

JIMMA UNIVERSITY
COLLEGE OF PUBLIC HEALTH AND MEDICAL SCIENCES
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MAGNITUDE OF ANEMIA AND ASSOCIATED FACTORS AMONG PREGNANT
WOMEN ATTENDING ANTENATAL CARE AT GAMBELLA HOSPITAL

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ABSTRACT

Back ground: In Ethiopia, anemia is the severe problem, affecting higher proportion of pregnant women. Despite the severity of the problem; no sufficient research data has been documented to disclose associated factors of anemia among ANC attendants in the study area.

Objectives: To assess magnitude and associated factors of anemia among pregnant women attending ANC in Gambella hospital.

Methods and materials: Cross-sectional study was conducted at Gambella hospital from April 2 to May 2, 2014. The required sample size (n) was 374. All women presenting at the hospital for antenatal care (ANC) independent of their stage of pregnancy and the number of visits were asked to participate in the study. Data was collected using pretested interviewer administered questionnaire and laboratory examination was also under taken. Univariate and bi-variate analysis was carried out and finally model was fit using multi-variate logistic regression.

RESULT: Three hundred and forty six pregnant women were surveyed in the study making a response rate of 93.5%. Mean HbC values of the pregnant women was 12.11 g/dl \pm 1.36. Majority of the participants 272(78.6%) found to be normal where as 67(19.4%) were having mild anemia and the rest 7(2%) had moderate anemia. Among the study participants malaria and ova parasite was found in 28(8.1%) and 25(7.2%) respectively. Final analysis in the multi-variate logistic regression indicated that pregnant women with malaria parasite infection were 15.8 (AOR= 15.7 95%CI: 5.09-48.411) more likely to be anemic than those free of infection. Pregnant women with helminthes infection were (AOR= 8.124, 95% CI: 2.754-23.96) more likely to be anemic than their counter parts. This study also indicated dietary habits with less than once consumption of vegetables per day were (AOR=4, 95%CI: 1.4-11.8) more likely to be anemic than their counter parts. It was also found that pregnant women who do not receive iron supplementation during their current pregnancy were (AOR=2.5, 95% CI: 1.195-5.128) more likely to anemic than their counter parts.

Conclusion: Based on the data presented in this study, parasitic infection such as malaria and helminthes, daily green leafy vegetable consumption and iron tablet supplementation were significantly associated with anemia among pregnant women.

Recommendation: we recommend to stakeholders to give emphasis to parasitic infection control such as malaria and helminthes control mechanisms and strengthen awareness creation activities to improve dietary habit of pregnant women on consumption of iron rich foods.

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ACRONYMS

ANC- Antenatal Care

BMI- Body Mass Index

CDC- Centers for Disease Prevention and Control

DALY- Disability Adjusted Life Years

EDHS- Ethiopia Demographic and Health Survey

g/dl- Gram Per Deci Litre

HBC- Haemoglobin Concentration

IDA- Iron Deficiency Anemia

IEC- Information Education Communication

ITN- Impregnated Treated Net

LBW- Low Birth Weight

MUAC- Mid Upper Arm Circumference

NGO- Nongovernmental Organization

PCV- Packed Cell Volume

RBC- Red Blood Cells

WHO- World Health Organization

10X- Ten Times Magnification

40X- Forty Times Magnification

CHAPTER ONE: INTRODUCTION

1.1 Background

Anemia is a critical health concern because it affects adversely growth and energy levels. Anemia is one of the world's biggest health problems. According to the World Health Organization (WHO), in 2005, anemia affected 1.62 billion people worldwide (24.8% of the global population). Preschool children were the social group that showed the highest prevalence of anemia, with an impact rate of 47.4%, followed by pregnant women at 41.8% (1).

Anemia is characterized by reductions in hemoglobin concentration (HbC), red blood cell count (RBC), packed-cell volume (PCV), and subsequent impairment in meeting the oxygen demands of tissues(2). The definition of anemia can be based on either hemoglobin or hematocrit concentration. It varies by age, sex, altitude, smoking, and pregnancy status(3)(4). According to the World Health Organization (WHO), anemia in pregnancy is defined as hemoglobin (Hb) concentration of less than 11 g/dL(1). There is an increased iron requirement during pregnancy due to greater expansion in plasma volume that results in a decrease in hemoglobin level (5).

Although anemia results from a wide variety of causes it is generally perceived that the most important contributing factor of anemia is iron deficiency, which accounts for about half of all anemia cases(2). Iron deficiency anemia is most prevalent and severe in young children (6-24 months) and women of reproductive age and pregnant women due to growth and the formation of new tissues , but is often found in older children and adolescents and may be found in adult men and the elderly(6). Blood losses during menstruation largely determine the iron requirements of non-pregnant women (6).

IDA prevalence indicates the nutritional status of a community. Considering the effects of IDA on maternal and fetal mortalities, physical function and child growth and development , it is regarded as one of the main health indicators(7). Recently, new studies of the economic costs of anemia have been completed indicating the massive cost burden of this disease and the cost effectiveness of reducing iron deficiency anemia rates in children and women. Such studies consider lost productivity, health costs and lifetime costs related to the permanently impaired cognitive development of young children who develop iron deficiency anemia (8).

The WHO/World Bank-supported analysis of the Global Burden of Disease ranked iron deficiency anemia as the third leading cause of loss of disability-adjusted life years (DALYs) for females aged 15–44 across the globe. Among men in this age group, iron deficiency anemia is ranked among the top 10 disease burdens globally, reflecting the debilitating effects of anemia even in this group. This factor was more important globally than war-related death and disability, and nearly as important as the global scourge of tuberculosis (8).

However, the interventions necessary to prevent iron deficiency anemia have high benefit/cost ratios, and are among the most cost effective in the realm of public health. The high economic costs of anemia and the low costs of interventions to address the problem should persuade national leaders to make the necessary policy decisions needed to fulfill these commitments (8).

1.2 Statement of the problem

Because of depleting their iron stores by the end of their pregnancy, losing a large amount of blood during childbirth, secreting a large amount of nutrients in their breast milk, and recurrent menstrual loss, women of reproductive age are most at risk of anemia(2) . According to WHO's estimate, the global prevalence of anemia in pregnant women is 68%. In Africa its prevalence is estimated to be 66.8% (5).

In Ethiopia, anemia is the severe problem, affecting higher proportion of pregnant women (22%) than women who are breastfeeding (19 %) and women who are neither pregnant nor breastfeeding (15%). According to EDHS 2011 the prevalence of anemia among pregnant women in Gambella is 20% (9), which is closer to the national prevalence.

Anemia is the most common hematological disorder during pregnancy(7)(10). Acute onset of anemia during pregnancy will greatly increase the risk of death because this can lead to rapid cardiac decomposition (11). During pregnancy any hemoglobin level below 11g/dl has been associated with premature labor, low birth weight , maternal mortality , and perinatal mortality (2)(10).

The aetiology of anemia in pregnancy is complex and multi-factorial (10)(13). In sub-Saharan Africa causes of anemia in pregnancy include infections such as malaria, hookworm and iron- and folate-deficient diet, maternal and reproductive factors as well as health service related factors and chronic illnesses such as chronic kidney disease (10).

Parasitic diseases, including helminthes infections and *P. falciparum*, have long been recognized as important contributors to anemia in endemic countries. Malaria due to *P. falciparum* clearly contributes to anemia throughout life and specifically during pregnancy(13). Infections with helminthes causing chronic blood loss are another major cause of iron deficiency anemia in areas where such infections are endemic (8).

Tea and coffee inhibit iron absorption when consumed with a meal or shortly after a meal(6). In addition to these diet-related causes, iron deficiency in women of childbearing age is also associated with repeated pregnancies and excessive menstrual bleeding(8). The consequences of low socio-economic status that effectively raise anemia rates include a lack of food security, inadequate or lack of access to health care and poor environmental sanitation and personal hygiene (8).

A varied array of interventions exists that are designed to prevent and correct iron deficiency anemia. These include dietary improvement, fortification of foods with iron, iron supplementation, and other public health measures, such as helminthes and malaria control as well as expansion of health services(14). Fortification of suitable food vehicles with absorbable forms of iron is a highly desirable approach to controlling iron deficiency (14).

