Prevalence of Soil Transmitted Helminthiasis and Malaria Coinfection and Associated Risk Factors among Pregnant Women in Gilgel Gibe dam Area, Southwest Ethiopia



By: Million Getachew (BSc)

Masters Thesis submitted to Department of Medical Laboratory and Pathology, Collage of Public Health and Medical Sciences, Jimma University; for partial fulfillment of the requirement for the Degree of Master Science in Medical Parasitology (MSc).

> October, 2011 JIMMA UNIVERSITY

Jimma University;

Collage of Public Health and Medical Sciences;

Department of Medical Laboratory Science and Pathology

Prevalence of Soil Transmitted Helminthiasis and Malaria Coinfection and Associated Risk Factors among Pregnant Women in Gilgel Gibe dam Area, Southwest Ethiopia

Candidate: Million Getachew (BSc)

Advisers: Mr. Ahmed Zeinedin (BSc, MSC, Assistant professor of Medical Parasitology) Mr. Delenasaw Yewhalaw (BSc, MSC, Assistant professor of Entomology)

> October /2011 JIMMA UNIVERSITY

Abstract

Introduction: Malaria and Soil Transmitted Helminthiasis (STH) are causes of significant morbidity, mortality, severe illness and poor economic development worldwide. These diseases are most of the time Co-endemic in Ethiopia with overlapping geographical distrubution.

Objectives: The main objective of this study was to determine the prevalence of malaria - soil transmitted helminths co-infection and anaemia, as well as to assess associated risk factors for the co-infection and anaemia among pregnant women in Gilgel-Gibe dam area, southwestern Ethiopia.

Materials and Methods: A cross-sectional community based study was conducted to assess malaria andsoil transmitted helminth co-infection in Gilgel Gibe area, southwestern Ethiopia. In this study 388 pregnant women were randomly selected from 6 rural and 6 urban kebeles found in three waredas around Gigel Gibe Dam area. Socio-demographic and socio-economic data were collected using semi-structured questionnaire. Stool samples were collected from all study participants and processed following the standard procedure of McMaster concentration technique. Thick and thin blood film were collected and examined microscopically following the methods described by Cheesbrough.Haematocrit value was determined using hematocrit centrifugation to determine the prevalence of anaemia.

Results: In the current study, the prevalence of soil transmitted helminths and malaria was 159 (41%) and 45(11.6%), respectively and the prevalence of soil transmitted helminths /malaria co-infection was 30(7.7%). The prevalence of anaemia in this study was 209(53.9%). Parasitic infection and anaemia were more prevalent in the rural than urban setting (p<0.05) mainly because of the increased exposure for parasitic infections in the rural resedence. Pregnant women with soil transmitted helminth were 3 times more likely to have malaria than women without helminthic infection (OR=3.3, 95% CI:1.72-6.40, p<0.001). Hook worm was the most prevalent 114(29.4%) soil transmitted helminthiasis infection followed by A. lumbricoides 58(15%) and T. trichuira 13(3.4%). Hookworm parasitic load was positively correlated with malaria parasitic load (r = 0.299, p<0.001) while A. lumbricoides parasitic load was negatively correlated with malaria parasitic load (r = -0.095, p < 0.001). There were a statistically significant association between soil transmitted helminthiasis and anemia (p < 0.001). Pregnant women coinfected with malaria and soil transmitted helminthaisis were 6 times more likely to be anaemic compared to those women infected with soil transmitted helminthes only (OR=5.9, 95% CI: 1.69-20.41, p=0.001). Habit of eating soil (p=0.008), Presence of stagnant water near study participants' house (p=0.012) and habit of using human feces as a fertilizer (p<0.001) were found to be risk factors for malaria and STH coinfection.

Conclusion: Malaria, STH and malaria/STH co- infection were found to be serious health problems among pregnant women in the study area with higher prevalence in the rural resedents. Infection with STH was found to increase malaria prevalence and eventhough infection with A.lumbricoides were found to reduce the severity of malaria; infections with the rest STH were associated with increased parasitic load. Moreover, the severity of anaemia was more pronounced in pregnant women with malaria/STH co-infection. Hence, pregnant women should be diagnosed for STH as well as malaria during their visit for ANC for prompt treatment of malaria and STH infection and to control anaemia associated with STH/malaria co-infection in pregnant women.

Key words: Co- infection, Malaria, Pregnant women, Soil transmitted helminths, Anaemia

Acknowledgment

In the very begining I would like to express my deepest gratitude to the almighty God, making every condition favorable for me to write this paper successfully.

I would like also to acknowledge my dear advisers Mr.Ahmed Zeinedin (BSc, MSC, Assistant professor of Parasitology), Mr.Delenasaw Yewhalaw (BSc, MSC, Assistant professor of entomology) for their great concern to facilitate all the necessary laboratory equipments and transportation during the data collection as well as for their valuable advice and comment on my work throughout the study. My acknowledgment also goes further to my friend Yaregal Asres (BSC) (instructor in Jimma University), for his kind cooperation by allowing me to use his personal computer to write this thesis paper.

Finally, I would like to thank our department, Medical Laboratory and Pathology, and the University as a as a whole for their permission to use the available resources of the University to prepare this paper.

Abstractii
Acknowledgmentiii
Table of Contents iv
List of Accronyms vii
List of Figures viii
List of tables ix
Chapter One Introduction1
1.1 Background1
1.2 Statement of the problem
1.3 Significance of the study
Chapter 2 Literature review
2.1 Malaria among Pregnant Women7
2.2 Soil Transmitted Helminthiasis among Pregnant Women
2.3 Malaria and STH co-infection among Pregnant Pomen8
2.3 Anaemia among pregnant women
Chapter Three Objectives
3.1. General Objective15
3.2. Specific Objectives
Chapter Four Methods and Materials16
4.1. Study area
4.2. Study design and period16
4.3. Population17
4.3.1 Source population17
4.3.2 Study population17

Table of Contents

4.3.3 Study Participants	17
4.4 Sample size determination and Sampling technique	17
4.4.1 Sample size determination:	17
4.4.2 Sampling technique:	
4.5 Inclusion and Exclusion Criteria	19
4.5.1 Inclusion criteria	19
4.5.2. Exclusion criteria	19
4.6 Data Collection and Processing	19
4.6.1 Data collection using Questionnaire	19
4.6.2 Blood sample collection & processing for malaria and anaemia	19
4.6.3 Stool sample collection and processing	20
4.7 Operational Definition of Terms	21
4.8 Data analysis	22
4.9 Quality Assurance	22
4.10 Limitation of the study	23
4. 11 Ethical Consideration	23
Chapter five Results and Discussion	24
5.1 Results	24
5.1.1 Socio demographic and socio economic characteristics of respondents	24
5.1.2 Parasite Prevalence and associated risk factors	27
5.1.3 Prevalence of anaemia and associated risk factors	36
5.2 Discussion	41
Chapter six Conlussions and Recommendations	45
6.1 Conclussions	45
6.2 Recommendations	45

Chapter seven References	47
Annex 1-Procedures of Parasitological Examination of Faeces	53
I. McMaster egg counting technique:	53
II. Microhaematocrit method	53
III. Procedure for Blood Film	54
Annex 2. Laboratory Result Format	56
Annex 3- Consent form in oromifa	57
Annex 4. Questinnaire in oromifa	58
Annex 5.concent form in Amharic	64
Annex 6. Questionnaire in Amharic	65
Annex 7- Consent form in English	72
ANNEX 8- Questionnaire in English	73
Annex 9- Laboratory Result Delivery Format	78
Annex 10- Data collection and processing pictures/Plates	79
Plate 1. The long distance journey on foot with HEW in the sunny day, to reach the he of randomly selected study participants in Gigel Gibe area, 2011.	
Plate 2. Health Extension worker (HEW) collecting data using semi- structured questionnaire in Gigel Gibe area, 2011.	79
Plate 3. Capillary blood collection during house to house survey in Gigel Gibe area, 2	
Plate 4. Blood sample collection at Burqa Asendabo health post, 2011	80
Plate 5. Transporting stool sample from the field to Jimma University Laboratory for processing 2011	80
Plate 6. Stool sample processing at Jimma University clinical laboratory 2011	80

List of Accronyms

CSA - Choriondroitin Sulfate A DALYs - Disability Adjusted Life Years HCG - Human Chorionic Gonadotropin Hgb- hemoglobin HCT- hematocrit IFN-7 -Interferon gama ITN- Insecticide-treated bed nets LBW - Low-Birth Weight NGOs- Non Governmental Organizations NO – Nitric Oxide PTD - Pre-Term Delivery RDT- Rapid Diagnostic Test SGA - Small-for Gestational Age **STH**-Soil-Transmitted Helminths Th1 -T helper cell 1 Th2 - T helper cell 2 WHO- World Health Organization

List of Figures

Figure 1. Map of the study area16	
Figure 2. Profile of study participants in Gigel Gibe area, 2011	
Figure 3. Age specific malaria prevalence among pregnant women in Gilgel Gibe	
hydropower dam area, southwest Ethiopia, August to September 2011	
Figure4. Proportion of malaria, STH infection and co-infection among pregnant	
women in Gilgel Gibe hydropower dam area, southwest Ethiopia,	
August to September 2011	•

List of tables

List of Tables Page
Table 1. Socio demographic characteristics of respondents in Gilgel Gibe hydropower damarea, southwest Ethiopia, August to September 2011
Table 2. Socio-economic characteristics and housing condition of respondents in Gilgel Gibehydropower dam area, southwest Ethiopia, August to September 2011
Table 3. Age-specific prevalence of soil transmitted helminths and risk factors of of STHinfection among pregnant women in Gilgel Gibe hydropower dam area, southwest Ethiopia,August to September 2011
Table 4. Parameter estimates from multivariable Logistic regression model predicting thelikelihood that a pregnant woman is infected with STH, Gilgel Gibe Dam area, southwestEthiopia, August to September 2011
Table 5. Age-specific prevalence of malaria and risk factors of of malaria among pregnantwomen in Gilgel Gibe hydropower dam area, southwest Ethiopia,August to September 201131
Table 6. Parameter estimates from multivariable Logistic regression model predicting thelikelihood that a pregnant woman is infected with malaria, Gilgel Gibe Dam area, southwestEthiopia, August to September 2011
Table 7. Age-specific prevalence of malaria/STH co-infection and risk factors of malaria/STHco-infection among pregnant women in Gilgel Gibe hydropower dam area, southwest Ethiopia,August to September 2011
Table 8. Parameter estimates from multivariable Logistic regression model predicting the likelihood that a pregnant woman is Co-infected with STH and malrai, Gilgel Gibe Dam area, southwest Ethiopia, August to September 2011
Table 9. Association between number of helminthes species and malaria infection rate among
pregnant women in Gilgel Gibe hydropower dam area, southwest Ethiopia, August to September
2011
Table 10. Association of anaemia with socio-demographic characteristics and environment related factors among pregnant women in Gilgel Gibe hydropower dam area, southwest Ethiopia,
August to September 2011

Table 12. Distribution of anaemia among different age groups of pregnant women with resp	ect
to place of residence in Gilgel Gibe hydropower dam area, southwest Ethiopia, August to	
September 2011	39
Table 13. Association of STHs and malaria with anaemia among pregnant women in Gilgel	Gibe
Dam area, 2011	. 40

.....

Chapter One

Introduction

1.1Background

Parasitic diseases are diseases which include infections by protozoa, helminths, and arthropods. They pose major obstacles to health growth and socio-economic development in developing countries. Parasitic diseases such as malaria are life threatening as well as the leading cause of mortality in endemic countries; more sever to some risky groups, pregnant women. Others such as soil transmitted helminthiasis, onchocerciasis, shistosomiasis and lymphatic filariasis cause debilitating symptoms, which are chronic, hinder health growth in children and also significantly reduce the productive life of adults and result in serious complications in pregnant women, immunocomprimized member of the community (1).

In pregnancy, there is a transient depression of cell-mediated immunity that allows fetal allograft retention but also interferes with resistance to various infectious diseases (2). Depression of cellular immune responses to *P. falciparum* antigens in pregnant women which let them to be more susceptible to malaria is the result of this natural transient immune depression (3). Pregnant women in malaria-endemic areas are highly prone to malaria, and this is true especially in primigravid women (4). As a result of this reality pregnant women are more vulnerable to malaria and other parasitic infections compared to the other group of the society (4, 5).

On the top of host, pregnant women, immunossupression; studies showed that immunological interactions between protozoan and helminth infection can intensify the impact of parasitic infection when they co-exist. There are many hypotheses that have been presented to explain potential co-infection with malaria and intestinal helminths. It has been suggested that in malaria-helminth co-infection, helminth infection stimulates the Th2 cytokine response which possibly predominates, and down-modulates Th1 cytokines. Inhibition of the Th1 response prevents protective effects of IFN-7/during the blood and liver stages of malaria infection (6). This could explain the worsening of anemia in malaria-helminth co-infection (7). Helminth infection thus creates a cytokine milieu favorable to the production of non-cytophilic antibodies, thus making individuals more susceptible to clinical malaria (8). The presence of T regulatory cells is amplified during helminth infection, and if present in sufficient numbers, could induce a nonspecific suppression, (9) making individuals susceptible to infections such as malaria; however, malaria may also exacerbate the helminth infection associated anaemia and other consequences (8). Based on the above hypothesis many studies have indicated that infection with STH increase the incidence and prevalence of malaria (10, 11, 12). On the other hand studies have also showed that helminthic infection have protective impact, as a result reduced prevalence as well as minimized severity of malaria were reported from individuals co-infected with intestinal helminth; especially with *A. lumbricoides* (13, 14). Therefore there is still a confussion about the association between plasmodiums and STH infections. The biological association between these parasitic infections determines the strength of their outcome including anaemia.

Anaemia, one of the malaria/STH infection outcomes, is well known to impaired cognitive development, reduced physical work capacity and in severe cases increased risk of mortality particularly during prenatal period (15). Anemia in pregnant women is defined hemoglobin levels less than 11g/dL (16).The connection between malaria and anemia in pregnancy is well established (17); hookworms have also been shown to be associated with anemia in pregnancy (18). *A.lumbricoides* and *T.trichuria* typically have little impact on iron status. Because the mechanisms by which malaria and intestinal helminth infections cause anemia differ, it is possible that their impact on anemia are additive (18, 19) and could exacerbate adverse birth outcomes (20). Although the specific biological processes are not clearly delineated, the contribution of moderate and severe anaemia to poor oxygen transport to the developing fetus is a likely mode of action for anemia's adverse effect on fetal growth. In addition, malaria-associated anemia in the mother likely has important consequences on her outcome whereby already anemic women are at increased risk of severe consequences such as hypotension, shock and death (21).

Now days, peoples are interested to know the biological association between plasmodium and helminth infection. Additonally, great attention is also given to determine the potential impact of co-infection on hemoglobin levels and the relevance of an integrated approach to controlling anaemia. However, the epidemiologic evidence base on co-infection is currently inadequate; therefore there is a need for intensive epidemiological and parasitological studies to exactly resolve key scientific questions.

Even though a lot of studies have been done in Ethiopia to determine malaria, STH infection and anaemia prevalence in different communities of the country (22, 24, 25, 26, 27, 28) there is no adquite published literature with regard to the prevalence and associated risk factors of malaria and helminthic co-infection as well as co-infection associated anaemia among pregnant women in Gilgel-Gibe dam area, southwestern Ethiopia.

1.2 Statement of the problem

Pregnant women are under-studied group, but vulnerable to many infections. As a result parasitic as well as non parasitic diseases can easily affect them. This is basically due to suppression of the immune system during pregnancy (2). In sub-Saharan Africa up to 24 million women may become pregnant each year (29). Because pregnancy is a time of high hormone activity which may exert immuno-suppressive effects on the child bearing woman, this high rate of pregnancy is often related to an increase in the susceptibility of pregnant women to parasitic infections (2).

As far as parasitic diseases are conserned, the most significant burdens of parasitic diseases are caused by soil-transmitted helminths (STHs): *A. lumbricoides, T. trichiura, N. americanus* and *A. duodenale* as well as the plasmodium species: *P. falciparum* and *P. vivax.* To date, approximately one third of the world's population is infected with at least one species of STH, with *A. lumbricoides* infecting 800 million people, *T. trichiura* 600 million, hookworm 600 million and resulting in up to 135,000 deaths annually (30). Recent World Health Organization (WHO) estimates indicate that 3.8 billion persons infected, 720 million clinical cases, and an estimated 135,000 deaths attributed to clinical complications annually (54). An estimated 40 million pregnant women were infected with STHs and schistosomes (32). In Ethiopia, the prevalence of hookworm was estimated to be 16% and the prevalence of *A. lumbricoides* and *T. trichiura* are 37% and 30%, respectively (31).

According to the World Malaria Report released by the WHO in 2008, there were 247 million malaria cases among 3.3 billion people at risk in 2006 from 109 countries resulting in an estimated 881,000 deaths. These deaths were primarily in Africa (91%) where a person dies of malaria every 10 seconds (33, 34). It is estimated that 40% of the world's pregnant women are exposed to malaria infection during pregnancy (35). It is also well known that, malaria is the leading cause of miscarriages among pregnant women and poses great danger to the mother and the unborn babies. The danger to the unborn is mainly because of the fact that *P. falciparum* strains exist which have specifically high affinity to placental receptors such as choriondroitin sulfate A (CSA) and as a result the parasites are sequestered in the placenta. Therefore, in addition to causing severe disease to the mother, malaria parasites hide in the placenta and interfere with the transfer of oxygen and nutrients (food) from the mother to her unborn baby; thereby causing nutrition and growth defects and other negative effects to the unborn baby. This is often fatal if not attended in time (36). It was also known to be the fourth

leading cause of death in children under the age of five years and pregnant women in developing countries such as Ethiopia(37).

