PREVALENCE AND RISK FACTORS ASSOCIATED WITH INTESTINAL HELMINTHIC INFECTIONS WITH SPECIAL EMPHASIS TO *SCHISTOSOMA MANSONI* AMONG FISHERMEN AT LAKE HAWASSA, SOUTHERN ETHIOPIA

BY:- TADESSE MENJETTA (BSc)

A THESIS SUBMITTED TO JIMMA UNIVERSITY COLLEGE OF PUBLIC HEALTH AND MEDICAL SCIENCES, DEPARTMENT OF MEDICAL LABORATORY SCIENCES AND PATHOLOGY, IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR DEGREE OF MASTER OF SCIENCE IN MEDICAL PARASITOLOGY

JIMMA UNIVERSITY COLLEGE OF PUBLIC HEALTH AND MEDICAL SCIENCES, DEPARTMENT OF MEDICAL LABORATORY SCIENCES AND PATHOLOGY

PREVALENCE AND RISK FACTORS ASSOCIATED WITH INTESTINAL HELMINTHIC INFECTIONS WITH SPECIAL EMPHASIS TO *SCHISTOSOMA MANSONI* AMONG FISHERMEN AT LAKE HAWASSA, SOUTHERN ETHIOPIA

BY: - TADESSE MENJETTA (BSc)

ADVISORS: 1. SERKADIS DEBALKE (MSc)

2. DANIEL DANA (MSc)

NOVEMBER 2013

JIMMA, ETHIOPIA

Abstract

Background: Schistosomiasis and other intestinal helminthiasis are among the most common parasitic infections in developing countries and their impact on public health has been underestimated even if they cause considerable morbidity and mortality. Schistosomiasis like other neglected tropical diseases is a disease of poverty. It particularly affects agricultural and fishing populations. In Ethiopia, many surveys have shown that schistosomiasis and helminthes infections represent a major public health concern.

Objective: The objective of this study was to determine the magnitude of *Schistosoma mansoni* and other intestinal helminthes infections among fishermen at Lake Hawassa, Southern Ethiopia

Methods: A community based cross-sectional study was conducted from April to June 2013 in Hawassa, Southern Ethiopia. A total of 243 study subjects were included and systematic random sampling method was applied. Data on socio demographic features and other predisposing factors were collected by using semi structured questionnaires. Stool samples were collected and processed using wet mount, Kato-Katz and formol-ether concentration techniques.

Result: Of the total 243 stool samples examined, 169 were positive for one or more of intestinal helminthes with an overall prevalence rate of 69.55 %. The overall prevalence rate of *S.mansoni* was 29.22%. The other most frequent intestinal helminthes were: *A. lumbricoides* 99 (40.74%), *T. trichiura* 87 (35.80%) and hookworm species 14(5.76%). The prevalence of *S. mansoni* was associated with factors such as swimming, frequency of swimming and frequency of water contact.

Conclusion: The prevalence of *S. mansoni* infection observed in this study indicates that the fishermen could become a potential source of infection and therefore are responsible for parasite transmission. This study had also identified risk factors like habit of hand washing after defecation and before meal and shoe-wearing habits that are associated with helminthes infections other than *S. mansoni*. Therefore, therapeutic intervention and health education are needed.

Key words: *Schistosomiasis, Fishing, Fishermen, Intestinal helminthiasis*

Acknowledgement

First and foremost, my sincere and deepest gratitude goes to my advisors Ms. Serkadis Debalke and Mr. Daniel Dana for their unreserved assistance in giving me timely, constructive comments and relevant guidance during the development of this research thesis.

My genuine appreciation is expressed to Jimma University for its financial support and other resources to conduct this study.

I also thank the Hawassa College of Health Sciences, for provision of some reagents and materials necessary for this study and allowing me the laboratory during examination of my samples.

My sincere thanks go to Mr. Gemechu Tadesse staff at EHNRI, who provided me with the Kato-Katz kit.

My deepest gratitude also goes to the Hawassa Fishermen association for their cooperation during data collection.

Last, but not least, my sincere thanks go to study participants and those whose names are not mentioned but contributed to the fulfillment of this study.

Table of contents

List of tables

List of figures

List of acronyms

AOR	Adjusted Odds Ratio
CI	Confidence Interval
COR	Crude Odds Ratio
EPG	Egg Per Gram
Km	Kilo meter
MDGs	Millennium Development Goals
NTDs	Neglected Tropical Diseases
P-value	Probability value
SOPs	Standard Operating Procedures
SPSS	Statistical Package for Social Sciences
SNNPR	Southern Nations Nationalities and People Region
WHO	World Health Organization

CHAPTER ONE: INTRODUCTION

1.1 Background

Schistosomiasis and intestinal helminthiasis are the major infections in developing countries and their impact on public health has been underestimated even though they cause significant morbidity (Thiongo *et al.*, 2001). Worldwide, it is estimated that 3.5 billion people are affected, and 450 million are ill as a result of these infections, the majority being children (Ayalew *et al.*, 2011).

The distribution and prevalence of various species of intestinal helminthes differs from region to region because of several environmental, social and geographical factors (Legesse *et al*., 2004). Intestinal helminthes are more prevalent throughout the tropics, especially among poor communities. Records show increasing trends in helminthes infections, particularly in developing nations (Tadesse, 2005). Parasitic helminthes infections are the second most predominant causes of outpatient morbidity in Ethiopia (Mengistu *et al*., 2007).

Schistosomiasis is a trematode infection caused by *Schistosoma mansoni (S. mansoni), S.haematobium, S.japonicum, S.mekongi, and S.intercalatum* (Ross *et al*., 1997). It was first described in 1851 by Theodor Bilharz, after whom the disease was named bilharziasis (Risikat *et al*., 2012). The first published record of the causative agent was after the post-mortem discovery of the worm in the mesenteric veins of a patient in Kasr el Aini Hospital in Cairo by Theodor Bilharz (Aginya *et al*., 2008). It is one of the ten tropical diseases especially targeted for control by the World Health Organization (WHO) and World Bank, (Ross *et al*., 2002).

Schistosomiasis is prevalent in 75 countries of the world and there are globally about 500-600 million people at risk of infection and 200 million peoples are infected with schistosomiasis. Majority of the cases, about 85%, are found in 41 countries of Africa (Essa *et al*., 2012). *S.mansoni* infection is endemic in Africa, the Middle East, South America, and a few Caribbean countries (Harrison, 2008). Worldwide, it is the second important parasitic disease of public health besides malaria (Mazigo *et al*., 2010).

Transmission of *Schistosoma* species begins when cercariae penetrates the skin while a person is fishing, bathing, washing clothes, or engaged in agricultural work or different activities involving contact with water that is contaminated with human faeces and that contains the snail hosts of the parasites. Inside the snail host the parasite undergoes multiplication and develops to an infective cercarial stage (Cheesbrough, 2009). In Ethiopia, *S. mansoni* is transmitted by *Biomphalaria pfeifferi* (*B. pfeifferi*) and *B. sudanica* (Terefe *et al.*, 2011), (Yami *et al.*, 2010)

There are different factors that contribute for transmission of schistosomiasis. Including: migration of people, emergence of new foci, socio-economic status, sanitation, water supply patterns and level of faecal contamination of water source (Okpala *et al.*, 2004). Different water development projects including water conservation, irrigation, and hydroelectric power have contributed to the spread of *S. mansoni* and changes in its distribution (Essa *et al.*, 2012).

Schistosomiasis in human is both acute and chronic disease (Okpala *et al.*,2004). During penetration of cercariae, some previously exposed and unexposed persons experience an itching sensation called cercarial dermatitis (Lambertucci, 2010). Patients infected with *S. mansoni* develop granuloma around the egg and the shistosoma antigens in the intestine and liver. The granuloma is followed by fibrosis, and chronic inflammation in the liver, leading to portal hypertension, causing liver disease, ascites, and oesophagogastric haemorrhage (Essa *et al.*, 2012).

There are different measures to control schistosomiasis. These control measures include provision of latrines and introduction of effective sewage disposal systems, wearing of shoes (Okpala *et al.*, 2004). One strategy that is used to prevent this debilitating disease is the primary health care approach in relation to human behaviors like improper waste disposal (Essa *et al.*, 2012).

Control of schistosomiasis infection has gained much international interest when WHO recommended that control of schistosomiasis contributes to the achievement of the Millennium Development Goals (MDGs) (Dabo *et al.*, 2011). The WHO strategy for schistosomiasis control focuses on reducing disease through periodic, targeted treatment with praziquantel (Bigwan *et al.*, 2012). In May 2001, the World Health Assembly passed resolution 54.19 supported that regular treatment of high-risk groups to be as the best means of reducing morbidity and mortality (Handzel *et al.*,2003).

1.2 Statement of the problem

Parasitic worms adversely affect the health of humans in many parts of the world. They continue to be a public health problem globally (Dejenie *et al.,* 2009). Inadequate water, sanitation and hygiene are responsible for a major proportion of the burden of disease and death in developing countries (Ayalew *et al.*, 2011).

In many of the developing countries, helminthes are causes of chronic gut infection in humans, particularly by that of *Ascaris lumbricoides (A. lumbricoides)*, *Trichuris trichiura(T. trichiura)* and hookworms and the blood flukes, schistosomes. Helminthes infections as a whole can be viewed as main indicators of the sanitation level of a community (Dejenie *et al.,* 2009). Many reports illustrated that *A. lumbricoides* is the most prevalent intestinal parasite in different communities (Mengistu *et al.*, 2007).

The global burden of schistosomiasis, an estimated 85% is found in sub-Saharan Africa. Infection with helminthes is responsible for many deaths and morbidities. Among tropical parasitic diseases, the yearly estimated deaths of 200,000 are making schistosomiasis second only to malaria as a cause of mortality (Terefe *et al.*, 2011). In the vast majority of developing tropical and subtropical regions of the world, parasitic infections particularly those caused by helminthes and schistosomes constitute major public health and developmental challenges (Uneke 2010).