Since Fetal demands for iron are maximal during the second and third trimester(15), Centers for Disease Control and Prevention (CDC) recommend screening for anemia in pregnant women and universal iron supplementation which is a proven and effective public health intervention to meet the iron requirements of pregnancy except in the presence of certain genetic disorders such as hemochromatosis (5).

The nutritional status of women in Ethiopia, like in other developing countries, is low while their daily workload is often enormous to ensure survival of their children(5). Thus pregnant Women should be supplemented with Iron and or folic acid during their ANC attendance at health facilities. But According to EDHS 2011 About six in every ten Ethiopian women (57%) did not receive any antenatal care for their last birth in the five years preceding the survey. In Gambella region due to many reasons, 42% of pregnant women did not receive any antenatal care which is still substantial percentage (9).

The severity of the problem in poor countries like Ethiopia is widespread and associated with socioeconomic status, dietary behavior inaccessibility of health care services and parasitic infections such as helminthes and malaria and other factors of the population. Since malaria is endemic in Gambella

region, it exacerbates the risk of anemia among pregnant women. Despite the wider scope of the problem, no sufficient research data has been documented to disclose associated factors of anemia among ANC attendants. Therefore, the objective this study was to determine factors associated with anemia among pregnant women attending ANC in Gambella hospital.

CHAPTER TWO: LITERATURE REVIEW

Globally it is believed that Anemia increased maternal morbidity and mortality. In developing countries, it is commonly said that over 50% of pregnant women are anemic(16). It has been reported that close to 500,000 maternal deaths occur every year, vast majority taking place in developing world. Anemia is thought to be the major contributory cause of death in 20 – 40% of these maternal deaths. Furthermore it has been estimated that 16–20% of all maternal deaths are associated with iron deficiency anemia (17).

2.1 Anemia

Severe anemia that occurs in developing countries are a major cause of maternal mortality and morbidity and attributed as a direct or indirect cause of about 26 % of maternal death in Africa(16). A study from Gondar, Azezo health center showed over all prevalence of anemia was 21%, among them 49% were with mild anemia, whereas moderate and severe anemia was recorded in 46% and 5% respectively (18).

A study conducted at rural community in India among pregnant women showed that 310 subjects were enrolled, of whom 74.8% were found to be anaemic. The majority (50.9%) demonstrated moderate anaemia while mild and severe anaemia was recorded in 30.17% and 18.9% respectively. All the pregnant women with past history of abnormal vaginal bleeding (11.4%) were anaemic and 76.9% had moderate or severe anaemia (19).

A study conducted among pregnant women Shalla woreda showed that Mean haemoglobin level was found to be 12.0 ± 1.5 gm/dl with prevalence of 36% in which majority of them were mild anemia(5). A very high prevalence (82.9%) of anaemia was observed among pregnant women in India. The study observed significantly higher prevalence of anaemia among those pregnant women above 26 years of age (97.7%) & those from below Class IV socio-economic status (90.7%)(20).A study conducted among pregnant women at Bushalo health center on southern Ethiopia showed that the overall prevalence of anaemia in this study population was 51.9%. Prevalence of anaemia among rural women was higher than prevalence of anaemia among urban women i.e., 54.8%and 41.6% respectively (21).

2.2 Obstetric and reproductive related factors

Study conducted at Bushalo health center in southern Ethiopia, the trend of anaemia showed that as gestational age increase, the occurrence of anaemia also increases(21). Study conducted in rural

community of eastern Ethiopia, also indicated that the magnitude of anemia decreases as the gap between previous birth and current pregnancy increases, i.e. mothers who delayed the subsequent pregnancy for more than two years had lower prevalence of anemia, than those who delivered in less than two years (5).

Among pregnant women in Indian, Higher proportions of anemic women were also observed among the pregnant women who gave 2 births, 3 births and 4 or more births than among those who did not give birth (22). The proportion of anemia, Study done in westmore land, Jamaica, was higher among pregnant women who had history of abortion, did not use contraceptive, and experienced excess menstrual bleeding prior to the current pregnancy, compared to their counterparts (11).

2.3 Parasite infection and chronic illnesses

2.3.1 Malaria parasite infection

Most malaria infections and the most severe morbidity and mortality are caused by *Plasmodium falciparum* world wide. The other three human malaria parasites (*P. vivax*, *P. malariae* and *P. ovale*) contribute to fewer infections and to more moderate disease and relatively few deaths (21). Malaria constitutes a major public health problem and impediment to socioeconomic development in Ethiopia. It is estimated that about 75% of the total area of the country and 65% of the population is estimated to be at risk of infection (23).

Malaria infection causes 3%–5% of maternal anemia and, worldwide, about 50 million women are exposed to malaria(13) ,especially in malaria-endemic areas, malaria is the cause for almost 25% of maternal deaths each year(24). 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 anaemia (haemoglobin < 80 g/L or hematocrit < 0.25) each year in sub Saharan Africa as a result of malaria infection making pregnant mothers susceptible to increased risk of mortality (4) .

Although the vast majority of women with malaria infections during pregnancy remain asymptomatic, infection increases the risk of maternal anemia and delivering a low-birth-weight (LBW) baby. *P. falciparum* is the only human malaria parasite that is more common in pregnant than in non-pregnant women and is the only human parasite with a clear and substantial adverse effect on pregnancy, nutrition during pregnancy and pregnancy outcome (21). Malaria is thought to be the primary cause of severe anemia (Hgb < 7 g/dl) in at least 50 % of subjects living in malaria-endemic areas. Studies conducted in

Mali and Benin, where there is stable transmission of malaria throughout the year, indicated significant association between malaria infection and anemia in pregnancy (25)(26).

A study conducted in Shalla woreda, showed that the prevalence of anemia in pregnant women who do not use bed net every night was 51%. Analysis showed that consistent use of bed net and history of acute febrile illness during pregnancy were significantly associated with the occurrence of anemia(5). In another study from Ouagadougou , Burkinafaso it was found that anemia was significantly more common in women infected with *P. falciparum* (33%) compared with the uninfected pregnant women (10%) (24).

2.3.2 Helminthes Infection

Anaemia is particularly common in individuals infected with soil-transmitted helminthes(27). Heavy infections of *T. trichiura* and *A. lumbricoides* have long been known to be associated with iron deficiency anemia. Especially hookworms cause severe anemia and malnutrition in developing countries of the tropics (28).

An association was found between the presence of hookworm eggs in stool in the last trimester of pregnancy and low hemoglobin concentration(21). Study conducted in Gondar, Azezo health center was found significant association between the presence of hookworm and anaemia in pregnant women. Anemia was higher among pregnant women with helminthes infection than their counterparts (18).

A study conducted among pregnant women in Jimma university hospital showed a statistical significance difference between anaemic and non-anaemic cases with hookworm infection(29). A study conducted in Mali indicated that prevalence of anemia was higher among pregnant women with helminthes infection than their counterparts(25). From infected pregnant women in Venezuela half of them had mixed infections, due to different parasite species, which represented a significant risk to have anaemia, almost twice than those women who did not (21).

Hemoglobin levels were lowest in pregnant women who had helminthes or malaria infections (30). Heavy hookworm infection was also significantly associated with a lower Hb level compared to light infection(31). A study in Cote d'Ivoire included 32 pregnant women treated with pyrantel pamoate and showed that the prevalence of hookworm decreased by 93% and Hb increased by 6 g/L over the course of the pregnancy (31).

2.4 Nutrition and dietary behavior

Among pregnant women in Indian, who had low intakes of Fe and several other nutrients, higher intakes of Ca and P (dietary components known to inhibit Fe absorption) were independently associated with a higher prevalence of anaemia (32).

Another Study done in westmore land, Jamaica showed that the risk of anemia was reduced among the women who consumed green leafy vegetables once or more in a week compared to those who did not(22). Similarly it was also showed that pregnant women with a Body Mass Index (BMI) below 25 and a Mid-Upper Arm Circumference (MUAC) less than 25 cm were more likely to be anemic(11). Pregnant women who reported that they take tea after meal were more likely to develop anemia compared to who took less than once per day (5).