In Ethiopia, malaria was reported as the primary cause of health problems where an estimated 68% (50 million people) of the population lives in areas at risk of malaria. The diverse eco-climatic condition in the country makes malaria transmission pattern seasonal and unstable usually characterized by frequent focal and cyclic widespread epidemics (38). In 2003 malaraia was the primary cause of reported morbidity and mortality accounting for 15.5% of outpatient visits, 20% of hospital admissions and 27% of hospital deaths (20). In 2005, malaria was still the first leading cause of health problem accounting for 48% of outpatient consultations, 20% admissions and 24.9% inpatient deaths (39). The annual average number of malaria cases reported over the period from 2001 - 2005 was 9.4 million (range 8.4 - 11.5) while the annual average number of confirmed cases was 487,984 (range 392,419 - 591,442). *P.falciparum* and *P.vivax* are the main species accounting for 60% and 40% of malaria cases, respectively in almost all regions of Ethiopia (40).

In Oromia regional state, 82% of the districts (*woredas*) are malarious and malaria threatens nearly 14.4 million (65.2%) of the population. Annually, about 1.5 million cases are reported from malaria control clinics and general health services; constituting an average of 20% of all diagnosis. Evidences from retrospective study of the magnitude of malaria admission and death review in Oromia revealed that out of total 302,035 admissions, 16,061 deaths were registered from 1995 to 2000 (41). Malaria in the region accounts for about 17% of outpatient visits, 15% of admissions and more than 25% of hospital deaths. More than 50% of the annual malaria cases and deaths in Oromia were reported mainly from East Shewa, Arsi, West Arsi and Jimma zones where intestinal helminth infection is also endemic(22).

With helminths and malaria infections endemic through most of Africa, communities often endure infections with a number of different parasite species and individuals are often co-infected with combinations of helminths and malaria parasites (23, 26). The rates of co-infection may not only depend on chance, but also upon the spatial distribution of environmental conditions that favor transmission of multiple species, as well as upon immunological interactions and common factors in genetic susceptibility or host behavior. Immunological factors are expected to influence rates of co-infection because helminths modulate host immune responses both to themselves, and to concurrent infections. That is why pregnant women, immunologically compromised, are highly susceptible for parasitic infections such as malaria and Soil transmitted helminthiasis (42).

Furthermore, as far as disease epidemiology and its consequences are considered, in sub-Saharan Africa, malaria in pregnancy contributes to 15% of maternal anemia, 14% of low-birthweight (LBW) infants, 30% of preventable LBW, 70% of intrauterine growth retardation, 36% of premature deliveries, and 8% of infant mortality (17). These all complications of malaria were more common in primigravidae than multigravidae (4, 17) also known to become more severe when pregnant women with malaria are also infected with helminths, even though it is found that helminths parasites reduce cerebral malaria (43, 44). Therefore pregnant women living in Ethiopia and other parts of the world where malaria and intestinal helminth are co-endemic are continuously facing this challenge.

In Ethiopia, malaria and intestinal helminthes cause hundreds of thousands of avoidable deaths each year. Therefore, malaria and helminthiasis are major public health problems in country, especially for pregnant women in whom the outcomes of co-infection including anaemia are more pronounced (26, 38). It has been estimated that, at any one time in developing countries, half of the population (mainly children and women of reproductive age) is affected by anemia (45). In Ethiopia, anaemia is the most frequent morbidity among pregnant women with the prevalence raging from 23–66.5%, in which anaemia among pregnant women was consistently higher in the rural women compared to the urban counterparts (24, 25).

In general Pregnancy and parasitic infection, which results in different complications including anaemia, are prevalent in Ethiopia where they frequently coexist. Many studies have indicated that malaria and helminths infections as well as anaemia are common in the country where there is only 27% ANC covrage(46), in Oromia region, especially Jimma zone which includes Gilgel Gibe dam area (22, 24, 25, 47, 48). However, there is no recent and aduqate community based study done with regard to the prevalence and risk factors of malaria and helminthic co-infection as well as co-infection associated to anaemia among pregnant women in Gilgel-Gibe dam area, southwestern Ethiopia. Thus, the main purpose of this study is to fill this information gap and generate baseline information on the prevalence and associated risks factors of malaria/soil transmitted helminthes co-infection and anaemia among pregnant women living around Gilgel Gibe area, where malaria and other helminthes parasites are co-endemic.

1.3 Significance of the study

Malaria and soil transmitted helminthes infections take the major share of Ethiopian public health problems; especially in those vulnerable community members, pregnant women. These parasitic infections have a lot of consequence in pregnant women which ranges from a simple abdominal disconfert up to death. Therefore there is a need to design effective and consistent strategies to solve pregnant women's health problems and to upgrade their health status.

Research findings are then the power to exactly understand the existing problems and to design strategies for solving/alleviating those problems, in this case pregnant women health problems. Therefore, the significance of this study is to avail important information about the current prevalence and associated risk factors of malaria and soil transmitted helminths co-infection as well as anaemia among pregnant women living around Gilgel Gibe dam area, south western Ethiopia. As a result, the findings of this study can help to design intervention strategies to control malaria, helminthic infection and anaemia in pregnant women. In fact this study also contributes to the existing body of knowledge on association between malaria and helminthiasis.

Chapter 2

Literature review

2.1 Malaria among Pregnant Women

Malaria is one of the major causes of morbidity and mortality in tropical regions like sub Saharan African and Asian countries. Because of the favorable and conducive environment, the parasite and the vector population sustain well in these regions. As a matter of facts, malaria prevalence is highest among the poorest sections of the society, since they cannot afford protection from malaria through improved housing, clean environment and are particularly vulnerable to the impact of ineffective diagnosis and treatment (3, 49). Despite progress in fighting malaria worldwide, the parasitic disease kills close to 800,000 People annually. The disease accounts for an estimated loss of 44.7 million disability adjusted life years (DALYs), more than 80% of which are currently concentrated in sub-Saharan Africa (49). Pregnant women and newborn children are the worst sufferer of malaria as these two groups are highly susceptible to malaria infection. Each year, approximately 50 million women living in malaria-endemic countries throughout the world become pregnant. An estimated 10,000 of these women and 200,000 of their infants die as a result of malaria during pregnancy, and severe malarial anaemia contributes to more than half of these deaths (50).

Study done on pregnant women attending ANC in Thai- Burmese border indicated that from the total of 829 pregnant women 186 (22.4%) were infected with plasmodium species(59). Similarly a cross sectional study done in Sudan indicated that from a total of 744 pregnant women attended the antenatal clinic of New Haifa Teaching Hospital 102 (13.7%) of women were infected with *P. falciparum*, 18(17.6%) of these were severe cases (jaundice and severe anaemia). Malaria prevalence and intensity (parasite count) were not significantly different amongst the different gravidity sub-groups (P > 0.05). The highest prevalence (18.3%) occurred in grandmultigravidae and the highest intensity (11,511 parasites/ μ L) was observed in primigravidae (79). Another cross-sectional study done in a sample of > 700 pregnant women in Ghana determined 36.3% malaria infection prevalence(58) while related study done on 2,104 near-term pregnant Nigerian women indicated that, 816 (38.8%) were found to be infected with malaria parasites(51).

In Ethiopia, malaria was reported as the leading cause of health problems in which an estimated 68% (50 million people) of the population lives in areas at risk of malaria (38).

The Ministry of Health (MOH) stratifies the country into 3 levels with respect to malaria transmission: 1) nonmalarious (25% of the country); 2) highland (also referred to as "unstable malaria"; approximately two-thirds of the country); and 3) endemic transmission with seasonal peaks (the remainder of the country) (52)In cross-sectional studies done to determine the prevalence of malaria among pregnant women at a national level including three zones of Ethiopia the overall prevalence of malaria within the stable transmission areas was 10.4%(53).

2.2 Soil Transmitted Helminthiasis among Pregnant Women

Soil-transmitted helminths (STHs) infections are major global health burden, with an estimated 3.8 billion persons infected, 720 million clinical cases, and an estimated 135,000 deaths attributed to clinical complications annually (54). Prevalence and distribution of helminth infection varies by place, with age and immunological status. Due to their weak immunity pregnant women are more susceptible to helminthes infection (2).

Intestinal helminthes are common sources of illness for pregnant women in which an estimated 44 million pregnant women are infected with hookworm worldwide, with 7.5 million in sub Saharan African alone. Hookworm infection is considered a major health threat to adolescent girls and women of reproductive age, with adverse effects on the outcome of pregnancy (55). Hotez and his colleagues indicated in their review that, 30% of Kenyan, 41% of Nepalese, and 53% of Vietnamese women of reproductive age had a hookworm infection (56). Interestingly a population based study done on prevalence and risk factors for soil-transmitted helminthes infection in mothers and their infants in Butajira, Ethiopia, also has indicated that; the general prevalence of STH infection in mothers was 43.5% in which hookworms were found in 36.1% and *A. lumbricoides* was found in 8.8% of mothers (26).

Related cross sectional study done to determine the prevalence of STH among Pregnant Women in Rural Western Kenya reviled high prevalence of helminthic infection, in which 76.2% of the study subjects were infected with at least one STH (57). Similar studies done in Nigeria, Ghana and Thailand had reported the prevalence of STH among pregnant women to be 48.3%, 25.7% and 70%, respectively (51, 58, 59).

2.3 Malaria and STH co-infection among Pregnant Pomen

Pregnant women living in malaria-endemic areas are highly susceptible to malaria, and this is true especially in primigravid women. A study carried out in Gabon, a state in west central Africa, reported *P. falciparum* prevalence of 57% in pregnant women, with primigravidae women having significantly higher prevalence (64%) than women in their second pregnancy (40%) (4). It was estimated that over a third of the world's population,

mainly in the tropics and sub-tropics, is infected with parasitic helminthes and Plasmodium species (34).

In sub-Saharan Africa, where intestinal helminthes infection and malaria are endemic the overlapping distribution of these parasitic infections results in high rates of co-infection (5). Hospital based studies were conducted to investigate the occurrence of Plasmodium/intestinal helminth co-infections among pregnant Nigerian women, and their effects on birthweights, anaemia and spleen size, in which prevalence of co-infection found to be more than 18%. In the study, from 2,104 near-term pregnant women examined, 816 (38.8%) were found to be infected with malaria parasites. Among the 816 parasitaemic subjects, 394 (48.3%) were also infected with intestinal helminths, 102 (12.5%) having mixed helminth infections. The prevalence of the helminth species found in stool samples of parasitaemic subjects examined was, *Ascaris lumbricoides* (19.1%), hookworm (14.2%), *Trichuris trichiura* (7%) *Schistosoma mansoni* (3.4%), *Enterobius vermicularis* (2%), Hymenolepis sp. (1.6%) and Taenia sp. (1%) (51).In a similar study done in Ghana prevalence of co-infecto was found to be16.6% (58).

In Ethiopia, similar study was done to determine the prevalence of malaria/helminth coinfection and the associated problems among febrile outpatients attending Alaba Kulito Health Center, southern Ethiopia in 2007. In the study a total of 1802 acute febrile patients were diagnosed for malaria in which 458(25.4%) of them were positive for malaria. Co-infection with Plasmodium and helminth parasites is associated with higher anaemia prevalence than single infection with Plasmodium parasites. And this difference was also significant for haemoglobin concentration, in which patients co-infected with *Plasmodium* and helminth parasites showed lower mean haemoglobin concentration (19).

As far as co-infection is conserned there are two factors in which co-infection of malaria and intestinal helminths depends; 1) the overall prevalence of individual species and 2) the degree of association between different species. If infection with *P. falciparum* and helminths are independent, occurrence of co-infection is simply determined by the relative frequency of individual species. Thus, the age patterns of co-infection will depend on the age-specific prevalence rates as predicted by simple probability. However, if co-infection is either synergistic or antagonistic, occurrence of both parasites would be significantly different from that predicted by individual chance encounters with either infection. Such associations may arise because of biologic associations, whereby the presence of one species promotes or inhibits the establishment and/or survival of the second species, potentially through immune modulation (19, 60). Alternatively, co-infection may reflect concurrence of common socio-economic and/or environmental risk factors promoting survival of both species (8, 10).

Having the above hypothesis as a base, recent studies have suggested that helminth infections may adversely influence susceptibility to other infections, including malaria (43). To further investigate this hypothesis in a sub-Saharan African setting, surveys of helminth infections were conducted in 2003 among individuals who had been under weekly active case detection for clinical malaria during the preceding 18 months in four villages in Kabale District, southwest Uganda. Overall, 47.3% of individuals had at least one intestinal nematode species infection: hookworm, *Ascaris lumbricoides* and *Trichuristrichiura* were detected in 32.1%, 17.4% and 8.1% of individuals, respectively. Evidence of significant household clustering of *A. lumbricoides*, *T. trichiura* and hookworm and clustering of heavy infection of each species were found. The association between helminth infection and clinical malaria was investigated in two villages and no evidence for an association was observed between the presence of infection or heavy infection and risk of malaria (61).

In contrast, another laboratory study indicated that there is evidence suggestive of both synergism and antagonism in nematode and protozoa infections, and this may have implications for the epidemiology of multiple parasite species in humans (43, 62) Early studies suggested that *Ascaris lumbricoides* infection might be protective against malarial disease, (13, 14) in contrast to several later reports, which suggested that STH infections may increase the risk of malaria infection (10, 12). Epidemiologic observations now suggest a range of scenarios in which helminth infections may increase susceptibility to *Plasmodium* infection but may also under certain circumstances protect against severe malaria (60).

The study done among Nigerian pregnant women indicated that when compared with uninfected women, co-infection was associated with a 3-fold, 2.6-fold, and 3.5-fold increased risk of low birth weight (LBW), pre-term delivery (PTD) and small-for gestational age (SGA), respectively in women with anemia, and was associated with an almost 3-fold increased risk of anemia. A comparison of women with co-infection women with women infected with intestinal helminths only showed that risks of LBW, SGA, and anemia were exacerbated by co-existent helminths. Similarly, when compared with women infected with malaria only, co-infection substantially increased the risk of anemia (51).

In Ethiopia, the study done in Alaba Kulito Health Center, southern Ethiopia, revailed that only few cases of helminth and malaria co-infected patients showed signs and symptoms of severe malaria as compared with severe manifestation in patients infected with plasmodium parasite alone(19). This indicates that helminth infection provides some degree of protection against the development of severe malaria which is also in agreement with the previous reports (43, 62).

In the above study, light infection with *A. lumbricoides* was associated with higher *Plasmodium* parasitaemia, while those with heavy *A. lumbricoides* infection were found to have lower parasitaemia. However, patients infected lightly with hookworm had significantly lower *Plasmodium* densities compared to heavily or moderately infected cases suggesting positive interaction between hookworm infections with *Plasmodium* densities. Nevertheless, increase in *Plasmodium* density was found to be strongly related to the number of helminth species rather than to the intensity of helminth infection. \the study showed that patients with two or more helminth species were associated with higher density of malaria parasites. Although their reports were for general helminth infections, the work of LeHesran *et al* (2004) also showed a positive association between worm load and Plasmodium densities. Thus, it appears that the type of helminth involved, the intensity of infection and age influence the relationships between malaria susceptibility and helminth infection (19, 63).

As far as the risk factors of malaria and intestinal helminths co-infection is concerned, the study done in Ghanaian pregnant women to identify risk factors for co-infection identified that, being single, young age, low income, short pregnancy interval and primigravidae were risk factors for malaria and intestinal helminth co-infection. Young age at pregnancy was strongly associated with an increased risk of co-infection. Single women and primigravid women also had an increased risk of co-infection. Young primigravid women had 5.2-fold increased rate of co-infection, whereas the rate of co-infection for multigravid women was 3.2. Weekly income of < 200,000 cedis was associated with 2.4-fold increased rate of co-infection among multigravid women and 1.6 times increased rate among primigravidae. Single primigravid women had 3.1-fold increased rate of co-infection for multigravid as the rate of co-infection for multigravid women (58). Findings from other studies also showed that younger and primigravidae women are the most susceptible to *P. falciparum* infection (4, 58, 64, 65).

On top of the above reality, study findings also showed that in many sub-Saharan African countries women consume soil during pregnancy; a study done in Kenya found that 73% of pregnant women ate soil regularly in which an overall prevalence of 25.7% was observed for intestinal helminth infections (66). Related cross sectional study done in Ghana revealed a prevalence rate of *A. lumbricoides* (12.3%) and hookworm (7.9%) which was higher than those of other helminthes compared to the observation that have been made in other studies in sub-Saharan Africa. The relatively high prevalence of helminth infections in Ghanaian population was considered to be indicative of poor sanitation and improper sewage disposal, as shown by the fact that 37.9% of the participants had no toilet facilities in their homes (58).

Despite lack of adequate data about the rate and risk factors of malaria and soil transmitted helminths co-infection among pregnant women in Ethiopia it is well known that poor sanitation, improper sewage disposal, lack of latrine and contaminated water and food are the common risk factors for the overall intestinal parasitic infections(16, 30). The common risk factors for malaria infection include improper sewage disposal, impropore ITN utilization and the presence of stagnant water. On top of these all factors the low economic status of the people and the climatic condition which fover the spread of helminthes, plasmodia and mosquito vectors, are also known to be risk factors for these parasitic infections in Ethiopia (19, 31, 39, 47, 68).

