

COLLEGE OF NATURAL SCIENCES DEPARTMENT OF BIOLOGY

Prevalence and Risk Factors of Soil-Transmitted Helminths among School Children at Goro Primary School, South West Shewa, Ethiopia

By: Tigist Tiruneh

A Thesis Submitted to Department of Biology, College of Natural Sciences, Jimma University in Partial Fulfillment of the Requirements for the Degree of Masters of Science in Biology

> January 2020 Jimma, Ethiopia

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> Advisors: TsigeKetema(Ph.D.) Co-advisor: GeletaGeshere(MSc)

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Approved by:		
1. Advisor		
Name	_Signature	_Date
2. Co-advisor		
Name	_Signature	_Date
3. External Examiner		
Name	_Signature	_Date
4. Internal Examiner		
Name	_Signature	_Date
5. Department head		
Name	_Signature	Date

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List of Acronyms

-	AIDS	Acquired Immune Deficiency syndrome
-	FMOH	Federal Ministry of Health
-	IPI	Intestinal Parasitic Infection
-	MDA	Mass Drug Administration
-	SAC	School-Age Children
-	SPSS	Statistical Package for the Social Science
-	SSA	Sub-Sahara Africa
-	STH	Soil-Transmitted Helminths
-	WHO	World Health Organization

Abstract

Soil-Transmitted Helminths (STHs), also known as Geohelminthes, are human parasitic nematodes thatneed soil contact for their egg development and become infectious. Soil-transmitted helminths are widespread in developing countries. In Ethiopia, the prevalence of STHs varies in different parts of the country. However, the prevalence of STHs infection in the present study area was not studied yet. The objective of this study was, therefore, to determine the prevalence of STH infections and associated risk factors among school children at Goro PrimarySchoolfromApril to June 2019. A cross-sectional study design was employed. Direct wet mount and formol- ether concentration techniques were utilized to detect the STHs in stool samples. The stool samples were collected from all study subjects with labeled, clean and leak-proof stool cup. Then the stool samples were examined immediately. Moreover, community and individual-level risk factors associated with STHs infection were assessed using a semi-structured questionnaire. Accordingly, the overall prevalence of soil-transmitted helminths infections observed in the collected fecal sample was 15.8% (n=61/387) and the most abundant STH parasite washookworms (n=35/61, 57.4%) followed byAscarislumbricoides(n=22/61, 36.06%) and double infection (n=4/61, 6.5%). Factors independently associated with soil-transmitted helminth infections were being illiterate (AOR= 2.3, 95% CI: 1.1-4.8, P= 0.021), lack of habit of wearing shoes (AOR= 4.1, 95% CI:2.0-8.5, P<0.001), lack of frequent hand washing before meal (AOR= 2.3, 95% CI:1.2-4.5, P= 0.019), use of unprotected drinking water (AOR= 39, CI:3.9-393, P= 0.002), and presence of dirt in fingernails (AOR= 3.5, 95%) CI:1.8-6.9, P<0.001) were significantly associated with infection of STHs is (P<0.05). In linewith World Health Organization (WHO) classification, STH infection observed in the study area classified into the low-risk area group calling for none or case-by-case treatment. Thus, enhancing awareness of the community in the study area on how to keep personal hygiene and environmental sanitation quite important to keep the burden on a controllable level.

Keywords: Ascarislumbricoides, Ethiopia, Hookworm, Prevalence, Soil-transmitted helminths, Trichuristrichiura

1. Introduction

1.1 Background

Soil-Transmitted Helminthes is also known as Geohelminthes are human parasitic nematodes that need soil contact for their egg development. There are five species of human STHs: roundworms (*Ascarislumbricoides*), whipworms (*Trichuristrichiura*), hookworms (*Necatoramericanus*and*Ancylostomaduodenale*) and threadworms (*Strongyloidesstercoralis*) (Brooker*et al.*, 2006). The STHs are a group of parasitic nematode worms causing human infection through contact with parasite eggs or larvae that thrive in the warm and moist soil of the world's tropical and subtropical countries. Adult worms of STHs live for years in the human gastrointestinal tract and produce numerous eggs in their lifetime (Bethony*et al.*, 2006; Anteneh*et al.*, 2008).

The disease, particularly morbidity associated with high worm burden of STHs infections, is causingsignificant public health problem in developing countries with deprived of personal hygiene and environmental sanitation. The transmission of STHs is higher in countries where poverty, poor nutrition, inadequate sanitation, lack of clean drinkingwater and minimal health care prevailed (Bethony*et al.*, 2006). Other possible risk factors include overcrowding, poverty, poor residential infrastructure, food prepared under unhygienic conditions, failure to put on footwear, presence of ditches that retain water/sewage around the house, lack of sanitation, improper sanitary habits (not washing the hands after using the toilets), geophagia (eating of sand) and poor socio-economic status (Bethony*et al.*, 2006; Nwokeocha*et al.*,2018). The highest rates of infection are often in children between the ages of 5 and 15 years due to their habits of playing in contact with soil (WHO, 2004). STHs infections are known to cause great morbidity in endemic areas. Effects of STH infections usually result in malnutrition, iron deficiency anemia, malabsorption syndrome, intestinal obstruction, chronic dysentery, rectal prolapse, respiratory complications, poor weight gain, impaired cognition in children (effect on school performance) and also hinder economic development (Bethony*et al.*, 2006).

Nearly 2 billion people of the globe are infected with at least one STHs species of which *Ascarislubricoides* accounted for about 1.2 billionand *Trichuris trichiura*0.8 billion cases (Parija*et al.*, 2017; WHO, 2012). Hookworm is also a common STHs infection in Sub-Saharan Africa (SSA). It is estimated that 198 million people are infected with hookworm in this region (29% of the region's population), including 40–50 million school-aged children. Hookworms are found in SSA, with the former representing the predominant hookworm species (Hotez and Kamath 2009). In Ethiopia, *Necatoramericanus* is more common than *Ancylostomaduodenale*, and hookworm infections are most prevalent in communities located between 800 and 1200m altitude (Anteneh*et al.*,2008). *Strongyloidesstercoralis* is patchily distributed in tropical areas particularly in Sub Saharan Africa and Southeast Asia. Strongyloidiasis estimated 200 million infected worldwide. In Ethiopia, Strongyloidiasis was found to be

more common in AIDS patients than non-AIDS(Anteneh*et al.*, 2008). The parasite is not highly prevalent in this country, but it occurs in the same geographic areas and communities as hookworm infection (Anteneh*et al.*, 2008).