2.5 Health care service and medication history

Study from Bushalo health center showed that Pregnant women who did not take iron during pregnancy were greater than two times likely to be anaemic than those who took iron supplementation(21). Similar study in Azezo health center also indicated that significant association between iron supplementation and anemia in pregnancy (18).

Moreover Pregnant women attending antenatal care regularly had less prevalence of anaemia comparing to those pregnant women who did not attend antenatal care, on their previous pregnancy (21). The finding were also similar with the study conducted at westmore land, Jamaica , that Pregnant women who had four or more antenatal care visits were less likely to be anemic than women who had less than four antenatal visits (11).

Generally parasitic infections such as Malaria and helminthes infections and nutritional and nutrient factors such as low BMI and MUAC, restrictive dietary behavior (low consumption of meat, vegetables and fruits in the diet), intake of iron absorption inhibiting factors like tea and coffee after meal frequently were found to be associated factors for anemia. Other obstetric and reproductive factors such as gestational age, history of abortion, contraceptive use, excess blood loss during menstrual period, and giving birth two or more were found to be associated factors for anemia. Furthermore socio demographic factors such as low income and maternal age and health care service like low ANC attendance (visit) and lack of iron supplementation were also found to be associated factors for anemia among pregnant women.

2.6 Conceptual frame work

The conceptual frame work was constructed after reviewing many published literatures and journals about factors of anemia in pregnancy. All associated factors has been grouped in to 5 categories which are parasitic infections and chronic illness, maternal and reproductive related factors, nutrition related factors, socio demographic and economic factors and health care services related factors.

Obstetric and reproductive history: obstetric and reproductive related factors can directly, in case of maternal blood loss, or indirectly by other mechanisms can be associated factors of anemia among pregnant women

Parasitic infection and chronic illness: malaria infection in pregnant women can lead to anemia especially in malaria endemic area like Gambella. A Helminthes infection such as hook worm greatly contributes for anemia in pregnant women. Illnesses such as chronic kidney disease can cause anemia in pregnant women

Nutrition and dietary behavior: intake of iron inhibiting foods like tea and coffee, low iron dietary consumption and less meat and vegetables consumption may lead to anemia in pregnant women. Less consumption of meat, vegetables and fruits may deteriorate the immune system of the body and as result malaria infection may occur.

Socio-demographic and economic factors: factors related to socio demographic are associated factors of anemia in pregnancy. Low income may be associated with less utilization of health services such as ANC. In addition to that low income may also be associated with less consumption of iron containing dietary foods. Low socio economic might be also be associated with susceptibility to parasitic infection and undesirable obstetric and reproductive history.

Health care services and medication history: less utilization of health care services can result in anemic status of pregnant women. Lack of iron supplementation and de-worming can result anemia in pregnancy. Not utilizing Contraceptive methods prior to the current pregnancy can also be associated with less health care services utilization.

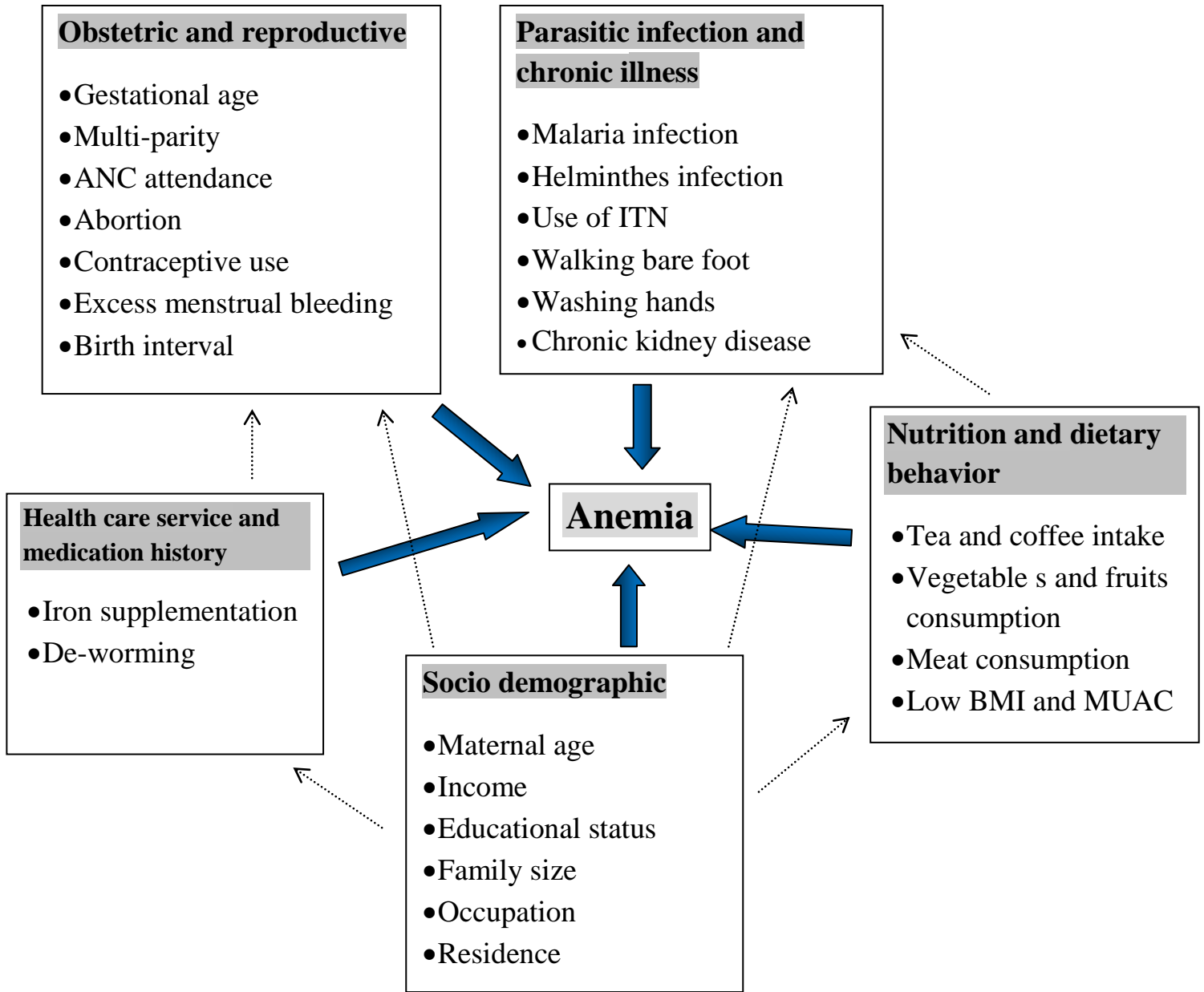


Figure 1: conceptual frame work of factors associated with anemia among pregnant women

Source: developed after reviewing different literatures

2.7 Significance of the study

This research was conducted to determine the magnitude and factors associated with anemia among pregnant women attending ANC in Gambella hospital. Since there was no previous data about magnitude and associated factors of anemia among pregnant women attending ANC in the study area, the finding of this study has illustrated the association and contribution of parasitic infections such as malaria and helminthes infection with anemia in pregnancy. This may provide statistics needed, to Gambella regional health bureau and other stake holders working on maternal and reproductive services, for planning parasitic infection control programmes that aims at reducing the prevalence of anemia.

The present study can also be used as basis for dietary related interventions that can be directed at dietary improvement of pregnant women that can be achieved through providing health educations and awareness creation activities on the use of diversified dietary habits. The finding can also be valuable to the ANC program and services to include malaria and helminthes infection screening for every pregnant woman in addition to hemoglobin measurement and to integrate iron supplementation with parasite treatment. There by to improve the service of ANC and improve the health status of pregnant women. More over the finding of the study can provide data for further investigation and research regarding anemia in pregnancy.