2.3 Anaemia among pregnant women

Anaemia is a condition in which the number of red blood cells or their oxygen-carrying capacity is insufficient to meet physiologic needs, which vary by age, sex, altitude, smoking, and pregnancy status. In its severe form, it is associated with fatigue, weakness, dizziness and drowsiness. Prevalence of deficiency anaemias in healthy teenage population can be an important indicator of general nutritional status. Iron deficiency is thought to be the most common cause of anaemia globally, although other conditions, such as folate, vitamin B12 and vitamin A deficiencies, chronic inflammation, inherited disorders and parasitic infections can all cause anaemia (11).

Parasitic diseases, including P. falciparum and helminth infections, have long been recognized as major contributors to anemia in endemic countries. Malaria contributes to reduce hemoglobin concentrations through a number of mechanisms, principally by destruction and removal of parasitized red cells and the shortening of the life span of nonparasitized red cells, and decreasing the rate of erythrocyte production in the bone marrow (12). Some of the mechanisms that cause anemia during malaria are associated more with the acute clinical states (e.g., hemolysis or cytokine disturbances), whereas chronic or repeated infections are more likely to involve dyserythropoiesis (69). The effects of infection with a single helminth species on the risk of anemia are also well documented, with risk correlated to infection intensity (70). Hookworm causes iron deficiency anemia through the process of intestinal blood loss through nitric oxide (NO) release. Because NO can reduce erythrocyte deformability, it could lead to increased red blood cell destruction (71). Ascaris lumbricoides and Trichuris trichuria typically have little impact on iron status. Because the mechanisms by which malaria and intestinal helminth infections cause anemia differ, it is possible that their impact on anemia are additive (18) and could exacerbate adverse birth outcomes (58).

Although the vast majority of women with malaria infection during pregnancy remain asymptomatic, infection increases the risk of maternal anemia and delivering a low-birth-weight baby. Malaria due to *P.falciparum* also clearly contributes to anemia throughout

life and specifically during pregnancy. It is estimated that in sub-Saharan Africa 23 million pregnant women are exposed to malaria infection annually and approximately 400,000 pregnant women develop moderate or severe anemia (hemoglobin <80 g/L or hematocrit <0.25) each year in sub-Saharan Africa as a result of malaria infection (72). Women in their first and second pregnancies living in an endemic area are at higher risk of acquiring malaria than non-pregnant or multigravidae, due to reduction of an appropriate immune response to the malaria parasite (4).

Soil transmited helminthes especially hook worm is well known to cause anaemia. Data from the early 1990s suggest that 44 million of the developing world's 124 million pregnant women harbored hookworm infection (73); with 7.5 million in sub–Saharan Africa alone. Hookworm infection is considered a major health threat to adolescent girls and women of reproductive age, with adverse effects on the outcome of pregnancy (55). Hookworm infections induce deficiencies of iron, total energy, protein and possible folate and zinc (74). Severe iron deficiency anaemia during pregnancy has been linked to increased maternal mortality, impaired lactation, prematurity and low birth weight (55).

Estimates in Kenya and Nepal suggest that hookworm infection causes 30% and 41%, respectively, of moderate or severe cases of anaemia among pregnant women (haemoglobin level, < 9 g per deciliter). The association between hookworm infection and anaemia is greatest in multigravidaes (55, 56). Studies in Africa and Asia reported a higher prevalence of anaemia and its association with women of age < 20 years, third trimester of pregnancy, rural residents and multi-parous women (58, 75).

Anaemia in pregnancy is also related to different socio-demographic, dietary and economic factors. Mother's age < 20 years, educational status, economic position, and antenatal care were significantly associated with anaemia during pregnancy in a study conducted in India (76).

In general anaemia is globally hot topic of research and it has been extensively studied throughout the world. The overall prevalence of anaemia is about 30% world wide and 36% for developing countries. Among all groups, young children and pregnant women are most affected with an estimated global prevalence of 43% and 51%, respectively. The prevalence of anemia in pregnant women reaches 65% in South east Asia and 63% in Africa (77). In Ethiopia, different anaemia prevalences were found among pregnant women attending in ANC in Jimma and Bushulo health centers, which includes; 41.9% and 51.9% respectively (27, 28).

In the study done in Jimma health center, Ethiopia of the overall prevalence of anaemia (41.9%), the prevalence of anaemia in rural and urban residents were 56.8% and 35.9%, respectively. The mean haemoglobin level was 10.9 gm/dl and 6.4 gm/dl for the whole group and anaemic women, respectively. The majority (74.3%) had moderate anaemia while 2.5% had severe anaemia. In this study, it was found that the rate of anaemia was higher among the illiterate and increased with parity.In the related study done to determine the prevalence of intestinal parasite and anaemia among pregnant women in Bushulo health center, Tula woreda, Southern Ethiopia; the total parasitic infection and anaemia were 58.2% and 51.9%, respectively. Moreover, mean haematocrite level was 34%, anaemic women were 14 times likely to have hookworm plus other intestinal helminthic infection and 2 times likely not to have shoe wearing habit during pregnancy (27, 28).

Generally, there is poor understanding of the association between malaria and helminths infection and thier effect on anaemia, as well as the basic factors for their co occurrence especially in pregnant women; in which outcomes of the findings can help to improve health status of these most valunrable groups. To achieve this goal there is a need to do extensive study on this area globally as well as at a national level. Despite of this fact, there is no published literature on the prevalence and associated risk factors for co-infection of malaria and intestinal hemminth in pregnant women in Ethiopia in general and in Gilgel Gibe area in particular. Therefore, this study is intended to investigate the prevalence of malaria and intestinal helminthes co-infection among pregnant women in Gilgel Gibe area and to assess the possible risk factors for these co-infections so as to fill the existing knowledge gap.

Chapter Three

Objectives

3.1. General Objective

The main objective of this study is to determine the prevalence of malaria/soil transmitted helminthes co-infection and anaemia among pregnant women in Gilgel Gibe area and to assess the associated risk factors for these infections and anaemia.

3.2. Specific Objectives

- To determine the prevalence of malaria among pregnant women in Gilgel Gibe dam area.
- To determine the prevalence of soil transmitted helminthes infection among pregnant women in Gilgel Gibe dam area.
- To determine the prevalence of malaria-helminth co-infection among pregnant women in Gilgel Gibe dam area.
- To determine the prevalence of anaemia among pregnant women in Gilgel Gibe dam area.
- To assess the association of malaria- soil transmitted helminth co-infection with anemia.
- To assess associated risk factors for malaria and soil transmotted helminths coinfection.

Chapter Four

Methods and Materials

4.1. Study area

The study area, Gilgel Gibe Dam area, is located 260 km south-west of the capital, Addis Ababa in Oromia Regional State, south-western Ethiopia near Gilgel-Gibe hydroelectric dam which started operating in 2004. It includes six urban kebeles and 6 rural kebeles which are located in Serbo (Kersa), Omo-Nada and Tiro-Afeta districts (*weredas*) with 10,800 households and a total population of 55,000. The study area lies between latitudes 7°42'50"N and 07°53'50"N and between longitudes 37°11'22"E and 37°20'36"E, at an altitude of 1,672-1,864 m above sea level. The area has a sub-humid, warm to hot climate, receives between 1,300 and 1,800 mm of rain annually and has a mean annual temperature of 19°C. The rainfall pattern of the area is similar to other parts of Ethiopia with the long rainy season starting in June and extending up to September, while the short rainy season begins in March and extends to April/May. (47).

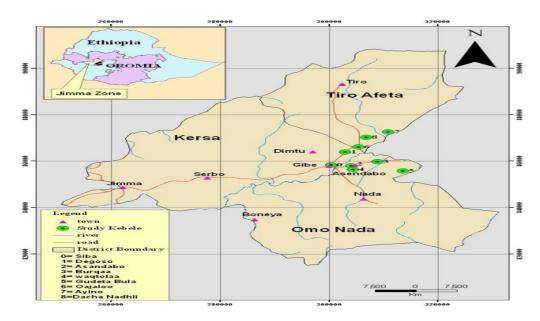


Figure 1. Map of the study area

4.2. Study design and period

A cross-sectional community based study was conducted in Gilge Gibe Dam area, southwestern Ethiopia from august to September/2011. Participants from rural and urban kebeles were assessed through house to house visit.

4.3. Population

4.3.1 Source population

All the pregnant women found in the study area, Gilgel-Gibe hydropower dam area, were the source population.

4.3.2 Study population

All the pregnant women in the randomly selected six rural and six urban kebeles were the study population.

4.3.3 Study Participants

All the pregnant women who fulfill the inclusion criteria; randomly selected from the sampling frame which contain list of pregnant women in the six randomly selected kebeles were the study participant. Those study participants were involved in the assessment of malria/STH co-infection and anaemia.

4.4 Sample size determination and Sampling technique

4.4.1 Sample size determination:

Sample size of the study was determined using a formula for a single population proportion with 95% level of confidence as follows:

$$n = (p.q \ (Z\alpha/2)^2) \ /d^2$$
$$\frac{(0.5)(1-0.5)(1.96)^2}{(0.05)^2} = \underline{385}$$

Applying the sample size correction formula using the estimated total pregnantwomen population of 4000, the corrected sample size was calculated to be:

$$(Fn=n/1+(n/N))=Fn=385/(1+(385/4000))$$

<u>388</u>

Where:-

n = The number of study participants; i.e. sample size of the study

P = Proportion of malaria and STH co infection among pregnant women (0.5)

- q = Compliment of P (1-p)
- Z = Standardized normal distribution curve value for the 95% CI(1.96)
- d = The margin of error taken (5%)
- 4.4.2 Sampling technique:

:

From the total of 23 kebeles found around Gilgel Gibe dam area, six rural and six urban kebeles were selected by stratified random sampling. The calculated sample size was proportionally distributed to the randomly selected 12 kebeles according to the total pregnant women with in each selected kebele as shown in figure 2. The study participants were then randomly selected using systematic random sampling from the sampling frame which was constructed after identifying the pregnant women in using physical examination and HCG test for those who are suspected.

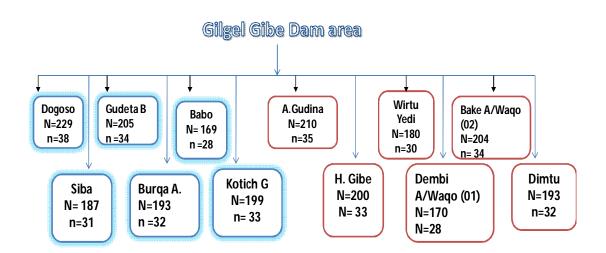


Figure 2. Profile of study participants in Gigel Gibe area, 2011.

4.5 Inclusion and Exclusion Criteria

4.5.1 Inclusion criteria

All the pregnant women randomly selected from the six town kebeles and six village kebeles who were at home during or who visit the health center during sample collection was included in the study.

4.5.2. Exclusion criteria

Pregnant women who take antimalarial treatment with in the past 28days and/or began antihelminth treatment in the past 14 days and those who did not give the consent were excluded from the study. In the urban set up pregnant women who came out of these randomly selected six town kebeles were excluded.

4.6 Data Collection and Processing

4.6.1 Data collection using Questionnaire

A semi structured questionnaire was developed in English version and then translated into Amharic and the local language of the study area, Afan Oromo. Comparisons were made on the consistency of the three versions. The questionnaire was further modified after a pretest by trained data collectors on 5% of participants and any ambiguous questions as well as repetitive ideas were corrected. Additional response categories were also added based on the pretest findings. (Annex 4)

The translated Oromiffa version was administered to identify the revalence and possible risk factors of malaria/ STH co-infection and anaemia in the study area. The questionnaires address the individuals' socio-demographic information, use of malaria and STH preventive measures, housing conditions, their knowledge and attitude concerning malaria and soil transmitted helminthes transmission and other related issues to assess some possible risk factors for the co-infection.

4.6.2 Blood sample collection & processing for malaria and anaemia

A. Blood sample collection & processing for malraia assessment

Within the same day when the participants donate their stool sample, capillary blood sample was also collected from all the 388 pregnant women following aseptic technique. During blood collection from individuals the glass slides were labeled in the field as well as in the health centers in such a way that the slide code could match with the stool container and questionnaire lebel. Then, the finger was cleansed with alcohol-moistened

swab, dried with a piece of dry cotton, punctured with a disposable blood lancet. Through wiping off the first drop of blood, thick and thin films were made on the same slide. After being air-dried in a horizontal position, the slides were placed in a slide box and carefully transported to Asendabo Health Center where parasitological test was carried by experienced laboratory technicians and technologists.

Before staining the blood films, the thin blood films were fixed in methanol for 30 sec. Then smears were stained with 10% Giemsa solution for 10 minutes. The staining techniques and blood film examination was conducted employing WHO guid line. Microscopic examination of thick films using high power magnification for the presence of parasites and parasite species identification using thin films under 100x oil immersion objective were done by experienced laboratory technicians and technologists. In addition to the qualitative examination, parasitic load was determined following WHO guide line.

B. Blood sample collection and processing for hematocrit measurment

Hematocrit is the volume of red cells expressed as a percentage of the volume of whole blood in a sample of capillary or venous blood. The purpose of this test is to determine the red blood cell mass by measuring space occupied by packed red blood cells. It is known to be the most reproducible hematological test (72).

The microhaematocrit method was carried out using heparinized hematocrit capillary tubes which are 75mm in length and having an internal diameter of about 1mm. Three-fourth of the capillary tubes were filled with capillary bood and sealed with wax in the other end of the tube. Blood sample with in the sealed hematocrit capillary tubes were centrifugated with 12,000 rpm speed for 5 minutes in semi-automatic centrifuge (HAMATOKRIT 210, Hettich ZENTRIFUGEN) to determine the packed cell volume, hematocrit. (annex 1)

4.6.3 Stool sample collection and processing

After getting their written consent stool samples were collected from participants. The study participants were provided with labeled screw caped stool cap and informed on how to collect about 5gram stool sample. The collected stool sample were soon transported to Jimma university clinical laboratory where it was processed following the standard procedure of McMaster concentration technique (annex 1). The type of parasite and parasitic load were recorded in the prepared laboratory format(annex1).

4.7 Operational Definition of Terms

- Anaemia A condition with HCT value lower than 33% for first and third trimester and less than 32% for second trimester
- **Co-infection** Infection with both malaria and intestinal helminth
- Gilgel Gibe Dam area- Area which includes 6 rural and six urban kebeles around Gilgel Gibe dam
- > Hand washing- washing hands before handling food
- Latrine service- having access to latrine which can be private or communal
- ▶ Mild anaemia- HCT value lower than33% and greater or equal to 30%
- Moderate anaemia- HCT value lower than 30% and greater or equal to 25%
- Severe anaemia- HCT value lower than 25%
- > **Prevalence** The proportion of infected pregnant women
- **Primery school** -1^{st} -8th grade
- **Secondary school** -9th and above
- **Resedance** –staying more than half a year
- Risk factors- Factors that enhance the chance of malaria and intestinal helminths co infection
- Soil transmitted helminthes- Helminth including *A.lumbricoides*, H.worm, *T.trichuira*, *E.vermicularis* and Hymenolopis spp.
- Stagnant water- Accumulated water less than 2 km from the respondants' house, in which mosquitoes can breed

4.8 Data analysis

The laboratory result formats and the questionnaire was, coded and checked for completeness and cleaned of any inconsistencies. It was entered to excel and analyzed by SPSS-16 database program. For the demographic data the descriptive statistics were used to give a clear picture of background variables like age, sex, residence and other variables in structured questionnaires. The frequency distributions of both dependent and independent variables were worked out and the association between each independent and dependent variables were measured and tested using Pearson's chi square, with 95 % confidence interval and 5% level of precision. To determine risk factors for co-infection, multiple logistic regressions were used analyzing only those predictors with statistically significant association with the dependent outcome. Finally, the data was described and presented using tables and charts.

4.9 Quality Assurance

To insure the representativeness of the study units sampling frame were constructed using house to house survey before data collection. Prior to data collection the questionnaire was translated to the local language, pre tested and the data collectors were trained well by the principal investigator how to go for data collection and collect appropriate raw data.

During data collection (interview) any unclear ideas and terms was explained well for the respondents in local language. For better quality of the laboratory results, fresh and sufficient quantities (5g) of stool samples as well as appropriate blood samples (capillary blood) were collected. The quality of Giemsa stain was also checked using known positive samples and more than one hundred high power fields of the thick film was examined at a magnification of 1000×, before identifying a slide as negative or positive.Parasitic load was counted following the WHO guide line by well expreinced laboratory technicaines and technologists.All the positive and 10% of randomly selected negative blood films were rechecked independently by blinded senior laboratory technologiest in Jimma University specialized hospital. Stool samples where processed with in the date of collection using appropriate floatation solution and results where rechecked by the principal investigator. The appropriateness of haematocrite centrifuge and reader was checked.

After data collection process, the supervisors and principal investigator checked the questionnaires for completeness and consistency. Any incomplete or misfiled questionnaires were sent back to the respective data collector for correction. Laboratory results of malria diagnosis and McMaster concentration stool examination were recorded

on well prepared format carefully and finally were attached with the questionnaire. The data were lastly coded, entered to excel and analyzed by SPSS-16 database program.

4.10 Limitation of the study

The main limitations of the study were:

- Eventhough the procedure followed to measure hematocrit value for the assessment of anaemia was recommended by WHO, there may be interpersonal variation in reading the value from the reference scale.
- Stool samples with parasite load less than 50 ova/gram are reported as negative due to the limitation of McMaster to detect less than 50 ova/gram of stool.

