varying magnitude. Improvement of sanitation and health education is required to reduce intestinal parasitic infection.*

Keywords: Intestinal parasite, Preschool children, Risk factors

1. INTRODUCTION

1.1 BACKGROUND OF THE STUDY

Protozoan and helminthic parasites have adverse consequence in health of human being. Parasitic protozoa are single-celled microorganisms that possess one, rarely two nuclei. Two most prevalent intestinal protozoa among preschool children are, Entamoeba histolytica lives in side the intestine as a trophozoite that means vegetative stage of protozoa showing motility and the ability to grow, feed, and reproduce. It produces resistant cysts by which it is transmitted. The other one is flagellates possesses at some stage of their life cycle one or more long hair-like flagella for locomotion. They reproduce asexually by binary fission. Gardia lamblia is an intestinal flagellate that is bilaterally symmetrical having two nuclei, two axonemes, and four pairs of flagella. Motile trophozoites can be found in faeces and also cysts by which G. lamblia is transmitted (Nyantekyi *et al.,* 2010)

Protozoan parasites are one of the causative agents of intestinal parasites. The main clinical manifestation of the disease caused by these parasites is diarrhea (Andualem *et al.,* 2014). The importance of parasites is due to its distribution in large population especially in children in most of the developing and poor countries, possibly because these parasites have easy way of transmission, such parasites are; Entamoeba histolytica, Giardia lamblia and Cryptosporidium infection happen via faecal contaminated food and water in most regions of the world. Intestinal protozoan parasites are among the most common human infections which are distributed throughout the world with high prevalence rates in developing countries (Shrestha *et al*., 2012)

Parasitic helminths include the nematodes (roundworms), trematodes (flukes) and the cestodes (tapeworms). Representatives of each of these groups are important parasites for preschool children. The adult worms, that inhabit the intestine, discharge their eggs or larvae in faeces. Nematode infections transmitted through soil contaminated by human faces, are causes for anemia, vitamin A deficiency, stunted growth, malnutrition, intestinal obstruction and impaired mental development (WHO, 2015).In Ethiopia high prevalence of helminth infection is attributable to factors associated with low socio-economic status. Such factors include poor personal hygiene, environmental sanitation, low household income, overcrowding and lack of clean water supplies. For instance, Ethiopia has one of the lowest quality drinking water supply and latrine coverage (Muhajir *et al.,* 2017)

Most infectious diseases are particularly prevalent in areas with warm climates in which man exerts himself least in developing sanitary protection and typically has a low threshold of resistance to invading organisms. Not all parasitic infections cause diseases of clinical significance. There are many factors that predispose to disease development, including host and parasite factors. The host factors include age, level of natural immunity at the time of

infection, life style , and presence of co-existing disease or a physical condition which reduces immune responses, e.g. pregnancy , under nutrition or malnutrition (Gelaw *et al.*,2013).

Intestinal parasites have their own characteristics; have different morphological and biochemical mechanisms to infect humans and animals. They are usually classified as protozoa and helminths. The most important intestinal protozoan pathogens are *Entamoeba histolytica*, *Cryptosporidium species*, *Giardia lamblia*, Cystoisospora (Isospora) belli, *Cyclospora cayetanensis*, and members of the phylum Microsporidia. The predominant intestinal helminths are Ascaris lumbricoides, Trichuris trichiura, Schistosoma mansoni, hookworm, *Hymenolepis nana, Enterobius vermicularis* and *Strongyloides stercoralis* (WHO, 2018).

Over 1.5 billion people are infected with one or more protozoan and Helminthic parasites; there are 700 million people infected with hookworm and 807 million people infected with ascariasis according to World Health Organization (WHO) estimates (WHO, 2018). Intestinal parasites are predominant in the developing countries mostly in sub-Saharan Africa including Ethiopia. Though affect all population, children are the most risk group. Intestinal parasite causes weight loss, intestinal obstruction and cognitive impairment (Mathers *et al.*, 2007). In Ethiopia intestinal parasitic infections are prevalent in varying magnitude among preschool children, school aged children and other age group. Toilet utilization , hand washing , and waking bare foot has impact in the prevalence of intestinal parasites (Abate et al.,2013), in addition mother's knowledge on the transmission of intestinal parasites play role in disease prevention and control (Nyantekyi *et al.*,2010).

Intestinal parasite infections are common among preschool children. There are different causes for the prevalence such as playing with soil, sucking fingers and defecation in open field. Maternal awareness for the prevention and control of intestinal parasite has its own impact on the prevalence. Ethiopia is affected by more prevalence of intestinal parasites that responsible for the major share of morbidity and mortality where there is poor sanitation, poor personal hygiene and absence of potable water. To reduce the impact of intestinal parasites increasing access to safe water, sanitation and health education are necessary and WHO recommends preventive chemotherapy, which is a periodic administration of anthelminthic medicines albendazole or mebendazole as a public health intervention (WHO, 2015).

Even though, several studies have been conducted on prevalence of intestinal parasites in Ethiopia, there are still several localities in the country including the study area, Kirkos sub city, for which information about the prevalence of intestinal parasitic infections was not available. Knowledge of residents about parasite transmission, personal and environmental hygiene and sanitation, proper use of latrine, checkup and treatment of children and the effect

of family size on children's infection with intestinal parasites were not studied in the present study area. Therefore, the purpose of this study is to obtain information about the prevalence of intestinal parasites and associated risk factors libbefana kindergarten children. Kirkos sub city, Addis Abeba, Ethiopia.

1.2. STATEMENT OF THE PROBLEM

There are a number of reports on protozoan and helminthic infections in different African countries. It is clear that unsanitary conditions, unclean water utilization and low health education service are common in African. A study conducted in Democratic Republic of Congo, shows the burden of intestinal parasite among pre-school children, an overall prevalence of 43%. According to their report the prevalence of protozoan and helminthic infection was higher among children aged between 1 and <5 years than those aged <1 year this might be to the exposure of neonate to contaminated food and water (Kandala *et al.*, 2013).

The prevention and control mechanism for intestinal parasites are mass drug administration, health education, sanitation and clean water supply. Globally in 2013, more than 266 million preschool-aged and 609 million school-aged children were estimated in need of preventive chemotherapy for soil transmitted helminths in 106 countries. In Africa more than 13.8 million preschool- aged children in need of treatment were treated. (WHO, 2015).

In Ethiopia main strategies are mass drug administration, case detection and transmission control. Information on the prevalence and distributions is incomplete and not updated periodically. There is lack of information on the prevalence of intestinal parasite and knowledge, attitude and practice of their mother on prevention and control in the present study area.

Human protozoan parasites are identified as causes of morbidity and mortality throughout the world particularly in developing countries including Ethiopia. They are more prevalent throughout the tropics, especially among poor communities. Records show that increasing trends of intestinal protozoan parasitic infections in developing countries. A high prevalence of intestinal protozoan parasite infections in human are positively correlated with poverty and poor personal hygiene, lack of safe water supply and contamination of the environment by human excreta and animal wastes. protozoan parasitic infections increase host`s susceptibility to other infections and diminish learning ability and growth especially in growing children (Mekonnen *et al.,* 2014). Intestinal protozoa Parasite species are responsible for some of the most devastating and prevalent diseases of humans. Protozoan parasite infections constitute a global health burden causing clinical morbidity in 450 million people; many of these were women of reproductive age and children in developing countries (Quihui, 2006)

1.3 SIGNIFICANCE OF THE STUDY

This study gives baseline information on the prevalence of protozoan and helminthic parasites among libbefana kindergarten children, factors associated with IPIs and mothers and care givers knowledge, attitude and practice on the prevention and control of intestinal parasites. The finding is also important for those who are working on the prevention and control of protozoan and helminthic parasitic infections among the stated age groups, in the study area and other similar areas. In addition this study could help those who want on the association of mothers' knowledge and managements practice on infectious disease and child health status for IPIs as baseline information for formulation appropriate control strategies.

2. LITERATURE REVIEW

2.1. Human protozoan and helminthic Parasites

Protozoan and helminthic parasitic infections are global health problems causing clinical illness in 450 million inhabitants in developing countries (Quihui *et al.,* 2006). Parasites found in the intestine can be categorized into two groups; as protozoan and helminths. The major intestinal parasites of global public health concern are the protozoan species such as *E. histolytica and G. intestinalis*, soil transmitted helminthes *A. lumbricoides, T. trichiura*, hookworm and schistosomiasis (WHO, 2018). Helminthic infections are enhanced by poor socio-economic conditions, lack of sanitary facilitates, improper disposal of human feces, insufficient supplies of potable water poor personal hygiene, poor housing conditions and lack of education (WHO, 2015).

IPI and helminths in particular, are associated with increased risks for nutritional anemia, protein energy malnutrition and growth retardation in children, poor increase in body weight in pregnancy, intrauterine growth retardation, and low birth weight (Tulu *et al* 2014). Children infected with soil-transmitted helminths (STHs) have poor educational level and performance at school and a high level of truancy, thus impacting on their future earnings and productivity (Mohammed *et al*., 2015).

2.1.1. Human protozoan infections

Protozoa are single celled organisms. The common protozoan parasites include Entamoeba histolytica/dispar, Giardia lamblia/intestinalis, Cryptosporidium and Cyclospora. Even though the majority of protozoa occur as free living organisms in the soil, moist, marine or fresh water environments, a substantial number also exists as mutual, commensal or parasite. Protozoan parasites are known to affect all species of vertebrates and many invertebrates. They are able to adapt to life in virtually all body sites of their hosts (Ngonjo *et al*., 2012).

Entamoeba histolytica/dispar is an intestinal parasite that characterized by possessing clear protoplasm which form pseudopodia. These pseudopodia are the means by which the organisms move and use for feeding purposes. The two species Entamoeba histolytica and Entamoeba dispar are morphologically identical but pathologically distinct (WHO, 2015). Amoebiasis is one of the health issues in many developing countries. It is the second most common cause of death due to parasitic infection after malaria as estimated by the World Health Organization (WHO, 2018). Approximately about 10% of the world population is infected with E. histolytica/dispar (Gelaw *et al*., 2013), but most infections occur due to the noninvasive species (Petri and Singh1999). Asymptomatic infection with E. histolytica is defined as the presence of cysts in stools in the absence of colitis or extra intestinal infections. These healthy carriers may pass millions of cysts in the stool per day as the trophozoites multiply in the intestinal lumen (Haftu *et al.,* 2014).Approximately, 90% of all

intestinal E.histolytica infections are asymptomatic. Clinical symptoms of acute intestinal Amoebiasis include diarrhea, bloody stool that may contain necrotic mucosa, abdominal pain, tenderness and fever (Firdu *et al.,* 2014).