Schistosomiasis and helminthes infections give rise to much suffering and death and also they contribute the continuation of poverty by diminishing the work capacity and productivity of adults (WHO, 2011). Both schistosomiasis and helminthic infections tend to be highly aggregated in that a small percentage of infected persons have very high worm burdens (Handzel *et al.*, 2003). In Ethiopia intestinal parasitosis is prevalent because of low level of living standards, poor environmental sanitation and ignorance of simple health promoting factors (Ayalew *et al.*, 2011).

Schistosomiasis causes both public health and socioeconomic problems in developing countries including Ethiopia. The mortality due to the disease is low; usually less than 1%, but because of the chronic nature of the disease and the morbidity affects farmers, laborers and fishermen causing seriously socioeconomic development of the disadvantaged developing country (Erko *et al.*, 2009). Schistosomiasis particularly affects agricultural and fishing populations (Bigwan *et al*., 2012).

Schistosomiasis is mostly prevalent in sub-Sahara Africa, where not only it overlaps with other few low priority diseases but also high priority diseases such as HIV, malaria, and tuberculosis (Essa *et al.,* 2012). Although estimates of schistosomiasis highlight the large numbers infected, the disease often receives less attention by health care personnel, national governments and international agencies than it merits. This is partly because not everyone infected will become ill (Kabatereine *et al.,* 2004). Schistosomiasis is a so-called neglected tropical disease, because it primarily affects poor rural communities in developing countries (Abou-zeid *et al.*, 2012).

Health strategy for achievement of effective parasitic disease control programs demand knowledge of magnitude of the disease. Hence, the present study provides current information on prevalence and intensity of infections as well as on associated risk factors of *S.mansoni* and other intestinal helminthes infections among fishermen at Lake Hawassa, Southern Ethiopia.

CHAPTER TWO: LITERATURE REVIEW

A study conducted on schistosomiasis among farmers and fisherman in the West Part of Burkina Faso, the prevalence of *S. mansoni* and hookworm species among fishermen were 16.35% and 4.8 % respectively (Zongo *et al.*, 2008). According to the study conducted in Tanzania (Handzel *et al.*, 2003), showed that the prevalence of *S. mansoni* was associated with proximity to Lake Victoria.

In a survey of human schistosomiasis that was done among the residents of Ndinjor district of Langtang in Nigeria, prevalence of *S. mansoni* among fisherman was 8.3% (Nanvya *et al.,* 2011). Another study conducted in Kariba, Zimbabwe, on transmission of schistosomiasis among the subsistence and commercial fisherman the prevalence of *S. mansoni* was 12.5% and 26.3% respectively (Chimbari *et al.,* 2003).

A study done at two Egyptian villages (Gharbia Governorate) to determine the prevalence and morbidity of schistosomiasis among rural fishermen revealed the prevalence of *S. mansoni* 72.4% (El-Hawey *et al.,* 1995). An epidemiological cross sectional study of *S.mansoni* conducted in two fishing villages (Rhino Camp and Obongi) of Uganda showed a prevalence of 81.5% and excreted 100 to 500 *S.mansoni* eggs per gram (EPG) (Odongo-aginya *et al.*, 2002).

Another study conducted in Kigungu fishing village, situated along Lake Victoria in Entebbe, Uganda, showed the prevalence of *S.mansoni* to be 47.4% (Odongo-aginya *et al.*, 2011) and in Kitubulu and Bugonga fishing villages the prevalence infection rate was 40.3% with mean egg count of 187 eggs per gram(epg) and 32.4% with 126 epg respectively (Lakwo 1991). In Piida fishing community at Butiaba, along Lake Albert, Uganda, a study revealed that *S. mansoni* is highly endemic with an overall prevalence of 72%, a mean intensity of 419.4 eggs per gram (epg) of faeces (Kabatereine *et al.*,2004).

In a study done at lakeside and island communities in Lake Victoria, Uganda, prevalence of *S. mansoni* was found to be 40.8% (Kabatereine *et al.*, 2011). According to a study done among primary school children in Kenya near Lake Victoria, where the majority of the population are subsistence farmers and fishermen, showed the prevalence of *S. mansoni* infection to be 16.3% (Handzel *et al.,* 2003).

A study done in Mbita and its adjacent islands of Lake Victoria, western Kenya, the prevalence of *S.mansoni*, hookworms, *A. lumbricoides* and *T. trichiura* infection was 60.5%, 8.4%, 3.3% 1.6% respectively (Odiere *et al.,* 2012). A study conducted in South Indian fishing village the prevalence of *A. lumbricoides*, *T. trichiura* and hookworm was 91%, 72%, and 54% respectively (Naish *et al.*, 2004).

Intestinal parasitism has been widespread in Ethiopia. Parasitic helminthes infections are the second most predominant causes of outpatient morbidity in the country (Mengistu *et al.*, 2007). In Ethiopia, many surveys carried out on intestinal schistosomiasis and helminthiasis has shown that helminthes infections represent a major public health concern. Results of epidemiological studies in different parts of Ethiopia showed that there is wide dispersion of schistosomiasis cases (Legesse *et al.,* 2010). A review of literatures on prevalence of *S. mansoni* in Ethiopia shows that it ranges from less than 1% up to more than 90% (Essa *et al.,* 2012).

A study conducted among students at Gorgora, Northwest Ethiopia, a small road side town on the shore of Lake Tana where the peoples' economy depends on different occupations including farming and fishing, prevalence of *S. mansoni* was found to be 20.6% with mean intensity of infection 125 eggs per gram of feces. In the same study the prevalence of *A. lumbricoides*, *Hymenolepis nana (H.nana)*, *T. trichiura*, hookworm species, and *Strongyloides stercoralis (S. stercoralis)* was 18.8%, 6.7%, 3.6 %, 2.2 %, and 1.4% respectively (Essa *et al.*, 2012).

According to a study that was done in Northern Gondar (Ayalew *et al.*, 2011), the prevalence of *A. lumbricoides*, *S. mansoni*, hookworm species, *T. trichiura*, *H. nana*, and *S. stercoralis* was 48%, 15.9%, 11.5%, 7%, 6.8 and 2.4% respectively. Another study done among urban dwellers in southwest Ethiopia, Jimma (Mengistu *et al.*, 2007) has also revealed the prevalence of *T. trichiura (*60.9 %), *A. lumbricoides* (40.9%), hookworm species (17.5%), *S. stercoralis (*2.1%), *H. nana* (5.0%), *S. mansoni* (14.8%).

A study done in Adwa Town (Legesse *et al*., 2010), Northern Ethiopia, has shown the prevalence of *S. mansoni, A. lumbricoides,* hookworm species, *H. nana,* and *S. stercoralis* to be 63.0%, 6.4%, 1.0 %, 8.1% and 0.3% respectively. Another study that was conducted in a rural area close to the Southeast of Lake Langano (Legesse *et al.,* 2004) has revealed the prevalence of hookworm (60.2%), *S.mansoni* (21.2%), *T. trichiura* (14.7%), *A. lumbricoides* (6.2%) and *S.*

stercoralis (5.8%), in that order. The prevalence of *S.mansoni*, *A. lumbricoides* , *T. trichiura* , hookworm species , *H. nana* , *S. stercoralis* , and *H. diminuta* in a study done in Babile town, Eastern Ethiopia (Tadesse 2005) was 4.3%, 3.9%, 3.6%, 6.7%, 10.1%, 1%, and 0.2%

According to study that was done among school children and other residents at Bushulo village, a small rural town, located along the shore of Lake Hawassa Southern Ethiopia, where most of inhabitants largely rely on water from Lake Hawassa for both domestic and laundering purposes, showed the infection rates of *S.mansoni*, trichuriasis, ascariasis and hookworm to be 73.7%, 41.5%, 37.2% and 28.4%, respectively. Other parasitic infections observed were *H.nana* (1.7%), Taenia species (1.4%), and *Enterobius vermicularis* (1.4%) (Terefe *et al.,* 2011).

A cross-sectional study involving school children in Tikur Wuha bordering Lake Hawassa, revealed the prevalence and intensity of *S.mansoni* to be 12% and 69 eggs per gram (epg) of stool, respectively (Mitiku *et al.*, 2010).

A cross-sectional study that was conducted among children engaged in fishing, and fish processing activities at Lake Hawassa, Southern Ethiopia, showed the prevalence of *S. mansoni*, (33%), *A. lumbricoides* (76.9%), hookworm species (62.7%), *T. trichiura* (60%) (Merid *et al.,* 2001).

Different studies including those done in Adwa Town, Northern Ethiopia (Legesse *et al.,* 2010), Jimma town , South Western Ethiopia (Mengistu *et al.*, 2011), Gorgora Town, Northwest Ethiopia (Essa *et al., 2012*), Kenya (Handzel *et al.*, 2003), Brazil (Massara *et al.*, 2004) have indicated that *S. mansoni* infection was significantly associated with swimming habit in lakes or rivers. It was also mentioned in studies conducted in Malaysia (Nasr *et al.*, 2013) and Honduras (Smith *et al.,* 2001) that *A.lumbricoides* and *T.trichiura* infections were found to have association with the availability of latrine. A study conducted in Babile town, Eastern Ethiopia (Tadesse 2005),Thailand (Jiraanankul *et al.*, 2011), India (Traub *et al.*, 2004) indicated that shoe wearing habit was significantly associated with hookworm infection.

The majority intensity of *S. mansoni* infection in studies done in Jimma (Mengistu *et al.*, 2011), Southern and Central Zones of Tigray (Dejenie *et al.*, 2009), Tikur Wuha area ,Southern Ethiopia (Mitiku *et al.*, 2010), Kenya (Handzel *et al.*, 2003) showed that it was light infection.

Therefore, periodic assessment of the status of helminthes infections especially that of *S.mansoni* among people that are engaged in fishing activities is required for control and prevention of the disease.

2.1 Significance of the study

Many people living around Lake Hawassa are engaged in fishing. Furthermore, they eat raw fish, swim in the lake and defecate in an open field which contributes to the wide distribution of intestinal parasites. Diseases related to these risk behaviors such as schistosomiasis, diphylobothriasis and other intestinal helminthes infections are expected to exist. However, current information is limited on the status of schistosomiasis and other intestinal helminthes infections even though it is needed to plan effective prevention and control measures.