In Ethiopia, numerous epidemiological surveys of STHs were conducted and varying infection rates were reported from different regions and communities. According to de Silva (2003), the prevalence of *Ascarislumbricoides* and *Trichuristrichiura* in Ethiopia ranges25-44.9% and the prevalence of hookworms in the country are 5-24.5%. The low economic standard, poor sanitation and ignorance of simple health promotion practices favor the wide distribution of intestinal helminths in Ethiopia (Kloss and Zein, 1993). Compared to the end of rainy seasonsSchool-Age Children (SAC) were shown to have a marginally higher prevalence of both *Trichuristrichiura* and hookworms, but relatively uniform *Ascarislumbricoides* infections during dry seasons (Mekonnen*et al.*, 2019). The country is fighting against the STHs by using only Mass Drug Administration (MDA) once or twice a year based on the prevalence of the disease. The MDA is underway for all school children in target schools using ant-helminthic drugs (albendazole or mebendazole) (WHO, 2002). The WHO recommendations on how frequently to deliver targeted treatment to high-risk groups in different endemic situations have recently been revised.

1.2. Statement of the Problem

Soil transmitted helminth infections remaina significant public health problem, particularly in developing countries. According to WHO (2012), the STHs of major concern to humans are *Ascarislumbricoides*, *Trichuristrichiura*, *N. americanus*, and *A. duodenale*. The latest estimates indicate that more than 2 billion people in the world are infected with at least one species of STHs (due to*Ascarislumbricoides*: 1 billion, *Trichuristrichiura*: 800million and hookworm: 740million) and 4 billion are at risk of acquiring the infections. The highest prevalence occurs in areas where sanitation is inadequate and water supplies are unsafe (WHO, 2012).

According to the FMOH (2016) both schistosomiasis and STHs are endemic in Ethiopia and represent significant health burdens. Soil-transmitted helminths infections are widely distributed throughout the country. The number of people living in STHs endemic areas is estimated at 81 million, which is comprised of 9.1 million pre-school-aged children, 25.3 million school-aged children and 44.6 million adults. The number of individuals living in areas qualifying for STHs Mass Drug Administration is 56.7 million, comprised of 4.6 million pre-school children, 17.7 million school-age children, and 31.32 million adults (FMOH, 2016).

According to a report by WHO (2012), the number of children who received preventive chemotherapy every year was only 200 million School-Age Children (SAC) out of 600 million SAC need to receive the treatment in 2010. Large-scale and successful control activities implemented during 2001–2010 demonstrate the feasibility of large-scale deworming, and these experiences have informed the development of tools to facilitate the work of control managers. The limitations of the national program to fight against the STHs include; lack of information about the prevalence of STHs in all district/localities, targeting only school children, lack of consistency and reliance only

on MDA (WHO, 2012). Therefore, the aim of this study was to contribute to providing information about the status of STHs in the study area which is not known yet.

1.3. Objectives of the study

1.3.1. General Objective

• To assess the prevalence of STHs infections and associated risk factors among school children at Goro PrimarySchool.

1.3.2. Specific Objectives

- To determine the prevalence of STHs infections among school children at Goro Primary School.
- To describe socio-demographic factors associated with the infections of STHs.
- To identify associated risk factors of STHs infections among the school children
- To identify the major STH parasite species in the study area.

1.4. Significance of the Study

School-age children are at higher risk of acquiring STHs infections and the main target of the national intervention focused on this group. The goal of this study was to determine the prevalence and risk factors of STHs among Goro Primary School students where such studies not conducted yet in the area. Identification of risk factors associated with STHs infections is vital to guide policymakers in designing a more focused preventive approach to control the disease. Therefore, the current study was conducted to determine the prevalence of STH infections and associated determinants among school children in Goro district, Southwest Shewa, Central Ethiopia.

2. Literature Review

2.1. Soil-Transmitted Helminths Situation in Ethiopia

Soil-transmitted helminth parasite infections are widely distributed throughout the tropics and sub-tropics. Recent estimates suggest that *Ascarislumbricoides* infect 1.221 billion people, *Trichuristrichuria* 795 million, and hookworms 740 million worldwide. The greatest numbers of STH parasite infections occur in tropical and sub-tropical regions of Asia, especially China, India, and southeast Asia, as well as Sub-Saharan Africa (De Silva *et al.*, 2003).

Soil-transmitted helminths have a high prevalence rate globally, especially in those societies living in impoverished conditions. In Ethiopia also, the disease affects millions of people as a result of several risk factors (Samuel, 2015). The most common STHs which found worldwide are *Ascarislumbricoides*, *Trichuristrichiura*, and the hookworms, (*N. americanus* and *A. duodenale*). Ethiopia has the second-highest burden rank of *Ascarislumbricoides* in SSA: 26 million people are infected, which covers 15% of the overall burden in SSA (Hotez and Kamath, 2009). And there are 21 million people infected with *Trichuristrichiura*, which accounts for 13% of the disease burden in SSA (Hotez and Kamath, 2009). It is estimated that there are 11 million people infected with hookworm in the country. Which makes the country to have the third highest burden rank in SSA (Anteneh*et al.*, 2008). *N. americanus* is more common than *A. duodenale* in the country, and hookworm infections are the most prevalent in communities located between 800 and 1200m altitude (Anteneh*et al.*, 2008).

In Ethiopia, like in other developing countries, intestinal parasitic infections are widely distributed. Several studies indicated that the prevalence of STHs infections washigh in the lower altitudes. Ethiopia has one of the lowest qualities of drinking water supply and latrine coverage in the world (Mengistu*et al.*, 2007). So the distribution and prevalence of various species of intestinal parasites differ from region to region because of several environmental, social and geographical and other factors (Haftu*et al.*, 2014).

In Ethiopia, numerous epidemiological surveys of STHs were conducted and varying infection rates were reported from different regions and communities (Mengistu*et al.*, 2007). Ethiopia is estimated to have the second-highest burden in terms of ascariasis, and the third-highest burden of hookworm. Infection like trichuriasis is also common. A third of Ethiopians are infected with ascariasis, one quarter is infected with trichuriasis and one in eight Ethiopians lives with hookworm (Deribe*et al.*, 2012).

2.2. Soil-Transmitted Helminths Prevention and Control Measures

Control measures such as school-based deworming programs, Mass Drug Administration (MDA), using antiparasitic therapy have been described as efficient, cost-effective and safe strategies for prevention and control

of parasitic infections in schoolchildren. The MDA is underway as nationalcontrol and elimination program (WHO, 2004 and Abera*et al.*, 2013). The strategy for control of soil-transmitted helminth infections is to control morbidity through the periodic treatment of at-risk people living in endemic areas. People at risk are:preschool children, school-age children, women of reproductive age (including pregnant women in the second and third trimesters and breastfeeding women) and adults in certain high-risk occupations such as tea-pickers or miners (WHO, 2019).