CHAPTER THREE: OBJECTIVES

3.1 General objective:

- To assess the magnitude and associated factors of anemia among pregnant women attending ANC in Gambella hospital

3.2 Specific objectives:

- To determine the magnitude of anemia among pregnant women attending ANC in Gambella hospital
- To assess factors associated with anemia among pregnant women attending ANC in Gambella hospital

CHAPTER FOUR: METHODS AND MATERIALS

4.1 Study area and period

Gambella Regional State is located at south west Ethiopia which is 766 kilometers away from Addis Ababa. It has a total area of 25,802 square kilometers. The Climate of the region is semi arid& arid. The average annual rain fall ranges from 900-2100 mm with an average mean temperature of 27-33⁰c. The rain season is from May to October & the dry season from November to April. The region has a total population of 418,298 consisting of 25% urban inhabitants. It has a total fertility rate of 4.0. The region is bordering Oromia region in the North & East, SNNPRS in the South & East, & south Sudan republic in West. The region has three administrative Zones (Nuer, Agnwua & Mejenger), one special woreda and one town administration with a total number of 14 woreda's in the region. The region has a total of 108 functional health posts and 31 functional health centers.

Means of livelihood for the Majority of the population is farming and livestock production, where as some sort of the population are employees and others engaged in small and medium scale trading. Maiz, sorghum and Teff are the main crops consumed throughout the region and Injerra, pourage as well as fish are staple food throughout the region.

The study was carried out in Gambella Hospital, which is the only hospital found in Gambella town, the capital of Gambella regional state. The altitude ranges from 300-600 meters above sea level. The town is divided in to five lowest level administrative kebeles. It has a total population of above 52,000. The study was conducted at Gambella hospital from 2 April- 2 May 2014.

4.2 Study design

Facility based cross sectional study was conducted among pregnant women attending ANC at Gambella hospital.

4.3 Population

4.3.1 Source population: All pregnant mothers attending ANC at the hospital.

4.3.2 Study population: pregnant mothers who came for ANC service during the study period were the study population.

4.4 Inclusion criteria:

A pregnant woman who came to Gambella hospital for antenatal care visits or follows up during the study period.

4.5 Exclusion criteria:

A pregnant woman revisits for ANC during the study period who were already interviewed and pregnant women visiting the hospital due to severe medical conditions.

4.6 Sample size and sampling techniques

4.6.1 Sample size determination

The required sample size (n) was determined using single population proportion formula at a confidence level of 95%, value of a standard normal distribution score using 0.05 level of significance; expected prevalence of anemia in pregnant women was 20%(9), at $d = 0.04$, degree of accuracy desired. Proportion of different variables has been taken into account and calculated to look for maximum sample size, at the end maximum sample size was obtained by taking the prevalence of anemia among pregnant women which was 20% according to EDHS. Thus applying the formula,

$$n = \frac{(z_{\alpha/2})^2 \cdot pq}{d^2}$$

- Where n= total sample size
- Z= confidence interval
- P= prevalence of anemia
- D= margin of error

The sample size was 384 pregnant women. Since the source population was $< 10,000$ or n/N was greater than 0.05, population correction formula was used to determine sample size to be 340. By adding 10% for non-response rate the final sample size became 374.

4.6.2 Sampling technique /Sampling procedures

Using consecutive sampling method all pregnant women presenting at Gambella hospital for antenatal care (ANC) during the study period independent of the number ANC visit and their stage of pregnancy were asked to participate in the study after they met the inclusion criteria. Interview was continued until the end of specified period of time.

4.7 Data collection procedures

4.7.1 Questionnaires

Data was collected using pretested interviewer administered questionnaire, which contains socio-demographic characteristics, parasitic infection related factors, reproductive and maternal factors, health service related factors and nutrition related questions that was used to assess the habitual dietary intake of the pregnant women's enrolled into the study. MUAC measurement was also taken from each pregnant mother from their upper middle left arm using standard MUAC tape. BMI measurement was done by calculating their weight divided to their height in meter square. All these measurements were taken during the interview.

Data was collected at MCH department when pregnant women came for ANC visit to the hospital on the working hours of the institution. And it was collected by two trained Bsc nurses, who know the local language, working at the hospital.

4.7.2 Laboratory examination

4.7.2a Stool specimen examination

Collection of stool specimen: Stool specimen containers was given to each pregnant woman with toilet tissue paper and clean applicator stick after questionnaire administration to brought fresh stool specimen. Orientation was given to the women on how to collect sufficient amount and contamination free stool specimen. Pregnant women were requested to bring the stool sample immediately to process and examine. The laboratory technicians were checking appropriateness of stool specimen using rejection criteria's during receiving of sample.

Direct microscopy: Stool smear was prepared using saline for direct microscopic identification of intestinal helminthes and protozoa infection. Two slides were prepared for each pregnant woman. Direct smear was examined by 10X and 40 X microscopic magnifications.

4.7.2b Blood film examination

Pregnant women were screened for the presence of malaria infection by collecting blood from finger prick. Giemsa stained of thick and thin smears were prepared to determine the presence or absence of malaria parasite.

4.7.2c Measurement of anemia

Venous Blood sample of 2 ml was collected from each study subjects using EDTA tube. The venous blood was used immediately for testing hemoglobin concentration (HBC) by automated hematology analyzer CBC machine (cell dyn 1800). For the purposes of this study, the World Health Organization standard (Hb <11g/dl) was used to determine anemia in pregnancy based on hemoglobin levels.

4.8 Study variables

4.8.1 Dependent variable:

- ❖ Anemia in pregnant women

4.8.2 Independent variables:

❖ Socio demographic and economic factors

- Maternal age
- Occupation
- Family size
- Family income
- Educational status
- Residence

❖ obstetric and reproductive related factors

- Gestational age
- Multi gravidity
- Contraceptive use
- Excess menstrual bleeding
- Birth interval
- Abortion

❖ Parasitic infection and chronic illness

- Malaria infection
- Helminthes infection
- Chronic kidney disease
- Drinking water source
- Use of ITN
- walking bare foot
- latrine availability
- Hand washing habit

❖ Nutrition and nutrition related factors

- Meat and animal product consumption
- Vegetables and fruits consumption
- Tea and coffee intake
- BMI and MUAC

❖ **Health service related factors**

- ANC attendance
- Iron supplementation
- De-worming

4.9 Data analysis procedures

Data was entered and cleaned in Epi data and exported to SPSS version 16 soft ware for analysis. Descriptive analysis such as frequency and mean was performed to summarize the socio demographic characteristics, parasitic infection and chronic illness, maternal and reproductive, nutrition and nutritional factors as well as health service related factors and laboratory findings of the study subjects. Chi square test was done to check counts on each cell for categorical data. Kolmogrove smirnov test was done to check normality distribution. Frequencies and percentages of variables were calculated by anemia status. The relationship between anemia and associated factors was first investigated through bivariate analysis. In the bivariate analysis variables with a p-value $< \text{or} = 0.20$ were chosen as candidates for multiple logistic regression analysis. The associations between anemia and its associated factors were examined by multiple logistic regression analysis using enter method and expressed as crude odds ratios (COR) and adjusted odds ratios (AOR) to finally fit the model. Model fitness of multi variate logistic regression was confirmed by Hosmer-lemshow and chi-saquare block test. For all statistical tests P-value < 0.05 was considered significant.

4.10 Data quality management

Pretest of the questionnaires was done on 5% of all questionnaires in Gambella health center. One day orientation was given to the interviewers to standardize questionnaires administration as well as for diploma senior laboratory technicians to apply standard operational diagnostic procedures two days before commencement of the survey. Quality control of testing materials and machine was done as per the standard operating procedure. During the survey, all data collectors and data collection procedures were closely monitored by their investigator and randomly examined. Data was entered twice in Epi data soft ware. Double-entered data sets were compared using Epi data and discrepancies were removed against the original records.

4.11 Ethical consideration

The purpose of the study was explained to the pregnant women and they were asked if they would like to participate. Interview was carried out after obtaining verbal informed consent. Participation in the study was voluntary and no incentives were provided. The study was conducted after approval by ethical clearance committee of Jimma University. To ensure confidentiality, the data was only known by data collectors and investigator. During the study pregnant mothers with anemia were notified to their health care provider and supplemented with iron tablet. In addition pregnant mothers with test result positive for helminthes or malaria parasites were also notified to their health care provider and treated with appropriate medication accordingly.