Therefore, this study will enable to obtain the magnitude of *S.mansoni* and other intestinal helminthes among fishermen at Lake Hawassa and the information can be used by concerned governmental and non-governmental bodies to guide intervention measures. Furthermore, the findings can serve as reference data for further research to be done in the area and can create awareness of community about schistosomiasis and intestinal helminthiasis and this would also contribute towards the disease control.

CHAPTER THREE: OBJECTIVES

3.1 General Objective:

To determine the magnitude of *S.mansoni* and other intestinal helminthes infections among fishermen at Lake Hawassa, Southern Ethiopia

3.2 Specific objectives:

- To determine the prevalence of *S.mansoni*.
- To determine the prevalence of other intestinal helminthes.
- To determine intensity of *S.mansoni* and other intestinal helminthes
- To assess associated risk factors of *S.mansoni* and other intestinal helminthes infections

CHAPTER FOUR: MATERIALS AND METHODS

4.1 Study area and period

The study was carried out at Hawassa. Hawassa is located in the Rift Valley, 270 km South of Addis Ababa via Bishofitu. Lake Hawassa measures 16 km long and 9km wide, with a surface area of 129 square kilometers and has a maximum depth of 10 meters with an elevation of 1,708 meters. The study was carried out from April to June 2013.

Figure 1: Fishing at Lake Hawassa, Southern Ethiopia.

(Source: httpwww.africaimagelibrary.compage1per_page=24andsearch=Awassa)

4.2 Study design

A community based cross-sectional study was conducted to determine the magnitude of *S.mansoni* and other intestinal helminthes infections among fishermen at Lake Hawassa, Southern Ethiopia.

4.3 Population

4.3.1 Source population

All people who are engaged in fishing activities at Lake Hawassa served as the source of population to this particular study.

4.3.2 Study population

Systematically selected subjects among people who are engaged in fishing during the study period were enrolled in the study.

4.4 Inclusion and Exclusion criteria

4.4.1 Inclusion criteria

Those fishermen who have contact with water at least for three months were included.

4.4.2 Exclusion criteria

Those fishermen who have taken antischistosomal and antihelminthic treatment with in the past six months were excluded.

4.5 Sample size and sampling technique

The minimum sample size required for analysis was calculated using the 95% confidence interval with 5% marginal error. Using the formula $n = \frac{z^2 p(1-p)}{d^2}$, where n= sample size, z = z statistic for a level of confidence (z =1.96 at 95% CI), p = prevalence (p = 0.50), d = precision (if 5%, d= 0.05), the sample size became 384. Since sampling is from finite population of less than 10,000, population correction was used.

$n_o = \frac{n}{1+\frac{n}{N}}$; Where n= the sample size from the finite population, N= the total number of study population, and n_o= the corrected sample size. Then $n_o = \frac{384}{1+\frac{384}{520}}$ = 221. Allowing 10% for non response, the final sample size became 243.

Systematic random sampling technique was used to select study participant. The following steps were followed in order to achieve a systematic random sample: The interval size, k = N/n = 520/243= 2.13~ 2. Then randomly selecting an integer between 1and 2 and then take every 2^{nd} unit from the study population until the final sample size is obtained.

4.6 Study variables

4.6.1 Independent variables

The independent variables were:

- Age
- Religion,
- Availability of latrine
- Shoe wearing habit
- Habit of swimming
- Number of water contact
- Finger nail status and
- Habit of eating raw fish

4.6.2 Dependent variables

The dependent variables were: Prevalence of *S.mansoni* and other intestinal helminthes infection.

4.7 Ethical consideration

Ethical clearance was obtained from the ethical review board of Jimma University, College of Public Health and Medical Sciences. Letters of permission was presented to all respected offices. Written informed consent was obtained from each study subject and from the parents of children prior to data collection after the purpose of the study is explained to respondent. Confidentiality of the information was assured and privacy of the respondent was maintained. Results of participants were kept confidentially and those participants with parasitic infection were referred to health facilities for treatment by an assigned nurse.

4.8 Data collection

Semi-structured questionnaire was used to collect demographic data and risk factors. Data was collected by laboratory technicians who were selected and trained for this purpose.

4.8.1 Stool specimen collection and processing

A fresh fecal specimen was collected using labeled, clean, dry, leak-proof container. The study participants were oriented on how to provide stool specimen by a trained laboratory technician. Every individual was instructed to bring his own sufficient amount (about 4 gm) of stool.

Sample rejection criteria: Those stool samples that were small in amount and those collected by containers other than they were provided with were rejected.

4.8.1.1 Processing of stool specimen

Stool specimen was processed by wet mount, Kato-Katz and formol-ether concentration technique following WHO guide (WHO 2003) (Annex one).

4.8.1.2 Direct smear

Direct smear was done by emulsifying about 2 mg of stool using physiological saline (0.85% NaCl solution) on a microscopic slide and was examined by 10x and 40x microscope objective (WHO 2003) (Annex one).

4.8.1.3 Formol-ether concentration

Stool samples were emulsified in formol water, the suspension was strained to remove large faecal particles, then ether was added, and the mixed suspension was centrifuged. The faecal debris was separated in a layer between the ether and the formol water. The supernatant was discarded and a small drop of sediment was placed on a microscopic slide and examined by 10x and 40x microscope objective (WHO 2003) (Annex one).

4.8.1.4 Kato-Katz thick technique

The stool specimen was processed using Kato-Katz technique employing a 41.7mg template. The number of eggs of each species was recorded and converted into the number of eggs per gram of stool (EPG) in order to analyze intensity of infection. EPG was calculated by multiplying the egg count by conversion factor that is 24 (WHO 2003) (Annex one).

4.8.2 **Quality control**

Pre-analytical stage: The expiry date of normal saline, ether, and formol was checked before stool sample preparation and examination. Microscopic slides, cover glasses were checked for cleanliness. Standard operating procedures (SOP) were followed during stool specimen collection, transportation and storing.

Analytical stage: Following a clearly written SOP during performing the test and appropriate health and safety measures were followed.

Post-analytical stage: Recording of test results was verified. The parasitological test results were kept confidentially. Interpretation of test results was checked. Moreover for quality control purpose 10% of the total slides were randomly selected and read by a second experienced laboratory technologist.

4.9 **Data processing and statistical analysis**

All questionnaires were checked for completeness. Completed data was coded; entered; cleaned and analyzed using SPSS version 16 software statistical packages. Results are presented using tables and figures. To assess associations between risk factors and *S.mansoni* and other intestinal helminthes infections, logistic regression analysis was used. Statistical significance was considered as *P* value < 0.05.

4.10 **Dissemination of findings**

The findings of this study will be disseminated to different organizations that have contribution to prevent and control *S.mansoni* and other helminthes infections in the town. Feedback will be given to Southern Nations Nationalities and People Region (SNNPR) Health Bureau. In addition an effort will be made to publish the finding of this study in a scientific journal.

4.11 **Operational definition**

- Fishing: occupation of catching fish
- Intensity of infection: Grading of helminthes infection as light, moderate and heavy by counting helminthes eggs excreted in faeces.
  - *S. mansoni* [Light (1-99 epg), moderate (100-399 epg), heavy (≥ 400 epg)];
  - *T. trichiura* [Light (1-999 epg), moderate (1000-9999 epg), heavy (≥10,000 epg)];
  - *A.lumbricoides*[Light (1-4999epg), Moderate(5,000-49,999 epg), Heavy (≥50,000 epg)]
- Eggs per gram (EPG): The number of helminthes eggs per gram of faeces,
- Contact with water: getting in touch with water during fishing.

CHAPTER FIVE: RESULTS

5.1 Socio-demographic characteristics

A total of 243 fishermen with an age range of 15 to 39 years had participated in this study, giving response rate of 100%. There were no female fishers encountered. The mean age of the study participants was 26.5 years. Out of these 142 (58.44%) were from urban and 101(41.56 %) were from rural areas. Majority of the study subjects (69.55%) were Christians and about 178 (73.25 %) of them have attended grade 1-8 (Table 1).

Table 1: Socio demographic characteristics of the fishermen (n= 243) at Lake Hawassa, Southern Ethiopia, 2013

Socio demographic Characteristics		Frequency	(%)
Age group/years	15-19	44	18.10
	20-24	63	25.93
	25-29	72	29.63
	30-34	41	16.87
	35-39	23	9.47
Religion	Christian	169	69.55
	Muslim	74	30.45
Level of education	No formal education	32	13.17
	grade 1-8	178	73.25
	grade 9-12	27	11.11
	grade >12	6	2.47
Residence	Urban	142	58.44
	Rural	101	41.56
Monthly income	Birr <500	56	23.04
	Birr 500-1500	177	72.84
	Birr >1500	10	4.12

Most of the fishermen, 190 (78.19%) had latrine and 174 (71.60 %) of them have practiced hand washing after defecation. About 208 (85.60%) have the habit of hand wash before meal (Table

4). During the interview about 64.20% of the fishermen's finger nails were trimmed. About 231 (95.06%) reported that they have swam in the lake and out of these, 114 (46.91%) swam in the lake daily while the rest 53.09% swam sometimes. In addition, 219 (90.12%) have had daily contact with the lake for fishing (Table 3). Out of 243 fishermen interviewed, 180 (74.07%) of them claimed to bath their body in the lake. About 118 (48.56%) of the fishermen reported that they have worn shoe regularly (Table 6).

5.2 Parasitological investigations

Microscopic examination of stool was done using wet mount, Kato-Katz and formol ether concentration methods. Of the total 243 stool samples examined, 169 were positive for one or more of intestinal helminthes (after pooling the results of the three diagnostic methods) giving an overall prevalence of 69.55 %. The overall prevalence rate of *S.mansoni* was 29.22%. The other most frequent intestinal helminthes encountered were: *A. lumbricoides* 99 (40.74%), *T. trichiura* 87 (35.80%) and hookworm species 9(9.47%). (Figure 2).