However, the main challenge of the program include: budget constrain, re-infection and reliance on MDA. Therefore it is important to integrate other programs such as health promotion. For instance, information dissemination on the advantage of washing hands after defecation and proper use of latrine should be taken into account to alleviate the problem (Tefera*et al.*, 2017). Increasing the target group will also enhance the control intervention. By promoting adults to receive essential anthelmintic drugs (albendazole, levamisole, mebendazole or pyrantel) in a specified interval may also contribute to reducingthe diseases burden. The drugs are safe even when given to uninfected people, and thus there is no need for individual screening (WHO, 2004).

The WHO recommendations on how frequently to deliver targeted treatment to high-risk groups in different endemic situations have recently been revised (WHO, 2002). The strategy recommended by WHO to control morbidity from STHs (defined as the elimination of infections of moderate and high intensity) involves the periodic administration of anti-helminthic medicines (mainly single-dose albendazole (400 mg) and mebendazole (500 mg)) to the populations at risk of the disease. The recommended treatment schedule of once or twice-annual administration is determined by the initial prevalence of infection with any **STHs** (Ascarislumbricoides, Trichuristrichiura or hookworms (N. americanus and A. duodenale) among SAC. The aim is to reduce and maintain low levels of infections and thus protect individuals at risk from morbidity caused by STHs (WHO, 2012).

Outside the developing world, the 20th century saw dramatic reductions in the prevalence of infection as a result of improvements in living standards and specific control programs. Improved sanitation is aimed at controlling transmission by reducing soil and water contamination (Hotez and Kamath, 2009). Sanitation is the only definitive intervention to eliminate STHs infections but should cover a high percentage of the population to be effective (Hotez*et al.*, 2006). Health education is aimed at reducing transmission and re-infection by encouraging healthy behaviors. For STHs infections, 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 in transmission. Health education can be provided simply and economically and presents no contraindications or risks. With this perspective, it is reasonable to include this component in all helminths control

programs (Hotez*et al.*, 2006). The survey of intestinal helminths and risk factors in the primary school population provides epidemiological information to design effective control programs (Abera*et al.*, 2013).

2.3. Prevalence and Risk Factors of STHs Infections

Soil-transmitted helminths are widely distributed in developing countries, and in Ethiopia the prevalence of STHs varies in different parts of the country (Tefera*et al.*, 2017). The STHs infections are distributed very widely throughout the country. The number of people living in STHs endemic areas is estimated at 81 million (FMOH, 2016). Nearly half of the school children examined were infected with one or more STHs species (Tefera*et al.*, 2017). Compared to the end of rainy seasons SAC wasshown to have a marginally higher prevalence of both *Trichuristrichiura* and hookworms, but relatively uniform *Ascarislumbricoides* infections during dry seasons. Overall, any STH infection was less prevalent in the rainy season compared to the dry season(Mekonnen*et al.*, 2019).

Those students who use latrine sometimes had a significantly higher risk to acquire *T. trichiura*, than those who use always. Students who did not wash their hands after defecation were three times more likely to develop*A*. *lumbricoides* infection than those who washed their hands after defecation (Haftu*et al.*, 2014). According to Haftu*et al.*(2014), the factors strongly associated with intestinal parasites infections werelow educational level of mothers/guardians of children, regular practice of hand washing before feeding of the study participant, and fingernail status of the children.

The major risk factor for high prevalence of STHs include; poor sanitation and hygiene, school-age children are typically at increased due to their habits of play on the ground or contact with soil (WHO, 2002). Soil-transmitted helminths depend on environments contaminated with egg-carrying fecesfor transmission. Consequently, helminths are intimately associated with poverty, poor sanitation, and lack of clean water (Samuel *et al.*, 2017).

The prevalence and distribution of intestinal helminthes among primary schools varied according to the children residence. The overall prevalence was significantly higher in rural schoolchildren than in urban schoolchildren (Abera*et al.*, 2013). There were also huge differences in the prevalence of intestinal helminth infections among rural schools, which ranged from 48.3% to 80%. These differences might be attributed to local risk factors (Abera*et al.*, 2013). The odds of having intestinal helminths among children who eat unwashed or undercooked vegetables in the stool of the children were about six times higher than those who eat washed or cooked vegetables (Wale *et al.*, 2014).

Both host-specific and environmental factors have been identified that may affect the risk of acquiring or harboring heavy intensity helminths infection (Hotez*et al.*, 2006). Specific occupations and behaviors influence the prevalence and intensity of STHs infections. Consequently helminthes 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 helminth infections. Adequate warm and moisture are key features for each of the STHs (Hotez*et al.*, 2006).

2.4 Life cycle of Soil Transmitted Helminths

2.4.1 Life cycle of roundworms (Ascaris lumbricoides)

Adult roundworms live in the lumen of the small intestine. A female Ascaris worm may produce up to 200,000 eggs per day, which are passed with the faeces. Fertile eggs embryonate and become infective after 18 days to several weeks, depending on the environmental conditions (optimum: moist, warm, shaded soil). After infective eggs are swallowed, the larvae hatch, invade the intestinal mucosa, and are carried via the portal, then systemic circulation to the lungs. The larvae mature further in the lungs (10 to 14 days), penetrate the alveolar walls, ascend the bronchial tree to the throat, and are swallowed. Upon reaching the small intestine, they develop into adult worms. Ittakesbetween 2 and 3 months from ingestion of the infective eggs for adult female. Adult worms can live 1 to 2 years (Helbig *et al.*,2012).

2.4.2 The life cycles of Hookworms (Ancylostoma duodenale and Necator americanus)

Eggs are passed in the stool, and under favorable conditions (moisture, warmth, shade), larvae hatch in 1 to 2 days. The released rhabditiform larvae grow in the faeces and/or the soil, and after 5 to 10 days they become filariform (third-stage) larvae that are infective. These infective larvae can survive 3 to 4 weeks in favorable environmental conditions. On contact with the human host, the larvae penetrate the skin and are carried through the veins to the heart and then to the lungs. They penetrate into the pulmonary alveoli, ascend the bronchial tree to the pharynx, and are swallowed. The larvae reach the small intestine, where they reside and mature into adults. Adult worms live in the lumen of the small intestine, where they attach to the intestinal wall with resultant blood loss by the host. Most adult worms are eliminated in 1 to 2 years, but research findings have recorded worms several years old (Anteneh *et al.*, 2008).