4.12 Operational definitions

Anemia: a pregnant women with HBC of <11 g/dl, according to WHO definition, was classified as anemic.

- ❖ **Mild anemia:** hemoglobin level from 9 -10.9 g/dl.
- ❖ **Moderate anemia:** hemoglobin level from 7- 8.9 g/dl.
- ❖ **Severe anemia:** hemoglobin level from 4- 6.9 g/dl.
- ❖ **Very severe anemia:** hemoglobin level below 4 g/dl.

MUAC measurement: it is a measurement taken from left mid upper arm circumference using standard MUAC tape.

BMI measurement: it is a measurement which was obtained by calculating the weight of women in Kg divided by height of the pregnant women in square meters.

Urban: an area which is classified by the local or regional government as town. Such as regional, zonal or district towns.

Rural: Those are places which are found near to or far away from the town, but classified by the local or regional government as rural kebeles.

Below poverty line: it is an international income classification with monthly income of less than 37.5 USD (731 ETB).

Low income: it is monthly income of between 37.5-86.20 USD (732-1681 ETB).

Middle and high income: it is classified as monthly income of more than 86.20 USD (1681 ETB).

4.13 Dissemination plan

The findings of the study will be presented to Jimma University College of public health and medical sciences department of epidemiology. It will also be disseminated to the concerned body such as Gambella regional health bureau, Gambella hospital and NGO's working regarding maternal and reproductive health in Gambella regional state. Finally Efforts will be made to publish the finding of the study.

CHAPTER FIVE: RESULT

5.1 Socio demographic characteristics of Study participants

Three hundred and forty six pregnant women were surveyed in the study making a response rate of 93.5%. Since my study period was one month, I have only collected data until the end of data collection period. The mean age of the study participants was 24 years with SD of 4.6 and maximum of 38 years and minimum of 15 years. The mean income of the participants was 1848 ETB in which majority of them 168(48.6%) were with an income of more than 1681 ETB (see table 1).

Table 1: socio-demographic characteristics of pregnant women attending ANC at Gambella hospital, April 2014

Characteristics	frequency	Valid percent (%)
Age of participants (in years)		
15-19	55	15.9
20-29	237	68.5
30-39	54	15.6
Job		
house wife	136	39.3
Merchant	44	12.7
government employee	99	28.6
Student	56	16.2
others *	11	3.2
Monthly income (ETB)		
<732 (under poverty)	49	14.2
732-1681 (low income)	129	37.3
>1681 (medium and high income)	168	48.6
Level of education		
unable to read and write	41	11.8
primary	103	29.8
secondary	127	36.7
more than secondary	75	21.7
Marital status		
married/living as married	340	98.3
others **	6	1.7
Religion		
protestant	155	44.8
Orthodox	119	34.4
Muslim	32	9.2
Catholic	22	6.4
Others ***	18	5.2

Ethnicity		
Agnua	75	21.7
Nuer	42	12.1
Oromo	66	19.1
Kembata	31	9
Amara	35	10.1
Others ****	97	28
place of resident		
Urban	310	89.6
Rural	36	10.4

*Others: working in private and NGO's ** others: widowed, divorced or not married

***Others: religion followers of betel sinod & 7th Adventist

****Others: ethnic group from tigre, guraghe, mejeng, wopo, komo, hadiya, wolayta

5.2 Obstetric and Reproductive history

The mean gestational age of the participants was 28 weeks with a minimum 9 weeks and maximum of 38 weeks. About 22(6.4%) were in the first trimester while 234(67.6%) were in the second trimester and 90(26%) in the third trimester. Among the participants 183(52.9%) have ever give birth before the current pregnancy. Among the participants 35(10.1%) had miscarried or aborted pregnancy in their life time at least once and 92(26.6%) of them had faced irregularity of menstrual period prior to the current pregnancy. During their last pregnancy 159(78.7%) pregnant women were having ANC visit at health facility (see table 2).

Table 2: obstetric and reproductive history of pregnant women attending ANC at Gambella hospital, April 2014

Characteristics	frequency	Valid percent (%)
gestational age		
<= 12 weeks	22	6.4
13-27 weeks	234	67.6
>=28 weeks	90	26
bleeding during current pregnancy		
yes	26	7.5
no	320	92.5
parity		
0	163	47.1
1	80	23.1
2	58	16.8
3 up to 4	40	11.6
>=5	5	1.4
ANC follow up during last pregnancy		
yes	144	41.6
no	159	78.7
NA	43	21.3
place for the last delivery		
health facility	85	46.6
home	95	51.8
other places *	3	1.6
birth interval		
NA	163	47.1
<2 years	37	10.7
2-4 years	104	12.1
>4 years	42	12.1
frequency of miscarried/abortion		
0(never)	311	89.9
1	33	9.5
2	2	0.6
contraceptive use		
yes	237	68.5
no	109	31.5
irregularity of menstrual period		
yes	92	26.6
no	254	73.4
excess bleeding of menstrual period		
yes	46	13.3
no	300	86.7

*Other places: indicates for the births happened simply in roads NA: no response

5.3 parasitic infections and chronic illness

The laboratory finding showed that 25(7.2%) of the participants were having malaria infection and 28(8.1%) were with helminthes infection. But Participants with history of malaria infection and helminthes infection during the past 3 months were 128(37%) and 41(11.8%) respectively. Among the study participants 30(8.7%) have habit of walking on bare foot most of the time and 38(11%) have replied as they have history of chronic kidney disease (see table 3).

Table 3: parasitic infection and chronic illness history of pregnant women attending ANC at Gambella hospital, April 2014

Characteristics	frequency	Valid percent (%)
presence of malaria		
yes	25	7.2
no	321	92.8
type of malaria		
PF	23	92
PV	2	8
presence of ova parasite		
yes	28	8.1
no	318	91.9
type of ova parasite		
Hook worm	8	28.6
A.lumbricoid	14	50
t.trichuria	4	14.2
t.gordia	2	8.2
history of malaria infection during past 3 months		
yes	128	37
no	218	63
ITN availability		
yes	300	86.7
no	46	13.3
ITN utilization		
yes	182	60.5
no	118	39.5
history of helminthes infection during past 3 months		
yes	41	11.8
no	305	88.2
walking bare foot		
most of the time	30	8.7
some times	134	38.7
never	182	52.6

latrine availability		
yes	322	93.1
no	24	6.9
history of chronic kidney disease		
yes	38	11
no	308	89
principal drinking water source		
pipe to dwelling/premise	173	50
stand pipe	159	46
protected well/spring	5	1.4
unprotected source	9	2.6

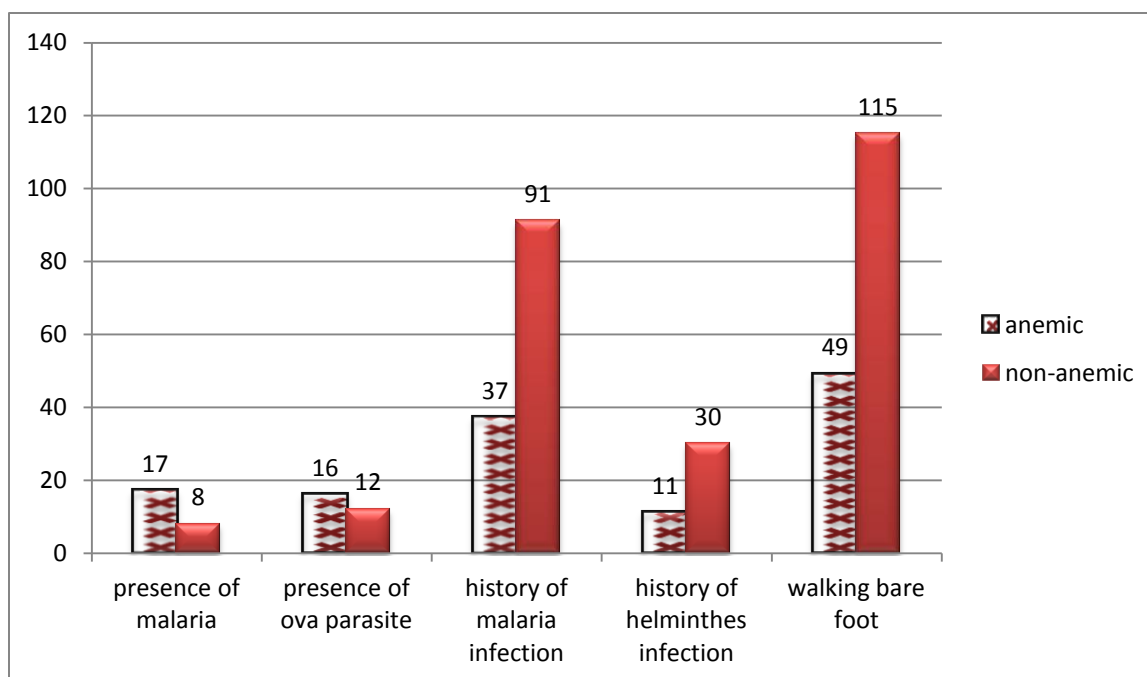


Figure 2: proportion of parasitic infection and related risk factors stratified by anemic status of pregnant women attending ANC at Gambella hospital April, 2014