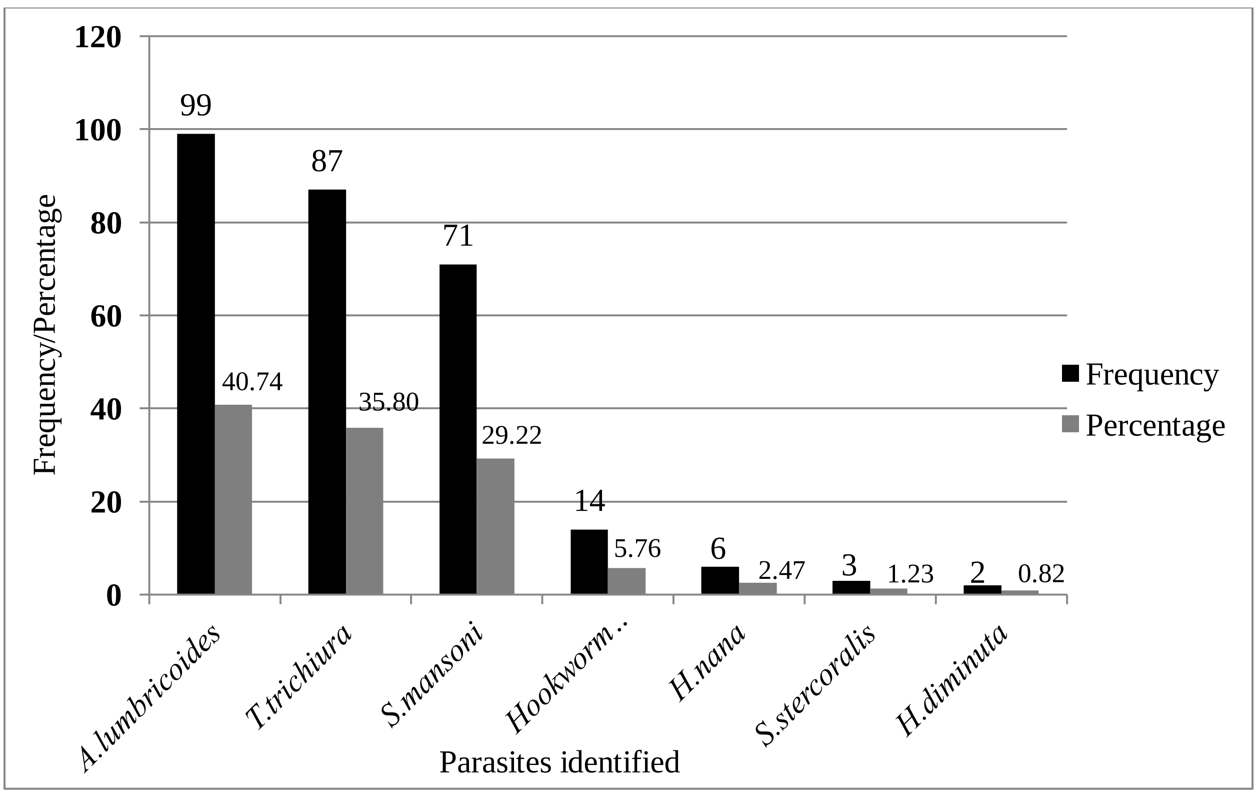

Figure 2: The type and frequency of intestinal helminthes identified among fishermen at Lake Hawassa, Southern Ethiopia, 2013

Among 169 positive fishermen, the majority, 74 (43.79%) of them had single infection while 95 (56.21%) were infected with more than one intestinal parasites. Moreover, 77 (45.56%) had double infection and 18(10.65%) had triple infections. The most common combinations of helminthes were double infection of *A. lumbricoides* and *T. trichiura* 48 (50.53%) which is followed by triple infection of *S.mansoni*, *A. lumbricoides* and Hook worm species 9(9.47%) (Table 2).

Table 2: Pattern of multiple intestinal helminthes infection among fishermen at Lake Hawassa, Southern Ethiopia, 2013

Parasites identified	**Frequency**	**%**
A. lumbricoides and *T.trichiura*	48	50.53
S.mansoni and *T.trichiura*	13	13.68
S.mansoni, A.lumbricoides and hookworm species	9	9.47
S.mansoni and *A.lumbricoides*	7	7.37
A.lumbricoides and hookworm species	5	5.26
T.trichiura and hookworm species	3	3.16
S.mansoni, T.trichiura and hookworm species	2	2.11
S.mansoni, A.lumbricoides and *T.trichiura*	2	2.11
A.lumbricoides, T.trichiura and *H.diminuta*	2	2.11
A.lumbricoides,T.trichiura and *H.nana*	1	1.05
S.mansoni, A.lumbricoides and *S.stercoralis*	1	1.05
A.lumbricoides and *H.nana*	1	1.05
A.lumbricoides and *H.diminuta*	1	1.05
Total	**95**	**100.00**

5.3 Risk factors associated with *S. mansoni* infection

In this study, 71 (29.22%) of the study participants had *S. mansoni* infection. By the bivariate analysis, *S. mansoni* infection among fishermen, was significantly associated ($P<0.05$) with swimming, frequency of swimming, bathing and frequency of water contact. On the other hand, no significant association ($P=0.946$) was found between *S. mansoni* infection and water source for both cooking and drinking.

Multiple logistic regression was performed for variables that were significantly associated with *S. mansoni* in the bivariate analysis. Accordingly, *S. mansoni* infection was found significantly associated with swimming (P=0.022), frequency of swimming (P=0.001) and frequency of water contact (P=0.001). Fishermen that were swimming daily in the lake were 2.92 times [95%CI: 1.554, 5.502] more likely to acquire *S. mansoni* infection. But frequency of water contact and swimming were not a risk even if they have association in the bivariate analysis. However no significant association (P=0.065) was found between *S. mansoni* infection and where the fishermen bath (Table 3).

Table 3: Bivariate and Multivariate logistic regression analysis of factors associated with *S. mansoni* infections among fishermen at Lake Hawassa, Southern Ethiopia, 2013

Factors	***S.mansoni***			***COR [95%CI]**	**P-value**	**AOR (95%CI)**	**P-value**
	Pos (%)	**Neg (%)**	**Total**				
Swimming							
Yes	63 (27.27)	168 (72.73)	231 (95.06)	0.188[0.055, 0.644]	0.008	0.203 [0 .052,0.796]	0.022
No	8 (66.67)	4 (33.33)	12 (4.94)	1			
Freq of swimming							
Always	43 (37.72)	71 (62.28)	114 (46.91)	2.185[1.242, 3.842]	0.007	2.924[1.554, 5.502]	0.001
Sometimes	28 (21.71)	101 (78.29)	129 (53.09)	1			
Bath					0.008		0.065
River	11 (61.11)	7 (38.89)	18 (7.41)	03.143[1.013, 9.752]	0.047	2.046[0.590, 7.101]	0.259
Lake	45 (25.00)	135 (75.00)	180 (74.07)	0.667[0.329, 1.350]	0.260	0.611[0.288, 1.295]	0.198
Home	15 (33.33)	30 (66.67)	45 (18.52)	1			
Water contact							
Daily	57 (26.03)	162 73.97)	219 (90.12)	0.251[0.106,0.597]	0.002	0.203[0.078,0.529]	0.001
Sometimes	14 (58.33)	10 41.67)	24 (9.88)	1			
Water source for cooking/ drinking					0.946		
Pipe	51 (28.81)	126 (71.19)	177 (72.84)	1			
River	17 (30.91)	38 (69.09)	55 (22.63)	1.105[0.572,2.134]	0.766		
Lake	3 (27.27)	8 (72.73)	11 (4.53)	0.926[0.236,3.632]	0.931		

*COR= Crude Odds Ratio, AOR=Adjusted Odds Ratio, CI=Confidence Interval, Pos=Positive, Neg=Negative

5.4 Factors associated with A. lumbricoides and T.trichiura infections

Out of the total study participants, 40.74% and 35.80 % were positive for *A .lumbricoides* and *T.trichiura*. Bivariate analysis showed that *A .lumbricoides* and *T.trichiura* infections were significantly associated (P<0.05) with the availability of latrine, the habit of hand wash after defecation and before meal, water source for cooking/drinking and finger nail status. However, in the multivariate analysis it was only the availability of latrine that showed association with *A .lumbricoides* and *T.trichiura* infections (P=0.001). Fishermen who did not have latrine were 28 and 7.5 times more likely to acquire *A .lumbricoides and T.trichiura* respectively (Table 4 & 5).