4. 11 Ethical Consideration

Ethical clearance of the study was obtained from the Ethic review board of Jimma University and a permission letter from Jimma zone health office, Nada wereda heath office, Asendabo health center, Nada health center and Dimtu health center. The objectives and the nature of the study were explained to the participants so as to get their oral and written consent to be involved in the study voluntarily. To insure the confidentiality the data collected and results of laboratory tests were used only for this research purpose. Results of participants with parasitic infections, malaria and/or intestinal helminth, and low HCT level were sent, as soon as possible, to nearby health facilities for treatment and medical consultation in the ANC. Those infected pregnant women were referred for treatment. (Delivery format Annex 9)

Chapter five

Results and Discussion

5.1 Results

5.1.1 Socio demographic and socio economic characteristics of respondents

Table 1 presents the socio-demographic characteristics of the respondents. The minimum and maximum ages were 16 and 40, respectively with mean age of 25 years. Only 1(0.3%) was divorced while the rest pregnant women 387(99.7%) were living with their hasband.During the study 88(22.7%), 157(40.5%), 143(36.8%) were in their first, second and third trimester, respectively. The proportion of primigravida (pregnant for the first time) and multigravida were 125(32.2%) and 263(67.8%), respectively. Majority of the participants 372(95.9%) were Oromo in ethnicity and more than 92% of the respondents were Muslims.

As far as their housing conditions are considered, summarized in table 2, 72% of the pregnant women from urban resedence live in thatched house with their animals; including the urban setting the number of thatched houses was 148(38.1%). Laterine coverage was 99.2% with more than 50% of the latrines not supplied water for hand washing after using the latrine. Even though 62(16%) were living in an area where there is stagnant water, more than 87% of the pregnant women reported that, there were mosquitoes inside their house.

Conserning their water source, 171(44.1%) use tap water and the rest use other sources (Table 2). Two handred one (75%) of the pregnant women's house was sprayed with insecticides. ITN coverage with atleast one ITN in the study area was 348(89.7%), however, 40(10.3%) pregnant women had no ITN at all.

Characteristics		No.	%	Characteris	stics	No.	%
Residence	Urban	193	49.7	Occupation	Housewife	191	49.2
	Rural	195	50.3		Farmer	176	45.3
Age	16-20	128	33		Daily laborer	4	1.1
	21-25	115	29.6		Civil servant	13	3.31
	26-30	110	28.3		Business man	4	1.1
	31-35	27	7	Educational			
	36-40	8	2.1	status	Illiterate Read and write only	263 5	67.8 1.3
Ethnicity	Oromo	372	95.9		Primary school	83	21.4
	Amhara	4	1.1		Secondary school	18	4.6
	Yam	7	1.8		Above Secondary school	19	4.9
	Gurage	4	1.1	Marital	Married	387	99.7
	kembata	1	0.2	status	Divorced	1	0.3
Religion	Muslim	359	92.5				
	Orthodox	21	5.4	Trimester	First	88	22.7
	Protestant	6	1.6		Second	157	40.5
	Catholic	2	0.5		Third	143	36.8
.	Primigravid	125	32.2				
Parity	Multigravid	263	67.8				

Table 1. Socio demographic characteristics of respondents in Gilgel Gibe hydropower dam area, southwest Ethiopia, August to September 2011

Characteristics			Number	Percent (%)
House condition	Type of house	Thatched	148	38.1
	Dwelling	Corrugated iron sheet Same human/ animal dwelling Separate human/	240 142 246	61.9 36.6 63.4
	Presence of stagnant water	animal dwelling Yes No	62 326	16 84
	Presence of	Yes	339	87.4
	mosquitoes in the house	No	49	12.6
	ITN supplied	Yes	348	89.7
		No	40	10.3
	Sprayed with insecticide	Yes	291	75.0
		No	97	25.0
	Distance From the	Less than/=10Km	236	60.8
	Dam	>10Km	152	39.2
Latrine condition	Latrine presence Presence of Water for hand wash	Yes	385	99.2
		No	3	0.8
		Yes	186	47.9
		No	199	51.3
	Presence of Latrine opening cover	Yes	203	52.3
	opening cover	No	182	46.9
Water Source	Source of drinking water	Tap water	171	44.1
	water	Spring	86	22.2
		Protected well	74	19.1
		River water	53	13.7
		Unprotected well	4	1
Waste management	Liquid waste	Open field	281	72.5
	management	Pit	103	26.5
		Toilet	4	1.00

Table 2. Socio-economic characteristics and housing condition of respondents in Gilgel Gibe hydropower dam area, southwest Ethiopia, August to September 2011

As far as the study participants' knowledge and practices are concerned, most (93.6 %) of the participants have heared about malaria; on the other hand 16.5% pregnant women have no idea about soil transmitted helminths. All respondents knew at least one of the sign and symptoms of malaria, 358(92.3%) of respondentss knew that malaria is transmitted by mosquito bite. Contrary to this 114(29.4%) of the pregnant women didn't know about the transmission of soil transmitted helminthes and only 153(39.4%) of the participants were aware of the fact that walking bare foot can expose them for soil transmitted helminthes. All the study participants had a habit of washing their hands before meal surprisigly only 336(86.6%) had a habit of washing hands after using toilet. Two handred seventy three (70.4%), 253(65.2%) and 22(5.7%) of the respondents had a habit of walking bare foot, eating uncooked food and habit of eating soil, respectively. Thirty two (8.2%) of the participants used human feces as a fertilizer.

5.1.2 Parasite Prevalence and associated risk factors

5.1.2.1 Prevalence and Associated risk factors of soil transmitted helminthes (STH) among pregnant women

Of the total of 388 study participants, 159(41%) were infected with one or more helminth parasites. Helminthic infection was higher 94(48%) in sudy subjects from rural setting than respondents from urban setting 65(33.9%) (p<0.05). Pregnant women in the age group of 20 years or less were the most infected women. Hook worm was found to be the most prevalent STH with a total prevalence of 114(29.4%) followed by *A.lumbricoieds* 58 (15%) and *T.trichuiria*13 (3.4\%).

One handred twenty seven (79.9%) of the total STH infected women had single helminth infection, while 27(17%) had double infection, and the rest 5(3.1%) had triple infection. It was observed that, place of residence (p=0.003), occupation (p=0.003), type of house (p<0.001), animal dwelling (p<0.001), lack of water for hand washing within the toilet (p=0.008), habits of walking bare foot (p<0.001) were significantly associated with STH infection. (Table 3). Multivariable Logistic regression indicated that habit of bare foot waking (p=0.041) and animals dwell in the house(P=0.018) were the main risk factors for STH infection; in which pregnant women having the habit of walking bare foot were 1.7 times more likely to be infected by STH(AOR= 1.76, 95% C.I: 1.02-3.02, p=0.041). Additionally, pregnant women living with their animals in the same house were three times more likely to be infected by STH (AOR= 2.97, 95% C.I: 1.20-7.33, p=0.018).(Table4)

Variables		Soil transmitted helminths infection		s infection	P - value
		Negative	Positive	Total	-
Residence	Rural	102	94	196	0.003
	Urban	127	65	192	
House type	Thatched	70	78	148	<0.001
	Corrugated iron sheet	159	81	240	
Animal	Yes	62	80	142	<0.001
doweling inside house	No	167	79	246	
Age group	16-20	69	59	128	0.270
	21-25	68	47	115	
	26-30	70	40	110	
	31-35	15	12	27	
	36-40	7	1	8	
Habit of bare	Yes	144	129	273	<0.001
foot walking	No	85	30	115	
Trimester	First trimester	50	38	88	0.890
	Second trimester	94	63	157	
	Third trimester	85	58	143	
Parity	Primigravida	75	50	125	0.437
	Multigravida	154	109	263	

Table 3. Age-specific prevalence of soil transmitted helminths and risk factors of of STH infection among pregnant women in Gilgel Gibe hydropower dam area, southwest Ethiopia, August to September 2011

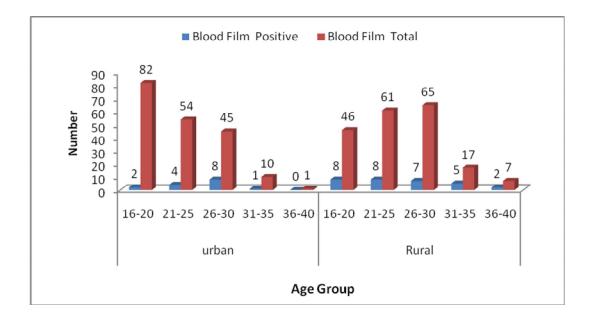
Predictors	AOR	95.0% C.I.		Р
		Lower	Upper	
Residence				
Rural	0.88	0.47	1.65	0.698
Urban	1.00			
Roof Type				
Thatched	0.76	0.30	1.92	0.567
Corrugated	1.00			
Bare foot waking				
Yes	1.76	1.02	3.02	0.041
No	1.00			
Animals dwell in the house				
Yes	2.97	1.20	7.33	0.018
No	1.00			

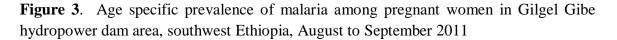
Table 4. Parameter estimates from multivariable Logistic regression model predicting the likelihood that a pregnant woman is infected with STH, Gilgel Gibe Dam area, southwest Ethiopia, August to September 2011.

AOR= Adjusted Odds Ratio, C.I= Confidence Interval

5.1.2.2 Prevalence and Associated risk factors of malaria among pregnant women

From the total of 388 blood films collected, 45(11.6%) were positive for malaria parasites. From the total malaria infection, infection with *P.falciparum*, *P.vivax* and both species were 32(71.1%), 7(15.6%) and 6(13.3%), respectively. Multigravida women were more infected 34(13%) with malaria compared to primigravida 11(8.8%) women. Malaraia was also found to be relatively more prevalent among rural 30(15.3%) than urban 15(7.8%) pregnant women. In rural setting pregnant women in the age group of less than or equal to 20 were the most infected group with prevalence of more than 17% as it is indicated in figure 3.





Minimum, maximum and mean malaria parasite load within the positive pregnant women were 80 plasmodium spp parasite / μ L, 36,480 plasmodium spp parasite / μ L and 3,877 plasmodium spp parasite / μ L respectively.

In this study, place of place of residence (p=0.021), presence of stagnant water near to study participants house (p=0.038), were significantly associated with malaria infection. (Table 5) In which only place of residence were found to be the main risk factor for malaria infection among pregnant women in the study area; where pregnant women living in the rural kebeles were two times more likely to be infected by malaria compaired to these pregnant women living in urban kebeles(AOR= 2.06, 95% C.I: 1.07-3.98, p=0.031). (Table 6)

Variables		Malaria			P - value
		Negative	Positive	Total	
Residence	Rural	166	30	196	0.026
	Urban	177	15	192	
House type	Thatched	126	22	148	0.080
	Corrugated iron sheet	217	23	240	
Presence of	Yes	50	12	62	0.038
stagnant water	No	293	33	326	
Age group	16-20	118	10	128	0.149
	21-25	103	12	115	
	26-30	95	15	110	
	31-35	21	6	27	
	36-40	6	2	8	
ITN utilization	Yes	308	40	348	0.507
	No	35	5	40	
Trimester	First trimester	77	11	88	0.107
	Second trimester	145	12	157	
	Third trimester	121	22	143	
Parity	Primigravide	114	11	125	0.154
	Multigravide	229	34	263	

Table 5. Age-specific prevalence of malaria and risk factors of of malaria among pregnant women in Gilgel Gibe hydropower dam area, southwest Ethiopia, August to September 2011

AOR		Р	
AOK			. 1
2.06	1.07	3.98	0.031
1.00			
0.49	0.238	1.03	0.059
1,00			
	2.06 1.00 0.49	AOR Lower 2.06 1.07 1.00 0.49 0.238	Lower Upper 2.06 1.07 3.98 1.00

Table 6. Parameter estimates from multivariable Logistic regression model predicting the likelihood that a pregnant woman is infected with malaria, Gilgel Gibe Dam area, southwest Ethiopia, August to September 2011.

AOR= Adjusted Odds Ratio, C.I= Confidence Interval

5.1.2.3 Prevalence of malaria and STH co-infection and associated risk factors among pregnant women

Overall, STH and malaria prevalence among pregnant women in the study area were 159(41%) and 45(11.6%), respectively where as the prevalence of STH and malaria co-infection among pregnant women was 30(7.7%) (Figure 4). STH and malaria co-infection were significantly higher (p<0.05) in pregnant women from rural setting 19(9.7%) than in pregnant women from urban setting 11(5.7%). Co- infection was more prevalent in those study participants within the age group 26-30 year. Presence of stagnant water (p=0.007), habit of eating soil (p=0.007) and habit of using human feces as a fertilizer (p=0.002) had statistically significant association with malaria and STH co-infection among pregnant women in Gilgel Gibe dam area. (Table 7). In which all the above three pridicators were the risk factors for the co-infection. Pregnant woman who lives in house which was near stagnat water is three times more likely to be co-infected (AOR= 2.99, 95% C.I: 1.28-7.00, p=0.012) (Table 8)

Variables		Malari	ia and STH co-ii	nfection	P - value
		Not co-infected	Co-infected	Total	
Residence	Urban	181	11	192	
	Rural	177	19	196	0.101
House type	Thatched	132	16	148	
	Corrugated iron sheet	226	14	240	0.058
Presence of	Yes	52	10	62	
stagnant water	No	306	20	326	0.011
Age group	16-20	120	8	128	
	21-25	105	10	115	
	26-30	100	10	110	0.826
	31-35	25	2	27	
	36-40	8	0	8	
Utilization of human feces as	Yes	25	7	32	0.007
fertilizer	No	333	23	356	0.007
Habit of eating soil	Yes	17	5	22	0.020
5011	No	341	25	366	0.020
Trimester	First trimester	81	7	88	0.675
	Second trimester	147	10	157	
	Third trimester	130	13	143	
Parity	Primigravide	117	8	125	0.324
	Multigravide	241	22	263	

Table 7. Age-specific prevalence of malaria/STH co-infection and risk factors of malaria/STH co-infection among pregnant women in Gilgel Gibe hydropower dam area, southwest Ethiopia, August to September 2011.

Predictors	AOR	95.0%	Р	
	non	Lower		1
Use of Human Feces as Fertilizer				
Yes	5.34	2.00	14.28	0.001
No	1.00			
Habit of Eating Soil				
Yes	4.64	1.50	14.36	0.008
No	1.00			
Presence of Stagnant water				
Yes	2.99	1.28	7.00	0.012
No	1.00			

Table 8. Parameter estimates from multivariable Logistic regression model predicting the likelihood that a pregnant woman is Co-infected with STH and malrai, Gilgel Gibe Dam area, southwest Ethiopia, August to September 2011.

AOR= Adjusted Odds Ratio, C.I= Confidence Interval

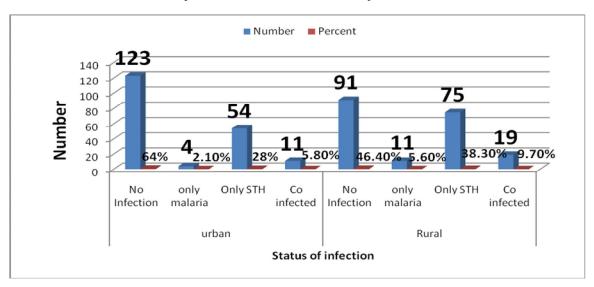


Figure4. Proportion of malaria, STH infection and co-infection among pregnant women in Gilgel Gibe hydropower dam area, southwest Ethiopia, August to September 2011.

Pregnant women infected with STH were likely to be three times more infected with malaria compared to these pregnant women with no STH infection (OR=3.3, 95% CI:1.72-6.40, p<0.001). Infections with Hook worm (OR=1.540, 95% CI:0.806-2.940), *A.lumbriciodes*(OR=3.954, 95% CI:1.982-7.887) and *T.trichiura*(OR=21.188, 95% CI:6.212-72.261) were found to increase malaria prevalence. There was statistically significant association between number of helminthes parasite species and malaria infection rate (p<0.001) and it was found that the rate of malaria infection was higher in those pregnant women infected with more than one helminth species.(Table 9) At an individual parasite load level, H. worm parasitic load was positively co-related to malaria parasitic load (r = 0.299, p<0.001), in contrary *A. lumbricoids* parasitic load was negatively co-related to malaria parasitic load (r=-0.095, p<0.001).

Table 9. Association between number of helminthes species and malaria infection rate among pregnant women in Gilgel Gibe hydropower dam area, southwest Ethiopia, August to September 2011.

	Total	Chi-	Р		
STH species	H species Malaria Negative Positive			square test Value	
				Value	
Single	117	10(7.9%)	127	4.79	<0.00 1
Double	25	2(7.4%)	27		
Triple	4	1(20%)	5		

5.1.3 Prevalence of anaemia and associated risk factors

The hemoglobine cut of value for anaemia according to WHO guide line is 11g/dl which is equal to 33% hematocrite value (20). Therefore those pregnant women in the first and third trimester with HCT value less than 33% and those in the second trimester with HCT value less than 32% were categorized as anaemic women. Anaemic women were further categorized as women with mild anaemia, moderate anaemia and severe anaemia which corresponds to HCT value of >=30 % < 33%, >=21% <30% and <21%, respectively.