According to World Health Organization (2015), the prevalence of amebiasis varies with the population of individuals affected, differing between countries and between areas with different socioeconomic conditions. Sometimes up to 50% of the population is affected in regions with poor sanitary conditions. Epidemiological studies have shown that low socioeconomic status and unsanitary conditions are significant independent risk factors for the infection. In addition, people living in developing countries have a higher risk and earlier age of infection than do those in developed regions. For example, in Mexico, 11% of the tested population aged 5 to 6 years was infected with amoeba.

In Ethiopia, more than 60% of the diseases are caused due to poor environmental health conditions arising from unsafe and inadequate water supply and poor hygienic and sanitation practices. According to the ministry of Health (2006), nearly 80% of rural and 20% of urban population have no access to safe water. A number of studies and routine diagnosis in Ethiopia indicate that amoebiasis is one of the most widely distributed diseases. In 50 communities of the central plateau of Northern Ethiopia the parasite was reported in 94% of the communities, with prevalence rate ranging from 3% to 55% (Alwabr *et al*., 2016). G.lamblia/intestinalis (also known as Giardia duodenalis or G. intestinalis) is a unicellular flagellated intestinal protozoan parasite of humans isolated worldwide and ranked among the top 10 parasites of man (Ayalew *et al*., 2011). Although symptomatic infection causes a broad spectrum of clinical manifestations, Giardia results in asymptomatic carrier state in a majority of cases. The asymptomatic infections are most common in children and people with prior exposure to a source of infection; Clinical symptom of giardiasis includes diarrhea, epigastric pain, wasting and impaired absorptions (Mathewos *et al.,* 2014).

G. lamblia is the most protozoan intestinal parasites isolated worldwide as causative agents of diarrhea. Epidemiological studies suggest that the parasite is responsible for about 5% of acute diarrhea and 20% of chronic diarrhea illness in the world. The incidence of diarrhea associated with Giardia is generally higher in developing countries in Africa, Asia, South and Central America where access to clean water and basic sanitation is lacking. The prevalence for Giardia lamblia in developed countries is around 2-5% but in developing countries may be up to 20-30% (Tilahun *et al*., 2010). Cryptosporidium species are very small intestinal protozoa. They dwell in the stomach or in the small intestine of mammals, birds and reptiles. This apicomplexa parasite infects humans and animals globally. Up to now eight valid Cryptosporidium species have been reported to be capable of infecting humans (Andualem *et al*., 2014). It can cause gastrointestinal illness in a wide variety of mammals, including humans, cattle, sheep, and goats, pigs and horses worldwide (Firdu *et al*., 2014).

Cyclospora species have been identified in animals. *Cyclospora cayetanensis* is the only species found in humans, and it is apparently restricted to this host. *Cyclospora cayetanensis* originally referred to as "cyanobacterium-like bodies" has been recognized as a waterborne pathogen and reclassified (Bahmani *et al*., 2017). It has been associated with several waterborne outbreaks worldwide.

2.1.1.1. Life cycle of human protozoan parasitise

There are two diagnostic life cycle stages commonly seen in parasites, the cysts and the adult trophozoite stage. The trophozoite stage can be detected directly on a slide without concentration. Cysts require concentration (Erismann *et al*., 2016). The life cycle of E. histolytica includes the motile and invasive trophozoite and the infective cyst. Infection is acquired primarily through the ingestion of infective cyst forms present in faecally contaminated water and food (Haftu *et al* 2014). The trophozoite measures 10-50 micro meter and contains a single nucleus; whereas, cyst is 10-15 micrometer in diameter and contains four nuclei. E. histolytica cysts are resistant to gastric juices present in the human stomach, chlorination desiccation and capable of surviving in a moist environment for several weeks (Liza *et al*., 2010).

After ingestion of contaminated water or food, the cyst wall is dissolved in the upper gastrointestinal tract and the organism excysts with in the lumen of the small intestine. During excystation, nuclear division is followed by cytoplasm division, giving rise to eight uni-nucleated trophozoites. Trophozoites *E. histolytica* epithelial cells line the gastrointestinal tract. Once penetration of the intestinal mucosa is achieved, dissemination to other organs, extra intestinal infections, usually the liver, can occur. Trophozoites which dwell in the colon multiply encyst and are passed in the stool from where further spread is possible (Matthys *et al*., 2011).

G. lamblia reproduces by binary fission. This is a type of reproduction in which one cell divides into two new cells by mitosis during the growth cycle. The components of the cell multiply so that each daughter cell is a complete copy of the parent cell. This parasite has a simple direct life cycle consisting of on infective cyst and a vegetative trophozoite. The cyst of *Giardia lamblia* is elliptically shaped and contains two to four nuclei (Alwabr *et al*., 2016).

The round or oval shaped cysts, which are the infective form of the protozoa, are approximately 11-14 m long and 7-10 m wide. After ingestion the cysts pass unharmed by gastric acid through the stomach to the small intestine. Excystation normally occurs in the duodenum. Infection with Giardia is usually confined to the upper small intestine but also has been observed in the bile duct and gall bladder of ill patients. The structure of the cyst makes the organism very resistant to environmental factors and disinfection and it is the transmittable form that causes the infection. Identification of the parasite is usually made by

microscopic examination of direct fecal smear for either trophozoite or cysts in the feces (Ngonjo *et al*., 2012).

Cryptosporidium completes its life cycle in a single host and culminates in the shedding of mature oocysts in the faeces. These are immediately infective for another susceptible host. The oocysts are 4–6 m in diameter (smaller than many other protozoa), and contain four crescent shaped infective structures of sporozoites. After ingestion the oocyst excysts in the small intestine, releasing the sporozoites. The sporozoites attach themselves to the gut epithelium, initiating the infection, which develops through further stages of asexual and sexual multiplication, zygote formation, oocyst formation, and sporulation. Each of the stages of the organism's life cycle is found within the cell but outside the cytoplasm, and after an incubation period of 2–10 days, the pathogen gives rise to symptoms in humans. No specific toxin is produced (Mohammed *et al*., 2015).

Cyclospora species may complete its life cycle within enterocytes. Cyclospora sp. oocysts are spherical, measuring 8– 10 m in diameter; they are excreted in the stool and sporulated to infectivity in the environment. Unpopulated oocysts contain a central morula-like structure consisting of a variable number of inclusions. Sporulated oocysts contain two ovoid sporocysts, within each of which there are two sporozoites (Speich *et al*., 2013).

2.1.2. Human helminthic parasitic infections

Parasitic helminthes (worms) that infect humans belong to two phyla, Platyhelminths and Nematoda. The common intestinal helminthes are trematodes (flukes) includes *Schistosomia mansoni*), nematodes (round worms) includes *A. lumbricoides, T. trichiura* and hook worms (*Necator americanus* and Ancylostoma duodenale) and cestodes (tape worms) includes *Hymenolepis nana, Taenia saginata* and *Taenia solium*. Helminthic infections are enhanced by poor socio-economic conditions, lack of sanitary facilitates, improper disposal of human feces, insufficient supplies of potable water, poor personal hygiene, poor housing conditions and lack of education (WHO, 2015). According to Masoumeh *et al*. (2012), at global burden, over one billion of the world's population is estimated to be infected with helminthes parasites and over two billion people are at risk.

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 21st century in many developing countries of the tropics. The disease is endemic in 74 tropical developing countries (Bahmani *et al.,* 2017). People become infected when coming in contact with water containing Schistosoma infected snails.

Nematodes: nematodes (round worms) are non-segmented helminthes, relatively simple structured organisms. They possess bilateral symmetry and a complete digestive tract with oral and anal openings; they taper to a relative point at both ends. They are also found to have separate sexes, with the male being smaller than the female, ranging in size from a few millimeters to over a meter in length. Nematodes infections have a wide spread distribution being found in both temperate and tropical climates. They can be found in fresh water, in the sea and in soil. About 85% of Nematodes infections are asymptomatic (Abera *et al*., 2014).Ascaris lumbricoides is the largest of the intestinal nematodes found in man. Crompton (1999) reported that *A. lumbricoides* infects at least one-fourth of the world's population. Annual morbidity associated with the parasite has been estimated by WHO at 60,000 with another 250 million peoples said to be at risk for acquiring the infection (Masoumeh *et al*., 2012). *A. lumbricoides* is a robust parasite. This is 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 (Ngonjo *et al*., 2012).

T. trichiura is commonly known as whip worm, due to the whip-like form of the body. T. trichiura infection is estimated to affect around 1049 million persons worldwide. Of these, 144 million are children of pre-school age and 233 million are of school age. These nematodes are most commonly seen in tropical climates and in areas where sanitation is poor (Shahrul *et al* 2012).

Cestodes: 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 *Hymenolepsis nana*, the former due to the custom of eating raw meat and the later due to unhygienic food consumption with contaminated hands and fingers that allow the ingestion of eggs from the faeces of an infected person (Bahmani *et al.,* 2017).

The adult worm of H. nana found in the intestine. It is very small, only a few centimeters long. The egg is unique in its appearance. It is small, measuring 30-47 μm 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, 2015).

2.1.2.1. Life cycle of human helminthic parasites

The life cycles of most helminthes follow the same pattern Adult worms of genera *Necator americanus* and *Ancylostoma duodenale* parasitise the upper part of the human small intestine, while roundworms parasitise the entire small intestine. Adult trichuris (whipworms) live in the large intestine, especially the caecum. 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 infections can also be acquired from pigs (Sierra *et al*., 2011).