Table 4: Bivariate and Multivariate logistic regression analysis of factors associated with *A. lumbricoides* infections among fishermen at Lake Hawassa, Southern Ethiopia, 2013

Factors	*A .lumbricoides*			*COR [95%CI]	P-value	AOR (95%CI)	P-value
	Pos (%)	Neg (%)	Total				
Latrine availability							
Yes	53 (27.89)	137 (72.11)	190 (78.19)		1		
No	46 (86.79)	7 (13.21)	53 (21.81)	16.987[7.217,39.983]	0.001	28.171[7.830, 101.354]	0.001
Hand wash after defecation							
Yes	54 (31.03)	120 (68.97)	174 (71.60)		1		
No	45 (65.22)	24 (34.78)	69 (28.40)	4.167[2.309,7.520]	0.001	0.919[0.316, 2.674]	0.876
Hand wash before meal							
Yes	73 (35.10)	135 (64.90)	208 (85.60)		1		
No	26 (74.29)	9 (25.71)	35 (14.40)	5.342[2.377, 12.006]	0.001	1.561[0.472, 5.170]	0.466
Water source for cooking/ drinking					0.011		0.100
Pipe	64 (36.16)	113 (63.84)	177 (72.84)		1		
River	32 (58.18)	23 (41.82)	55 (22.63)	2.457[1.325, 4.554]	0.004	0.315[0.098, 1.015]	0.053
Lake	3 (27.27)	8 (72.73)	11 (4.53)	0.662[.170, 2.585]	0.553	0.347[0.063, 1.927]	0.226
Finger Nail Trimmed							
Yes	51 (32.69)	105 (67.31)	156 (64.20)		1		
No	48 (55.17)	39 (44.83)	87 (35.80)	2.534[1.478, 4.344]	0.001	1.515[0.736, 3.119]	0.259

*COR= Crude Odds Ratio, AOR=Adjusted Odds Ratio, CI=Confidence Interval, Pos=Positive, Neg=Negative

Table 5: Bivariate and Multivariate logistic regression analysis of factors associated with *T.trichiura* infections among fishermen at Lake Hawassa, Southern Ethiopia, 2013

Factors	*T.trichiura* Pos (%)	Neg (%)	Total	COR[95%CI]	P-value	AOR (95%CI)	P-value
Latrine availability							
Yes	47 (24.74)	143 (75.26)	190 (78.19)		1		
No	40 (75.47)	13 (24.53)	53 (21.81)	9.362[4.616, 18.988]	0.001	7.453[2.834, 19.603]	0.001
Hand wash after defecation							
Yes	45 (25.86)	129 (74.14)	174 (71.60)		1		
No	42 (60.87)	27 (39.13)	69 (28.40)	4.459[2.470, 8.050]	0.001	1.410[0.528, 3.764]	0.493
Hand wash before meal							
Yes	62 (29.81)	146 (70.19)	208 (85.60)		1		
No	25 (71.43)	10 (28.57)	35 (14.40)	5.887[2.668, 12.988]	0.001	2.073[0.734, 5.850]	0.169
Water source for cooking/ drinking					0.003		0.504
Pipe	52 (29.38)	125 (70.62)	177 (72.84)		1		
River	30 (54.55)	25 (45.45)	55 (22.63)	2.885[1.549, 5.371]	0.001	0.670[0.262, 1.712]	0.403
Lake	5 (45.45)	6 (54.55)	11 (4.53)	2.003[.585, 6.854]	0.268	1.651[0.423, 6.444]	0.471
Finger Nail Trimmed							
Yes	46 (29.49)	110 (70.51)	156 (64.20)		1		
No	41 (47.13)	46 (52.87)	87 (35.80)	0.469[.272, .808]	0.006	0.948[0.460, 1.952]	0.884

*COR= Crude Odds Ratio, AOR=Adjusted Odds Ratio, CI=Confidence Interval, Pos=Positive, Neg=Negative

5.5 Factors associated with hookworm infection

In hookworm infection logistic regression analysis showed that no shoe wearing habit was a risk factor (P=0.003). Those fishermen who had never worn a shoe were 10.45 times (95%CI: 2.430, 44.973) at risk of having hookworm infection. But there was no association with the availability of latrine (Table 6).

Table 6: Bivariate logistic regression analysis of factors associated with hookworm infection among fishermen at Lake Hawassa, Southern Ethiopia, 2013

Variables	**Hookworm species**			***COR[95%CI]**	**P-value**
	Positive (%)	Negative (%)	Total (%)		
Latrine availability					
Yes	10 (5.26)	180 (94.74)	190 (78.19)		1
No	4 (7.55)	49 (92.45)	53 (21.81)	1.469[0.442, 4.887]	0.530
Shoe wearing habit					0.003
Regularly	3 (2.54)	115 (97.46)	118 (48.56)		1
Sometimes	5 (5.15)	92 (94.85)	97 (39.92)	2.083[0.485, 8.947]	0.324
Never	6 (21.43)	22 (78.57)	28 (11.52)	10.455[2.430, 44.973]	0.002

*COR= Crude Odds Ratio, CI=Confidence Interval

5.6 Intensity of infection with S. mansoni, A.lumbricoides, T. trichiura and hookworm species among fishermen at Lake Hawassa, Southern Ethiopia, 2013

Out of 71 fishermen who were positive for *S. mansoni* infection, the intensity of *S. mansoni* was predominantly (52.11%) light infection. The rest 43.66% and 4.23% were moderate and heavy infections respectively. The mean intensity of *Schistosoma mansoni* infection among fishermen was 158.88 epg. Egg load ranged from 24 to 504 per gram of stool. In ascariasis and trichuriasis, majority of cases were light infections, 81.82% for *A.lumbricoides*, and 91.95% for *T. trichiura* infections and there was no heavy infection. The intensity of hookworm infection was only light infection. The mean intensity of infection for *A. lumbricoides, T. trichiura* and hookworm were 1349.04 epg, 246.24 epg and 99.36 epg, respectively. The intensity of helminthes infection in this study has been assessed using faecal egg count obtained by the Kato-Katz stool examination technique. They were also categorized as light, moderate and heavy infections based on WHO threshold (WHO 2002) (Table 7).

Table 7: Intensity of *S. mansoni, A. lumbricoides, T. trichiura* and hookworm infections using Kato-Katz smear technique among fishermen at Lake Hawassa, Southern Ethiopia, 2013

Intensity of Parasite (EPG)	Frequency	%
S. mansoni		
Light (1-99)	37	52.11
Moderate (100-399)	31	43.66
Heavy (≥ 400)	3	4.23
A. lumbricoides		
Light (1-4,999)	81	81.82
Moderate (5,000-49,999)	18	18.18
Heavy (≥ 50,000)	0	
T. trichiura		
Light (1-999)	80	91.95
Moderate (1000-9,999)	7	8.05
Heavy (≥10,000)	0	0
Hookworm species		
Light (1-1,999)	14	100
Moderate (2,000-3,999)	0	0
Heavy (≥4000)	0	0

CHAPTER SIX: DISCUSSION

The overall prevalence of helminthes among fishermen in this study was 69.55 %. It is higher than studies in Babile town, Eastern Ethiopia (27.2%) (Tadesse 2005), Southern and Central Zones of Tigray (56.5%) (Dejenie *et al.,* 2009), Waja, North Ethiopia (65.62%) (Dejenie *et al.,* 2009), and Nigeria (45.5%) (Emeka 2013). However it was lower than that of Hawassa, South Ethiopia (92.7%) (Merid *et al.,* 2001). The reason for difference overall prevalence between the present study and other studies could be geography, the occupation and category of the study population, the time of study, sample size and socio-economic conditions of the study subjects.

In the present study the overall prevalence of *S. mansoni* among fishermen at Lake Hawassa, Southern Ethiopia was found to be 29.22%. It is an indication of a public health problem of *S. mansoni* in the study area. In addition, the presence of schistosome infected dwellers represent a risk for the introduction of a new transmission where the snail hosts might be available. This finding was comparable to a study done in Hawassa (33%) (Merid *et al.*,2001), and in Zimbabwe (26.3%) (Chimbari *et al.*, 2003). However it was lower than the findings in Adwa (63%) (Legesse *et al.,* 2010), Bushullo village, Southern Ethiopia (73.7%) (Terefe *et al.,* 2011) and Uganda 47.4% (Odongo-Aginya *et al*., 2011). It was higher than those studies conducted in Tikur Wuha, Southern Ethiopia (12%) (Mitiku *et al.,* 2010), near Lake Langano (21.2%) (Legesse *et al.,* 2004), Gondar (15.9%) (Ayalew *et al*., 2011), Jimma (14.8%) (Mengistu *et al.*, 2007), Babile (4.3%) (Tadesse 2005) and Gorgora (20.6%) (Essa *et al*., 2012). It was also higher than studies in other part of the world including Kenya (16.3%) (Handzel *et al*., 2003) , Nigeria (8.3%) (Nanvya *et al*., 2011) and Burkina Faso (16.35%) (Zongo *et al*., 2008). This difference could be due to differences in occupation, the study subjects, environmental factors that influence snail distribution and geographic condition.

In this study *S. mansoni* infection was found to be significantly associated with swimming which is in agreement with study done in Adwa Town, Northern Ethiopia (Legesse *et al.,* 2010), Jimma town , South Western Ethiopia (Mengistu *et al.,* 2011), Gorgora Town, Northwest Ethiopia (Essa *et al., 2012*), Kenya (Handzel *et al.,* 2003), Brazil (Massara *et al.*, 2004), frequency of swimming and frequency of water contact and fishermen that were swimming daily in the lake were 2.9 times more likely to acquire *S. mansoni* infection.

In this study, the prevalence of *A. lumbricoides* infection was 40.74%. This was comparable to the study done in Jimma (40.9%) (Mengistu *et al.*, 2007) and Bushulo village, Southern Ethiopia (37.2%) (Terefe *et al.,* 2011). However it was lower when compared with the study in Hawassa (76.9%) (Merid *et al.*,2001) and North Gondar (48%) (Ayalew *et al*., 2011) and it was also lower than those of studies in India 91% (Naish *et al*., 2004) but higher than that of Adwa (6.4%) (Legesse *et al*., 2010) , Gorgora (18.8%) (Essa *et al*., 2012), near Lake Langano (6.2%) (Legesse *et al.*, 2004).

The current study also showed the prevalence of *T. trichiura* to be 35.80% which is below the findings in Hawassa (60%) (Merid *et al.*,2001), Jimma (60.9%) (Mengistu *et al.*, 2007), Bushullo village, Southern Ethiopia (41.5%) (Terefe *et al.*, 2011) and India 72% (Naish *et al*., 2004) and above those of Gorgora (3.6%) (Essa *et al*., 2012), Gondar (7%) (Ayalew *et al*., 2011), Langano (14.7%) (Legesse *et al.*, 2004) and Babille**,** Eastern Ethiopia (3.6%) (Tadesse 2005). The contradictory report on the prevalence of ascariasis and trichuriasis between the different studies and the present study may be due to the difference in geography, environmental sanitation, personal hygiene, socioeconomic status, the category of the study population, the methods employed for stool examination. *A .lumbricoides* and *T.trichiura* infections were found to have association with the availability of latrine which is in agreement with studies done in Malaysia (Nasr *et al.*, 2013) and Honduras (Smith *et al.*, 2001). Fishermen that did not have latrine were 28 and 7.5 times more likely to acquire *A .lumbricoides* and *T.trichiura* respectively.