Of the 388 study participants, 209(53.9%) were anaemic. The minimum, maximum and mean HCT value were 18%, 48% and 32.7%, respectively. Of these 209 anemic women; 115(55%), 88(42.1%), 6 (2.9%) were with mild, moderate and sever anaemia, respectively.ITN utilization, habite of walking bare foot, using human feces as afertilizer, place of residence and occupation have statistically significant associated with anaemia (p < 0.001). In which water source, utilization of human feces as a fertilizer and ITN utilization were the main risk factors for anaemia in the study area (Table 10). Pregnant woman who use spring water was two times more likely to be anaemic compared to those who use pipe water (AOR= 2.08, 95% C.I: 1.06-4.10, p=0.033). Pregnant woman who used human feces as fertilizer were 3.6 times more likely to be anaemic compared to those who didn't use human feces as fertilizer(AOR= 3.62, 95% C.I: 1.43-9.14, p=0.006). Pregnant woman who was not using ITN during the study period was three times more likely to be anaemic compared to those who didn't use human feces as fertilizer(AOR= 3.62, 95% C.I: 1.43-9.14, p=0.006). Pregnant woman who was not using ITN during the study period was three times more likely to be anaemic compared to those who didn't use human feces as fertilizer(AOR= 3.62, 95% C.I: 1.43-9.14, p=0.006). Pregnant woman who was not using ITN during the study period was three times more likely to be anaemic compared to those who were using ITN (AOR= 3.45, 95% C.I: 1.58-7.55, p=0.002) (Table 11).

Characteristics		Ana	amia			
		Anaemic	Non Anaemic	n	Chi-square	Р
		N=209 n (%)	N=179 n (%)	-		
Place of Residence	Urban	87(45.3%)	105(54.7%)	192	11.19	0.001*
	Rural	122(62.2%)	74(37.8%)	196		
Age group	16-20	69(53.9%)	59(46.1%)	128	5.55	0.235
	21-25	62(53.9%)	53(46.1%)	115		
	26-30	54(49.1%)	56(50.9%)	110		
	31-35	17(63%)	10(37%)	27		
	36-40	7(87.5%)	1(12.5%)	8		
Occupation	Housewife	86(45%)	105(55%)	191	19.10	0.001*
	Farmer	114(45.3%)	62(54.7%)	176		
	Daily laborer	0(0%)	4(100%)	4		
	Civil servant	7(53.8%)	6(46.2%)	13		
	Business man	2(50%)	2(50%)	4		
Parity	Primigravide	66(52.8%)	59(47.2%)	125	0.08	0.772
	multigravide	143(54.4%)	120(45.6%)	263		
	Third trimester	90(63%)	53(37%)	143		
Water Source	Pipe	76(44.5%)	95(55.5%)	171	13.13	0.01*
	Protected well	41(55.4%)	33(44.6%)	74		
	Unprotected Well	3(75%)	1(25%)	4		
	River/pond	32(60.4%)	21(39.6%)	53		
	Spring	57(66.3%)	29(33.7%)	86		
Human Feces as Fertilizer	Yes	25(78.1%)	7(21.9%)	32	8.26	0.004*
	No	184(51.7%)	172(48.3%)	356		
ITN utilization	Yes	181(52%)	167(48%)	348	4.67	0.031*
	No	28(70%)	12(30%)	40		
Habit of walking bare foot	Yes	158(57.9%)	115(40.1%)	273	5.958	0.010*
	No	51(44.3%)	64(55.7%)	115		

Table 10. Association of anaemia with socio-demographic characteristics and environment related factors among pregnant women in Gilgel Gibe hydropower dam area, southwest Ethiopia, August to September 2011.

* Significant at (p < 0.05),

Predictors	AOR	95.0%	% C.I.	Р
	AUK	Lower	Upper	F
Residence				
Rural	0.91	0.39	2.10	0.820
Urban	1.00			
Occupation				
Housewife	1.00			
Farmer	1.73	0.74	4.04	0.209
Daily laborer	0.00	0.00		0.999
Civil servant	1.56	0.48	5.07	0.462
Business man	1.69	0.23	12.47	0.606
Water Source				
Pipe	1.00			
Protected well	1.12	0.61	2.05	0.724
Unprotected well	2.91	0.28	30.04	0.370
River/pond	1.25	0.57	2.71	0.577
Spring	2.08	1.06	4.10	0.033
Use of Human Feces as Fertilizer				
Yes	3.62	1.43	9.14	0.006
No	1.00			
Current ITN utilization				
No	3.45	1.58	7.55	0.002
Yes	1.00			
Habit of barefoot walking				
Yes	1.18	0.69	2.03	0.546
No	1.00			

Table 11. Parameter estimates from multivariable Logistic regression model predicting the likelihood that a pregnant woman is anemic, Gilgel Gibe Dam area, southwest Ethiopia, August to September 2011.

AOR= Adjusted Odds Ratio, C.I= Confidence Interval

Even though age group (P=0.232) and parity (P=0.428) were not significantly associated with anaemia in the study area, the study result showed that multigravida women (table 6) urban pregnant women within the age group 16-20 and rural pregnant women within the age group 36-40 were more likely to be anamic (Table 12).

Table 12. Distribution of anaemia among different age groups of pregnant women with respect to place of residence in Gilgel Gibe hydropower dam area, southwest Ethiopia, August to September 2011.

Age group	% of Anaemia			
Age group	Urban	Rural		
16-20	39(47.6%)	30(65.2%)		
21-25	23(42.6%)	39(64%)		
26-30	21(46.7%)	33(50.8%)		
31-35	4(40%)	13(76.5%)		
36-40	0(0%)	7(100%)		

In this study malaria was found to be strongly associated (p < 0.001) with anaemia in which 100% of the pregnant women infected with malaria were anaemic. Additionally, the risk of developing anaemia was higher by 6 fold in pregnant women co-infected with malaria and soil transmitted helminths compared to those women infected with only soil transmitted helminthes (AOR=5.9, 95% CI: 1.69-20.41, p=0.001).Hookworm infection was also found to be associated with anaemia compared to other STH (P<0.001) (Table 13).

Characteristics				Chi- squar	
		Tota l	Anaemic	e	Р
STH infection	T.trichuira	13	11(84.6%	5.12	0.021
	A.lumbricoids	58	32(55.2%	0.05	0.472
	H.worm	114	, 78(68.4%	13.76	<0.00 1
STH and malaria infection	Only STH	129	, 78(60.5%)	3.39	0.041
	Only malaria	15	15(100%)	13.36	<0.00 1
	Malaria/STH co- infection	30	27(90%)	17.08	<0.00 1

Table 13. Association of STHs and malaria with anaemia among pregnant women in Gilgel Gibe Dam area, 2011

Assessment was also done to determine the effect of H.worm, malaria and the whole STH parasite load, number of parasite spp and co-infection on the severity of anaemia. The study finding indicated that H.worm (P=0.002), malaria (p<0.001), number of parasite species (P=0.001) and co-infection (p<0.001) have significant association with the level of anaemia. There was a significant correlation between increasing Hookworm parasite load(r = -.110, P < 0.001), *A.lumbricoids* (r = -.122, P < 0.001) and *T.trichuira* (r = -.025, P < 0.001) and decreasing hematocrite value. There was increased rate of severe anaemia level in H.worm load more than 1,000 ova/gram of stool. Interestingly rate of severe anaemia was high (83.3% of all sever cases) among pregnant women with single helminth infection than those of pregnant women infected with more than one helminthes infection (0%).

5.2 Discussion

Most of the time malaria and STH infections share endemicity in Ethiopia and the prevalence of malaria/STH co- infection among the pregnant women in our study were 7.7% with rate of co infection higher in rural (9.7%) than urban (5.7%) pregnant women. Co-infection was more prevalent in those within the age group range of 26-30 year which was different from the previous report from Nigeria where pregnant women less than 20 were the most co-infected group (51). The prevalence of co-infection was lower than the prevalence of co-infection reported from Thialand (59), Ghana (58) and Nigeria (51).

There was strong association between malaria and STH infection in which those pregnant women infected with STH were 3.3 times likely to be infected with malaria which is almost similar to a study from Ghana where women with intestinal helminth infection(s) were 4.8 times more likely to have malaria infection (58) and Senegal (81). There was a statistically significant association between number of helminths parasite species and malaria infection rate and it was found that the rate of malaria infection was higher in those pregnant women infected with more than one helminth species. This result was similar to the study done in Alaba Kulito Health Center, southern Ethiopia (19). In this study hook worm parasitic load was positively co-related to malaria parasitic load and this result was comparable with the previous studies which suggested that STH infections may increase the risk of malaria infection (10, 62) and especially H.worm aggravates the severity of malaria (19, 60). In contrary A. lumbricoids parasitic load was negatively corelated to malaria parasitic load; therefore as the parasitic load of A. lumbricoids increases the severity, parasitic load, of malaria decreases. This finding was comparable with the studies which showed the protective effect of A. lumbricoids to reduce the severity of malaria (19, 59).

In the current study, the overall STH prevalence, 159(41%), was lower than the findings from Bushulo health center, Southern Ethiopia 58.2 %(28) and Thailand 70 %(59) but was higher than the findings reported from Kenya 25.7 %(66) and Ghana 36.3% (58). Hook worm was found to be the most prevalent STH. The prevalence of Hook worm, *A.lumbricoieds* and *T.trichuiria* were 29.4%, 15% and 3.4%, respectively. This result was different from the prevalence of national report (31) as well as study done in Nigeria (51). This difference may be due to the difference in the subjects of the studies in which in the national reports all community members were used; and different group of a community has different risk of infection.In addition to that the difference in soil type among these three studies may have changed the prevalence of infection. Prevalence of H. worm was similar to prevalence H.worm reported from Butajira, Ethiopia (26) and Kenya (66) but lower than the prevalence reported from Thailand 42.8 % (59) and Peru 47.22% (35).

Malaria is the most serious public health problem in Ethiopia where it is highly distributed throught the country. In our study malaria prevalence in Gilgel-Gibe dam area

was 11.6% with *P.falciparum* and *P.vivax* prevalence of 71.1% and 15.6%, respectively; whereas mixed infection with both species and maximum parasitic load were 13.3% and 36,480 parasites / μ L, respectively. Malaria prevalence found in this study was by far lower than the results of similar studies done in Nigeria (51) and Burkina Faso (80), but almost similar to malaria prevalence reported from Sudan (79). This difference may be because of climatic difference and life style of the communities. The malaria parasite species distribution was different from the study done in the same area with different study subjects, where *P.vivax* was reported to be more prevalent than *P.falciparum* (47).

Risk factors for parasitic infection may differ from site to site so identifying the risk factors of a given area helps to develop appropiate prevention and control strategy of infection. This study indicated that, habit of bare foot waking and animals dwell in the house were the main risk factors for STH infection. Pregnant women who have habit of walking bare foot and having same dwelling with animals were the most infected groups compared to the pregnant women who didn't face for those risk factors. This could be because of the fact that all the factors listed above have the tendency to increase the women's exposure to STH infection in which the main means of transmission is through feco oral contamination (1, 78). Place of residence were also associated risk factor for STH infection of mothers in the study done in Butajira, Ethiopia (26). Another related study done in Ghana showed that being young age less than 20 years and low weekly income were the factors for either helminth or malaria infection in pregnant women(51). However, none of these factors were significantly associated with STH infection in this study. This difference in risk factors between Ethiopian and Ghanian pregnant women may be due to the difference in their socio-economic level as well as the difference in the study participants between the two studies; in which the participants in the above study was pregnant women following ANC but in the current study participants were selected from the community.

In the case of malaria, place of residence were found to be the main risk factor for malaria infection among pregnant women in the study area; where pregnant women living in the rural kebeles were two times more likely to be infected by malaria compaired to these pregnant women living in urban kebeles. As in the case of STH infection, malaria was more prevalent in rural residents (15.3%) than urban (7.8%) residents. This is mainly because rural residents are areas where people are increasingly exposed to malaria parasite infection because of their life style; highly related with farming, and the presence of wide range of breeding sites in addition to the main reservoir dam which is located close to these villages.

Even though significant association was not shown between parity and malaria, multigravida women were more infected (13%) with malaria compared to primigravidae (8.8%). This finding was in agreement with the study done in Sudan where parity was not associated with malaria (79). On the other hand this result was not comparable with many

studies which showed party is associated with malaria and these less immuned primigravide women were more likely to be infected with malaria than multigravide (4, 51, 58, 80). In the rural setting, higher proportion of those pregnant women in the age group of less than or equal to 20 were infected with prevalence of 17% which is consistent to the study done in Ghana (58). This high prevalence in those young age pregnant women, most of them are primgravide, could be attributed to low immunity.

Compared to the study done in Thailand, Ghana and Nigeria the possible risk factors for malaria/STH co infection in the study area were different, in which presence of stagnant water, habit of eating soil and habit of using human feces as a fertilizer were associated risk factors for malaria and STH co-infection among pregnant women in Gilgel Gibe areain which pregnant woman who lives in house which is near stagnat water was three times more likely to be co-infected; while parity, low income, being young age and marital status (being single) were the associated risk factors in other studies (51, 58, 59). Therefore, targeting the possible risk factors identified in this study may reduce malaria and STH co-infection in pregnant women.

Both malaria and STH infections result in different complications. The complications may be more complicated and severe where they co-exist in an individual, especially in children and pregnant women. Anaemia is one of the most known outcomes of malaria and STH co-infection. The prevalence of anaemia was 53.9%; with minimum, maximum and mean HCT value of 18%, 48% and 32.7%, respectively. From the total of 209 anemic women; 115(55%), 88(42.1%), 6 (2.9%) were with mild, moderate and severe anaemia, respectively. This result was almost similar to that of anaemia prevalence in Bushulo health centers, Ethiopia 51.9%(28) but slightly higher than the anemia prevalence previously reported from Jimma health center 41.9% (27), Asendabo health center 23%(24), and Peru 47.31% (35).

Even though age group and parity were not associated with anaemia in the study area, the result of the study showed that multigravida women within the age group 16-20 and 36-40 were more likely to be anamic in urban and rural settings, respectively. This finding was in agreement with the study conducted in India (76). Water source, utilization of human feces as a fertilizer and ITN utilization were the main risk factors for anaemia in the study area in which pregnant woman who use spring water was two times more likely to be anaemic compared to those who use pipe water; pregnant woman who used human feces as fertilizer were 3.6 times more likely to be anaemic compared to those who didn't use human feces as fertilizer. Pregnant woman who was not using ITN during the study period was three times more likely to be anaemic compared to those who were using ITN.

In the current study those pregnant women which had a habit of walking bare foot have high anaemia prevalence (57.8%). This could be attributed to walking bare foot is one route of H.worm transmission. H.worm infection rate was also statistically associated with anaemia in which those pregnant women infected with H.worm have 2.4 times increased risk of developing anaemia, in which 68.4% of the pregnant women infected with H. worm were anaemic. This finding is similar to the findings of other related studies (28, 59). There was a significant correlation between increasing H.worm parasite load, A.lumbricoids and T.trichuira and decreasing hematocrite value. Therefore, as the helminth parasitic load increases the hematocrit level decreases; which mean the risk of developing anaemia increases. This result was comparable to the study result from Peru, where there was a significant correlation between increasing hookworm egg counts and decreasing hemoglobin levels (35). Anaemia was six times higher in malaria and STH coinfected pregnant women than those infected with STH only which is in agreement with the findings of many studies (28, 51, 58, 59). These findings are good indications for the fact that preventing malaria/STH co-infection is preventing anaemia in case of the risk groups, pregnant women; therefore there is a need to design strategies which help to diagnose pregnant women for malaria and STH co-infection during their ANC visit instead of testing for Hgb level and blood group only.

Chapter six

Conlussions and Recommendations

6.1 Conclussions

- Malaria and STH co-infection and anaemia are currently serious health problem of pregnant women living in Gigel Gibe Dam area. In which the rate of co-infection with malaria and STH was higher in the rural areas.
- Presence of stagnant water, habit of eating soil and habit of using human feces as a fertilizer were the risk factors for malaria and STH co-infection among pregnant women in Gilgel Gibe
- The presence of STH infection has an influence on the rate of malaria. In which malaria prevalence is increased in those pregnant women with STH, compare with those free of STH infection.
- Hook worm load is positively correlated with malaria parasitic load; while *A.lumbriciodes* parasitic load is negatively correlated with malaria parasitic load.
- The severity of anaemia is pronounced more when pregnant women infected with STH are also co-infected with malaria.
- H.worm has strong association with anaemia and as the parasitic load increase anaemia severity also increases.

6.2 Recommendations

- ✓ Currently there is high rate of STH and malaria infection among the pregnant women in Gigel Gibe area. The prevalence of anaemia was also high. Therefore responsible bodies including the zonal health office and other nongovernmental organizations has to be co-operated to supply the necessary treatments for these pregnant women suffering from parasitic infections and anaemia.
- ✓ It is better if pregnant women are diagnosed for STH as well as malaria during their visit for ANC purpose to solve the problem of malaria/soil transmitted related anaemia.

- ✓ Compared to their knowledge about malaria; many pregnant women know nothing about the ways of STH transmission, there is a need to enhance the rural as well as urban pregnant women's knowledge about STH infection.
- ✓ There is a need to do related parasitological as well as immunological studies to exactly determine the association b/n STH infections and malaria.