The soil-transmitted helminthes vary greatly in size, and female worms are larger than males. After mating, each adult female produces thousands of eggs per day, which leave the body in the feces. People become infected with *T. trichiura* and *A. lumbricoides* by ingesting the fully developed eggs (Yami *et al*., 2011). *A. lumbricoides* infections in humans occur when an ingested infective egg releases a larval worm that penetrates the wall of the duodenum and enters the blood stream. From here, it is carried to the liver and heart, and enters pulmonary circulation to break free in the alveoli, where it grows and molts. In 3 weeks, the larvae pass from the respiratory system to be coughed up, swallowed, and thus returned to the small intestine, where they mature to adult male and female worms. Fertilization can now occur and the female produces as many as 200,000 eggs per day for a year. These fertilized eggs become infectious after two weeks in soil; they can persist in soil for 10 years or more (Matthys *et al.,* 2011).

2.2. Epidemiology of human protozoan and helminthic Infections in preschool children

Protozoan and helminthic parasites are present throughout the world in varying degrees of prevalence. The burden of the infection remains one of the greatest health problems in the developing world, with over one billion people estimated to be infected (Emile *et al.,* 2013), while most infections remain asymptomatic, the clinical spectrum of disease widespread, ranging from mild gastrointestinal symptoms to death from disseminated infection. In East Africa including Somalia, Ethiopia, Eritrea, Kenya and Sudan more than 50% of people are at risk of illness from parasitic diseases. This is due to deteriorated health service, armed conflict, famine and economic hardship (Aleka *et al*., 2015). Preschool age children are one of the groups at high risk for intestinal parasitic infections. The adverse effects of intestinal parasites among children are adverse and alarming. Intestinal parasitic infections have a great effect on the survival, appetite, growth and physical fitness, preschool children cognitive performance of school age children (Mekonnen *et al*., 2014).

Intestinal protozoan infections are endemic worldwide. In developed countries the prevalence of human intestinal parasitic protozoan infection 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 is also a cause of "travellers' diarrhea, in which the disease is sometimes also called beaver fever. Entamoeba 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, 2015).

Soil transmitted helminthes *A. lumbricoides, T. trichiura*, hookworm and schistosomiasis (WHO, 2015). The incidence and prevalence of these parasitic pathogens vary between and within the countries. This is due to ecological and socio economic factors and difference in human behaviors and sanitations. Over population deficient sanitary facilities and shortage of potable water are the major risk factors particularly in the tropics and sub tropics (WHO, 2018).

2.2.1. Global distribution of human IPIs in preschool children

Global prevalence of helminthic infections in school age children is estimated at about 35% caused by Ascaris, 25% caused by trichuris and about 26% are caused by hookworms. Heavy infection of intestinal parasite can cause malnutrition, loss of appetite, interfering food absorption and weight loss. Majority of intestinal parasites are more likely to make the children ill. And, this can lead to the children's missing of school (WHO, 2015). Globally, the intestinal parasitic infections have been recognized as one of the most significant cause of illnesses and diseases especially among disadvantaged communities. With an average prevalence rate of 50% in developed world and 95% in developing countries, it is estimated that intestinal parasitic infection results in 450 million illnesses (WHO, 2018). These infections are ubiquitous with high prevalence among the poor and socio economically deprived communities where overcrowding, poor environmental sanitation, low level of education and lack of access to safe water are prevalent.

2.2.2. Human protozoan and helminthic infections in Ethiopia

Intestinal parasites have been widely distributed in Ethiopia, as in most of African countries. They are more prevalent in the poor segments of the population with low household income, poor handling of personal and environmental sanitations, overcrowding and limited access to clean water (Aleka *et al.,* 2015).

Parasite helminthic infections are the second most predominant causes of outpatient morbidity in the country. Several studies stated that, Ethiopia has one of the lowest qualities of drinking water supply and latrine coverage in the world. According to Masoumeh *et al*., (2012), more than half a million annual visits of the outpatients' services of the health institutions are caused by intestinal parasitic infections. This estimate is inaccurate, because most of the health institutions lack appropriate diagnostic tools to detect low level of parasite burden. Still data on intestinal parasites in Ethiopia is inadequate (Mathewos *et al*., 2014). As a result of low level standards of living, poor environmental sanitations and ignorance of simple health promoting factors, intestinal parasitism is very high. Even though the prevalence of individual parasite varies in different parts of the country, *Ascaris lumbricoides* is the most prevalent intestinal parasite. Many reports in Ethiopia indicated that the most

prevalent soil transmitted helminthes are A. lumbricoides followed by *T. trichiura* (Wegayehu *et al*., 2013).

As Abera *et al*., (2014) reported, *A. lumbricoides* and *T. trichiura* are co-existing and highly prevalent with increase in altitude. The highest prevalence of infection was recorded at an altitude more than 2400m above sea level and it was also known by commonly affecting school age children. Of 301 school children who were studied in south western Ethiopia, 68.4% harbored one or more parasites. In that study from the total ten species identified, *Ascaris lumbricoides* was the leading (52.2%) followed by *T. trichiura* (18.6%), while *Schistosoma mansoni* was the least (0.3%) (Abossie *et al*., 2014).

Another study was conducted in Babile town; eastern Ethiopia on 415 school children and Hymenolepis nana was the most prevalent followed by hook worms (Gelaw *et al*., 2013). In Ethiopia, infection of intestinal parasite remains among the most ubiquitous and serious health problems with strikingly high prevalence rates of the major protozoan and helminthic infections. According to Andualem *et al.,* (2014), intestinal parasitism accounts for 8.5% of all male and 10.4% of all female outpatients' visit in the country.

2.3. Factors that affect the epidemiology of protozoan and helminthic infections

2.3.1. Behavior, household clustering and occupation

Specific occupations, household clustering, and behaviors influence the prevalence infections particularly for hookworm, in which the highest intensities occur among adults (Brooker *et al*., 2010). Engagement in agricultural activities, for example, remains a common denominator for hookworm infection.

2.3.2. Poverty and sanitation

Parasitic infections depend for transmission on environments contaminated with egg-carrying feces. Consequently, intestinal parasites are intimately associated with poverty, poor sanitation, and lack of clean water. The provision of safe water and improved sanitation are essential for the control of parasitic infections. The populations in developing countries live in conditions that are highly conducive to the acquisition of parasitic infestation. Poor hygiene, crowded household conditions, dietary habits, education level of the community and deficient sanitation mark their day-to-day life (Valiathan *et al*., 2016).

2.3.3. Climate, water and season

Intestinal parasitic infections are highly prevalent in warmer and moister areas. This is because hook worm, Ascaris, Trichuris and S. mansoni ova require humid environments Many helminths infections are more common in children than in adults. In addition to this,

multiple infections can also play a role because children tend to live more closely with nature and with their pets (Valiathan *et al.*, 2016). Not all parasitic infections cause diseases of clinical significance. There are many factors that predispose to disease development, including host and parasite factors. The host factors include age, level of natural immunity at the time of infection, life style, and presence of co-existing disease or a condition which reduces immune responses, e.g. pregnancy, under nutrition or malnutrition (Gelaw *et al*., 2013). Soil transmitted helminthes are highly affected by surface temperature altitude, soil type, and rainfall (Wale *et al*., 2014). Adequate warmth and moisture are key features for each of the soil transmitted helminthes.

2.3.4. Age dependency

The high prevalence rate of intestinal infection in children is attributed to many factors, particularly environmental and personal hygiene. For reasons not well understood, school aged children (including adolescents) and pre-school children tend to harbor the greatest number of intestinal worm. As a result they experience growth stunting and diminished physical fitness as well as impaired memory and cognition (Valiathan *et al*., 2016).

2.4. Morbidity and health effect of protozoan and helminthic parasitic Infections

Protozoan and helminthic infections are the most important human parasites at global scale. Morbidity and mortality due to intestinal parasitic infections are usually more pronounced in children compared to adults due to their higher nutritional requirements and less mature immune systems (Gelaw *et al*., 2013). Furthermore, the risk for poor clinical outcomes is reported to be increased in those children who were already malnourished prior to becoming infected (Strunz *et al*, 2014).

Preschool children and infants are reported to be most vulnerable to the adverse nutritional effects of the intestinal parasitic infections. The main reason of this is that, they often suffer from an increased intestinal parasitic infections burden associated with a greater exposure to these infections' agents by virtue of unsanitary practices associated with child development, such as playing in contaminated dirt and water, sucking on dirty fingers and other objects.

Growing children also have high nutritional requirements (Strunz *et al*, 2014). Asymptomatic Amoebiasis which occurs after initial acute infections is highly prevalent in both children and adults residing in developing countries. Acute E. histolytica infection is associated with malnutrition. The rapid transit time associated with both frequent diarrheal episodes is responsible for decreased nutrient absorption. In addition, children who suffer from liver abscess and other forms of extra-intestinal amoebic infections are reported to experience metabolic alterations, which also affect their nutritional status ((Strunz *et al,* 2014).

2.5. Prevention and Control of protozoan and helminthic Infections

According to World Health Organization guidelines (WHO, 2018), any health program aiming at controlling morbidity of intestinal parasitic infections should have evidence based estimates of this problem. Human intestinal parasitic protozoan infections can be controlled through proper treatment and disposal of row swage and maintaining clear water supply including the protection of open wells, springs and rivers from contamination with swage and feces. The risk for infection can also be reduced via the adequate boiling of drinking water or treatment of water with chlorine or iodine. The exterior of row vegetables and fruits should be washed with soup and soaked in vinegar for some minutes before conception (Shahrul *et al*., 2012).

A well-structured control strategy needs to be based on local and accurate data concerning the epidemiology, definition of targets, definition of appropriate chemotherapy and health education campaigns, sanitation, monitoring and evaluation programs (Abera *et al*., 2014). All these components need to be integrated into the prevailing system of primary health care and must be based on multi sectorial collaboration (WHO, 2015), a goal often difficult to carry out in practice, which is why it is common to find control programs based on some of these elements and with limited results. There are three types of basic control programs,

2.5.1. Improved sanitation

Prevent faecal contamination of the environment by using latrine and protecting water supply from faecal contamination. Control programs based on sanitation aim to reduce or interrupt transmission, prevent reinfection and gradually reduce worm loads (Bahmani *et al*., 2017). However, to be effective in a short period of time they need to be combined at their first stage with chemotherapy. Long term sanitary control programs need to add elements to improve the economic conditions of a region, to ensure a reliable and permanent sanitation system and have permanent health education programs (Gelaw *et al*., 2013).