2.4.3 Life cycle of whipworms (Trichuris trichiura)

Unembryonated eggs are passed with the stool. In the soil, the eggs develop into a 2cell stage, an advanced cleavage stage, and then embryonate, thus, eggs become infective in 15 to 30 days. After ingestion (soil-contaminated hands or food), the eggs hatch in the small intestine, and release larvae that mature and establish themselves as adults in the colon. The adult worms live in the caecum and ascending colon. The adult worms are fixed in that location, with the anterior portions threaded into the mucosa. The females begin to mature 60 to 70 days after infection. Female worms in thecaecum shed between 3,000 and 20,000 eggs per day. The life span of the adults is about 1 year (Helbig *et al.*, 2012).

2.4.4 Life cycle of Strongyloides stercoralis

This parasite undertakes two types of life cycles namely external and internal autoinfection patterns. In the former, female adult worms produces eggs which hatch in the bowel and most of them are passed in the faeces as non-infective rhabditiform larvae ready to penetrate exposed skin. Small proportions of larvae in the bowel however, penetrate the large intestinal mucosa or the perianal skin and by the process of autoinfection perpetrate infection indefinitely. In some circumstances the external cycle can be omitted o Filariform larvae develop internally and reinvade bowel or skin, enter circulation, migrate to lungs, penetrate alveoli and travel up trachea and swallowed and reach intestine (Helbig *et al.*, 2012).

3. Materials and Methods

3.1 Study Area

GoroPrimary School located in GoroWeredafound in Southwest Shewa Zone, Oromia Regional State, Ethiopia. It is located 135kms away from Addis Ababa to thesouthwestern Ethiopia, and 21kms from Welisotown.TheWereda is bordered on the south by Gurage Zone(SNNP), the North by WenchiWereda, on the east WelisoWereda and on the west AmeyyaWereda. The livelihood of the inhabitants mainly depends on subsistence farming, withteff and maize being commonly produced cereal. The average altitude of the area is 1660-1906meter above sea level, with average temperature ranging from 9°C to 30°C. The area soil is characterized by 30% lime, 60% clay and 10% silt. The average annual rainfall is 800-1600mm. There are four health centers, four private clinics and nineteen health posts.

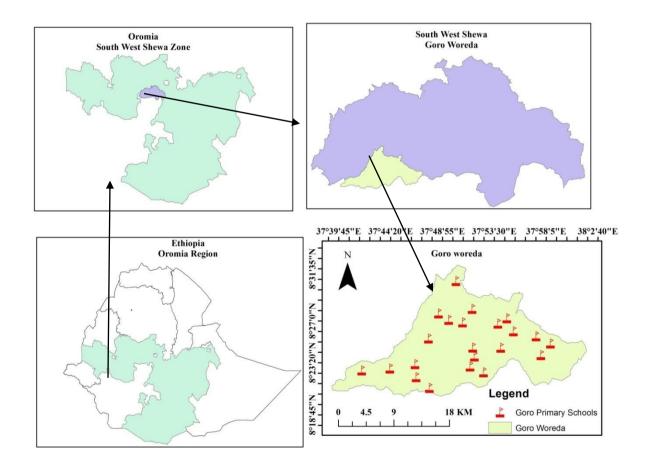


Figure1.Map of Study Area from Ethio-GIS

3.2 Study Design and Period

School age children based cross-sectional study design was conducted to determine prevalence and risk factors of soil transmitted helminthes from April to June 2019 at Goro Primary School, GoroWereda, and South-West Shewa.

3.3 Population

3.3.1 Source Population

All school children inGoroWereda, South-West Shewa, were the source population.

3.3.2. Study population

Children enrolled at Goro Primary School in the 2018/19 Academic Year were the study population. The total population of grade 1-8 students enrolled during the 2018/19 academic year in Goro Primary School was 1242, of these, 628 were males and 614 were females

3.4. Sample Size Determination and Sampling Technique

3.4.1. Sample size determination

The sample size (n) was determined using single population proportion (Daniel, 1999).

$$n = Z_{\alpha/2}^{2*}P^{*}(1-P)/d^{2} = (1.96)^{2*}0.5^{*}(1-0.5)/(0.05)^{2} = 384$$

Where; Z $\alpha/2$ is the critical value of the Normal distribution at $\alpha/2$ at 95%=1.96, P is prevalence= 50% (considering other highest report from the country) and d² is the margin of error=±0.05. By adding 5% non-response rate, the final sample size was403 students.

3.4.2. Sampling technique

The school was selected purposively. To select the study subjects, the students were first stratified according to their educational level (Grade 1 to Grade 8). Then, the study subjects were selected using systematic random sampling by using class attendance sheets the sampling frame. Brief description of students enrolled and not enrolled in the study is expressed in the following figure (Table 1).

School Name	Grades	Student population		Sample population			
		М	F	Т	М	F	Т
	1	102	81	183	30	30	60
	2	105	88	193	34	29	63
	3	92	77	169	17	38	55
Goro Primary	4	52	77	129	25	17	42
School	1-4	351	323	674	106	114	220
	5	98	105	203	25	41	66
	6	68	80	148	27	21	48
	7	65	46	111	18	18	36
	8	46	60	106	18	15	33
	5-8	277	291	568	88	95	183
Total	1-8	628	614	1242	194	209	403

Table 1Total population of the school and sample size of the study participants 2019

3.5. Eligibility

Inclusion criteria

All selected Goro primary school children, who consented to participate in the study and attended the school during the study period.

Exclusion criteria

Students who have taken any anti-helminthic drug before two weeks of the study period and students who refused to participate were not included in the study.

3.6. Study variables

The dependent variable was detection of geohelminthic parasitic infection in stool samples and the independent variables were socio-demographic factors like: age, sex, residence, mothers level of education, and other associated factors like:shoes wearing habit, Use latrine or not, Fingers nail status of children, water source, hand washing habit before meal, hand washing habit after defecation and play with soil.

3.7. Data collection

3.7.1. Questionnaire data

Socio-demographic characteristics and associated risk factors were collected by trained data collectors using pretested questionnaires from selected students under monitoring of the Principal Investigator (PI). The questionnaire was first prepared in English and then translated into the local language, AfaanOromoo, which was administered by researcher.

3.7.2. Laboratory data

Stool sample were collected from all volunteer study subjects with labeled, clean and leak proof stool cup. Study participantswere informed to collect thumb sized stool samples of their own and to use tissue paper to protect possible contamination. Laboratory procedures were performed in Goro Health Center by two laboratory technologists.