Chapter seven

References

- WACIPAC, Why parasitic disease control? 2008, available at http://98.130.228.222/wacipac/index.php, accessed on January 10/2010.
- 2. Trowsdale J, Betz AG., Mother's little helpers: mechanisms of maternal-fetal tolerance. *Nat Immunol*, 2006; 7:241–246.
- 3. Riley EM., Schneider G., Sambou I., Greenwood BM., Suppression of cellmediated immune responses to malaria antigens in pregnant Gambian women. *Am J Trop Med Hyg*, 1999; 40: 141–144.
- 4. Bouyou-Akotet MK., Ionete-Collard DE., Mabika-Manfoumbi M., Matsiegu PB, Mavoungou E, Kombila M., Prevalence of *Plasmodium falciparum* infection in pregnant women in Gabon. *Malar J*, 2003; 2: 18.
- 5. Adegnika AA., Agnandji ST., Chai SK. *et al.*, Increased prevalence of intestinal helminth infection during pregnancy in a sub-Saharan African community. *Wien Klin Wochenschr*, 2007; *119*: 712–716.
- 6. Torre D., Speranza F., Martegani R., Role of proinflammatory and antiinflammatory cytokines in the immune response to *Plasmodium falciparum* malaria. *Lancet Infect Dis*, 2002; 2: 719–720.
- Basavaraju SV., Schant P., Soil-transmitted helminths and *Plasmodium falciparum* malaria: epidemiology, clinical manifestations, and the role of nitric oxide in malaria and geohelminth coinfection. Do worms have a protective role in *P. falciparum* infection? *Mt Sinai J Med, 2006; 73:* 1098–1105.
- Mwangi TW., Bethony J., Brooker S., Malaria and helminths interactions in humans: an epidemiological viewpoint. *Ann.Trop Med Parasitol, 2006; 100:* 551– 570.
- 9. Yazdanbakhsh M., Van der Biggelaar A., Maizels RM., Th2 responses without atopy: immunoregulation in chronic helminths infections and reduced allergic disease. *Trends Immunol*, 2001; 22: 372–377.
- 10. Tshikuka JG., Scott ME., Gray-Donald K., Kalumba ON., Multiple infection with Plasmodium and helminths in communities of low and relatively high socioeconomic status. *Ann Trop Med Parasitol, 1996; 90:* 277–293.
- 11. Haemoglobin concentrations for the diagnosis of anaemia and assessment of severity. Vitamin and Mineral Nutrition Information System. Geneva, World Health Organization, 2011 (WHO/NMH/NHD/MNM/11.1) (http://www. who.int/vmnis/indicators/haemoglobin)). accessed on January 10/2010.
- 12. McDevitt MA., Xie J., Gordeuk V., Bucala R., The anemia of malaria infection: role of inflammatory cytokines. *Curr Hematol Rep*, 2004; 3: 97–106.

- Murray MJ., Murray AB., Murray MB., Murray CJ., Parotid enlargement, forehead edema, and suppression of malaria as nutritional consequences of ascariasis. *Am J Clin Nutr*, 1977; 30: 2117–2121.
- Murray J, Murray A., Murray M., Murray C., The biological suppression of malaria: an ecological and nutritional interrelationship of a host and two parasites. *Am J Clin Nutr*, 1978; 31: 1363–1366.
- 15. World Health Organization /UNICEF. Iron deficiency anaemia, assessment, prevention and control: (2001) a guide for programme managers.
- World Health Organization. Report of the WHO informal consultation on hookworm infection and anaemia in girls and women. (1996)Geneva: WHO, CTD/SIP/ 96.1.
- 17. Steketee RW., Nahlen BL., Parise ME., Menendez C., The burden of malaria in pregnancy in malaria-endemic areas. *Am J Trop Med Hyg*, 2001; 64: 28–35.
- Ndomugyenyi R., Kabatereine N., Olsen A., Magnusses P., Malaria and hookworm infections in relation to heamoglobin and serum ferritin levels in pregnancy in Masindi district, Western Uganda. *Trans R Soc Trop Med Hyg*, 2002; *102*: 130–136.
- Abraham Degarege, Abebe Animut, Mengistu Legesse and Berhanu Erko., Malaria and helminth co-infections in outpatients of Alaba Kulito Health Center, southern Ethiopia: a cross sectional study. *BMC Research Notes* 2010; 3:143.
- 20. Health and Health related Indicators; planning and programming Department, MOH, 2003/04.
- Ismail R., Ordi J., Menendez C., Ventura J., Aponte J., Kahigwa E., Hirt R., Cardesa A., Alonso L. Placental pathology in malaria: a histological, immunohistochemical, and quantitative study. Hum. *Pathol* 2000; 31: 85–93.
- 22. Addis Continental Institute of Public Health (ACIPH), Qualitative Study on Malaria Prevention and Control in Oromia and Amhara Regional States in Ethiopia; Report Submitted to Academy for Educational Development (AED) and NetMark, 17 July, 2009.
- 23. Booth M., Bundy DA., Estimating the number of multiple-species geohelminth infections in human communities. *Parasitol.* 1995; 111:645–53.
- 24. Yonas T., Prevalence of anaemia among ANC attendants of Assendabo Teaching Health Center (dissertation). Jimma Institute of Health Sciences, Jimma, 1996.
- 25. Gebremedin S. Prevalence of anaemia and its predisposing factors in pregnant mothers attending antenatal care in Mettu Carl hospital. (2004).
- 26. Yeshambel B., Girmay M., Alemayehu A., *et al.* Prevalence and risk factors for soil-transmitted helminth infection in mothers and their infants in Butajira, Ethiopia: a population based study. *BMC Public Health* 2010, 10:21.
- 27. Desalegn S. Prevalence of anaemia in pregnancy in Jima town, southwestern Ethiopia. Ethiop Med J. 1993;31(4):251-8.

- 28. Tadege B., determinants of anaemia in pregnant women with emphasis on intestinal helminthic infection at bushulo health center southern Ethiopia, Addis Ababa University Libraries Electronic Thesis and Dissertations: 2009 AAU-ETD.
- 29. Bundy DAP, Chan MS, Savidi I. Hookworm infection in pregnancy. *Trans R* Soc *Trop Med Hyg*, 1995; 89: 521-522.
- 30. Hotez PJ. One World Health: Neglected Tropical Diseases in a Flat World. *PLoS Negl Trop Dis.* 2009;3(4):
- Tadesse Z, Hailemariam A, Kolaczinski JH: Potential for integrated control of neglected tropical diseases in Ethiopia. *Trans R Soc Trop Med Hyg* 2008; 102:213-214.
- 32. Hotez P., The Global Burden of Parasitic Disease in the New Millennium. Abstr Intersci Conf Antimicrob Agents Chemother Intersci Conf Antimicrob Agents Chemother. 2001; 41: abstract no. 1131.
- WHO, World Malaria Report, 2008. Available at http:// www.who.int/malaria/wmr2008/ accessed on January 10/2010.
- 34. Snow RW., Guerra CA., Noor AM., Myint HY., Hay SI., The global distribution of clinical episodes of *Plasmodium falciparum* malaria. *Nature*, 2005; *434*: 214–217.
- 35. Larocque R., Casapia M., Gotuzzo E. and W. Gyorkos T., Relationship between intensity of soil-transmitted helminth infections and anemia during pregnancy Am J Trop Med Hyg., 2005;73: 783-789.
- 36. Fried M, Duffy PE. Adherence of *Plasmodium falciparum* to chondroitin sulfate A in the human placenta. *Science*, 1996; 272: 1502–1504.
- 37. Rowe AK., Rowe SY. and Snow RW., The burden of malaria mortality among African children. International Journal of Epidemiology, 2006; 35:691–704.
- 38. CSA. The Ethiopian Demographic and Health Survey 2005. Addis Ababa: Central Statistical Agency; 2006.
- 39. National five- year strategic Plan for malaria prevention & control in Ethiopia; 2006-2010, FMOH, 2006.
- 40. Ministry of Health, Malaria diagnosis and treatment guidelines for health workers in Addis Ababa, Ethiopia, 2007.
- 41. Deressa W, Chibsa S and Olana D. Magnitude of malaria admissions and deaths at hospitals and health centers in Oromia, Ethiopia. Ethiopian Journal Health Development, 2004; 42: 237-246.
- 42. Okoko BJ, Enwere G, Ota MO. The epidemiology and consequences of maternal malaria: a review of immunological basis. *Acta Trop.*, 2003; 87(2):193-205.
- 43. Nacher M, Singhasivanon P, Traore B, *et al.* Helminth infections are associated with protection from cerebral malaria and increased nitrogen derivatives concentrations in Thailand. *Am J Trop Med Hyg.*, 2002; *66*: 304–309.

- 44. Stephen D. Hillier, Mark Booth, Lawrence Muhangi, *et al.*, *Plasmodium falciparum* and helminth co-infection in a semi-urban population of pregnant women in Uganda. *J Infect Dis.* 2008; 198(6): 920–927.
- 45. Hercberg S. and Galan P., Nutritional anaemia Baillieres Clin Haematol, 1992; 5:143 168.
- 46. Ethiopia: demographic and health survey 2000. Addis Ababa, Ethiopia: Central Statistical Authority and ORC Macro; 2001.
- 47. Yewhalaw D, Legesse W, Van Bortel W, *et al.*, Malaria and water resource development: the case of Gilgel-Gibe hydroelectric dam in Ethiopia. *Malar J.*, 2009; 8: 21.
- Shargie EB., Gebre T., Ngondi J., *et al.*, Malaria prevalence and mosquito net coverage in Oromia and SNNPR regions of Ethiopia.*BMC Public Health*, 2008, 8:321.
- 49. Staedke SG, Nottingham EW, Cox J, Kamya MR, Rosenthal PJ, Dorsey G: Proximity to mosquito breeding sites as a risk factor for clinical malaria episodes in an urban cohort of Ugandan children. *Am J Trop Med Hyg*, 2003; 69:244-246.
- 50. WHO. Global Malaria Programme-Pregnant Women and Infants. World Health Organisation, 2007a) Geneva, Retrieved Feb 18,2007 from <http://www.who.int/malaria/pregnantwomenandinfants.html>
- 51. Egwunyenga AO., Ajayi JA., Nmorsi OPG., Duhlinska-Popova DD., *Plasmodium* /intestinal helminth coinfections among pregnant Nigerian women. *Mem Inst Oswaldo Cruz, Rio de Janeiro*, 2001; 96: 1055–1059.
- 52. National five year strategic plan for malaria control in Ethiopia Addis Ababa, Ethiopia: Ethiopia Ministry of Health; 2001.
- 53. Newman RD, Hailmariam A, Jimma D, Degefie A, Kebede D, Rietveld AC, Nahlen BL, Barnwell JW, Steketee RW, Parise ME: Burden of malaria during pregnancy in areas of stable and unstable transmission in Ethiopia during a nonepidemic year. *J Infect Dis*, 2003, 187:1765-1772.
- 54. World Health Organization Prevention and control of schistosomiasis and soiltransmitted helminthiasis: report of a WHO expert committee. Geneva (2002), Switzerland: World Health Organization.
- 55. Peter J., Simon B., Phil J., Bethony M., Maria E., Alex L., Shuva X. Hookworm infection, *N Eng J Med*, 2004; 351(8): 799 807.
- 56. Hotez JP., Brooker S., Bethony JM., Bottazzi ME., Loukas A., Xiao S., Hookworm. *Infect Eng J Med*, 2004; 351:799-807.
- 57. Van Eijk AM, Lindblade KA, Odhiambo F, *et al.*, Geohelminth Infections among Pregnant Women in Rural Western Kenya; a Cross-Sectional Study. *PLoS Negl Trop Dis* 2009; 3(1):
- 58. Nelly J. Yatich, Jiang Yi, Tsiri Agbenyega, *et al.*, Malaria and Intestinal Helminth Co-infection Among Pregnant Women in Ghana: Prevalence and Risk Factors. *Am. J. Trop. Med. Hyg.*, 2009; 80(6): 896-901.

- 59. Boel M, Carrara VI, Rijken M, *et al.*, Complex Interactions between Soil-Transmitted Helminths and Malaria in Pregnant Women on the Thai-Burmese Border. *PLoS Negl Trop Dis.*, 2010; 4:
- Hartgers FC, Yazdanbakhsh M. Co-infection of helminths and malaria: modulation of the immune responses to malaria. *Parasite Immunol*, 2006; 28: 497–506.
- 61. Adrienne E. Shapiro, Edridah M. Tukahebwa, Jennifer Kasten, Si^an E. Clarke, Pascal Magnussen, Annette Olsen, *et al.* Epidemiology of helminth infections and their relationship to clinical malaria in southwest Uganda. *Trans. R. Soci. Trop. Med. Hyg*, 2005; 99: 18-24.
- 62. Nacher, M., Singhasivanon, P., Yimsamran, *et al.* Intestinal helminth infections are associated with increased incidence of *Plasmodium falciparum* malaria in Thailand. *J. Parasitol.*, 2002b; 88, 55-58.
- 63. Le Hesran JY., Akiana J., Ndiaye el HM., Dia M., Senghor P., Konate L., Severe malaria attack is associated with high prevalence of *Ascaris lumbricoides* infection among children in rural Senegal. *Trans R Soc Trop Med Hyg, 2004; 98:* 397–399.
- 64. Shapiri AE., Tukahebwa EM., Kasten J., Clarke SE., Magnussen P., Olase A., *et al*, Epidemiology of helminth infections and their relationship to clinical malaria in Southwest Uganda. *Trans R Soc Trop Med Hyg*, 2005; 99: 18–24.
- 65. Muhangi L., Woodburn P., Omara M., Omoding N., Kizito D., Mpairiwe H., *et al.* Associations between mild-to-moderate anaemia in pregnancy and helminth, malaria and HIV infection in Entebbe, Uganda. *Trans R Soc Trop Med Hyg*, 2007; 101: 899–907.
- 66. Geissler PW., Prince RJ., Levene M., *et al.*, Perceptions of soil eating and anemia among pregnant women on the Kenyan coast. *Soc Sci (Kans), 1999; 488:* 1069–1079.
- 67. Reproductive Health and Research Publications: Making Pregnancy Safer. World Health Organization Regional Office for South-East Asia. 2009. Available at http://www.searo.who.int/EN/Section13/Section36/Section129/Section396_1450. htm. Retrieved 7 December 2009.
- 68. Federal Democratic Republic of Ethiopia MoH: National Five Year Strategic Plan for Malaria Prevention and Control in Ethiopia. Addis Ababa, Ethiopia; 2006.
- 69. Menendez C., Fleming AF., Alonso PL., Malaria-related anaemia.*Parasitol Today*, 2000; 16: 469–476.
- 70. Stephenson LS., Latham MC., Kurz KM., Kinoti SN., Oduori ML., Crompton DW., Relationships of *Schistosoma hematobium*, hookworm and malarial infections and metrifonate treatment to hemoglobin level in Kenyan school children. *Am J Trop Med Hyg*, 1985; 34: 519–528.

- Dondorp AM., Angus BJ., Chotivanitch K., *et al.* Red blood cell deformability as a predictor of anemia in severe falciparum malaria. *Am J Trop Med Hyg.* 1999; 60: 733–737.
- Guyatt L. and Snow W., The epidemiology and burden of Plasmodium falciparum related anemia among pregnant women in sub-Saharan Africa. *Am. J. Trop.Med. Hyg*, 2001; 64 (S1–2), 36–44.
- Huddle M., Gibson S., Cullinan R., The impact of malaria infection and diet on the anaemia status of rural pregnant Malawian women. *European J Clin Nutr*. 1999; 53: 792 – 801.
- 74. Nurdia S., Sumarni S., Suyoko., Hakim M., Winkvist A., Impact of intestinal helminthes infection on anaemia and iron status during pregnancy: a community based study in Indonesia. *SouthEast Asian J Trop Med Pub Health*, 2001; 32 (1):14 – 22.
- 75. Singh B., Fong F, Arikumeron S. Anaemia in pregnancy, a cross sectional study in Singapore, *European J Clin Nutr*, 1998; 52(1) 65-70.
- Bechuram M., Vikal T., Ranjan G. Risk factors of anaemia during pregnancy among the Garo of Meghalaya, India. *Human Ecology Special Issue*, 2006; 14(1):27-32.
- 77. WHO. Report of a technical working group on prevention and management of severe anaemia in pregnancy, WHO, Geneva, 20-22 may 1991:10.
- 78. Monica Cheesbrough. District laboratory practice in tropical countries, (*Cambridge University Press*) 2000; 1:244-247.
- 79. Adam I., Khamis H A.and Elbashir I M. Prevalence and risk factors for *Plasmodium falciparum* malaria in pregnant women of eastern Sudan. *Malaria Journal*. 2005; 4:18
- Sheick Oumar Coulibaly, Sabine Gies, and Umberto D'Alessandro. Malaria Burden Among Pregnant Women Living in the Rural District of Boromo, Burkina Faso. Am. J. Trop. Med. Hyg, 2007, 77:56–60.
- 81. Spiegel A, Tall A, Raphenon G, Trape JF, Druilhe P, Increased frequency of malaria attacks in subjects co-infected by intestinal worms and *Plasmodium falciparum* malaria. *Trans R Soc Trop Med Hyg*, 2003; 97: 198–199.

Annex 1-Procedures of Parasitological Examination of Faeces

I. McMaster egg counting technique:

The McMaster technique uses a counting chamber which enables a known volume of faecal suspension ($2 \ge 0.15$ ml) to be examined microscopically.

Procedure;

1. Weigh 2 grams of faeces and place into a container.

2. Add 30 ml of concentrated NaCl(33% w/v) flotation fluid.

3. Stir the contents of the beaker thoroughly with a fork, tongue depressor or spatula.

4. Filter the faecal suspension through a tea strainer or double layer of cheesecloth or dental napkin into the second container.

5. Homogenaze the filtrate in container two by transfering it in to other container (10 times) and withdraw a sub-sample using the pipette.