2.5.2. Treatment

Iodoquinol is used to treat asymptomatic infections and Metronidazole is used for symptomatic and chronic Amebiasis, including extra-intestinal disease. In acute clinical cases of giardiasis the disease is often self-limiting and therefore may require only supportive therapy, which usually consists of fluids to compensate for fluid losses in the diarrhea. Metronidazole is the drug of choice for Giardia infection. Recommended drugs used in the treatment of soil-transmitted helminths are albendazole, mebendazole; and older drugs including pyrantel, tiabendazole and niclosamide (Heelan, 2004).

2.5.3. Health education

Health education and promotion of healthy behaviors can play a key role in reducing the incidence of human intestinal parasitic infections. However, the effectiveness of those activities in reducing transmission of infection varies according to different reports. In some cases, health education can decrease costs, increase levels of knowledge, and decrease reinfection rates. Health education efforts can build trust and engage communities in aspects that are crucial to the success of public health initiatives (Mbae*et al*., 2014

3. OBJECTIVES

3.3.1 General objective

To assess the major intestinal parasite species and determine their prevalence rate in Libbefana kindergarten children, Addis Ababa during Oct. 2017- Jan. 2018

3.3.2 Specific objectives

- To identify the major intestinal parasite species among Libbefana kindergarten children.
- To determine the prevalence of intestinal parasite species among Libefana kindergarten children.

4. MATERIALS AND METHODS

4.1. Description of the study area

The study was conducted in Kirkos or cherkos sub-city Health Center, Addis Abeba the capital city of Ethiopia (Figure 1).According to the new city administration classification, Addis Ababa is divided into ten sub-cities, Among the Ten sub cities, Addis Ketema, Lideta and Kirkos are the most disadvantaged, that is to say majority of the inhabitants are poor. The majority of population (60-70%) of such disadvantaged areas suffered from food insecurity (kidist, 2010). Out of these disadvantaged areas, Kirkos sub city has been selected purposively.

Kirkos is one of the ten sub cities of Addis Abeba, the capital of Ethiopia as of 2011population was of 235,441, male110,069, female 125,372 population density per square 16,104,specific location of the study area around kazanchis.

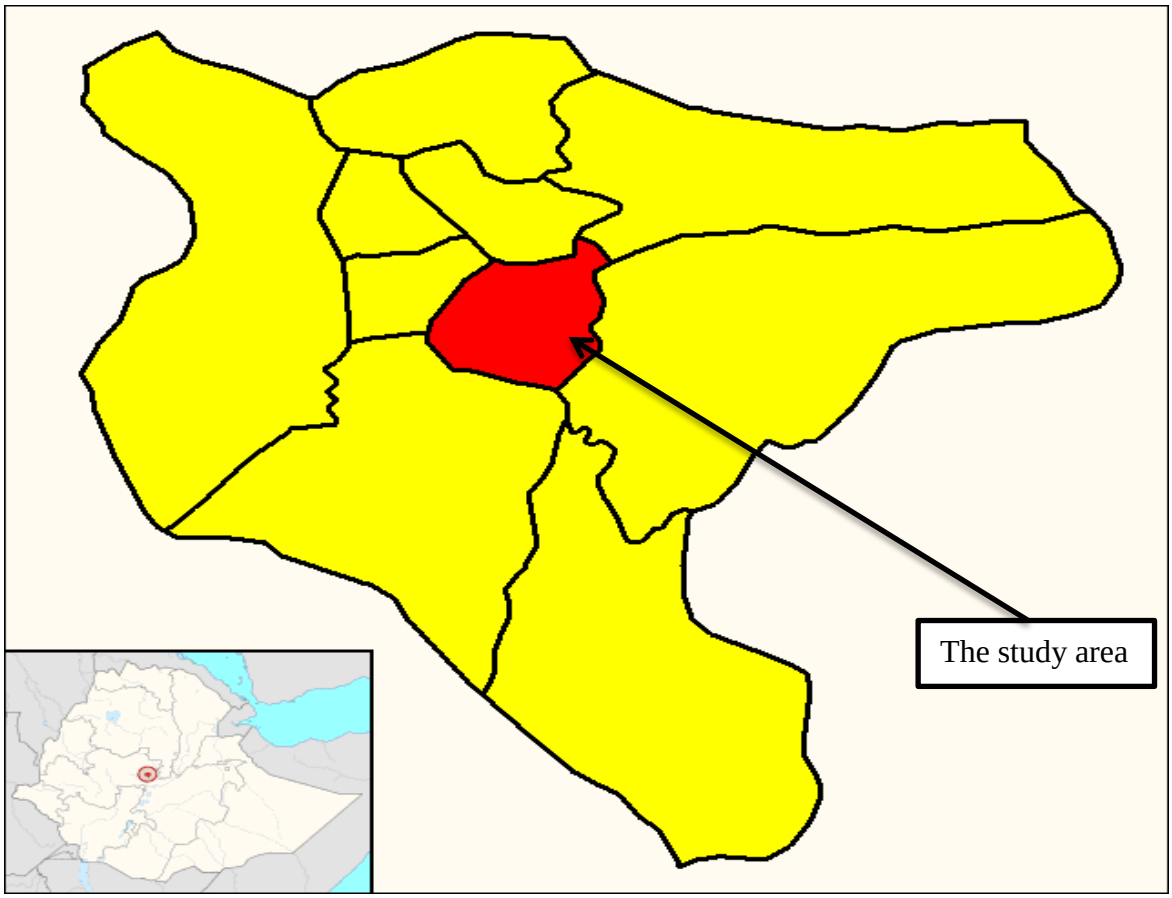


Figure 2: Map of the study area
Source:Https://en.m.Wikipedia.org'wiki'kirkos

4.2. The study design

The study design was a survey on intestinal parasitic infections and major risk factors among Libbefana kindergarten School children at Kirkos sub city It was conducted from Sep.2017 to Jan.2018.

4.3. The study participants

Preschool children those were found in Libbefana Kindergarten children during 2017/20182018 were included in the present study.

4.4. Exclusion criteria

All preschool children who started anti parasitic drugs and also completed treatment before three days were not included in the study. Furthermore, those patients from the surrounding Kirkos sub city healthy and school aged children were excluded in this study.

4.5. Sample size determination

The sample size was estimated using the following statistical formula (Daniel, 1999):-

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

Where:

n= sample required

Z= 95% confidence interval (1.96)

d= margin of error (5%)

P= prevalence rate.

Since the overall prevalence rate (p) of intestinal parasites is not known for the study area, prevalence rate was taken to be 30%..For the calculation, 95% confidence level (z) and 5% sampling error (d) was used. Therefore, three hundred twenty three (323) preschool children from were participated in the present study.

4.6. Sampling Technique

A sampling technique used in this study was convenience sampling technique which is used to include all consented patients requested for stool examination during the study period.

4.7. Method of Data Collection

4. 7. 1. Stool Sample collection

Each fecal sample specimen was consisting of 10-20g of fresh stool which was obtained with the cooperation of the family of less than five years old children. The collected samples were kept in a plastic container and transported to the laboratory for examination within one hour of delivery. At the time of sampling; date of sampling, age, sex, presence or absence of

intestinal parasitic infections, and code number was recorded for each child on the record format.

4. 7.2. Questionnaire Survey

A pre-tested standardized questionnaire based on intestinal parasitic infection was developed. The questionnaire was constructed in English and then translated into Amharic. Then, the children's families were interviewed in Amharic. This standardized questionnaire was used together with relevant general information on demographic and socioeconomic data of the children and their parents in the study area. The questionnaire was administered and observations on physical situations of each child attending at health center were recorded by the investigator and an oriented health assistant.

4.8. Laboratory Parasitological Procedures

4.8.1. Wet Mount Method

A direct wet mount with normal saline (0.85% NaCl solution) was prepared at study site and observed for the presence of motile intestinal parasites, trophozoites and eggs under light microscope at 10X and 40X magnification. Lugol's iodine staining was also used to observe cysts of intestinal parasites (WHO, 2004).

4.8.2. Formol-Ether Concentration Method

Using an applicator stick, approximately 5g or pea sized fecal materials were placed in a centrifuge tube containing10 ml of 10% formalin. After emulsifying the feces in the formalin, it was filtered through the nylon filter into the test tube. The filtrate was washed to discard any lumpy residue with a normal saline solution. Then after, the filtrate was washed again, by transferring into a test tube containing 7 ml of ether/ethyl acetate. The tube was closed with a stopper and it was shacked vigorously to mix. The stopper was removed and it was centrifuged at 1500 rpm for 2 minutes. The tube was rested in stand for five minutes. Four layers became visible with the top layer consist of ether, second was a plug of debris and the third was a clear layer of formalin and the fourth was the sediment. The plug of debris from the side of the test tube was removed with the cotton swab and poured off the liquid leaving a small amount of formalin for suspension of the sediment. Then after, the sediment was removed with a pipette. Then, a drop of fluid was added on the slide for examination under a cover slip. Some drop of iodine solution was added on the second glass slide. A 10x and 40x objectives was used to examine the whole of the deposit for ova and cysts and trophozoites.

4.9. Data Quality Control (DQC)

To ensure quality control, all the laboratory procedures including collection and handling of specimens were carried out in accordance with standard protocols (WHO, 2015). To ensure general safety, disposable gloves were worn and universal bio-safety precautions (NCCLS, 2002) were followed at all times. For QC of the direct wet mount method, preserved stool specimens known to contain parasite ova, cyst and larvae were included in each batch of samples to ensure that the procedures are precise.

The calibration factors for the 10x and 40x objectives were posted on the microscope for easy access; and the weight scales were checked at the beginning of each working day. To ensure accurate identification of parasite species, bench aids for the diagnosis of intestinal parasites WHO (2015), and diagrams of various parasite ova and larvae from the parasitological manual were reviewed.

4.10. Ethical Considerations

The official permission letter for ethical clearance was obtained from Addis Ababa University institution of review board. Further permission was obtained from Kirkos sub-city healthy center. The libbefana kindergarten children's family were informed about the objective and purpose of the study and verbal consent was obtained from each family before starting to distribute the questionnaires. The information obtained at each course of study was kept confidential.