Direct Wet Mount Microscopic Examinations for Detection of Intestinal parasites

A drop of saline and a small amount of faces were placed on the microscopic slide using applicator stick, then mixed and covered by cover slip. Then, the samples were examined microscopically under the light microscope at a magnification power of 100x and 400x. The remaining portion of the stool samples (\sim 1 g) werepreserved in 10% formalin to repeat the tests by concentration technique when the result of direct wet mount turned negative (Tadesse*et al.*, 2008).

Formol-Ether Concentration Technique for Stool Sample Examination

The formalin preserved specimen was thoroughly stirred and a sufficient quantity was strained through gauze into a 15mL pointed centrifuge tube to get the desired amount of sediment. Then saline was mixed and centrifuged at 2000-2500 rpm for 1 minute. The supernatant was decanted and washed again with tape water. About 10mL of 10 percent formalin was added to the sediment and mixed thoroughly. Then, 3mL of ether was added and shacked vigorously in an inverted position for 30 seconds. The resulting solution was centrifuged at 1500 rpm for about 1 minute, and four layers were produced. The three top layers were decanted carefully, and adhering debris were removed from the top with a cotton swab. Finally, the samples were examined microscopically at under high power magnification(Tadesse*et al.*, 2008).

3.8. Data quality control

- Data collectors were trained prior to data collection on questionnaire contents, sample collection and aim of the study.
- All collected data was rechecked by the principal investigator for completeness.

- The questionnaire and laboratory materials were pretested in the randomly selected students on 5% of total sample size.Pre-test was conducted by randomly selected students on 5% of total sample size. Possible adjustment on the data collection tool and the data collection procedure will be made during the pre-test.
- Standard operating procedures were used for specimen collection and processing for maintaining a good quality study.

3.9. Data management and analysis

The questionnaire was checked for completeness, coded and entered into excel and cleaned. It was exported to and analyzed using SPSS (statistical package for social sciences) version 20.0 statistical software. Descriptive statistics was utilized to summarize the socio-demographic profile of the study participants. Bivariate and multivariate logistic regression models wereused to determine the association between dependent variable and independent variables. P-Value 0.05 was considered as statically significant.

3.10. Ethical considerations

Ethical approval was obtained from ethical review committee of College of Natural Sciences, Jimma University. Informal verbal consent was obtained from students' parent/guardians, class teacher and directors. The students' privacy during the interview and stool collection was maintained and the data obtained from them was strictly kept confidential. All individuals with positive geo-helminthic parasitic infection were treated with the standard regimen for free in collaborating with GoroWereda Health center.

4. Results

4.1 Socio-Demographic Characteristics of the Study Participants

Out of 403 randomly selected students, 387 (96.3%) were volunteer to participate in the study and provided stool samples. The largest number of study participants, 268(70%) were from age groups of 11–16 years and the remaining 119(30%) were found in age range from 6-10 years. The pupils consisted of 188 (48.6%) were males and 199 (51.4%) females. The number of pupils seen in school with regard to gender was shown below. Out of these respondents 185 (47.8%) were urban residents and 202(52.2%) were live in the rural area. Regarding the grade level, 215(55.6%) were in grades 1- 4 and 172 (44.4%) were in grades 5 - 8. Majority of the students household, 275(71.1%) had greater than five family size, and 112(28.9%) of them had less than five family size (Table 2).

Variable	Alternative	Frequency
		(%)
	6-10	119 (30.0)
Age	11-16	268 (70.0)
	Female	199(51.4)
Sex	Male	188(48.6)
	≤5	112(28.9)
Family numbers	>5	275(71.1)
	Urban	185(47.8)
Residence	Rural	202(52.2)
	1-4	215(55.6)
Grade	5-8	172(44.4)

Table 2Socio-demographic characteristics of the study participants, amongSchool age children of Goro Elementary

 School inGoroWereda, 2019

4.2 Prevalence of Soil Transmitted Helminths

Of the 387 stool specimens examined, prevalence of soil transmitted helminthic parasite was observed in 61(15.8%) school children, from which 34(17.1%) were females and 27 (14.4%) were males. With regards to specific age, school children in the age group of 6-10 years were more infected (18.5%) than those in age range from 11 to 15 years of the study subjects (14.5%) (Table3).

Table 3 Prevalence of STHs among school children of Goro Primary School in Goro Wereda 2019

Age and sex		Participants	No. of positive	Frequency
				(%)
Male	6-10	56	12	21.4
	11-16	132	15	11.4
	Sub-total	188	27	14.4
Female	6-10	60	10	16.7
	11-16	139	24	17.3
	Sub-total	199	34	17.1
Both sex	6-10	116	22	18.9
	11-16	271	39	14.4
Total		387	61	15.8

Out of the 61 STHs infected students, 57 were infected with either Hookwormsor*A. lumbricoides*, only 4 students had double helminthic infection (Hookworms and *A. lumbricoides*). Hence the prevalence of single and double infection by the STHs was 14.8% and 1.0%, respectively (Figure 3).

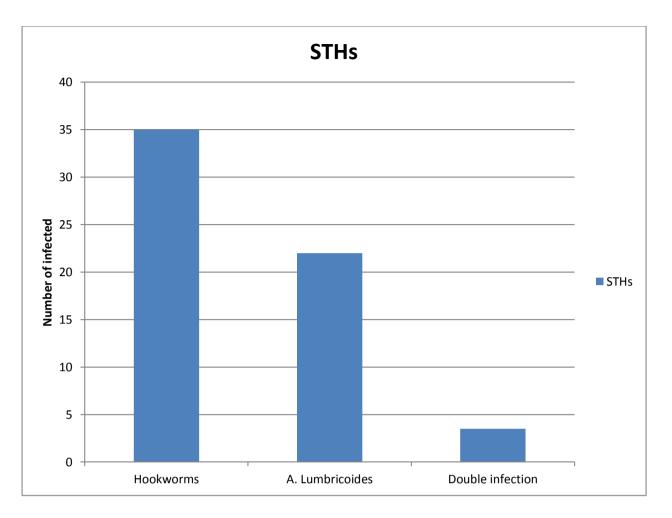


Figure2. Prevalence of STHs among School age children atGoro Elementary school in GoroWereda, South West Shewa, 2018/19

4. 3. Factors associated with STHs infections

Individual level characteristics includingsocio-demographic and associated risk factors such as age, sex, residence, religion, grade level and latrine availability were not significantly associated with infection of STHs (P>0.05). Other variables includingmother's educational level, shoe wearing habit, hand wash before meal, hand wash after toilet, drinking water source, presence of dirt in finger nails, and playing with soil were significantly associated with infection of STHs is (P<0.05) and considered as individual-level risk factors (Table4).