6. Fill the first compartment of the McMaster counting chamber with the sub sample.

- 7. Homogenize fluid again and fill second chamber with another sub sample.
- 8. Allow the counting chamber to stand for 5 minutes.
 - It is important to leave the chamber to stand to allow the eggs to float to the surface and the debris to go to the bottom of the chamber.

9. Examine the subsample of the filtrate under the compound microscope at 10 x 10 magnification.

II. Microhaematocrit method

- **A.** Fill the capillary tube by placing one end in the blood and lowering the other end and leave at least 15 mm of the tube unfilled.
- **B.** Remove the capillary tube from the blood, and wipe any traces of blood from its outer surface with a tissue.
- C. Seal the opposite end by plugging with a plastic sealing compound.
- **D.** Place the capillary tube in one of the radiating groove on the base plate of the microhaematocrit centrifuge.

- **E.** Place the upper (safety) place over the base plate and screw it down into position to prevent jumping of the tube out of their grooves during centrifugation.
- F. Close the centrifuge lid and centrifuge for 5 min. at 10.000 rpm.
- G. When the centrifuge gas stopped rotating, open the lid, remove the safety plate and take out the tubes.
- H. Read the PCV through a special reader as follows:
- Place the bottom of the RBCs column (which is just above the sealed end) exactly on the base line of the reader (0).
- Move the reader until the top of the plasma layer exactly means 100.
- Observe a line on the reader, which passes across the top of the RBCs layer, read the P.C.V. from the scale at the point where this line meets it.

III. Procedure for Blood Film

- 1. With the patient's left hand, palm upwards, select the third finger. Use cotton wool lightly soaked in alcohol to clean the finger.
- 2. With a sterile lancet puncture the ball of the finger using a quick rolling action. By applying gentle pressure to the finger, express the first drop of blood and wipe it away with dry cotton wool.
- 3. Apply gentle pressure to the finger and collect a single small drop of blood on the middle of the slide. This is for thin film. Apply further pressure to express more blood and collect two or three larger drop on to the slide about 1cm from the drop intended for thin film. Wipe the remaining blood away from the finger with cotton wool.
- 4. Thin film: using another clean slide as a "spreader", and with the slide with the blood drops resting on a flat, firm surface, touch the small drop with the spreader and allow the blood to run along its edge. Firmly push the spreader along the slide, away from the larger drops, keeping the spreader at an angle of 45°.
- 5. Thick film: always handle slides by the edges or by a corner, to make a thick film as follows:

Using the corner of the spreader, quickly join the larger drops of blood and spread them to make an even, thick film.

6. Allow the thick film to dry in a flat, level position protected from flies, dust, and extreme heat. Label the dry film with a pen or marker pencil by writing across the thicker portion of the thin film the patients' name or number and date.

- 7. Wrap the dry slide in clean paper, and dispatch with the patient's record form to the laboratory as soon as possible.
- 8. The slide used for spreading the blood films must be disinfected and could then be used for the next patient, another clean slide from the pack being used as a spreader.
- 9. Fix the thin film by adding three drops of methanol, or by dipping it in a container of methanol for a few seconds.
- 10. Place the slides back to back in a staining dish.
- 11. Pour the stain gently in to the dish, until the slides are totally covered.
- 12. Allow to stain for 10 minutes out of sunlight by 10% Giemsa.
- 13. Pour clean water gently in to the dish to float off the iridescent scum on the surface of the stain.
- 14. Gently pour off the remaining stain, and rinse again in clean water for a few seconds. Pour the water off.
- 15. Remove the slides one by one and place them in a slide rack to drain a dry, film side downwards, and making that the film does not touch the slide rack.
- 16. Then clean back side of slide and examine by 100x objective.

Annex 2. Laboratory Result Format

Participant ID. No. _____ Name of the village/kebele _____

Type of	Result			Remark
Tests	Type of parasite/s	Qualitative	Quantitative/density	
1.Stool Examinatio n (McMaster)		1. Positive	(/gm)	
	Hook worm	2. Not seen		
		1. Positive	(/gm)	
	A.lumbricoide	2. Not seen		
		1. Positive	(/gm)	
	T. trichiura	2. Not seen		
		1. Positive	(/gm)	
		2. Not seen		
		1. Positive	(/gm)	
		2. Not seen		
		1. Positive	(/gm)	
	-	2. Not seen		
	Total parasite type ()		Total parasite Density	
			(/gm)	
2. Blood Film Examinatio n	P.vivax	1. Positive	/gill/	
			-	
		2. Not seen	-	
	Dfoloinorum	1. Positive	-	
	P.falciparum	2. Not seen 1. Positive		
	Other haemoparasite/s	2. Not seen	-	
		2. NOT SEEN	Total Malaria parasite	
	Type of malaria	1. Single	Density	
			Donorty	
	infection	2. Mixed	(/µl)	
4. HCT				
			%	

Annex 3- Consent form in oromifa

Guca walii galttee

Haadholii ulfaa naannoo Gilgel Gibe jiraatan irratti faca'insa fi sababoota dhukkuba busaafi raammolee garaa ilaalchisee waa'ee qo'annoo taasifamuu irratti afaan danda'uun ibsi naaf kennamee, qo'nnicharratti akkan hirmaadhuuf haaluman gaafatameen kaayyoofi faayidaa qo'anichaa hubadhee kaffalttii malee odeeffannoo barbaachisaa ta'e kennuuf eyyemamaa ta'uu kootii mallotoo kootin na mirkaneessa.

Mallottoo hirmaataa_____

Guyyaa _____

Annex 4. Questinnaire in oromifa

Gaaffilee

<u>Seensa</u>: Gaaffilee kun qoronnoo waa'ee faca'insa dhukkuba busaafi raammolee garaa fi sababoota ka'umsa isaanii haadholii ulfaa naannoo gilgel gibe jiraatan irratti adeemsfamuuf yuunivarsiitii Jimmaatti barumsaa digrii lammaffaatiif qophaa'ee dha.Bu'aan qoronnoo kanaa jiraattota naannoo sanaa keessattuu; dhukkubichaaf salphaatti saaxilama kan ta'an, haadholii ulfaa, fayyan saanii eeguudhaaf ni fayyada jedhamee waan amanamuuf gaaffiwwan deebisuuf harmaannaa gootaniif dursinee isin galatooffanna.Odeeffannoo nuuf kenniitan kamiiyyuu qoronnoo kanaaf qofa kan ooluu yoota'u, odeeffannoo isinirraa argamu iccitiidhan qabama.

Kutaa I. Odeeffannoo waliigalaa

I.Teessoo

Maqaa waradaa	

Maqaa baadiyyaa/gandaa _____

Maqaa Goxii _____

Lakkoofsa maatii _____

Lakkoofsa manaa _____

Fageenyaa giddugaleessa hidhaa Gilgal Gibee irraa _____km

II. Odeeffannoo hawaasummaa

201.Umurii _____

202.Garee sabaa

1. Oromoo 2. Amaaraa 3. Kafaa 4. Yamm 5. Daawroo 6. Tigree

7.Guraagee 8.Kan biraa yoo taatan_____

203. Amantii

1.Musliima 2.Ortodoksii 3.Pirootestaant	ii					
4. Kaatoolik 5. Kan biraa yoo taatan						
204.Нојіі						
1. Haadha manaa 2. Qotee bulaa 3. Hojjetaa guyyaa 4	1. Barataa					
5.Hojii mootumaa 6.Hoji daldalaa 7. Kan biraa yoo ta	.'e					
205.Galii ji'aa(Qarshii)						
1.< 500						
206. Haala fuudha fi heerumaa						
1.Hin fuune						
2.Fuudhe						
3.Kan hike						
4.Abbaa manaa kan du'e						
207.Baay'ina ijoollee kan dhalatan(Kan du'an dabalatee- yoo jira ta'e)						
208.Manaa						
1. Kan Dhuunfaa2.Kireeffataa						
209.Sadarkaa baruumsaa						
1. Kan hin baranne2. Dubisufi bareessuuf qofaa						
3. Sadarka 1 ^{ffaa} 4. Sadarka 2 ^{ffaa} 5. Sadarka 2 ^{ffaa} ol						
210.Haala ulfaa yeroo qo'annoon adeemsifamu						
1.Marsaa 1 ^{ffaa} (hanga ji'a 3)						
2. Marsaa 2 ^{ffaa} (ji'a 3 –ji'a 6)						
3. Marsaa 3 ^{ffaa} (ji'a 7 ol)						
III. Haala manaa jireenyaa						

301.Meeshaalee ijoo baaxii(xaaraa) manaa

1.Citaa 2.Qorqoorroo 3.Kan biro,_____

302. Meeshaalee ijoo lafa keessa manaa

1.Lafa 2.Simiintoo 3.Mukaa 4.Kan biro,_____

303. Akaakuu giddgidaa mana

1.Citaa(migira) 2.Suphee 3.Siinto 4.Kan biro,_____

304. Beelladanni manaa maatii waliin manaa keessa jiraatuu?

1.Eeyyee 2.Hinjiraatan

305.Yoo beelladanni manaa maatii waliin manaa keessa jiruu ta'a, maal fa'aadha?

306.Toora mana jireenyaa keessan mooraan loowwanii/horii jiraa?

1.Eeyyee 2.Hinjiruu

307.Bishaan kuufamaan bookeen busaa irratti horuu dandeessumooraa keessaa nijiraa?

1.Eeyyee 2.Hinjiruu

308. Gaafi 307 dhaaf deebiin kee eyyee yoo ta'e, fageenyi manaa iddoo bookeen itti walhoran irraati hangam fagaata?

1.<100m 2.>100-<500m 3.500-1000m 4.>1000m

309.Manaa jireenyaa keessaa bookeen busaa jirtii?

1.Eeyyee 2.Hinjirtu

310. Manni fincaanii qabbdhaa?

1.Eeyyee 2.Hinjqabuu

311.Yoo manni fincaanii jiraate goosa kam

1.Ammayya 2.Aadaa

312.Iddoo argama manni fincaanii?

1.manaa keessa 2.manaa ala garuu mooraa keessa 3.mooraan ala

313. Manni fincaanii qadaada qaba?

1.Eeyyee 2.Hin qabu

314. Manni fincaanii kun yoo qadaada qabaate itti fayyadamttu?

1.Eeyyee

2.Hin fayyadamu

315. Manni fincaanii bishan harka dhiqannaa qabaa?

1.Eeyyee 2.Hin qabu

316. Fageenyi naannoon jireenyaa buufata fayyaa irraa qabu?

1. <500m 2. 500-1000m 3.>1000m

Kutaa lama- Gaffilee beekumsa dhibeetiin wal-qabatan

401. Waa'ee dhukkuba buusaa dhageessee beektaa?

1. Eyyee 2. Hin dhageenye

402.Gaafi 401 dhaaf deebiin kee eyyee yoo ta'e dhukkuba buusaanamarra namatti akani dabra bekktuu?

1. Eyyee	2.Hin beeku
----------	-------------

403. Gaafi 402 dhaaf deebiin kee eyyee yoo ta'e karaalee dadaarbiinsaa maal faadha?

1. Cinninnaa bookee busaa 2. Dhiiga namarraa fudhachu(Blood Transfusion)

3.Dhukkubsataa busaa tuqudhaan 4. Harma hoosisuudha 4.Kan biro,___

404. Gaafi 401 dhaaf deebiin kee eyyee yoo ta'e mallottoolee dhukkubni buusaa maal faadha?

1. hoo'insa 2.hurgufuu 3.dhukkubbii mataa 4.teessissu 5.hirina feedhii nyaataa

405. Waa'ee dhukkuba raammolee garaa dhageessee beektaa?

1. Eyyee 2. Hin dhageenye

406. Gaafi 405 dhaaf deebiin kee eyyee yoo ta'e dhukkuba raammolee garaa namatti akani dabra bekktuu?

1. Eyyee 2. Hin beeku

407. Gaafi 406 dhaaf deebiin kee eyyee yoo ta'e karaalee dadaarbiinsaa maal faadha?

1. Nyata dheedhii nyachu2.harka oosoo hindiqatiin nyachu3.mila duwwaa deemuu4.Bobo'AA nama akka xaa'oo fayyadamun5. Kan biro,_____

Kutaa- Sadii-- Gaffilee gocha/raawwidhan wal-qabatan

501. Yeroon meqaa harka kee nyaata duraa

1. Yeroo hundaa bishaaniifi saamunaan

2. Yeroo hundaa bishaan qofaan

3. Yeroo hundaa bishaanii, darbee darbee bishaaniifi saamunaan

4.Darbee darbee harka koo hin dhiqadhu

502. Deebiin kee gaafi 501 "4" yoo ta'e sababnni kee nyta duraa yeroo hundaa hin dhiqanneef maal dha?

1.Bishan dhabamuu

2. Yeroon itti dhiqadhu dhabamuu

3.Harki koo xuraa baannan nan dhiqadha jedhe hin yaadu

4.Dhimma dhabuu

5. Yeroo hunda maaliif akkan hindhiqanne hin beeku

6. Kan biro yoo jirate _____

503. Manni fincaaniin booda harka keessan hangam dhiqatti?

1. Eeyyee /Yeroo hunda 2. Eeyyee /Darbee darbee 3. Hin dhiqadhu

504. Mucaa keessan erga qulqullessitanii booda (segerashe/sa) harka keessa nidhiqattuu? (mucaa yoo qabaattan)

1. Yeroo hundaa bishaaniifi saamunaan

2. Yeroo hundaa bishaan qofaan

3. Yeroo hundaa bishaanii, darbee darbee bishaaniifi saamunaan

4.Darbee darbee harka koo hin dhiqadhu

505. Hamam kuduraa fi muduraa dheedhii nyaateta ji'a lamma kan darrban?

1. yeroo hundaa 2. yeroo tokko tokko 3. Dheedhiidhan nyadhen hin beeku

506. Boba'aa nama akka xaa'oo fayyadamtu?

1,Eeyyee

2 .Hin fayyadamnu

507.Madda bishaan dhugaatii

1.Ujummoo 2.Haroo dalla qabu 3. Haroo dallaa hin qabanne

4. Laga	5. Kan biraa	

508. Xuraawa dhangala'oo essati dhabamsiisttu?

1.Dirree irratti 2.Boolla keessaatti 3. Manni fincaanitti 4.Karaa biroon,_____

509.Bookee busaa ittisuuf saaphana siree nifayyadamttuu?

1,Eeyyee 2 .Hin fayyadamnu

510. Saaphana siree meeqaa manaa kesaa qabduu?

511. Amaa saaphana siree kesaa eenyu cisaa?

1.Ejoolee wagaa 5 gadii 2. Ejoolee wagaa 5 ol 3. Haadha ulfaa 4. Aabaawarraa

5. Misensaa matii hundaa saaphana siree nifayadha 6. Saaphana siree kanfayadhu hinjiruu

512. Saaphana siree keessaan yeroodhaan daawwaa nicuuphama?

1,Eeyyee 2.Hincuuphamu

513.Manaa jireenyaa keessanitti daawaan ilbilsotaa nibiifamaa(gannaa kan kesaa)?