4.11. Study Variables

Protozoan and helminthic parasitic infection positivity used as dependent variable. Whereas use of water and its handling, refers to whether the water contaminated or not. Parents education level, this variable indicates the levels of education, such as primary education, secondary education and had diploma and above. Hand washing after defecation before eating, this refers to hand washing practice of children. Personal hygiene refers to generally keeping oneself clean and awareness to parasites infection.

4.12. Data analysis

The data was computerized using Excel 2007, cleaned and checked against original document before analysis. All statistical analyses were performed using SPSS for windows version 20 statistical package. The prevalence of intestinal parasites was determined by Pearson p-value) test verifying the relationship between independent factors and protozoan and helminthic infection. The 95% CI was used to show the accuracy of data analysis. Probabilities less than 5% ($P < 0.05$) was considered statistically significant.

5. RESULTS

5.1. Socio-Demographic Characteristics of the Study Participants

Socio-demographic characteristics of the study participants are summarized Table 1. A total of 323 subjects were participated in this study, among them, 156 (48%) were males and 167 (51. 7%) females. Children`s age ranged between ≤3 was 64(19.8%,) 3.1-4: 79(24.5%), 4.1-5; 84(26.0%), 5.1-6: 96(29.7%).

Table1 Socio-demographic characteristics of study participants at Libefana kindergarten children

Parameters	Category	Frequency n=323	Percent
Sex	Male	156	48.3
	Female	167	51.7
Age group	≤3	64	19.8
	3.1-4	79	24.5
	4.1-5	84	26.0
	5.1-6	96	29.7
Religion	Orthodox	198	61.3
	Muslim	72	22.3
	Protestant	53	16.4
Parent educational level	Illiterate	61	18.9
	Primary school	113	35.0
	Secondary school	93	28.8
	Diploma and above	56	17.3
Water consumption	By boiling	61	18.9
	As it is (pipe water)	262	81.1
Availability of latrine	Public	198	61.3
	Private	125	38.7
Personal hygiene	Poor	246	76.2
	Good	77	23.8
Parent awareness to parasitic Infection	Poor	238	73.7
	Good	85	26.3

From 232 population participated in this study, 198(61.3%) were Orthodox, 72(22.3%) were Muslims, 53(17.3%) were Protestants. 246(64.1%) respondents said that they were used boiled water supply whereas 262(81.1%) have a protected water supply for domestic use. With regard to parents' education, 61(18.9%), 113(35%), 93(28.8%), and 56(17.3%) said that they were illiterate, primary school, secondary school and had diploma and above respectively. 271(83.9%) of the children households had public latrines. The remaining 52(16.1%) had private latrines in close vicinity of their homes. 246(76.2%) and 77(23.8%) were with poor and good personal hygiene, respectively. About 238(73.7%) participants' were with poor life skills and the remaining 85(26.3%) were with good life skills.

5.2. Prevalence of protozoan and helminthic Infections among libbefana kindergarten Children

The prevalence of intestinal protozoan and helminth parasite infections among preschool children is summarized in Table 2. The prevalence rate of protozoan parasite infections and helminthic parasite infections among preschool children of both sex and all age groups was 51(15.8%) and 18(5.6%), respectively; with 69(21.4%) overall ratio. Out of these, the prevalence rate of protozoan parasitic infection and helminthic parasitic infection among male children was 23(14.7%) and 11(7.0%) respectively; with 34(21.7%) overall infection rate. Similarly, the prevalence rate of protozoan parasitic infection and helminthic parasitic infection among female children was 28(16.8%) and 7(4.2%), respectively; with 35(20.9%) overall prevalence rate. Although there was no statistically significant ($p>0.05$) difference, the prevalence rate of protozoan parasitic infection in female preschool children was greater than that of males. To the contrary the prevalence rate of helminthic parasitic infection in male children among preschool children was greater than that of females. This might be male children have a chance to play out of their homes and increased the chance of infection with soil transmitted helminths

As shown in Table 2, Among 323 children participated in this study, 69 (21.4%) libbefana kindergarten children were infected with at least one or more parasites. Children were infected with protozoan and helminth parasites with a prevalence 51 (15.8%) and 18 (5.6%) respectively. Children age ranged between ≤3 was 6(9.4%), 3.1-4: 12(15.2%), 4.1- 5: 16(19.04%), 5.1- 6: 35(36.5%), 5.1-6:35(36.5%) were infected with human protozoan and helminthic parasites. The low rate of prevalence was observed in aged ≤3 year old and high rate of infections was for the children aged from 5.1-6 years old. Children in 5.1-6 years old were highly infected with both protozoa and helminth infections and indicating a risk for acquiring intestinal parasite infections. The prevalence rate of intestinal parasite and species diversity in the study site revealed that increment with age group as the age increased; particularly it was highest in age group from 5.1-6 years old 36.5% .This might be due to the common childhood behaviors of eating soil, neglecting to wash hands after defecation eating meal, using non-purified water and improper cleaning of child dinning utensil. The

prevalence rate of infection intestinal parasites in this study were not statistically significant between male and female children of different age group (p>0.05).

Table 2: Prevalence of protozoan and helminthic infections among libbefana kindergarten children

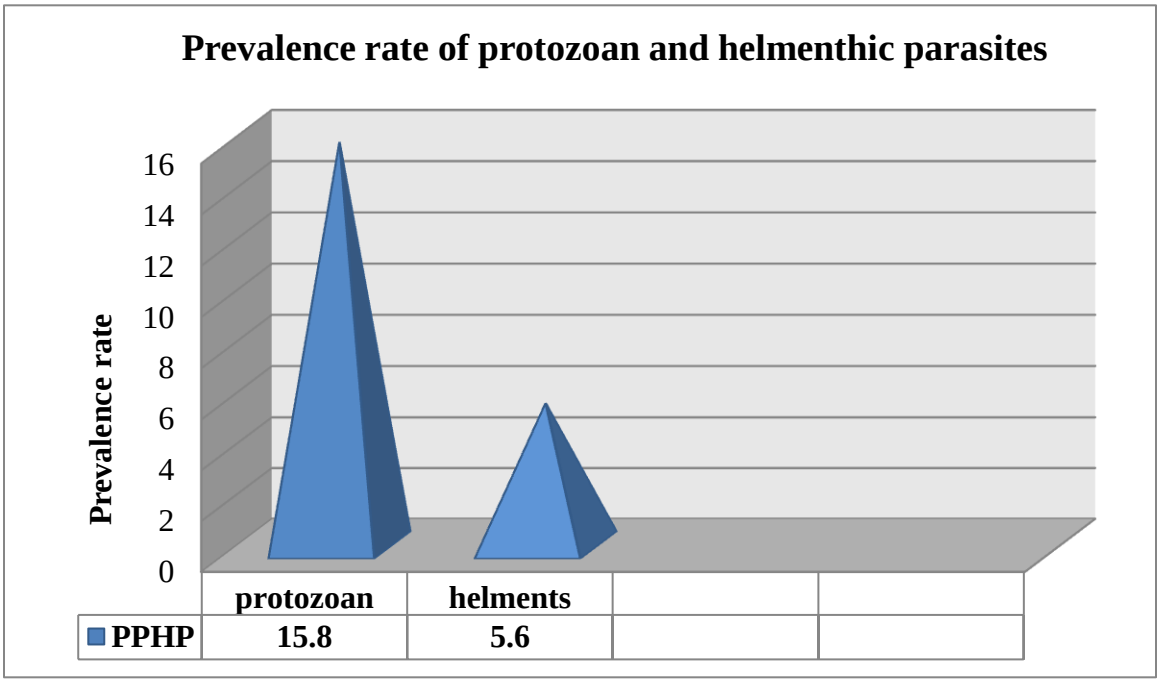
Age Group (years) Sex	No of Examined	Protozoan Parasitic Infection		Helminthic Parasitic infection		p-value
		Positives		Positives		
		No	%	No	%	
≤3						
Male	31	2	6.5	1	3.2	1.000
Female	33	3	9.1	0	0	
3.1-4						
Male	41	5	12.2	1	2.4	0.533
Female	38	4	10.5	2	5.3	
4.1-5						
Male	39	4	10.2	3	7.7	1.000
Female	45	7	15.6	2	4.4	
5.1-6						
Male	45	12	26.7	6	13.3	1.000
Female	51	14	27.4	3	5.9	
All age						
Male	156	23	14.7	11	7.0	1.000
Female	167	28	16.8	7	4.2	
Total	323	51	15.8	18	5.6	

Among 31 male Libbefana kindergarten children age ≤3, 2(6.5%) and 1(3.2%) children infected with protozoan and helminthic parasites. Similarly from33 female children age ≤3, 3(8.6%) infected with protozoan parasitic infection but none is infected with helminthic parasites. Out of 41 male preschool children age range between3.1-4, 5(12.2%) and 1(2.4%) children infected with protozoan and helminthic parasites but 4(10.5%) and 2(5.3) female children were infected with protozoan and helminthic parasitic infection. There was statistically insignificance association between protozoan and helminthic parasitic infection and sex of children (p>0.05)

In the present study out of 41 male libbefana kindergarten children age 4.1-5, 4(10.2%) and 3(7.7%) children were infected with protozoan and helminthic parasites. About 45 female children age 4.1-5, 7(15.6%) and 2(4.4%) preschool children were infected with protozoan and helminthic parasites respectively. out of 45 male preschool children age 5.1-6, 12(26.7%) infected with protozoan parasites whereas 6(13.3%) infected with helminthic parasites. Similarly from 48 female children of the same age group, 14(29.2%) and 3(6.3%) positive for protozoan and helminthic parasitic infection of the same age group. There was statistically insignificance association between protozoan and helminthic parasitic infection and sex of children ($p>0.05$)

To sum up, from 69(21.4%) of positive cases of surveyed in Libefana kindergarten school, the prevalence rate of protozoan and helminthes parasitic infections were found to be 51(15.8%) and 18(5.6 %,) respectively (Table 2). Thus, it was interesting to find that the intestinal protozoan infection seemed to be more problem than the helminthes parasites. The high prevalence of these infections is closely correlated with poverty, poor environmental hygiene and impoverished health services (Gelaw et al., 2013) There was statistically no significance correlation between protozoan and helminthic parasitic infection and sex of children ($p>0.05$)