The prevalence of hookworm species in this study was 5.76% and it was in agreement with the findings from Babile**,** Eastern Ethiopia (6.7%) (Tadesse 2005) and Burkina Faso 4.8% (Zongo *et al*., 2008) but lower than those of Hawassa (62.7%) (Merid *et al.*,2001), Langano 60.2% (Legesse *et al.*, 2004), Bushullo (24.8%) (Terefe *et al.*, 2011), and India (54%) (Naish *et al*., 2004) and higher than Adwa (1%) (Legesse *et al*., 2010) and Gorgora (2.2%) (Essa *et al*., 2012).

The contradictory report on the prevalence of hookworm species between the present study and the other studies may be explained by variations in geography, environmental sanitation, time of study and personal hygiene. This study showed that shoe wearing habit is significantly associated with hookworm infection which was in agreement with studies done in Babile town, Eastern Ethiopia (Tadesse 2005),Thailand (Jiraanankul *et al.*, 2011), India (Traub *et al.*, 2004) and those fishermen who had never worn a shoe were 10.45 times at risk of having this infection. In addition, the insanitary excreta disposal practices of the fishermen around the lake may as well have influenced the spread of hookworm infection.

Analysis of the classes of intensities of *S. mansoni* infection in these study showed that the majority of infection was light infection which is in agreement with studies done in Jimma (Mengistu *et al.*, 2011), Southern and Central Zones of Tigray (Dejenie *et al.*, 2009), Tikur Wuha area ,Southern Ethiopia (Mitiku *et al.*, 2010), Kenya (Handzel *et al.*, 2003).

Strengths and limitations of the study

Strengths:

- The study has focused on one of the most vulnerable groups (the fishermen) of the population especially to *S. mansoni* infection.
- The non-response rate in the study was zero.

Limitation:

There are only few studies done on prevalence of *S. mansoni* and other intestinal helminthes infection among fishermen that could be used to compare the findings of the current study.

CHAPTER SEVEN: CONCLUSIONS AND RECOMMENDATIONS

The present study showed that *S. mansoni* and other intestinal helminthes infection was prevalent among fishermen at Lake Hawassa, Southern Ethiopia. The prevalence of *S. mansoni* was associated with factors such as swimming, frequency of swimming and frequency of water contact. Moreover, the prevalence of *S. mansoni* and other intestinal helminthes infection rate observed in this study indicates that the fishermen could become a potential source of infection and therefore are responsible for parasite transmission in the study area and calls for immediate measures. This study had also identified a number of risk factors like habit of hand washing after defecation and before meal, dirty material in the finger nail and shoe-wearing habits that are associated with helminthes infections other than *S. mansoni*. Therefore, the following recommendations are forwarded to health planners and decision makers to give serious consideration for control of these diseases through:

- Mass chemotherapy for fishermen directed against the *S. mansoni* and other intestinal helminthes parasite to reduce the worm burden.
- Health education program about the transmission of *S. mansoni* and other intestinal helminthes infection and how to improve hygienic practices and reduce fecal pollution of the environment must be given to fishermen in particular, as it plays a significant role in changing human behavior.
- Health education on the importance of wearing shoes regularly to the prevention and control of hookworm and parasites with similar modes of transmission.
- Construction of latrines around the lake as most of the fishermen used to defecate in the bush which contaminates both the lake and the environment.
- Besides, further studies should be done in the area.

REFERENCES

Abou-zeid, A.H.A., Abkar, T.A. and Mohamed, R.O., 2012. Schistosomiasis and soil-transmitted helminths among an adult population in a war affected area , Southern Kordofan state , Sudan. *Parasites and Vectors*, 5(133).

Aginya, I.O. and Doehring, E., 2008. Epidemiology , of bilharzias (schistosomiasis) in Uganda from 1902 until 2005. *African Health Sciences*, 8(4), page 239–243.

Ayalew, A., Debebe, T. and Worku, A., 2011. Prevalence and risk factors of intestinal parasites among Delgi school children , North Gondar , Ethiopia. *Journal of Parasitology and Vector Biology*, 3(5), page 75–81.

Bigwan, E., Tinja, B. and Damen, J.G., 2012. Prevalence of schistosomiasis among secondary school boarding students in Potiskum metropolis, Yobe state, North- Eastern Nigeria. *Bayero Journal of Pure and applied Sciences*, 5(1), page 155–158.

Cheesbrough, M., 2009. *District Laboratory Practice in Tropical Countries, Part 1* Second Edi., Cambridge University Press.

Chimbari, M.J. *et al.*, 2003. Transmission of schistosomiasis in Kariba , Zimbabwe , and a cross-sectional comparison of schistosomiasis prevalences and intensities in the town with those in Siavonga in Zambia. *Annals of Tropical Medicine and Parasitology*, 97(6), page 605–616.

Dabo, A. *et al.*, 2011. Urinary schistosomiasis among preschool-aged children in Sahelian rural communities in Mali. *Parasites and Vectors*, 4(21).

Dejenie, T., Asmelash, T. and Teferi, M., 2009. Intestinal Helminthes Infections and Re-Infections with Special Emphasis on Schistosomiasis Mansoni in Waja, North Ethiopia. *Momona Ethiopian Journal of Science*, 1(2), page 4–16.

Dejenie, T. and Petros, B., 2009. Irrigation Practices and Intestinal Helminth Infections in Southern and Central Zones of Tigray. *Ethiopian Journal of Health Development*, 23(1), page 48–56.

El-Hawey, A. *et al.*, 1995. Prevalence and morbidity of schistosomiasis among rural fishermen at two Egyptian villages (Gharbia Governorate). *Journal of the Egyptian Society of Parasitology*, 25(3), page 649–57.

Emeka, L.I., 2013. Prevalence of Intestinal Helminthic Infection among School Children in Rural and Semi Urban Communities in Nigeria . *Journal of Dental and Medical Sciences*, 6(5), page 61–66.

Erko, B. *et al.,* 2009. Diagnostic efficacy of indirect hemagglutination test in relation to Kato method for diagnosis of Schistosomiasis mansoni. *Ethiopian Journal of Health Sciences*, 19(2), page 61–65.

Essa, T. *et al.,* 2012. Current Status of Schistosoma mansoni Infections and Associated Risk Factors among Students in Gorgora Town , Northwest Ethiopia. *International Scholarly Research Network Infectious Diseases*, 2013.

Handzel, T. *et al.,* 2003. Geographic distribution of schistosomiasis and soil-transmitted helminths in Western Kenya : Implications for anthelminthic mass treatment. *American Journal of Tropical Medicine and Hygiene*, 69(3), page 318–323.

Harrison, 2008. Harrison's Principles of internal medicine.

Jiraanankul, V. *et al.*, 2011. Incidence and risk factors of hookworm infection in a rural community of central Thailand. *The American journal of tropical medicine and hygiene*, 84(4), page 594–8.

Kabatereine, N. and Kemijumbi J, Ouma JH, Kariuki HC, Richter J, Kadzo H, Madsen H, Butterworth AE, Ørnbjerg N, V.B., 2004. Epidemiology and morbidity of Schistosoma mansoni infection in a fishing community along Lake Albert in Uganda. *Royal Society of Tropical Medicine and Hygiene*, 98(12), page 711–8.

Kabatereine, N.B. *et al.,* 2011. Integrated prevalence mapping of schistosomiasis , soil-transmitted helminthiasis and malaria in lakeside and island communities in Lake Victoria, Uganda. *Parasites and Vectors*, 4(1), p.232.

Lambertucci, J.R., 2010. Acute schistosomiasis mansoni : revisited and reconsidered. *Mem Inst Oswaldo Cruz, Rio de Janeiro*, 105(4), page 422–435.

Legesse, L., Erko, B. and Hailu, A., 2010. Current status of intestinal Schistosomiasis and soil-transmitted helminthiasis among primary school children in Adwa Town, Northern Ethiopia. *Ethiopian Journal of Health Development*, 24(3), page 191–197.

Legesse, M. and Erko, B., 2004. Prevalence of intestinal parasites among schoolchildren in a rural area close to the southeast of Lake Langano , Ethiopia. *Ethiopian Journal of Health Development*, 18(2), page 116–120.

Massara, C.L. *et al.,* 2004. Factors Associated with Schistosomiasis Mansoni in a Population from the Municipality of Jaboticatubas , State of Minas Gerais , Brazil. *Mem Inst Oswaldo Cruz, Rio de Janeiro,* 99, page 127–134.

Mazigo, H.D. *et al.,* 2010. Co-infections with Plasmodium falciparum, Schistosoma mansoni and intestinal helminths among schoolchildren in endemic areas of northwestern Tanzania. *Parasites and Vectors*, 3(44), page 1–7.

Mengistu, A., Gebre-selassie, S. and Kassa, T., 2007. Prevalence of intestinal parasitic infections among urban dwellers in southwest Ethiopia. *Ethiopian Journal of Health Development*, 21(1), page 12–17.

Mengistu, M. *et al.*, 2011. Human intestinal schistosomiasis in communities living near three rivers of Jimma town , South Western Ethiopia. *Ethiopian Journal of Health Sciences*, 21(2), page 111–118.

Merid, Y. *et al.*, 2001. Intestinal helminthic infection among children at Lake Awassa Area, South Ethiopia. *Ethiopian Journal of Health Development*, 1(15), page 31–38.

Mitiku, H. *et al.*, 2010. Transmission of Schistosoma mansoni in Tikur Wuha area ,Southern Ethiopia. *Ethiopian Journal of Health Development*, 24(3), page 180–184.

Naish, S., McCarthy, J. and Williams, G.M., 2004. Prevalence , intensity and risk factors for soil-transmitted helminth infection in a South Indian fishing village. *Acta Tropica*, 91, page 177–187.

Nanvya, T.N., Dakul, D.A. and Mwansat, G.S., 2011. Schistosomiasis in Ndinjor district of Langtang North Local Government Area of Plateau State , Nigeria. *Nigerian Journal of Parasitology*, 32(2), page 209–213.