5.4 Nutrition and dietary behavior

Among the study participants 250(72.3%) eat meat or fish at least once per week and 85(24.6%) consume green leafy vegetables at least once per day. Among the participants 114(32.9%) and 139(40.2%) have habit of drinking tea and coffee after meal at least once per day respectively. The mean MUAC and BMI values of the participants were 23.9 and 20.8 respectively (see table 4).

Table 4: nutrition related factors of pregnant women attending ANC at Gambella, April 2014

Characteristics	frequency	Valid percent (%)
frequency of eating meat or fish		
at least once a week	250	72.3
less than once a week	90	26
not at all	6	1.7
eat meat or fish during the past 24 hours		
yes	149	43.8
no	191	56.2
frequency of eating an egg		
at least once per week	146	42.2
less than once per week	114	32.9
not at all	86	24.9
eat an egg during the past 24 hours		
yes	53	20.2
no	209	79.8
frequency of vegetable consumption		
at least once per day	85	24.6
less than once per day	251	72.5
not at all	10	2.9
eat vegetables during the past 24 hours		
yes	202	60.1
no	134	39.9
frequency of fruit		
at least once per day	82	23.7
less than once per day	261	75.4
not at all	3	1.2
eat fruits during the past 24 hours		
yes	164	47.8
no	179	52.2
staple food		
injera with 'wot'(cereals)	194	56
pourage	110	31.8
pourage with fish	25	7.2
injera with wott and pourage	17	5
iron rich foods		
pouage with fish	25	7.2
other staple foods	321	92.8
frequency of tea		
at least once per day	114	32.9
less than one per day	74	21.4
not at all	158	45.7

frequency of coffee		
at least once per day	139	40.2
less than one per day	99	28.6
not at all	108	31.2
MUAC measurement		
17-20.9	47	13.6
>=21	299	86.4
BMI measurement		
<18.5	89	25.7
18.5-24.9	212	61.3
>=25	45	13

5.6 health care service and medication history

In their current pregnancy 247(71.4%) and 86(24.9%) of them had taken iron tablet and de-worming respectively (see table 5).

Table 5: health service related characteristics of pregnant women attending ANC at Gambella April 2014

Characteristics	frequency	Valid percent (%)
taking iron tablet		
yes	247	71.4
no	99	28.6
taking de-worming		
yes	86	24.9
no	260	75.1

5.2 Hemoglobin concentration of study participants

All 346 study participants had taken all laboratory examinations such as stool and blood film examinations as well as and HBC measurement. Mean HBC values of the pregnant women was 12.11 g/dl with SD of ± 1.36 having the maximum value 14.5 g/dl and minimum of 7.40 g/dl. Majority of the participants 272(78.6%) found to have a normal HBC level where as 67(19.4%) were having mild anemia and the rest 7(2%) had moderate anemia.

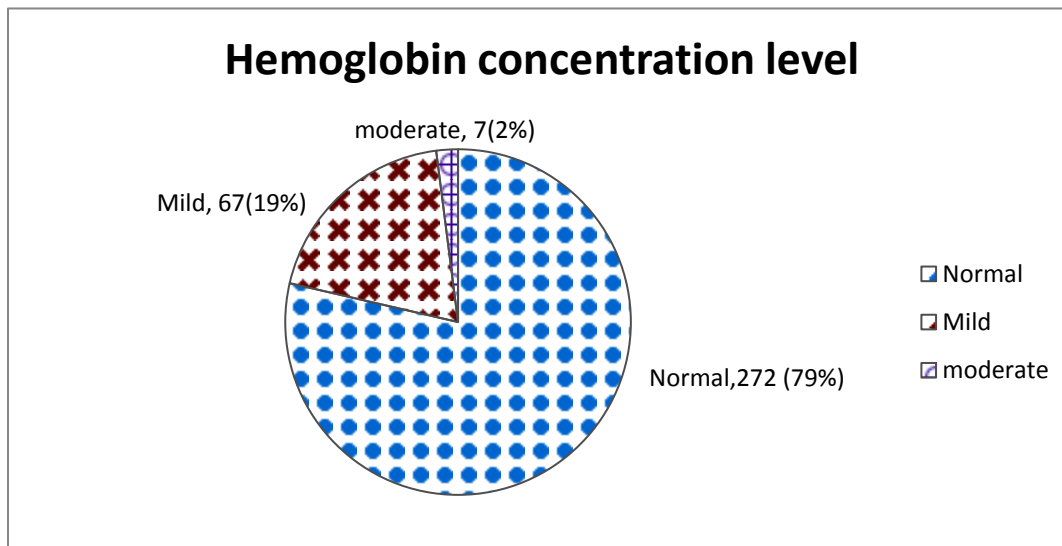


Figure 3: level of hemoglobin concentration of pregnant women attending ANC at Gambella hospital, April 2014

Association between socio-demographic characteristics and anemia

In the bi-variate analysis there was statistically significant difference among anemic and non anemic pregnant women in their place of residence ($p=.003$) and average monthly income ($p=.007$). But the remaining socio demographic characteristics did not show significant association with anemia. Both average monthly income and place of residence of study participants were selected as candidates for multiple logistic regressions.

Table 6: bi-variate analysis of association between socio-demographic characteristics and anemia

characteristics	Anemic		COR(95% CI)
	yes n (%)	no n (%)	
Age of participants (in years)			
15-19	12(3.5%)	43(12.4%)	1
20-29	50(14.5%)	187(54%)	.958(.470-1.953)
30-39	12(3.5%)	42(12.1%)	1.024(.414-2.534)
Level of education			
unable to read and write	12(3.5%)	30(8.7%)	1.8(.761-4.259)
primary	25(7.2%)	78(22.5%)	1.442(.713-2.918)
secondary	21(6.1%)	92(26.6%)	1.027(.50-2.11)
more than secondary	16(4.6%)	72(20.8%)	1
family size			
1 up to 2	20(5.8%)	74(21.4%)	1
3 up to 4	24(6.9%)	120(34.7%)	.740 (.382-1.432)
>=5	30(8.7%)	78(22.5%)	1.423(.744-2.723)
average monthly income (ETB)			
<732(under poverty)	17(4.9%)	32(9.2%)	2.656(1.3-5.428)*
732-1681 (low income)	29(8.4%)	100(28.9%)	1.450(.812-2.588)
>1681 (middle & high income)	28(8.1%)	140(40.5%)	1
place of residence			
urban	62(17.9%)	248(71.7%)	1
rural	12(3.5%)	24(6.9%)	2.0(1.28-4.22)*

1= reference group

*significant at p-value<0.05

Income classification: source from UNICEF poverty line income and World Bank income category

Association between obstetric history and anemia

In the bi-variate analysis gestational age (p=.048), irregularity of menstrual period (p=.022) and ANC follow up during last pregnancy were significantly associated with anemia. Gestational age, ANC follow up during last pregnancy, irregularity of menstrual period and excess bleeding during menstrual period prior to the current pregnancy were selected candidates for multiple logistic regression.