Figure 2: prevalence rate of protozoan and helminthic parasites

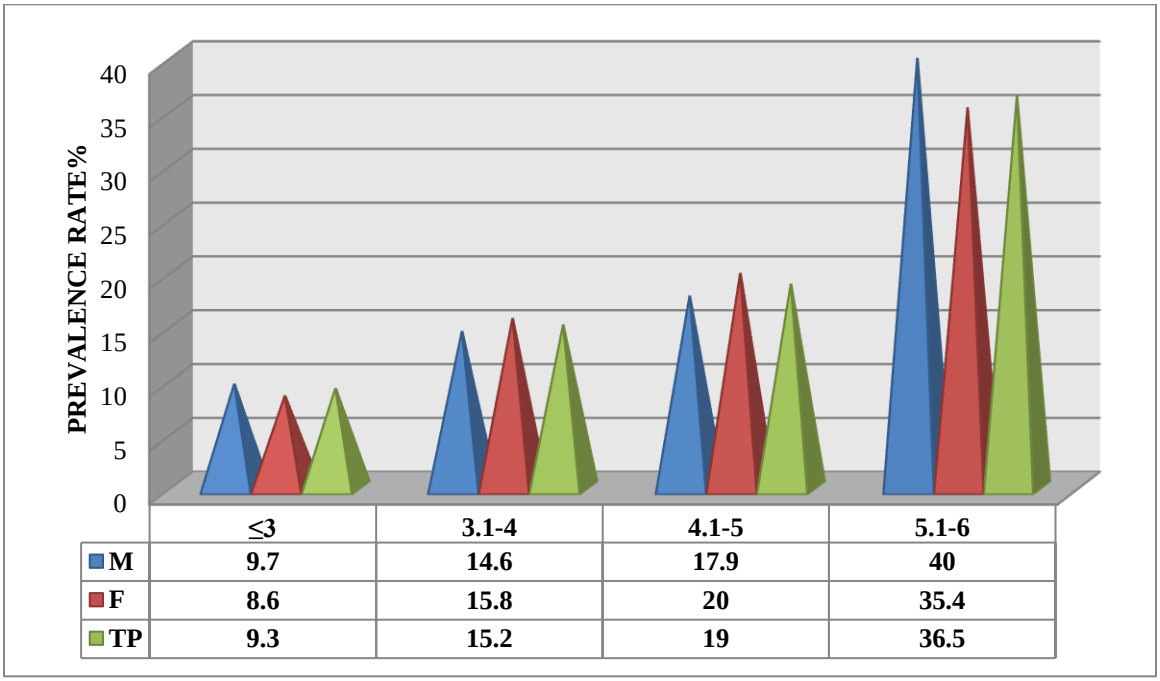


PPHP=Prevalence of protozoan and helminthic parasites

The above figure shows that, 323 children whose stool samples were examined 51 (15.8%) and 18(5.6%) were infected with protozoan and helminthic parasites respectively. Pathogenic

protozoa infections were higher prevalent then intestinal helminths infection. The transmission of these parasites occurs by ingestion of cysts through the fecal–oral route, either directly, via person to person contact or indirectly, via contamination of surface water or food. In the present study, the highest prevalence of protozoa parasites could be due to contaminated water and food, as this study has shown that significantly higher protozoan parasitic infections 51(15.8%) were found among preschool children. Highest prevalence of pathogenic protozoa infections are known to cause diseases in children (CDC, 2010). The transmission of these parasites occurs by ingestion of cysts through the fecal-oral route, either directly, via person to person contact or indirectly, via contamination of surface water or food (Bahmani *et al.*, 2017).Soil transmitted helminthes infections is the cause of common health problems in tropical and subtropical areas, with the greatest numbers occurring in sub-Saharan Africa, the Americas, China and East Asia(WHO, 2018).Younger children are predisposed to heavy infections with intestinal parasites since their immune systems are not yet fully developed, and they also habitually play in faecally contaminated soil. In addition to considerable mortality and morbidity, infection with intestinal protozoan helminthes parasites have been found to profoundly effect on a child's mental development, growth and physical fitness while also predisposing children to other infectious agents (Mbae *et al.*,2013).

Figure 3; prevalence of intestinal parasites at Libbefana kindergarten children of different age group during October 2017 to July 2018



	≤3	3.1-4	4.1-5	5.1-6
M	9.7	14.6	17.9	40
F	8.6	15.8	20	35.4
TP	9.3	15.2	19	36.5

M=Male, **F**=Female, **TP-=** Total prevalence

Figure 3 shows that The prevalence rate of protozoan and helminthic parasite and species diversity in the study site revealed that increment with age group as the age increased; total

prevalence rate of protozoan and helminthic parasites in age group ≤3; 6(9.2%), 3.1-4; 12(15.2%), 4.1-5;16(19%) and 5.1-6;35(36.5%) particularly highest infection rate was recorded in age group from 5.1-6 years old 35(36.3%).This might be due to the common childhood behaviors of eating soil, poor hands washing after defecation and before eating, using non-purified water and improper cleaning of child dinning utensils.

5.3. Prevalence rate major protozoan and helminthic parasite species identified libefana kindergarten Children

Protozoan and helminth parasites identified in the stool samples examined among preschool children less than presented in Table 3. The result of parasitological investigations showed that, from 323 specimens of preschool children in Libbefana kindergarten children, 69(21.4%) were positive for one or more intestinal parasites. Of these, 34(21.8) and 35(21.0) were males and females, respectively.

Table 3 Prevalence protozoan and helminthic parasite species identified among libbefana kindergarten children

Age Group (years)	No of Examined	Protozoan parasites		Helminthic parasites			Multiple Parasites	P value
		Gl	Eh/d	Al	Tt.	Hn.		
		No pos. (%)	No pos. (%)	No pos. (%)	No pos. (%)	No pos. (%)	No pos. (%)	
≤3								
Male	31	1(3.2)	1(3.2)	1(3.2)	0	0		1000
Female	35	2(5.7)	1(2.9)	0	0	0		
3.1-4								
Male	41	3(7.3)	3(7,3)	1(2.4)	0	0		0.533
Female	38	2(5.3)	1(2.6)	0	1(2.6)	0		
4.1-5								
Male	39	3(7.7)	1(2,6)	2(5.1)	0	1(2.6)	1(2.6)	1.000
Female	45	4(8.9)	3(6.7)	1(2.2)	1(2.2)	0	0	
5.1-6								
Male	45	6(6.7)	5(6.7)	3(6.7)	2(4.4)	1(2.2)	2(4.4)	1.000
Female	48	8(9.5)	7(9.5)	1(2.1)	1(2.1)	2(4.2)	1(2,1)	
All age								
Male	156	13(8.3)	10(6.4)	7(4.5)	2(1.3)	2(1.3)	3(1.3)	
Female	167	16(9.6)	12(7.2)	2(1.2)	3(1.8)	2(1.2)	1(1)	
Total	323	29(8.9)	22(6.8)	9(2.8)	5(1.5)	4(1.2)	4(1.2)	

G. lamblia and *E. histolytica/dispar* the major protozoan parasites identified from children under five years with the prevalence of 29(9.0%) and 22(6.8%), respectively. A higher prevalence of *G. lamblia* 29(9.0%) was found among libbefana kindergarten children as compared with *E. histolytica/dispar* 22(6.8%) (Table3).The major helminth parasites identified in the stool samples of the preschool children were *A. lumbricoides, T. trichiura, H. nana* with the prevalence of 9(2.8), 5(1.5) and 4(1.2) respectively. *A. lumbricoides* was the most prevalent helminth parasites in the study sites.

Among 31 male libbefana kindergarten children age less than or equal to three protozoan and (≤ 3), 1(3.2%) and 1(3.2%) were positive for *G. lamblia* and *E. histolytica/dispar* respectively whereas from helminthic parasites only *A, lumbricoides* 1(3.2%) positive case identified in stool sample. Out of 35 female children in this age group, 2(5.7%) and 2(5.7) were positive for *G. lamblia* and *E. histolytica/dispar*

In the present study from 41 male children in age group 3.1-4, 3(7.3%) were positive for *G.* lamblia and *E. histolytica/dispar* similarly among soil transmitted helminths only 1(2.4%) positive for *A. lumbricoides*. From 38 female children of these age group, 2(5.3%) and 1(2.6%) were positive for *G. lamblia* and *E. histolytica/dispar* respectively while only 1(2.6%) T. trichiura were positive cases.

A total of 39 male children in age group 4.1-5, 3(7.7%) and 4(8.9%) were positive for *G.* lamblia and *E. histolytica/dispar* similarly among helminthic parasites 2(5.1%) and 1(2.6%) were positive for *A. lumbricoides* and *H. nana* respectively. From 45 female children of these age group, 4(8.9%) and 3(6.7%) were positive for G. lamblia and *E. histolytica/dispar* respectively. while among soil transmitted helminthes *A. lumbricoides* 1(2.2%) and *T. trichuira* 1(2.2%) were positives.

To sum up, out of 323 parasitological investigation of stool sample of libbefana kindergarten children, 69 (21.4%) were positive for one or more intestinal parasites. Out of these, 34(21.8) and 35(21.0) were males and females, respectively. *G. lamblia* and *E. histolytica/dispar/* were the major protozoan parasites identified from preschool children with the prevalence of 29(9.0%) and 22(6.8%), respectively. A higher prevalence of *G. lamblia* (8.9%) was found among libbefana kindergarten children as compared with *E. histolytica/dispar/* (6.8%) (Table3). Similarly, the major prevalent helminth parasites identified in stool samples of the children were *A.lumbricoides, T. trichuira, H. nana* with the prevalence of 9(2.8%), 5(1.5%) and 4(1.2%) respectively. *A.lumbricoides* was the most prevalent helminth parasites in the study sites.

5.4. Association of Intestinal Parasitic Infections with socio demographic characteristic at libbefana kindergarten children

This study has also analyzed correlation between socio-demographic factors of the preschool children and the prevalence of intestinal parasitic infections. The overall prevalence of each protozoan and helminthic parasite species diagnosed in the study of pupils and the proportion of different socio-demographic factors are presented in (Table 4).