1,Eeyyee 2.Hinbiifama

514. Hamam mila duwwaa deemtu?

1.yeroo hundaa 2.yeroo tokko tokko 3. mila duwwaa deemen hin beeku

515. Wayitii ulfaa keessan baratama biyee qama'uu qabduu?

1,Eeyyee 2.Hin qabu

Annex 5.concent form in Amharic

የስምምነት/የውል ቅጽ

በግለገል ግቤ አካባቢ የሚኖሩ ነፍሰጡር እናቶች የወባና የሆድ ትላትል በኽታ ስርጭት እና መንስኤዎች ላይ ስለሚደረግ ጥናት በምችለው ቋንቋ መብራርያ ተሰዋቶኝ በጥናቱ እንድሳተፍ በተጠየቅኩት መሰረት የጥናቱ ዓላማና ጥቅም በመረዳት ያለምንም ክፍያ አስፈላጊ የሆነውን መረጃ ለመስጠት ፍቃደኛ መሆኔን በፌርማየ አረ.ጋግጣለሁ፡፡

የታሳታፊው ፌርማ _____ ቀን

64

Annex 6. Questionnaire in Amharic

መጠይቅ

<u>መግቢያ</u>: ይህ መጠይቅ በግልገል ግቤ አከባቢ በሚኖሩ ነፍሰጡር እናቶች ያለውን የወባ እና የሆድ ትላትል በሽታዎችን ስርጭት እና መንስኤዎች ለማጥናት በጅማ ዩኒቨርሲቲ ለድህረ ምረቃ ጥናት የተዘጋጀ ነው፡፡የጥናቱ ውጤትም የአከባቢው ነዋሪዎችን በተለይም በቀላሉ ለህመም ተጋላጭ የሆኑትን ነፍሰጡር እናቶች ጤንነት ለመጠበቅ ይረዳልተብሎ ስለሚታመን መጠይቁን በመመለስ ለሚያደረጉት ትብብር በቅድሚያ እናመሰግናለን፡፡የሚሰጡን ማነኛውም መረጃ ለጥናቱ ዓላማ ብቻ የሚውል ሲሆን በምሥጢርም ይያዛል፡፡

ክፍል አንድ - ጠቅሳሳ መረጃ

¦.አድራሻ

የወረዳው ስም_____

የገጠፋ/ቀበሌው ስም _____

የንዋ ስም _____

የተሳታፊዋ ቤተሰብ መለይ ቁዋር_____

ቤት ቁኖር_____

ከግድቡ ያልው አማካኝ ርቀት ____(ኪ.ሜ)

||.<u>ማሕበራዊ ጉዳዮች</u>

201.**ዕድሜ** _____

202.**ብሔር**

1.ኦሮሞ	2.አማራ	3 . ከፋ	4 . የም	5.ዳውሮ
6.ትግሬ	7 . ጉራጌ	8.ሌላ ከሆ	ን	

203.**ሃይማኖት**

1. እስልምና 2. ኦርቶዶስ 3. ፕሮቴስታንት 4. ካቶሊክ 5. ሌላ ከሆኑ 204.*ሥ*ራ 1.የቤት እመቤት 2.ገበሬ 3.የቀን ሥራተኛ 4.ተማሪ 5.የመንግስት ሰራተኛ 6.**ነ.ጋዴ** 7.ሌላ ከሆነ 205.የቤተሰብ አማካኝ የወር ገቢ 1.< 500 2.500 - 1000 3.>1000 206.**የተዳር ሁኔታ** 1. ደሳገባች 2. ደገባች 3. የፌታች 4. ባል የሞተባት 207.የተወለዱ ልጆች ብዛት _____(የሞተ/ቱ ልጆች ካሉ - በፀይወት የልሉትም ጨምሮ) 208.**የቤት ሁኔታ** 1.**የ**ማል 2.ከራይ 209. የትምህርት ደረጃ 1.ያለተማረች 2.መፃፍ እና ማንበብ ቢቻ 3.1^ኛ ደረጃ 4.2^ኛ ደረጃ 5.**ከ** 2^ኛ ደረጃ በሳይ 210. የዕርግዝናው ደረጃ 1.የመጀምርያ ሦስት ወራት $2.h4^{*}$ እስከ 6^{*} ወር 3.ከ7^ኛወር በላይ |||.የመኖርያ ቤት ሁኔታ 301. **የቤቱ ዓይነት**

1.የሳር ጎጆ 2.የቆርቆሮ ቤት 3.ሌላ ከሆነ_____ 302.የምድር ቤቱ ዓይነት 1.አራር 2.ሲሚንቶ 3.እንጨት 4.ሌላ ከሆነ_____

303. የግድግዳው ዓይነት

1. ሳር 2.*ቄቃ* 3.ስሚነቶ 4.ሌላ ከሆነ _____ 304. መኖርያ ቤትዎ ውስጥ የሚኖሩ እንስሳት አሉ

1**. አዎ** 2 . የለ·ም

305.ስተራ ቁጥር 13 መልሶዎት አዎ ከሆነ እነስሳቱ ይጥቀሱ _____

306. መኖርያ ቤትዎ አጠገብ የእንስሳት በረት አለ? 1.አዎ 2.የሉም

307. ጊቢዎ ውስጥ የወባ ቲንኞች ሊራቡበት ሚችሉ የተጠራቀመ ወሃ አለ?

1. አዎ 2. የስም

308. ለዋያቄ 307 መልሶ 1 ከሆነ መኖርያ ቤትዎ የወባ ቲንኞች ከሚራቡበት ቦታ ያለው ርቀት

1.< 100 °C 2. >100 - < 500 °C 3. 500 - 1000 °C 4. >1000 °C

309. ቤትዎ ውስዋ የወባ ቲንኞች አሉ?

1.አዎ 2.የሉም

310.ሽንት ቤት አልዎት?

1.አዎ 2.የስም

311. ሽንት ቤት ካልዎት ምን ዓይነት ነው?

1.ባህሳዊ 2.ዘመናዊ

312. ሽንት ቤት ካልዎት ያለበት ቦታ የት ነው?

1. መኖርያ ቤትዎ ውስጥ 2. መኖርያ ቤትዎ ውጭ/ጊቢ ውስጥ 3. ከጊቢ ውጭ

313. ሽንት ቤት ካልዎት ሽንት ቤቱ መክደኛ አለው?

1.አዎ 2.የለውም

314. ሽንት ቤቱ መክደኛ ካለው ይጠቀሙበታል?

1.አዎ 2.አልጠቀምበትም

315. ሽንት ቤት ካልዎት ሽንት ቤቱ የእጅ መታጠብያ ወሃ አለው?

1.አዎ 2.የለውም

316.መኖርያ ቤትዎ ቅርብ ከሆነው ከጤና ጣቢያው/ጤና ኬላ ያለው ርቀት

1.< 500 °C 2.500 - 1000 °C 3. >1000 °C

ክፍል ሁለት – በወባ እና የሆድ ትላትል በሽታዎች ላይ

ያሳቸው እውቀት እና አመለካከት በተመለከተ

401.ስለ ወባ ሰምተው ያውቃሉ?

1.አዎ 2.አሳውቅም

402.ለተራ ቁጥር 401 መልሶዎት አዎ ከሆነ ወባ ተሳሳፊ በሽታ መሆኑ ያውሉ?

1.አዎ 2.አሳውቅም

403.ስተራ ቁጥር 402 መልሶዎት አዎ ከሆነ መተሳለፍያ መንገዶቹ ምን ምን ናቸው?

 1. የትንኝ ንክሻ
 2. ደም በ
 3. ከበሽተኛው ጋር በሚድረግ

 ቀጥታ ነክኪ
 4. ጡት በማጠባት

 5. ሌላ ካለ ይግልው_____

404.ለተራ ቁጥር 401መልሶዎት አዎ ከሆነ የሽታው ዋና ዋና ምልክቶች ምን ምን ናቸው?

1.ትኩሳት 2. ማንቀጥቀጥ 3. ራስ ምታት 4. ተቅማዋ

5. የምግብ ፍላጎት መቀነስ 6. ሌላ ካለ ይግልፁ_____

405.ስለ ሆድ ትሳትል በሽታዎች ሰምተው ያውቃሉ?

1.አዎ 2.አሳውቅም

68

406.ለተራ ቁጥር 405 መልሶዎት አዎ ከሆነ የሆድ ትሳትል በሽታዎች ተሳለፌ መሆናቸውን ያውቃሉ?

1.አዎ 2.አሳውቅም

407. ለተራ ቁዋር 406 መልሶዎት አዎ ከሆነ መተሳለፍያ መንገዶቹ ምን ምን ናቸው?

1. ያል በሰለ ምግብ መመገብ 2. እጅ ሳይታጠቡ መመገብ 3. የለጫጣ መሂድ

4. አየነ ምድርነ እነደ መዳበረያ መጠቀም 5. ሌላ ካለ ይግልፁ_____

ክፍል ሦስት -የተሳታፊዎች ባህርይ በተመለከተ

501.ምግብ ከመመገብዎ በፊት ምን ህል እጅዎትን ይታጠባሉ?

1.ሁሌ በውኃና ሳሙና እታጠባለህ

2.ሁሌ በውኃ ቢቻ እታጠባሎህ

3. ሁሌ በውኃ/አንዳንዴም በሳሙና እታጠባሎህ

4.አንዳንዴ ሳልታጠብ አመገባለሁ/ምግብ አዘጋጃለሁ

502.ለተራ ቁዋር 501መልሶዎት `መ' ከሆነ ሁሌ በው**ታና ሳሙና እንዳይታ**ጠቡ ምክንያቶት ምንድነው?

1.ውኃ ማጣት 2.እጄ የምታጠብበት ግዜ ማጣት

3.እጄ ላይ የሚታይ ቆሻሻ ከሌለ መታጠብ አለብኝ ብየ ስለማሳስብ

4. ማድየለሽነት 5. ለምን ሁሌ በውኃና ሳሙና እንደማልታጠብ አሳውቅም

6.ሌላ ከሆነ _____

503. ከሽንት ቤት በኃላ ይታጠባሉ?

1.አዎ/ሁሌ 2.አዎ/አንዳነዴ 3.አልታጠብም

504. ልጅዎን ካፀዳዱ በሗሳ እጅዎን የታጠባሉ? (ሀፃን ካልዎት)

69

1.ሁሌ በውኃና ሳሙና እታጠባለህ 2.ሁሌ በውኃ ቢቻ እ*ታ*ጠባሎህ 3.ሁሌ በውኃ/አንዳንዴም በሳሙና እታጠባሎሀ 4.አንዳንዴ አልታጠብም 505. ይለብሰሉ የቅጠላ ቕጠል ምግቦች እና ፍራፍሬዎች ባለፉት ሁለት ወራት ተመግበዋል? 1.አዎ/በየቀኑ 2.አዎ/አንዳነኤ 3.ተመግቤ አላውቅም 506. የሰው ዓይነ ምድር ለመዳበርያነት ይተቀማሉ? 1.አዎ 2.አልጠቀምም 507.የመጠዋ ውኃ የሚያገኙት ከየት ነው? 1.የቧንቧ 2.የታጠረ ምንጭ 3.የልታጠረ ምንጭ 4. ወንዝ 5. የዝናብ ወሃ 6.ሌላ ከሆነ _____ 508.የፌሳሻ ቆሻሻ የት ያስወግዳሉ? 1.ክፍት ሜዳ ላይ 2.ንድንድ ውስጥ 3.ሽንት ቤት ውስዮ 4.ሌላ ከሆነ _____ 509.አምበርን የወባ ቲንኞች ንክሻ ለመከላከል ይጠቀማሉ? 1.**አ**ዎ 2.አልጠቀምም 510. **ስንት አኰበር አልዎት**? 511. በወቅቱ አኰበሩን የሚጠቀምበት ማን ነው? ሀ ከአምስት አመት በታች ህናናት በከአምስት አመት በላይ ህናናት ሐ ነፍሰጡር እናት መ አባዎራ ሥ. ሁሉም የቤተሰብ አባል ሪ.አምበር ሚጠቀም የቤተሰብ አባል የለም 512. አምበሩን በየጊዜው በመድሓኒት ይነከራል? 1.አዎ 2. አየነስርም 513. መኖርያ ቤትዎ በዚህ ክረምት በፀረ ወባ ቲንኞች መድሓኒት ተረጭተዋል? 1.**አ**ዎ 2.አልተረጨም

514. **ያለ ጫጣ የመሂድ ልምድ**

1.ሁል ጊዜ 2.አንዳንድ ጊዜ 3. ያለ ጫማ ሀጄ አላውቅም

515. በእርግዝና ጊዜዎ አፈር የመብሳትን እንደ ባህላዊ መደሃኒትነት ይጠቀማሉ?

1.አዎ 2.አልጠቀምም

የመጠይቁ ቁዋር ____

መጠይቁ የሞሳው ሰው ስም _____

Annex 7- Consent form in English

I am requested to participate in the study by being informed of the objective of the study, which is to determine the current prevalence of malaria and intestinal helminths co-infection among pregnant women in Gilgel Gibe area and to assess the possible risk factors for these co-infections.

Because I believe that the results of this study have importance to be used as a guide for the prevention and/or control malaria and intestinal helminths in Gilgel Gibe area and the country as a whole, I have agreed voluntarily to provide the necessary information answering all the questions included on the questionnaire; without requesting any costs from the researcher.

Signature of participant: date

ANNEX 8- Questionnaire in English

Introduction -This questionnaire, developed for master's thesis, is designed to collect data about prevalence and risk factors of malaria and intestinal helminths co infection among pregnant women living around Gilgel Gibe area. The result of this research is believed to enhance the health status of the community; especially the most vulnerable community members, pregnant women. So we, in the very begging, are gland to acknowledge your innocent response.

Part I: General Information

I. Address

Name of wereda/ District	
Name of Village/Kebele	
Name of Got	
Participant ID No	
House number	
Average distance from the Dam Kn	1

II. Sociodemographic information

- 201. Age:_____
- 202. Ethnic group:
 - 1.Oromo 2.Amhara 3.Kefa 4.Yem 5.Dawro 6.Tigre
 - 6.Gurage 7.Other specify

203. Religion

- A. Muslim
- B. Orthodox
- C. Protestant
- D. Catholic
- E. Others_____

204. Occupation

1.Housewife 2.Farmer 3.Daily laborer 4.Student 5.Civil servant6.Business man 7.Other specify______

205. Family monthly income

- 1.< 500 2.500 1000 3.>1000
- 206. Marital status
 - 1.Single 2.Married 3.Divorced 4.Widowed

- 207. Number of children _____(Including alive children(if any))
- 208. Home 1. Owen 2. Rent
- 209. Educational status
 - a. Illiterate
 - b. Read and write only
 - c. Primary school
 - d. Secondary school
 - e. Above Secondary school

210. Trimester

- 1. First trimester (First 3 months)
- 2. Second trimester $(4th 6^{th} month)$
- 3. Third trimester (Above 7th month)

III. Condition of Household

- 301. Main material of the roof
 - 1. Thatched
 - 2. Corrugated iron sheet
 - 3. Other, specify_____
- 302. Main material of the floor
 - 1.Earth
 - 2. Cement
 - 3.Wood
 - 4.Other, specify_____

303. Main material of the wall

2. Grass 2.Mad 3.Cement 4.Other, specify_____

304. Are there animals living with the family inside the household?

1. Yes 2. No

305. If there animals living inside the household; specify_____

306. Is there cattle's shelter around your house?

1. Yes 2. No

- 307. Is there stagnant water around your compound?
 - 1. Yes 2. No

308. If yes to Q307, Distance of your house from stagnant water

1. < 100M 2. > 100 - < 500 M 3. 500 - 1000M 4. > 1000M

309. Are there mosquitoes in this house?

1. Yes 2. No 310. Do you have latrine? 1. Yes 2. No 311. If there is latrine facility what type? 2. Pit 1. Flush toilet 3. Other, specify_ 312. Location of the latrine 1. In the house 2. Outside the house but in compound 3. Outside the compound (Communal) 313. Does the latrine have cover? 1. Yes 2. No 2. No 314. If the latrine has cover, do you use it? 1. Yes

315. Is the toilet provided with water supply for hand washing? 1. Yes 2. No

316. Distance of your house from the nearby health facility

1. < 500M 2.500 - 1000M 3. > 1000M

Part two –Questions related to Knowledge and attitudes towards the diseases

401. Have you heard about human malaria?

1. Yes 2. No

402. If yes for Q.401, Do you know that malaria can be transmitted from person to other person?

1. Yes 2. No

403. If your response for question number 402 is yes, what are the possible ways of transmissions?

404. If your response for question number 401 is yes, what are the major sign and symptoms of the disease?1.Fever2. Shivering3. Headache4. Diarrhea

5. Loss of appetite 6. If other please specify;

405. Have you heard about human intestinal helminth?

1. Yes 2. No

406. If your response for question number 405 is yes, do you know that intestinal helminthaisis can be transmitted from person to other person?

1. Yes 2. No

407. If your response for question number 406 is yes, what are the possible ways of transmissions?

1. Eating uncooked food 2. Eating without washing hands 3. Walking bare foot

4. Using unfertilized soil as a fertilizer 5. If other please specify;_____

Part three - Questions related to practice

501. Habit of hand washing before meal.

- 1. Always with water and soap
- 2. Always with only water
- 3. Always with water and sometimes with water & soap
- 4.I sometimes eat without washing my hands

502. If your response for question number 501 is 4, what is/are your reason(s) not to always wash your hands you eat/handle food?

1. Lack of water

2. Lack of time to wash hands

3.I don't think that I must always wash my hands unless contaminated with visible dirty

- 4. Carelessness
- 5.I don't know why I don't always wash
- 6. Others please specify_____

503. How frequently do you wash your hand after latrine use?

1. Always 2. Sometimes 3. Not at all

504. If you have child do you wash your hands after clean your child's feaces?

1. Yes, Always with water and soap

2. Yes, Always with only water

3. Yes, Always with water and sometimes with water & soap

4.I sometimes forget washing my hands

505. Habit of eating uncooked vegetables and fruits during the last two months

1. Daily 2. Some times 3. I never consumed uncooked vegetables and fruits 1. Yes 506.Do you use human feaces as a fertilizer? 2. No 507.Source of drinking water 1. Tap 2. Protected well/spring 3. Unprotected well/spring 4. River/pond 5. Rain 6.Others, please specify____ 508. Where do you dispose your liquid wastes? 4. Other, specify_____ 1. On field 2. Pit 3. Septic tank/toilet 509. Do you use ITN to prevent mosquito bite? 1. Yes 2. No 510. How many ITN do you have at home?_____ 511. Who is using the ITN currently? 1.Children under 5 year 2.Children above 5 year 3.pregnant women 5. All family members 4. Husband 6.No body is using ITN 512. Are the ITN impregnated on time? 1. Yes 2. No 513. Is your house sprayed with insecticide in this summer? 1. Yes 2. No 514. Habit of walking bare foot 2. Sometimes 3. Not at all 1. Always 515. Do you have the habit of eating soil during your pregnancy period as a traditional medicine? 1. Yes 2. No

Annex 9- <u>Laboratory Result Delivery Format</u> Name of the health facility _____

			Laboratory Result			
Sr.No	Participant ID.					
		Stool Examination		Blood Film Examinatio	HCT Normal(
		Qualitative result	Total parasite load/g	n	37-47%)	
						-
Name a	and Signature o	f the investigator- Milli	oŋ ₈ Getachew(MSc C	andidate)_		

Annex 10- Data collection and processing pictures/Plates.



Plate 1. The long distance journey on foot with HEW in the sunny day, to reach the house of randomly selected study participants in Gigel Gibe area, 2011.



Plate 2. Health Extension worker (HEW) collecting data using semi- structured questionnaire in Gigel Gibe area, 2011.



Plate 3. Capillary blood collection during house to house survey in Gigel Gibe area, 2011.



Plate 4. Blood sample collection at Burqa Asendabo health post, 2011



Plate 5. Transporting stool sample from the field to Jimma University Laboratory for processing 2011



Plate 6. Stool sample processing at Jimma University clinical laboratory 2011