	Alternative	STHs Status		COR(95%CI)	P-value
Variable		Participants	Positive (%)		
		(%)			
Age	6-10	119 (30.7)	22(18.5)	1.4(0.8-2.5)	0.259
	11-15	268 (69.3)	39(14.5)	Ref	Ref
Sex	Female	199(51.4)	34(17.1)	1.2(0.7-2.1)	0.463
	Male	188(48.6)	27(14.4)	Ref	Ref
Mother's	Literate	254(65.6)	43(16.9)	Ref	Ref
Education	Illiterate	133(34.4)	18(13.5)	2.3(1.1-4.8)	0.021*
Family size	<u><</u> 5	112(28.9)	10(8.9)	1.3(0.7-2.4)	0.385
	>5	275(71.1)	51(18.6)	Ref	Ref
Residence	Urban	185(47.8)	26(14.1)	Ref	Ref
	Rural	202(52.2)	35(17.3)	1.3(0.7-2.2)	0.378
Grade	1-4	215(55.6)	40(18.6)	1.6(0.9-2.9)	0.088
	5-8	172(44.4)	21(12.2)	Ref	Ref
Latrine available	No	39(10.1)	10(25.6)	2.0(0.9-4.4)	0.079
	Yes	348(89.9)	51(14.7)	Ref	Ref
Shoe wearing	Sometimes	154(39.8)	46(29.9)	6.2(3.3-11.6)	P<0.001*
habit	Always	233(60.2)	15(6.4)	Ref	Ref
Hand wash before	Sometimes	96(24.8)	28(29.2)	3.2(1.8-5.7)	P<0.001*
meal	Always	291(75.2)	33(11.3)	Ref	Ref
Hand wash after	Not at all	152(39.3)	31(20.4)	5.6(1.9-16.5)	0.002*
toilet	Only with water	143(36.9)	26(18.2)	4.9(1.6-14.5)	0.004*
	With water & soap	92(26.8)	4(4.4)	Ref	Ref
Drinking water	Protected	382(98.7)	57(14.9)	Ref	Ref
source	Unprotected	5(1.3)	4(80.0)	22.8(2.5-207.8)	0.006*
Dirt in finger nail	No	221(57.1)	20(9.1)	Ref	Ref
	Yes	166(42.9)	41(24.7)	3.3(1.8-5.9)	P<0.001
Play with soil	No	210(54.3)	21(10.0)	Ref	Ref
	Yes	177(45.7)	40(22.6)	2.6(1.5-4.7)	0.001*

 Table 4 Prevalence of STHs and individual level associated risk factors amongSchool age children of Goro

 Elementary School inGoroWereda, 2019

NB: COR: Crude Odds Ratio; CI: Confidence Interval

Individual-level risk factors were further evaluated by multivariable analysis for their predictive potential. Accordingly, among these characteristics, family size, grade of children, availability of latrine, habit of hand washing after toilet, and playing with soil didn't show significant association (P>0.05) with STHs infection (Table 4). Prevalence of the STHs infections was significantly (P< 0.05) associated with shoe wearing habits in which children infrequently wearing shoeswere 3 times more likely to acquire the STH infection as compared to children who wear shoes always(AOR: 4.1, 95%CI: 2.0–8.5, P<0.001). The irregular hand washing before meal had also significant association with STH infection as compared to individuals regularly washing their handsbefore meal (AOR=2.3; 95%CI: 1.2-4.5; P = 0.019). Children infrequentlywashed their handsafter toilethad also significant higher risk to have STHs infection (COR=5.6; 95%CI: 1.9-16.5; P< 0.05), but didn't show significant association

in multivariable analysis (AOR=1.5; 95%CI: 0.4-5.2; P> 0.05)(Table 4).Mother's Educationhad also significant association with STH infection (COR=2.3; 95%CI: 1.1- 4.8; P< 0.05), but didn't show significant association in multivariable analysis (AOR=2.1; 95%CI: 0.90 - 4.6; P> 0.05). The STHs infection was 3 times higher (AOR: 3.5, 95% CI: 1.8–6.9) in children who had dirt in their finger nail as compared tothose whodid not. Drinking water from unprotected source had strong association with STHs infection in which individuals who uses of unprotected water are more likely to be STHs positive (AOR=39.0; 95%CI=3.9 - 393; P<0.05).STHs infection was significantly associated with frequent habit of playing with soil (COR: 2.6,95% CI:1.5–4.7; P<0.05), but didn't show significant association in multivariable analysis (AOR=1.5,95% CI: 0.7-3.2, P>0.05) (Table 5).

Variables		Participants (%)	Positive (%)	P value	AOR(95%CI)
Mother's	≤5	112(28.9)	10(8.9)	Ref	Ref
Education	>5	275(71.1)	51(18.6)	0.072	2.1(0.9-4.6)
Grade	1-4	215(55.6)	40(18.6)	0.841	1.0(0.4-2.0)
	5-8	172(44.4)	21(12.2)	Ref	Ref
Latrine available	No	39(10.1)	10(25.6)	0.426	1.4(0.6-3.6)
	Yes	348(89.9)	51(14.7)	Ref	Ref
Shoe wearing	Sometimes	154(39.8)	46(29.9)	0.000*	4.1(2.0-8.5)
habit	Always	233(60.2)	15(6.4)	Ref	Ref
Hand wash	Sometimes	96(24.8)	28(29.2)	0.019*	2.3(1.2-4.5)
before meal	Always	291(75.2)	33(11.3)	Ref	Ref
Hand wash after	Not at all	152(39.3)	31(20.4)	0.560	1.5(0.4-5.2)
toilet	Only with water	143(36.9)	26(18.2)	0.097	2.7(0.8-9.0)
	With water and soap	92(26.8)	4(4.4)	Ref	Ref
Drinking water	Protected	382(98.7)	57(14.9)	Ref	Ref
source	Unprotected	5(1.3)	4(80.0)	0.002*	39.0(3.9-393)
Dirt in finger	No	221(57.1)	20(9.1)	Ref	Ref
nail	Yes	166(42.9)	41(24.7)	0.000*	3.5(1.8-6.9)
Play with soil	No	210(54.3)	21(10.0)	Ref	Ref
	Yes	177(45.7)	40(22.6)	0.337	1.5(0.7-3.2)