Table 4 Association of intestinal parasitic infections with socio demographic characteristic at libbefana kindergarten children'

Parameters	Category	Frequency	Intestinal Protozoan	Intestinal Helminthes	p- value
			Pos. No (%)	Pos. No (%)	
Parents educational Level	Illiterate	61	18(29.5)	5(8.2)	0.034
	Primary school	113	17(15.0)	7(6.2)	
	Secondary school	93	11(11.8)	4(4.3)	
	Diploma and above	56	5(8.9))	2(3.6)	
Availability of latrine	Public	198	39(19.7)	13(6.6)	0.784
	Private	125	12(9.6)	6(4.8)	
Personal hygiene	Poor	246	42(16.7)	14(5.6)	0.024
	Good	77	9(11.7)	4(5.2)	
Parents awareness to parasitic infection	Poor	238	39(16.4)	12(5.04)	0.462
	Good	85	12(14.1)	7(8.2)	
Hand washing habits	Sometimes	216	38(17.6)	16(7.4)	0.001
	Always	107	13(12.1)	2(1.9)	
Finger cleanness	Clean	95	6(6.3)	3(3.2)	0.013
	Not clean	228	45(19.7)	17(7.5)	
Eating uncooked Food and salads	Sometimes	104	5(4.8)	7 (6.7)	0.036
	Always	219	46(21)	11(5)	

Among the 323 participants of the study, 61 participants of the study were with illiterate parents, 18(29.5%) protozoan and 5(8.2%) helminthes parasitic infections. 113 participants of the study were parents who complete primary school, 17(15%) protozoan and 7(6.2%) helminthes parasitic infections, From 93 participants of the study whose parents completed secondary education, 11(11.8%) were found to be positive for protozoan and 4(4.3%) for helminthes infection. The remaining 56 participants' of the study parents who had diploma and above, 5(8.9%) were found to be positive for protozoan and 2(3.6%) for helminthes infection. Parents educational level statistically significance with intestinal parasitic infection (p=0.034).

Out of 198 participant of who have public latrine, 39(19.7%) were positive for protozoan parasites whereas 13(6.6%) positive for helminthic parasite, 125participants who have private latrine 12(9.6%) were positive for protozoan parasites and 6(4.8%) positive for helminthic parasite. There was statistically insignificance association between intestinal parasitic infection and toilet availability (p=0.784).

In the present study, out of 246 participants of the study who were with poor personal hygiene, 42(16.7%) and 14(5.6%) were found to be positive for protozoan and helminthes parasites infections, respectively (Table4). About 77participants of the study who were with good personal hygiene, 9(11.7%) and 4(5.2%) were found to be positive protozoan and helminthes parasites infections, respectively (Table4). Children personal hygienic condition statistically significance with intestinal parasitic infection (p=0.024)

A total of 238 children parents poor awareness to parasitic infection, of these, 39(16.4%) and 12(5.04%) were positive for protozoan and helminths parasitic infection. About 85 parents and care giver with good life skill 11(13.4%) and 9(10.5) were positive for protozoan and helminths infection. About 298 parents with poor awareness of parasitic infection, out of these good awareness to parasitic infection 12(14.1%) and 7(8.2%) were positive for protozoan and helminths infection. Parents awareness to parasitic infection were statistically significance with intestinal parasitic (p=0.462)

From 216 participants of the study who were with poor hand washing habit after defecation and before eating, 38(17.6%) and 16(7.4%) were found to be positive for protozoan and helminthes parasites infections, respectively (Table4). About 107 participants of the study who were with good hand washing habit after defecation and before eating, 13(12.1%) and 2(1.9%) were found to be positive protozoan and helminthes parasites infections, respectively (Table4). Hand washing habit after defecation and before eating, statistically significance with intestinal parasitic infection (p=0.001)

Among 95 participants of the study clean their finger, 6(6.3%) were found to be positive for protozoan parasites and 3(3.2%) for helminthic infection. From 228participants of the study who do not clean their finger, 45(19.7%) were found to be positive for protozoan and

17(7.5%) for helminthic infection. in this study, finger cleanness were the major risk factors for the prevalence rate of intestinal parasites p=0.013 (Table4).From 104 participants of the study sometimes eat uncooked food and salads, 5(4.8%) were found to be positive for protozoan parasites and 7(6.7%) for helminthic infection. Out of 219 participants of the study that always eat uncooked food and salads, 46(21%) were found to be positive for protozoan and 11(5%) for helminthic infection. in this study, eating uncooked food and salads were the major risk factors for the infection rate of intestinal parasites p=0.046 (Table4).

To sum up, the prevalence rate of intestinal parasitic infections were statistically significant ($p<0.05$) with some risk factors such as parents educational level, hand washing habits, water consumption, personal hygiene, parents awareness to parasitic infection were the major factor for intestinal protozoan and helminthic parasitic infection. However in this study toilet availability was statistically insignificance correlation between intestinal parasitic infection and toilet availability (p=0.784).

6. DISCUSSION

Intestinal parasites are one of the leading causes of death among children in the developing countries. Hence, adequate information about the prevailing state is an important epidemiological tool in evaluating existing or new intervention programs. In this study, the overall prevalence of intestinal parasitic infection was 21.4%. This is higher findings from Gondar Ethiopia 17.3% (Aleka*et al* 2013) and from Tanzania (15.1%) (Vargas *et al* 2004). The finding is lower as compared to studies done in Wondo Genet (85.1%) (Gelaw *et al*., 2013), in Yergalem hospital (49.5%) (Firdu *et al.,* 2014), and in Kenya (25.6%) (Mbae *et al.*, 2013), this variation could be due to different geographical distribution of the parasites, timeline and implementation of different prevention and control measures. However, the finding of our study is still higher according to the national safe environment strategy in the extension program in Ethiopia. Pathogenic protozoa infections are known to cause diseases in children]. The transmission of these parasites occurs by ingestion of cysts through the fecal–oral route, either directly, via person to person contact or indirectly, via contamination of surface water or food (WHO, 2011).

The prevalence of both intestinal helminthic and protozoan infections among preschool children at Kirkos sub city healthy center, Addis Abeba, Ethiopia. The results of the study revealed the presence of various intestinal parasitic infections in varying degrees among preschool children. From 323 children participated in this study, 21.4% preschool children were infected with at least one or more parasites. 15.8% Children were infected with protozoan and 5.6% helminth parasites. *G. lamblia* and *E. histolytica/dispar* were the major protozoan parasites identified from preschool children with the prevalence of 9.0% and 6.8%, respectively. A higher prevalence of *G. lamblia* 9.0% was found among preschool children as compared with Entamoeba histolytica/dispar 6.8%.The major helminth parasites identified in the stool samples of the preschool children were *A.lumbricoides* 2.8, *T. trichiura* 1.5, *H. nana* 1.2. *A.lumbricoides* was the most prevalent helminth parasites in the study sites.

In the present study, *G. lamblia* was the predominant protozoan parasite with a prevalence of 9%. This causes malaise, abdominal cramps, weakness, weight loss, distention, and flatulence. Children are more liable to massive infection with severe clinical manifestations (CDC, 2011). The more chronic stage is associated with vitamin B12 mal-absorption, disaccharides deficiency and lactose intolerance (WHO, 2015). The organism may invade the liver, lung and brain where it produces abscesses that result in liver dysfunction, pneumonitis, and encephalitis (Wegayehu *et al*., 2013). Malaise, weakness, weight loss, abdominal cramps, distention, and flatulence can occur. Children are more liable to clinical giardiasis than adults. Immunosuppressed individuals are especially liable to massive infection with severe clinical manifestations. Symptoms may continue for long periods (Alwabr *et al.*, 2016). Early symptoms include flatulence, abdominal distension, and nausea and foul-smelling bulky, explosive, and often watery, diarrhea. The stool contains excessive

lipids but very rarely any blood or necrotic tissue. The more chronic stage is associated with vitamin B12 malabsorption, disaccharides deficiency and lactose intolerance (WHO, 2015). This study also found that *E. histolytica* was the second prevalent (6.8%) protozoan parasite among the study participants.

The major soil transmitted helminthic parasites identified in the stool samples of the preschool children were *A.lumbricoides, T. trichiura, H. nana* with the prevalence of 2.8, 1.5 and 1.2 respectively. *A.lumbricoides* was the most prevalent helminth parasites in the study sites. It is found in association with poor personal hygiene, poor sanitation, and contamination of hand by human faces which contain infective egg. Preschool children infected with ascariasis may cause intestinal muscle become damage and absorption impaired, protein digestion or absorption impaired, absorption of fat decrease and infective children are often vitamin K deficiency (WHO, 2018).*T. trichiura* (1.5%) is the second most abundant soil transmitted helminths in this study. Preschool children develop *Trichuris* dysentery syndrome, characterized by abdominal pain and dysentery, children develop weight loss and become emaciated. Anemia is common and results from both mucosal bleeding secondary to capillary damage

The highest prevalence of protozoa parasites could be due to contaminated water as this study has shown that significantly higher parasitic infections (15.6%) were found among preschool children who drank untreated water. Public health interventions such as the provision of clean water, community health education, observation of food hygiene, and maintenance of functioning sanitation systems are fundamental to long-term intestinal parasite control (Harhay, 2010). In this study, about 16.7% of the parasitic infection of the children was due to poor hand washing practice of mothers. This problem is also reported in University of Gondar community school (Gelaw *et al*., 2013). Moreover, preschool children born from illiterate mothers were more infected (37.7%). Similarly, nail hygiene and educational level of mothers were closely associated with the prevalence of intestinal infections .All these evidences have shown that there should be effective implementation of intervention activities to control the spread of intestinal parasitic infections in the setting.

7. CONCLUSION AND RECOMMENDATIONS

7.1 Conclusion

The major protozoan and helminthic parasite species diagnosed in the libbefana kindergarten children of Kirkos sub city were *G.lamblia, E. histolytica, A.lumbricoides, T. trichiura* and *H. nana*. The findings in the present study showed that intestinal parasitic protozoan infections were the major public health problems in the libefana kindergarten children of kirkos sub city, Addis Ababa Ethiopia. *G.lamblia* and *E. histolytica,* infections were common protozoan infection in the study area. *A.lumbricoides* and *T. trichiura* and H. nana were found as a dominant species of intestinal helminth parasitic infection.