Nasr, N. a *et al.*, 2013. Towards an effective control programme of soil-transmitted helminth infections among Orang Asli in rural Malaysia. Part 1: prevalence and associated key factors. *Parasites and vectors*, 6(1).

Odiere, M.R. *et al.*, 2012. High prevalence of schistosomiasis in Mbita and its adjacent islands of Lake Victoria , western Kenya. *Parasites and Vectors 2012,*, 5(278).

Odongo-Aginya, E. *et al.*, 2011. Evidence of long term benefit of morbidity reduction due to praziquantel treatment against schistosoma mansoni in Kigungu fishing village in Entebbe, Uganda. *African Journal of Infectious Diseases*, 5(2), page 33–39.

Odongo-aginya, E.I. *et al.*, 2002. High prevalence and morbidity of Schistosoma mansoni along the Albert Nile in Uganda. *African Health Sciences*, 2(3), page 99–106.

Okpala HO, Agwu E,Agba MI, Chimezie OR, Nwobu GO, O.A., 2004. A survey of the prevalence of Schistosomiasis among pupils in Apata and Laranto areas in Jos , Plateau State, Nigeria. *Online Journal of Health and Allied Sciences*, 1(1).

Oyewole, F. *et al.*, 2007. Helminthic reduction with albendazole among school children in riverine communities of Nigeria. *Journal of Rural and Tropical Public Health*, 6, page 6–10.

Risikat, S.A. and Ayoade, A.A., 2012. Correlation analysis between the prevalence of schistosoma haematobuim and water conditions : A Case Study among the School Pupils in

Southwestern Nigeria. *International Journal of Research and Reviews in Applied Sciences*, 13(1), page 160–165.

Ross, A.G.P. *et al.*, 2002. Schistosomiasis. *New England Journal of Medicine*, 346(16), page 1212–1220.

Ross, A.P. *et al.*, 1997. Epidemiologic features features of schistosoma japonicum among DongtingLake region (Hunan province) of China. *American Journal of Tropical Medicine and Hygiene*, 57(3), page 302–308.

Smith, H. *et al.*, 2001. Prevalence and intensity of infections of Ascaris lumbricoides and Trichuris trichiura and associated socio-demographic variables in four rural Honduran communities. *Memórias do Instituto Oswaldo Cruz*, 96(3), page 303–314.

Tadesse, G., 2005. The prevalence of intestinal helminthic infections and associated risk factors among school children in Babile town , Eastern Ethiopia. *Ethiopian Journal of Health Development*, 19(2), page 140–147.

Terefe, A. *et al.*, 2011. Schistosomiasis mansoni and soil-transmitted helminthiasis in Bushulo village , southern Ethiopia. *Ethiopian Journal of Health Development*, 25(1), page 46–50.

Thiongo, F., Luoba, A. and Ouma, J.. H., 2001. Intestinal helminths and schistosomiasis among school children in a rural district in Kenya. *East African medical journal*, 78(6), page 279–282.

TL, L. and Aginya, O., 1991. The intensity of Schistosoma mansoni in the fishing villages of Bugonga and Kitubulu in Entebbe Peninsula, Uganda. *East African medical journal*, 68(5), page 372–7.

Traub, R.J. *et al.*, 2004. The prevalence, intensities and risk factors associated with geohelminth infection in tea-growing communities of Assam, India. *Tropical medicine and international health*, 9(6), page 688–701.

Uneke, C.J., 2010. Soil transmitted helminth infections and schistosomiasis in school age children in sub-Saharan Africa: Efficacy of chemotherapeutic intervention since World Health Assembly Resolution 2001. *Tanzania Journal of Health Research*, 12(1).

WHO, 2003. *Manual of basic techniques for a health laboratory* Second edi., World Health Organization, Geneva, Swizerland.

WHO, 2002. Prevention and control of Schistosomiasis and Soil-Transmitted Helminthiasis. *WHO Technical Report Series 912*.

WHO, 2011. *WHO,Helminth control in school age children A guide for managers of control programmes, 2nd ed.*, Yami, A., Kebede, S. and Mamo, Y., 2010. Impact assessment of Gibe hydroelectric dam on schitosomiasis : a cross sectional study in Southwest Ethiopia. *Ethiopian Journal of Health Sciences*, 20(2), page 129–136.

Zongo *et al.*, 2008. Schistosomiasis among Farmers and Fisherman in the West Part of Burkina Faso (West Africa). *Journal of Biological Sciences*, 8(2), page 482–485.

Annex one: Laboratory techniques

1.1 Wet mount /direct examination of faecal specimens

1. Place one drop of 0.85% NaCl on the slide.
2. Take a small amount of faecal specimen and thoroughly emulsify the stool in saline
3. Place a 22mm cover slip at an angle into the edge of the emulsified faecal drop.
4. Systematically scan the entire 22mm cover slip with the 10x objective.
5. Switch to high dry (40X objective) for more detailed study of any suspect eggs.

1.2 Kato-Katz technique

1. Soak the cellophane strips in the glycerol–malachite green (or methylene blue) solution for at least 24 hours before use.
2. Transfer a small amount (approximately 0.5 g) of faeces on to a piece of scrap paper (newspaper is ideal).
3. Press the screen on top of the faecal sample.
4. Using the applicator stick, scrape across the upper surface of the screen to sieve the faecal sample.
5. Place the template on a clean microscope slide. Transfer the sieved faecal material into the hole of the template and level with the applicator stick.
6. Remove the template carefully so that all the faecal material is left on the slide and none is left sticking to the template.
7. Cover the faecal sample on the slide with a glycerol-soaked cellophane strip.
8. If any glycerol is present on the upper surface of the cellophane, wipe it off with a small piece of absorbent tissue.
9. Invert the microscope slide and press the faecal sample against the cellophane on a smooth surface (a piece of tile or flat stone is ideal) to spread the sample evenly.
10. Do not lift the slide straight up or it may separate from the cellophane. Gently slide the microscope slide sideways while holding the cellophane. Wipe off any excess glycerol with a piece of absorbent tissue to ensure that the cellophane stays fixed.

1.3 Formol-ether concentration technique

1. Using a stick, emulsify an estimated 1g of faeces in about 4ml of 10% formol water contained in a screw –cap bottle or tube.
2. Add further 3-4ml of 10% formol water, cap the bottle and mix well by shaking.
3. Sieve the emulsified faeces, collecting the sieved suspension in a beaker.
4. Transfer the suspension to a conical tube and add 3-4 ml of diethyl ether.
5. Stopper the tube and mix for 1 minute.
6. With a piece of wrapped around the top of the tube, loosen the stopper.
7. Centrifuge immediately at 3000 rpm for 1 minute.
8. Using a stick, loosen the layer of faecal debris from the side of the tube and invert the tube to discard the ether, fecal debris and formol water.
9. Return the tube to its upright position and allow the fluid from the side of the tube to drain to the bottom. Tap the bottom of the tube to re-suspend and mix the sediment.
10. Transfer the sediment to the slide, and cover with cover glass.
11. Examine the preparation microscopically using the 10x objective with the condenser closed sufficiently to give good contrast. Use 40X objectives to examine cysts.

Annex two: Questionnaire

Questionnaire/English version

Jimma University collage of public health and medical sciences department of medical laboratory science and pathology

Questionnaire to assess socio-demographic and associated factors for *S.mansoni* and intestinal helminthes infections among fishermen at Lake Hawassa, Southern Ethiopia

A. Participants identification

1. Code __________________
2. Age __________
3. Religion 1. Christian 2. Muslim 3. Others (Specify)___________

B. Risk factors

4. Level of education 1. Illiterate 2. Literate /specify grade attained_________
5. Residence 1. Urban 2. Rural
6. Income/year ________________________
7. Do you eat raw fish? 1. Yes 2. No
8. Do you have latrine? 1. Yes 2. No
9. Do you wash your hands after defecation? 1. Yes 2. No
10. Do you wash your hands before meal? 1. Yes 2. No
11. Water source for drinking/cooking 1. Pipe 2. River 3. Lake 4. Others/specify_______
12. Number of water contact 1. Daily 2. Sometimes 3. None
13. Do you swim in the lake? 1. Yes 2. No
14. How often do you swim in the lake? 1. Always 2. Sometimes
15. Shoe wearing behavior: 1. Regularly 2. Sometimes 3. never wear shoe
16. Where do you bath? 1. River 2. At Lake. 3. at home
17. Are finger nails trimmed? 1. Yes 2. No
18. Did you take antihelmenthic drugs in the last six monthes? 1. Yes 2. No

Name of the interviewer ____________________Signature __________Date _______________

Name of investigator ____________________Signature __________Date _______________

Questionnaire /Amharic version

ጅማ ዩኒቨረሲቲ የህብረተሰብ ጤናና ሕክምና ሳይንስ ኮሌጅ የሜዲካል ላቦራቶሪ ሳይንስና ፓቶሎጂ ትምሕርት ክፍል

ይህ ቅፅ አሳ አጥማጆችንና ከአሳ ጋር ንክኪ ያላቸዉ ሰዎች ለአንጀት ዉስጥ ትላትሎች ተጋላጭ የሚያደርጉ ምክንያቶችን መሰብሰቢያ ነዉ።

ክፍል አንድ፡ የተሳታፊው መለያ

1. የተሳታፊው ልዩ መለያ ____________________

2. ዕድሜ________________________________

3. ሐይማኖት 1) ክርስቲያን __________ 2) ሙስሊም ________ 3) ሌላ (ይግለጹ)______

ክፍል ሁለት: የአጋላጭ ምክንያቶች መረጃ

4.የትምህርት ደረጃ 1. ያልተማረ _______ 2. የተማረ /የትምህርት ደረጃ ይግለዑ____

5.መኖሪያ 1. ከተማ 2. ገጠር

6.የገቢ መጠን/በአመት _______________

7.ጥሬ አሳ □መገባሉ? 1. አዎ___ 2. አይደለም_________

8.ሽንት ቤት አለዎት? 1. አዎ___ 2. አይደለም_________

9.ከሽንት ቤት ሲመለሱ እጅዎን ይታጠባሉ 1. አዎ__ 2. አይደለም_________

10. ከምግብ በፊት እጅዎን ይታጠባሉ? 1. አዎ_____ 2. አይደለም_________

11. ለመጠጥ ና ለምግብ የሚውል ውሃ በዋናነት የሚያገኙት ከየት ነው?