Table 7: bi-variate analysis of association between reproductive and obstetric history with anemia

characteristics	Anemic		COR(95% CI)
	yes n (%)	no n (%)	
gestational age			
1 st trimester	5(1.4%)	17(4.9%)	1
2 nd trimester	40(11.6%)	194(56.1%)	1.306(.369-4.624)
3 rd trimester	31(9%)	59(17.1%)	3.328(2.913-12.124)*
bleeding during current pregnancy			
yes	4(1.2%)	22(6.4%)	.649(.217-1.947)
no	70(20.2%)	250(72.3%)	1
parity			
0	28(8.1%)	132(38.2%)	1
1	18(6.1%)	61(17.6%)	1.326(.695-2.532)
2 up to 3	21(5.2%)	64(18.5%)	1.464(.785-2.728)
4 and above	7(2%)	15(4.3%)	.568(.123-2.612)
Birth interval			
<2 years	14(7.7%)	23(12.6%)	2.587(.936-7.154)
2-4 years	21(11.5%)	83(45.4%)	1.075(.434-2.663)
>4 years	8(4.4%)	34(18.6%)	1
ANC follow up during last pregnancy			
yes	31(15.3%)	128(63.4%)	1
no	15(7.4%)	28(13.9%)	2.212(1.056-4.63)*
history of abortion			
yes	8(2%)	27(7.5%)	1.100(.477-2.534)
no	66(19.1%)	245(70.8%)	1
contraceptive use			
yes	49(14.2%)	188(54.3%)	.876(.507-1.512)
no	25(7.2%)	84(24.3%)	1
irregularity of menstrual period			
yes	27(7.8%)	63(18.2%)	1.906(1.084-3.337)*
no	47(13.6%)	209(60.4%)	1
excess bleeding during menstrual period			
yes	14(4%)	32(9.2%)	1.750(.879-3.485)
no	60(17.3%)	240(69.4%)	1

1= reference group

*significant at p-value<0.05

Association between parasitic infection and chronic illness with anemia

Bi-variate analysis shows that statistically significant association between presence of malaria parasite ($p < 0.001$) and presence of ova parasite ($p = .001$) with anemia among pregnant women. It was also found that history of malaria infection during the past three months ($p = .010$) and habit of walking on bare foot ($p = .030$) have significant association with anemia. Presence of ova and malaria parasite, history of malaria infection as well as habit of walking bare foot were selected as candidates for multiple logistic regressions.

Table 8: bi-variate analysis of association between parasitic infection and chronic illness related factors with anemia

characteristics	Anemic		COR(95% CI)
	yes n (%)	no n (%)	
presence of malaria parasite			
yes	17(4.9%)	8(2.3%)	9.842(4.051-23.913)*
no	57(16.5%)	264(76.3%)	1
presence of ova parasite			
yes	16(3.5%)	12(8.1%)	10.650(4.33-26.228)*
no	58(16.8%)	260(75.1%)	1
history of malaria infection			
yes	37(10.7%)	91(26.3%)	1.989(1.182-3.3487)*
no	37(10.7%)	181(52.3%)	1
ITN availability			
yes	66(19.1%)	234(67.6%)	1
no	8(2.3%)	38(11%)	.749(.332-1.678)
ITN utilization			
yes	31(10.3%)	121(50.2%)	1
no	36(12%)	113(27.6%)	1.243(.734-3.876)
history of helminthes infection			
yes	11(3.2%)	30(8.7%)	1.408(.669-2.965)
no	63(18.2%)	242(69.9%)	1
habit of walking on bare foot			
most of the time	11(3.2%)	19(5.5%)	3.636(1.548-8.542)*
sometimes	38(11%)	96(27.7%)	2.486(1.413-4.374)*
never	25(7.2%)	157(45.4%)	1
history chronic kidney illness			
yes	10(2.9%)	28(8.1%)	1.584(0.746-3.367)
no	64(18.5%)	244(70.5%)	1

1= reference group

*significant at p-value < 0.05

Association between nutrition and nutrition related with anemia

Bi-variate analysis indicated that meat consumption weekly (p=.0045), green leafy vegetable consumption daily (p=.001), coffee drinking after meal every day (p=.001), low BMI values (p=.023) and MUAC (p=.03) were having significant association with anemia among those pregnant women. Meat consumption weekly, egg consumption weekly, fruit consumption daily, coffee intake daily after meal, low MUAC and BMI values were selected as candidates for multiple logistic regression.

Table 9: bi-variate analysis of association between nutrition and dietary behavior with anemia

characteristics	Anemic		COR(95%CI)
	yes n (%)	no n (%)	
frequency of eating meat or fish			
at least once a week	44(12.7%)	208(60.1%)	1
less than once a week	27(7.8%)	61(17.6%)	2.127(1.153-3.922)*
frequency of egg			
at least once per week	24(6.9%)	123(35.5%)	1
less than once per week	50(14.5%)	149(43.1%)	1.720(1.00-2.957)**
frequency of vegetable consumption			
at least once per day	6(1.7%)	79(22.8%)	1
less than once per day	64(18.5%)	187(54%)	4.639(1.934-11.126)*
frequency of fruits			
at least once per day	13(3.8%)	69(19.9%)	1
less than once per day	61(17.6%)	203(58.7%)	1.595(.826-3.080)**
staple food			
pourage with fish	4(1.2%)	21(6.1%)	1
other staple food	70(20.2)	251(72.5%)	1.464(.487-4.405)
frequency of tea			
at least once per day	27(7.8%)	87(25.1%)	1.222(.714-2.091)
less than once per day	47(13.6%)	185(53.5%)	1
frequency of coffee			
at least once per day	44(12.7%)	95(27.5%)	4.217(1.936-9.182)*
less than one per day	30(8.7%)	177(51.2%)	1
BMI			
<18.5	23(6.6%)	66(19.1%)	3.259(.989-10.735)*
18.5-24.9	47(13.6%)	165(47.7%)	2.475(.804-7.624)
>=25	7(2%)	38(11%)	1
MUAC measurement			
<21	18(5.2%)	29(8.4%)	2.693(1.398-5.190)*
>=21	56(16.2%)	243(70.2%)	1

1= reference group

*significant at p-value<0.05

Association between health care services and medication history with anemia

Bi-variate analysis indicated that taking or receiving iron tablet ($p < 0.001$) in the current pregnancy showed significant difference among anemic and non anemic pregnant women (see table 10).

Table 10: bi-variate analysis of health services related factors with anemia

characteristics	Anemic		COR(95%CI)
	yes n (%)	no n (%)	
iron supplementation/intake			
yes	36(10.4%)	211(61%)	1
no	38(11%)	61(17.6%)	3.216(1.857-5.572)*
receiving de-worming			
yes	20(5.8%)	67(19.4%)	1
no	54(15.6%)	205(59.2%)	.882(.493-1.580)

1= reference group

*significant at $p\text{-value} < 0.05$

5.7 Multi- variate logistic regression analysis

Final analysis in the multi-variate logistic regression indicated that parasitic infections such as helminthes and malaria infection have significant influence on anemia in pregnancy. Among the participants pregnant women with malaria parasite infection were 15.8 (AOR= 15.7 95%CI: 5.09-48.411) more likely to anemic than their counter parts. The finding of this study also revealed that Pregnant women with helminthes infection were (AOR= 8.124, 95% CI: 2.754-23.96) more likely to be anemic than those pregnant women who were free of helminthes infection

This study also indicated dietary habits of pregnant women such as vegetable consumption has an effect on their anemia status. Pregnant women with less than once consumption of vegetables per day were (AOR=4, 95%CI: 1.4-11.8) more likely to be anemic than those consuming vegetables at least once daily. Health services provision like iron supplementation during pregnancy has an influence on anemia in pregnancy. This study indicated that pregnant women who do not receive iron supplementation during their current pregnancy were (AOR=2.5, 95% CI: 1.195-5.128) more likely to anemic than their counter parts.