NB: AOR: Adjusted Odds Ratio; CI: Confidence Interval; *significant at p < 0.05

5. Discussion

Due to differences with the risk factors atdifferent localities, environmental sanitation, culture of the community among regions, the prevalence and distribution of Intestinal Parasitic Infection(IPI) varies in Ethiopia (Birmekaetal., 2017). The study findings showed that STHs infections are still major public health concern among children in Goro district with 15.8% of participants infected with at least with one STH species. Thus out of the four STH species, hookworm infection was the most predominant (9.1%), followed by A. lumbricoides infection (5.7%). There were no infections observed ue to T. trichiura and S. stercoralisspecies. There were double infections 4 (1.0%) with Hookworm and A.lumbricoides. The prevalence of the STHs infection was relatively low. According to WHO (2012) STH endemic areas classifications, thereare three categories in line with application of Mass Drug Administration (MDA): 1. Hightransmission (where prevalence is > 50%), 2. moderate transmission (where prevalence is between 20% - 50%), and 3. low transmission (where prevalence is < 20%). Accordingly, the study area would beclassified into the low risk area group calling for none (case-by-case treatment). In fact in the study areamass chemotherapy which is done twice a year. The deworming programme only targets two- five years children hence leaving the older children vulnerable to infection. As children interact with the other children, their playful behavior exposes them to contaminated soil, water, and food hence the risk of re-infection is high in older children. In the study area 2018/19,5405 of children between two – five years old received helminthic drug in the deworming campaign. MDAis a means of delivering safe and inexpensive essential medicines based on the principles of preventive chemotherapy, where populations or sub-populations are offered treatment without individual diagnosis (WHO, 2004).

This study indicated that Hookworms (9.1%), *A. lumbricoides* (5.7%), were STHs infecting school children of Goro Primary School. The prevalence of hookworm in the current study was significantly higher than other intestinal helminthes which might be due to the fact that most infected children are rural and they wear shoes sometimes and played or walked over loamy soils and cultivated fields without shoe. As a result, children walking barefoot on the soil contaminated with faecal matter could be exposed to the infective larval stages of the hook worm parasite (Abera*et al.*, 2013). Moreover, study conducted in Gurage Zone also reported high prevalence of hookworms than other STHs. This study area, Gurage Zone, is adjacent to GoroWereda with almost the same soil type and environment (Birmeka*et al.*, 2017; Weldesenbet*et al.*, 2019).

The current findings indicated that the prevalence of STHs (15.8%), was relatively low compared to a previous study carried out on schoolchildren in other place but the same country,Jimma town, where the prevalence of STHs was (45.6%), Durbete Town (54.9%) and Tilili town(44.2%)(Abera and Nibret 2014; Alelign*et al.*, 2015; Tefera*et al.*, 2017).In contrast, the overall prevalence was higher than the results from Gurage zone(9.5%), Ambo

Town (12.6%)andBabile Town (13.8%) with prevalence of STH (0.47%)(Weldesenbet*et al.*, 2019;Samuel *et al.*, 2017; Tefera*et al.*, 2015). The most common combination of STHs in this study is hookworms and *A. lumbricoide* which agrees with other report byAbera*et al* (2013). This is because Ascaris and hookworms are the most abundant and the main species that infect people (Ng'ang'a*et al.*, 2016; Samuel *et al.*, 2017; Tefera*et al.*, 2017; WHO, 2019).

Effects of high hookworm and Ascaris infections usually result in malnutrition, iron deficiency anemia, mal absorption syndrome, intestinal obstruction, respiratory complications, poor weight gain, impaired cognition in children (WHO, 2019; Bethony*et al.*, 2006).The study identified key predictors of STH in the children, which included associated risk factors, use of unprotected water, not washing hands before feeding, walk with barefoot and dirt in finger nail. The present study showed that the majority of students were not washing hands after toilet useand before feeding. This is probably due to low knowledge of children and their parents about the faecal-soil-oral transmission of the helminthes through their unwashed hands, or due to lack of hand washing facilities (Ahmed*et al.*, 2017).Hand washing is one of the important parameters which intervene with fecal-oral transmission of diseases. So, hand washing must be practiced well before meal and after toilet use.The overall prevalence of intestinal parasitosis is greater among 5-14 year olds. This is because at such age school children indicate changes in exposure to conditions that favor the transmission of the infective stages of most of the helminths; other investigators showed that hand washing habit of this age group is very poor (Curtis and Cairncross,2003).

The other factors associated with infection in this study werewalk with barefoot. This might be due to children more participated in agricultural activities to help their parents that may exposed to infective (Birmeka*et al.*, 2017). Hookworm infections were increased in children not wearing shoes. This indicated that wearing shoes might have great importance in protecting hookworm infection(Bethony*et al.*, 2006). Using unprotected water had a significant association with Ascaris infection. These result might indicated that using protected water and avoiding contamination and keeping self-hygiene could contribute to reduction of ascaris infection. This may be due to unprotected water are not usually covered, and infective stage of ascaris being introduced into the water through flood or sewage direct dumping of dirt into the water, hence, are susceptible to contamination with human and animal faeces containing infective stages which might expose the students to helminthic infection. On the other hand, spring water could be easily contaminated with flood water and fecal matter contained in dust carried by wind. This is similar to the study in Gurage Zone (Birmeka*et al.*, 2017). The present study showed thatdirt in finger nail was risk factors for *A. lumbricoides* infections which is similar to study done in Jimma Town (Bajiro*et al.*, 2018).

Limitation

This study has certain limitations that need to be considered while interpreting the results. Most of the questions were self-reported and therefore that cannot possible to obtaining accurate information. This study misses some personal questions such as household income, weight and height; this may missing some values and might have affected the power of the study.

6. Conclusion and Recommendation

6.1 Conclusion

This study ascertains that some STHs infections were detected among school children at Goro Primary School. The most common species was Hookworm followed by *A. lumbricoides*. The school children were at low risk of infection with STHs based on WHO (2012) prevalence classification, where prevalence observed was< 20% that needs case-by-case treatment. The risk factors associated with worm infections wereshoe wearing habit, lack of hand wash before meal, unprotected water sources and dirt in finger nails.

6.2 Recommendations

The findings of this study showed that low prevalence of STH infections. But it is important to further minimize the diseases burden in the study area. This finding suggests improvement in water resources and sanitation infrastructure in the area. This result supports the need for the establishment of a health program for the control of the helminths in these communities, through health and hygiene education. Also, good personal hygiene and environmental sanitation must be encouraged as well as wearing shoes, use of safe drinking water, trimming finger nail and use of latrines. Strengthening health education program to increase the knowledge, attitude and practice of Schoolchildren and parents on how STHs are transmitted and prevented. Measures including education on personal hygiene and environmental sanitation should be taken into account to reduce the prevalence. However, further studies should be done by increase the sample size to detect the remaining species and understand current status of the infection.