1. ከቧንቧ 2. ከወንዝ 3. ከሃይቅ 4.ከሌላ (ግለጽ)______________________

12. ምን ያህል ጊዜ ከዉሀ ጋር ንክኪ ያደርጋሉ? 1.ሁል ጊዜ 2.አንዳንድ ጊዜ 3. በፍፁም

13. ሀይቅ ዉስጥ ይዋኛሉ? 1. አዎ___ 2. አይደለም_________

14. ምን ያህል ጊዜ ይዋኛሉ? 1.ሁል ጊዜ 2.አንዳንድ ጊዜ

15. ጫማ ያደርጋሉ? 1 .ሁል ጊዜ 2.አንዳንድ ጊዜ 3. በፍፀም

16. ገላዎን የት ይታጠባሉ? 1. ወንዝ 2.ሀይቅ 3. ቤት

17. የጣት ጥፍሮች ተቆርጦአል (በማየት የሚሞላ) 1. አዎ__ 2. አይደለም_________

18. ባለፉት ስድስት ወራት ለሆድ ዉስጥ ትላትል የሚሆን መድሀኒት ወስደዋል?

1. አዎ_ 2. አይደለም_________

የጠያቂው ስም _______________ፊርማ ________________ቀን__________

የዋና ተመራማሪ□ ስም_______________ፊርማ________ቀን___________

Annex three: Information Sheet and consent form

Information Sheet and consent form for Study participants

1. Information sheet

1.1 Title of the study

Intestinal helminthic infections among fishermen with special emphasis to *Schistosoma mansoni* at Lake Hawassa, Southern Ethiopia

1.2 Purpose of the study

The objective of this study was to assess the magnitude of schistosomiasis and other intestinal helminthic infections among fishermen at Lake Hawassa.

1.3 Procedures to be carried on the study subjects

Each individual was provided with labeled clean plastic sheet, toilet tissue paper and pieces of applicator sticks to collect stool sample.

1.4 Risks associated with the study

There is no risk on the study subject

1.5 Benefits of the study

The laboratory findings would be used to initiate appropriate treatment for the said infections. The study findings would also be used to design and implement control strategies in the study area in the future. This study will provide important current information for future studies among people engaged in fishing.

1.6 Confidentiality of your information

All information obtained will be kept private

1.7 Voluntary participation

Participation is only by willingness. I would also like to inform you that this study will be approved by Jimma university research ethical board. The address is:

Coordinator of research ethical review board, Jimma University
P.O. Box- 378, Jimma, Ethiopia
Tel. 0471120945
The address of investigator: Tadesse Menjetta, Mobile: 0916023356
The address of advisors of this study is: Serkadis Debalke: phone no. 0916829327
Daniel Dana: phone no. 0917800188
Jimma University
College of public health and medical sciences
Department of medical laboratory sciences and pathology

2. Consent form for study subjects

Name: ______________________________________

I have read the information sheet (or it has been read to me); I have understood that it involves about *Schistosoma mansoni* and other intestinal helminthic infections among people engaged in fishing at Lake Hawassa. And also I have understood about the purpose of the study, procedures to be carried out, risks associated with the study, benefits of the study and confidentiality of the information. Therefore, I am willing to give the requested information. I have asked some questions and clarification has been given to me. I have given my consent freely to participate in the study.

I______________________________________ hereby give my consent for giving of the requested information and a stool sample for the study.

Participant's signature ______________________ Date_______________

Investigator's signature _____________________Date _______________

3. Consent form for study subjects /Amharic Version/

ክፍል አንድ፡- ለጥናቱ መረጃና ተሳታፊነት መግለጫ ቅጽ

ዋና ተመራማሪ *ታደሰ መንጀታ*

አማካሪዎች *:* ወ/ት ሰርክአዲስ ደባልቄ

አቶ ዳንኤል ዳና

የድርጅቱ ስም: ጅማ ዩኒቨርሲቲ

የገንዘብ ድጋፉን የሰጠው ድርጅት፡ ጅማ ዩኒቨርሲቲ

የጥናቱ ርዕስ : የአንጀት ዉስጥ ትላትሎች ስርጭት በአሳ አጥማጆችና ከአሳ ጋር ንክኪ ባላቸዉ ሰዎች፡፡

ጤና ይስጥልኝ፡፡ እኔ __________________________ እባላለሁ፡፡ ይህንን ጥናት የምሰራው በጅማ ዩኒቨርሲቲ የድህረ ምረቃ ተምህርቴ ለማጠናቀቅ ማሙአያ እንዲሆነኝ ነዉ።

የጥናቱ ዓላማ

የአንጀት ዉስጥ ትላትሎች ስርጭት በአሳ አጥማጆችና ዉስጥ ለማዎቅ የሚረዳ ጥናት ነው፡፡

የአሰራሩ ሂደት

በዚህ ጥናት ዉስጥ ይሳተፉ ዘንድ እንጠይቆታለን፡፡ለመሳተፍ የሰገራ ናሙና መስጠትና ከበሽታው ጋር ተያያዥ የሆኑ መጠይቆችን መመለስ ይጠበቅብዎታል።

በጥናቱ ሊከሰቱ የሚችሉ ተያያዥ ችግሮች

በጥናቱ መሳተፍ ምንም አይነት ተያያዥ ችግር አያስከትልም።

በጥናቱ በመሳተፍ የሚገኝ ጥቅም

በሽታው ከተገኘ የህክምና እርዳታ እንዲያገኙ ይደረጋል፡፡

ክፍያን በተመለከተ

በጥናቱ በመሳተፍ ምንም አይነት ክፍያ አይከፈልዎትም።በአንጻሩ ከልብ ልናመሰግንዎት እንወዳለን፡፡

የጥናቱ መረጃዎች ሚስጥራዊነት

በጥናቱ ውስጥ የተሰበሰቡ ማናቸውም መረጃዎች ሚስጥራዊነታቸው የተጠበቀ ይሆናል፡፡

ከጥናቱ ስለመውጣትና ስለማቋረጥ

ይህ ጥናት በፈቃደኝነት ላይ የተመሰረተ እንደመሆኑ መጠን በማንኛውም ወቅት በፈቃድዎ ከጥናቱ መዉጣት ይችላሉ፡፡ ከጥናቱ ቢወጡም እንኳን የተለመደውን የህክምና እርዳታ በጤና ተቋሙ ውስጥ በማንኛውም ጊዜ የማግኘት መብት አለዎት፡፡

ከጥናቱ ጋር በተያያዘ ማናቸውም ጥያቄ ቢኖርዎት በሚከተው አድራሻ ጥያቄዎን ማቅረብ ይችላሉ።

አቶ ታደሰ መንጀታ (ስልክ:0916023356)፣ ጅማ ዩኒቨርሲቲ፣ ጅማ ፣ ኢትዮጵያ፡፡

<u>ክፍል ሁለት-የስምምነት ቅጽ</u>

የተሳታፊው መለያ ቁጥር____

እኔ _______________________________________ በዚህ ጥናት እነድሳተፍ ተጠይቄዐለሁ፡፡በክፍል አንድ ዉስጥ ያለዉ መረጃ ተነቦልኛል፡፡መጠየቅ የምፈልገዉን ጥያቄ በአግብቡ ጠይቄ አስፈላጊዉን ምላሽ በአጥጋቢ ሁኔታ ተመልሶልኛል፡፡ ስለሆነም በፈቃደኛነት በጥናቱ ተሳታፊ ለመሆን መፍቀዴን በፊረማዬ አረጋግጣለሁ።

ፊርማ ____________________

የዋና ተመራማሪ ፊርማ____________________

<u>ክፍል ሶስት-ልጆችን በተመለከተ</u>

የተሳታፊው መለያ ቁጥር____

የተሳታፊው ሙሉ ስም____

የተሳታፊው ህጻን ወላጅ/አሳደጊ ሙሉ ስም__________________________________

እኔ የ______________________________ ወላጅ/አሳዳጊ ልጄ በዚህ ጥናት እነዲሳተፍ እና የሰገራ ናሙና እንዲሰጥ ተጠይቄዐለሁ፡፡በክፍል አንድ ዉስጥ ያለዉ መረጃ ተነቦልኛል፡፡መጠየቅ የምፈልገዉን ጥያቄ በአግብቡ ጠይቄ አስፈላጊዉን ምላሽ በአጥጋቢ ሁኔታ ተመልሶልኛል፡፡ ስለሆነም በፈቃደኛነት ልጄ በጥናቱ ተሳታፊ እንዲሆን እና የሰገራ ናሙና ሰጥቶ የአንጀት ዉስጥ ትላትሎች ምርመራ እንዲደረግበት መፍቀዴን በፊረማዬ አረጋግጣለሁ።

የወላጅ/ያሳዳጊ ፊርማ ____________________

የዋና ተመራማሪ ፊርማ____________________

Assurance of Principal Investigator

I, the undersigned, declare that this research thesis is my own original work and it has not been presented in other universities, colleges or other institutions for similar degree or other purpose.

I agree to accept responsibility for the scientific ethical and technical conduct of the research project and for provision of required progress reports as per terms and conditions of the college of Public Health and Medical Sciences in effect at the time of grant is forwarded as the result of this application.

Name of the student	Signature	Date
Tadesse Menjetta, BSc	_____________	_____________

Approval of the Advisors

This thesis has been submitted with our approval as University advisor.

Approval of the first advisor

Name	Signature	Date
Serkadis Debalke, MSc	_____________	_____________

Approval of the second advisor

Name	Signature	Date
Daniel Dana , MSc	_____________	_____________

Approval of the internal examiner

Name	Signature	Date
Abdissa Biruksew	_____________	_____________

Head of the Departement

Name	Signature	Date
__________________________	_____________	_____________