7.2 Recommendations

The findings of the present study showed that intestinal protozoan and helminthic parasitic infections were prevalent in libbefana kindergarten children. Therefore, the major preventive measures would include: the improvement of general standards of sanitation through preventing the environment from faecal contamination. Generally, health authorities should make concerted efforts to ensure the prevention of these parasitic protozoan and helminthic from infecting people.

Long term control measures to improve libbefana kindergarten children health and growth condition including mass treatment for the effective control of intestinal parasitic infections. Increasing health education related to intestinal parasitic infection shows transmission of intestinal parasitic infection is prevented such as improvement of personal hygiene and environmental sanitation. For the control of these parasitic infections, a half-yearly repeated anti parasitic treatment is recommended for children as well as the community to reduce re-infection. This is because if one is de-wormed at intervals, there is a possibility of killing most parasitic helminthes and protozoan parasite cysts in the intestine before they cause heavy infestation leading to severe health consequences.

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Appendix-I English questioners

CONSENT FORM

An ensuring format of parents whose children are not old enough to begin schooling:

My respectful greetings go to you here

I am a post graduate student from Biology department, Addis Abeba University. I am here to study the current status of intestinal parasitic infection in Libefana kindergarten children. The information provided by you in this questionnaire will be used for research purposes. It will not be used in a manner which would allow identification of your individual responses. Stool examination may be necessary if your child has fever, or the stool is mucoid and bloody. I am planning to study the rate of Intestinal parasite by examining stool. On your agreement I would like to examine the stool of your child. If you agree to participate in this study you may expect the following:

- ❖ Stool samples will be taken processed for direct microscopy and concentrated techniques. The result will be informed to the respective physician.
- ❖ Despite your willingness, the researcher kindly request you to give your genuine response for each question

If the above conditions are acceptable to you, please sign on this form

Investigator	Parent/ Guardian
Signature	Signature
Date: _____	Date: _____

Questionnaires format

1. Code No._____
2. Age: _____months. Sex: _____ (M / F). Child lives with parents: _____ (Yes / No).
3. Father Age _____ years. Health status _____
   A=Healthy
   B=Sick
   C=Died).
4. Mother Age _____years. Health status _____
   A=Healthy
   B=Sick
   C=Died).
5. Maternal Education: _____
   A=Illiterate C=secondary education
   B=primary education D =diploma and above
6. Duration of diarrhoea _____days. History of fever _____ days.
7. Stool character: _____(1=bloody mucoid, 2= mucoid, 3=not 1 and 2).
8. Does your child wash or do you wash after toilet and before eating?
   A. Yes B. No
9. Do you eat unwashed fruits and vegetables? A) Always B) sometimes C) never
10. Which latrine type do you use?
    A. Private B. Public
11. How often you clean the latrine?
    A) Always B) once per week C) twice per week D) three times per week
    E) Not at all
12. Water consumption;
    A. By boiling C. as it is
13. Your child meal;
    A. always fresh B. sometimes fresh C. not fresh
14. Your child playing ground is:
    A. clean B. not clean
15. Child nails status A. Clean B. Not clean
16. Did you get information and training about personal and environmental hygiene and sanitation respectively before? A) Yes B) No
17. Do you wash your hands before meal and after latrine use?
    A. yes B. No

Appendix-IL-Amharic questioners

በጎፈቃደኝነት ማረጋገጫ

እድሜያቸው ለትምህርት ያልደረሱ ህጻናት ወላጆች መረጃ መስጫ ና በጎፈቃደኝነት ማረጋገጫ ቅፅ

ጤና ይስጥልኝ !!

እኔ በአዲስ አበባ ዩኒቨርስቲ ባዮሎጂ ዲፓርትመንት የድህረም ረቃተማሪ ነኝ::እዚህ የተገኘሁበት ዋና አላማ እድሜያቸው 5 አመት በታች የሆኑ ህጻናትን ስለሚያጠቃው የአንጀት ጥገኛ ተህዋስ በሽታ ለማጥናት ነው ::ለመጠየቁ የሚሰጡት መረጃ ለጥናታዊ ምርምር የሚዉል ነው::ይሁንና የእርሶን ማንነት ለማጥናት አይውልም ::በጎፈቃደኛ ከሆኑ ልጅዎ ትኩሳት፣ሰገራው ደም ያዘለ ወይም ንፍጥ መሰል ዝልግልግ ነገር ካለበት ሰገራ እንዲሰጥ ሊጠየቅ ይችላል፡ ፡የተሰበሰቡ የሰገራ ናሙናዎችን በማጥናት የአንጀት ጥገኛ ህዋስ በሽታን ስርዐት ማጥናት የዚህ ጥናታዊ ፅሁፍ ዋነኛ እቅድ ነው::በዚህ ጥንታዊ ጽሁፍ ለመካተት ከተስማሙ የሚከተሉትን እንዲያሟሉ ይጠየቃሉ::

- ☞ ልጅዎ ከላይ የተዘረዘሩ ምልክቶች ካለበት የሰገራ ናሙና እንዲሰጥ ይጠየቃል::
- ☞ መጠየቆቹ በሚያዙት መሰረት እውነተኛ እና ትክክለኛ መልሶችን እንዲሰጡ በትህትና ይጠየቃሉ::

ከላይ በተዘረዘረው ማብራሪያ መሰረት በዚህ ጥናታዊ ፅሁፍ ለመሳተፍ ከተስማሙ እባክዎ ከታች በተጠቀሰው ቅፅ ይፈርሙ::

መጠየቁን ያደረገው	ያጠነከረው የወላጅ/ያሳዳጊ
ፊርማ	ፊርማ
ቀን	ቀን

መጠይቅ

1. መለያ ቁጥር
2. የልጁ/ጇ እድሜ ወር ጾታ/ ወ/ሴ ልጅ/ጇ ከወላጆቹ ጋር ነው የሚኖረው /አዎ/ አይደለም
3. የእናት ትምህርት ሁናቴ

   ሀ= ያልተማሩ ለ= የመጀመሪያ ደረጃ ትምህርትየተማሩ

   ሐ.=ሁለተኛ ደረጃ የተማሩ መ=ዲፕሎማና ከዚያ በላይ የተማሩ
4. የተቅማጥ የጊዜ ቆይታለ ቀን፡፡ከዚህ በፊት የትኩሳት ሁናቴ ለ_____ ቀን፡፡
5. የሰገራ ሁናቴ

   ሀ= ንፍጥ እና ደም የቀላቀል ለ= ንፍጥመሰል ነገርያ ለው ሐ= ሁለቱም የሌለው
6. ልጅዎ ከተጸዳዳ/ች በኋላ ይታጠባል/ለች

   ሀ=ሁሌም ለ= አልፎአልፎ ሐ= አይታጠብም
7. በአግባቡያልታጠቡና ያልበሰሉ አትክልት እና ፍራፍሬ ትመገባላችሁ

   ሀ=ሁሌም ለ= አልፎአልፎ ሐ= በፍጹምአንመገብም
8. የምትጠቀሙት የመጸዳጃ አይነት ምንድንነው

   ሀ= የግል ለ= የህዝብ
9. በምንያል የጊዜ ልዩነት መፀዳጃ ቤት ትጸዳዳላችሁ

   ሀ=ሁሌም ለ= በሳምንት አንዴ ሐ= በሳምንት ሁለቴ

   መ=በሳምንት ሶስቴ ሠ=በጭራሹ አንጠቀምም
10. የውሃ አጠቃቀም ምን ይመስላል

    ሀ= በማፍላት ሐ= እንዳለ
11. የልጅዎ ምግብ

    ሀ. =ሁል ጊዜትኩስ የሆነ ለ= አልፎ አልፎ ትኩስ ሐ= ብዙ ጊዜ የዋለ
12. የልጁ/ጇ የመጫወቻ ቦታ

    ሀ= ፅዳት ያለው ለ= ፅዳቱ የጓደለ
13. የልጅዎ ጥፍር በሚየድግበት ጊዜ ይቆርጣሉ;

    ሀ= ፅዳት ያለው ለ= ፅዳቱ የጓደለ
14. 1ከዚህ በፊት ስለግል ና አካባቢ ንጽህና ትምህርት አግኝተው ያውቃሉ? ሀ/አዎ ለ/ የለም
15. እጅዎትን ምግብ ከመመገብዎ በፊት እና ከሽንት ቤት መልስ ይታጠባሉ? ሀ. አዎ ለ. አልታጠብም

Appendix-III Data collection format for parasitological analysis

Lab. Code No	Sex	Age	Parasite	Single infection	Double Infections	Multiple infections	Direct Microscopy	Conc. Method	Remark

Appendix-IV Result report for parasitological examination

Code No. ____ Sex _____ Age _____

Address Woreda ___ Kebele _____ Tel. _____
Type of specimen _____ Appearance _____
Date of collection _____
Ova/parasite Positive_____ Negative _____ Parasite identified:
Direct method Concentration method
1. _____ _____
2. _____ _____
3. _____ _____

Remark:_____
_____
_____
Date Reported_____ Signature _____

Appendix-V Laboratory data collecting format

Code	sex	Age	pos. neg.	Date dd.mm .yr.	parasitic identified in stool sample				
					Gl	Ehdm	Al	Tt	Hn

Key.

Ehdm= *Entamoeba histolytica/dispar/moshkovisi*
Gl= *Giardia lamblia*
Al= *Ascaris lumbercoides*
Tt= *Trichuris trichuira*
Hn= *Hymenolepis nana*
Pos. positive **Neg**. negative

Declaration

I, the undersigned declare that thesis is my original work in partial fulfillment of the requirement for the master of general Biology. I also declare that it has never been presented in this or any other university and that all resource and materials in the proposal have duly acknowledged.

Student name Birhanu Megersa
Signature _____
Date of submission _____

This thesis has been submitted for examination with my approval as a university advisor.
Advisor Name _____
Signature _____
Date of submission _____