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Annex I- Information sheet

Name of principal investigator: TigistTiruneh Name of organization: Jima University

Back ground - Soil Transmitted Helminthes are also known as Geohelminthes are human parasitic nematodes which need soil contact for their egg development. This information sheet is prepared by investigator with main objective of identifying determinants of geohelminthic parasitic infection detected by using cross sectional detection method. The group includes principal investigator (MSc graduate in general Biology summer program) and advisers from Jima University, college of Natural Science Biology Department. Data collectors and supervisors are also part of the team collecting and reporting quality data for study.

Purpose this study is to identify prevalence and risk factors of geo-helminthic parasitic infection among primary school children. Those there is need for intensive and habitual health education for behavioural changes related to personal hygiene and mass treatment will be effective for control of soil transmitted intestinal parasitic in the concerned area.

Procedure-we kindly invite you to participate in this research with the aim to identify prevalence and risk factors of geohelminthic parasitic infection among school children in Goro. If you agree to participate in this study, you will need to understand and sign agreement form. You will be asked some questionnaires and only your answer will be filled on questionnaire sheet. Stool sample for the laboratory diagnosis will be collected. Your questionnaire data and sample will be used only for this study and coding will be used for your confidentiality. If the result of your sample will be positive for geohelminthic parasitic infection, you will be treated according to the national drug guideline.

Benefits –in this study you will get direct benefit for diagnosis and treatment of geohelminthic infection. You participation also has an input for identifying prevalence and risk factors of geohelminthic infection among school aged children.

Incentive-There are no incentives for participating in this study.

Confidentiality- the information that we will collect in this research will be kept confidential and stored in a file. Code numbers will be used instead of your name and the file will be kept in a key and lock, and will not be accessed by anyone except the principal investigator.

Right to refuse and withdraw- you have full right to refuse or withdraw from participation in this study. Your refusal will not compromise your service in the school or in the health facilities in any way at any time.

Annex II- Consent form

I have been informed about a study have read all the information stated in the introductory part and I have an opportunity to ask any question about the study. I got satisfactory answer for all of my questions. For this study I have been requested to give stool sample and give information for the study. I have full understood and give my consent to give the stool specimen and reply for questionnaires. It is therefore my willing to give my informed consent and cooperate at my will in the course of the conduct of the study.

Name of participant(parent) ------Date------Date-------

Name of data collector------Date------Date-------

Consent form (Afaan Oromoo)

Guca waligalte

Waa'ee qorannoo kanaa ilaalchisee hubannoon gahaan naaf kennamee jira. Wanta anaaf hingalles akkan gaafadhu carraan naaf laatamee deebiin quubsaan naaf kennamee jira. Kanaafuu odeeffannoo fi boobbaa akkan laadhuuf gaafatameen jira. Anis guutummaan guutuutti waa'ee qorannoo kanaa hubadhee boobbaa fi odeffannoo barbaachisu kenneenjira. Kunis kan ta'e fedhakootiin ta'uuisaa mallattoo kootiin nanmirkaneessa.

Maqaa Hirmaataa qorannoo (Gudiftu)_____Mallattoo____ Guyyaa _____

Maqaa Odeffanno funaanu ______Mallattoo_____ Guyyaa _____

Annex III – Questionnaire

Name	Kebele	_Gere/Gott	
Reg.No. /code			
Health centerHI	H leader name		
Lab. Result –Not seen			
_Yes (1) Ascarieslumb	ricoides		
(2) Trichuristrich	uira		
(3) Hookworm			
(4)Strongloidesste	rcoralis		
Name of Lab. Personnel (species ide	entified by)	Sign	_ Date//2019

Date__/_/2019

Socio-demographiccharacteristics

No.	Question	Response
101	Age	
102	Sex	(1) Male(2) Female
103	Number of family member	
104	Residence	(1) Urban(2) Rural
105	Grade level	

Odefanno Namootaa

Koodii	Kebele	Gere/gott	
--------	--------	-----------	--

Lakk	Gafii	Debii
101	Umuri	
102	Saala	Dhi.
		Du.
103	Lakkofsa misseensa maatii	
104	Bakka jireenya	Magaala Baadiyyaa
105	Kuta	

Risk factors

Q. NO.	Question		Response	
201	Mother education		(1) Literate	
			(2) Illiterate	
202	Presence of latrine		(1) Yes	
			(2) No	
203	Shoe wearing		(1) Yes	
			(2) No	
			(3) Some times	
204	Hand washing before feeding		(1) Yes	
			(2) No	
205	Hand washing after defecting		(1) No	
			(2) Water only	
			(3) Water & soap	
206	Source of drinking water		(1) Tap water	
			(2) River	
			(3) Hand dug well	
207	Dirt matter under the nail		(1) Yes	
			(2) No	
	Habit of play with soil		(1) Yes	
208			(2) No	
Name of o	data collector	Sign	Date/	/2019
Name of S	Supervisor	Sign	Date//20	019

Madda Rakko

Lakk	Gafii	Debii
201	Barnotahadha	Kanbaratte
		Kanhinbaranne
202	Manafincaaniqabdu	Еууее
		Lakki
203	Koopheenikaawwata	Еууее
		Lakki
		Yeerotokkotokko
204	Yeroomaranyaataduraharkkanidhiqataa	Еууее
		Lakki
205	Manafincaaniergafayyadamtebodahaarkkanidhiqataa	Lakki
		Bishaanqofaan
		Bishaanifsaamunaan
206	Maddabishaandhugaatikeeti	Bonbaa
		Laga
		Bartaa /paampiiharkaa
207	Xurriiqeenssakeessa	Еууее
		Lakkii
208	Biyyeennitaphataa	Еууее
		Lakkii

Maqaa odefanno funaane _____ Mallatto _____ Guyyaa __/__/2019.

Maqaa to'ataa _____ Mallatto _____ Guyyaa __/__/2019.

Annex IV- Stool testing Procedures

Reagents and Materials

- Microscopes
- Microscope slides and cover slips
- Stool cup
- Applicator sticks
- Beaker
- 15ml conical centrifuge tube
- Dropping bottles containing physiological normal saline (0.85% w/ v)
- Pasteur pipette
- Ether
- 10% formalin solution

Collection and handling of stool Specimen

The specimen containers should be leak-proof, clean, dry and free from traces of disinfectant and not contaminated with urine or feces. A large teaspoon amount of feces is adequate for about 10 ml of a fluid specimen. The stool specimen must arrive in the laboratory soon after they are collected.