Table 11: multivariate logistic Regression analysis of association between factors associated with anemia among pregnant women attending ANC at Gambella hospital, April 2014

characteristics	Anemia (frequency)		COR(95%CI)	AOR(95% CI)
	yes n (%)	no n (%)		
place of residence				
urban	62(17.9%)	248(71.7%)	1	1
rural	12(3.5%)	24(6.9%)	2.0(1.28-4.22)	1.566(.534-4.591)
average monthly income (ETB)				
<732(under poverty)	17(4.9%)	32(9.2%)	2.656(1.3-5.428)	2.567(.914-7.207)
732-1681 (low income)	29(8.4%)	100(28.9%)	1.450(.812-2.588)	1.095(.507-2.366)
>1681 (middle & high income)	28(8.1%)	140(40.5%)	1	1
gestational age				
1 st trimester	5(1.4%)	17(4.9%)	1	1
2 nd trimester	40(11.6%)	194(56.1%)	1.306(.369-4.624)	1.112(.214-5.773)
3 rd trimester	31(9%)	59(17.1%)	3.328(2.913-12.124)	2.967(.586-15.01)
ANC follow up during last pregnancy				
yes	31(15.3%)	128(63.4%)	1	1
no	15(7.4%)	28(13.9%)	2.212(1.056-4.63)	1.349(.421-4.327)
irregularity of menstrual period				
yes	27(7.8%)	63(18.2%)	1.906(1.084-3.337)	1.661(.780-3.534)
no	47(13.6%)	209(60.4%)	1	1
excess bleeding during menstrual period				
yes	14(4%)	32(9.2%)	1.750(.879-3.485)	1.855(.336-3.233)
no	60(17.3%)	240(69.4%)	1	1
presence of malaria parasite				
yes	17(4.9%)	8(2.3%)	9.842(4.051-23.913)	15.7(5.09-48.411)*
no	57(16.5%)	264(76.3%)	1	1
presence of ova parasite				
yes	16(3.5%)	12(8.1%)	10.650(4.33-26.228)	8.124(2.754-23.96)*
no	58(16.8%)	260(75.1%)	1	1
history of malaria infection				
yes	37(10.7%)	91(26.3%)	1.989(1.182-3.3487)	1.738(.817-3.698)
no	37(10.7%)	181(52.3%)	1	1
habit of walking on bare foot				
most of the time	11(3.2%)	19(5.5%)	3.636(1.548-8.542)	1.746(.207-3.695)
sometimes	38(11%)	96(27.7%)	2.486(1.413-4.374)	1.258(.557-2.839)
never	25(7.2%)	157(45.4%)	1	1

frequency of eating meat or fish				
at least once a week	44(12.7%)	208(60.1%)	1	1
less than once a week	27(7.8%)	61(17.6%)	2.127(1.153-3.922)	1.597(.780-3.269)
frequency of egg consumption				
at least once per week	24(6.9%)	123(35.5%)	1	1
less than once per week	50(14.5%)	149(43.1%)	1.720(1.00-2.957)	1.886(.413-2.901)
frequency of vegetable consumption				
at least once per day	6(1.7%)	79(22.8%)	1	1
less than once per day	64(18.5%)	187(54%)	4.639(1.934-11.126)	4.451(1.496-13.25)*
frequency of fruits consumption				
at least once per day	13(3.8%)	69(19.9%)	1	1
less than once per day	61(17.6%)	203(58.7%)	1.595(.826-3.080)	1.903(.394-3.055)
frequency of coffee drinking				
at least once per day	44(12.7%)	95(27.5%)	4.217(1.936-9.182)	1.685(.823-3.45)
less than one per day	30(8.7%)	177(51.2%)	1	1
BMI				
<18.5	23(6.6%)	66(19.1%)	3.259(.989-10.735)	2.128(.54-8.251)
18.5-24.9	47(13.6%)	165(47.7%)	2.475(.804-7.624)	2.11(.588-7.567)
>=25	7(2%)	38(11%)	1	1
MUAC measurement				
<21	18(5.2%)	29(8.4%)	2.693(1.398-5.190)	1.616(.631-4.136)
>=21	56(16.2%)	243(70.2%)	1	1
iron supplementation/intake				
yes	36(10.4%)	211(61%)	1	1
no	38(11%)	61(17.6%)	3.216(1.857-5.572)	2.19(1.087-4.414)*

1= reference group

* significant association at p-value <0.05

CHAPTER SIX: DISCUSSION

This work aimed to study magnitude of anemia and different factors associated with anemia among pregnant women attending ANC services. The results indicate that parasitic infections, vegetable consumption and iron tablet intake were important factors significantly associated with anemia in this study population.

Anemia is serious problem which increased maternal morbidity and mortality and leading to different bad health consequences to the fetus (17). In our finding the prevalence of anemia in pregnancy was 21.4% and this finding is consistent with the study conducted in Gondar at Azezo health center (21.6%)(18) and Prevalence of anemia among pregnant women in Gambella region EDHS 2011(20%)(9), but it's much lower than the study conducted at Shalla woreda and India(82%) (5)(20). The lower prevalence of anemia in this study area of Gambella town may be explained by the consumption of common food types which includes fish and porrage, which is mostly prepared from germinated corn, in which this dietary habit contain improved bioavailability of iron and may have an influence on anemia in pregnancy (33).

In this study among anemic women 90.5% and 9.5% were mild and moderate respectively which is similar with study conducted in shalla woreda in which majority of pregnant women were with mild anemia, but study from India indicated that moderate anemia was higher than mild anemia among pregnant women. Iron deficiency generally develops slowly and is not clinically apparent especially in the mild stage until anemia is severe even though functional consequences already exist. Where iron deficiency anemia is prevalent, effective control programs may yield benefits to the pregnant women.

In the present study we did not find significant association between gestational age and the birth interval between pregnancies with anemia although other studies have shown the presence of significant association. This might be explained by receiving of iron tablet by majority of the study participants in which notable differences might not observe across the gestational age. The birth interval for the majority of the study participants were more than two years and women having two or more years between pregnancies, they are more likely to enter the subsequent pregnancy with adequate iron status (24).

In agreement with other studies in sub-Saharan Africa, malaria was one of the main factors associated with anemia. Plasmodium falciparum directly destroys erythrocytes and later causing anemia (26). In our finding malaria infection was significantly associated with anemia and 23% of anemia was attributable to

malaria infection. Anemia was much higher in pregnant women with malaria infection (68%) compared to pregnant women free of malaria infection (18%). This is consistent with the study conducted in Shalla woreda and Burkinafaso, Ouagadougou where anemia was more common in women infected with *p.falciparum* compared with the uninfected pregnant women(24). The finding was also similar with the study conducted in Mali and Benin, where there is stable transmission of malaria throughout the year, indicated significant association between malaria infection and anemia in pregnancy(25)(26). Since malaria infection is endemic in Gambella town, *Plasmodium falciparum* malaria causes a profound anemia during and after acute infection. Anemia is caused by hemolysis of red cells combined with suppression of erythropoiesis. For these reasons, detecting and treating malaria are essential for treating anemia as well as the use of insecticide-impregnated bed nets with other malaria controlling mechanisms to decrease the prevalence of anemia in pregnant women (33).

Anemia is the most common outcome of parasitic infection such as malaria and helminthes infection especially in sub-Saharan countries like Ethiopia. Many studies showed that prevalence of anemia is higher among pregnant women with helminthes infection. In our population, 21.6% of anemia was attributable to helminthes infestations. The finding of this study also indicated that magnitude of anemia in pregnant women with helminthes infection (57%) was higher than anemia in those women free of infection (18%). This is consistent with the study conducted at Bushalo health center, Azezo health center and Jimma university hospital(4)(18)(29). More over studies in Mali and Venezuela also indicated prevalence of anemia was higher among pregnant women with helminthes infection than their counterparts(25)(21). Helminthes infections such as hook worm can cause iron deficiency anemia especially in pregnancy by causing chronic blood loss, putting the mother and the fetus at high risk for anemia(26). Considering the seriousness of these infections during pregnancy, providing anti-helminthes drugs during the second trimester is, thus, part of public health intervention package against anaemia in pregnancy. But the present study showed that the proportion of pregnant women taking de-worming was low coverage. It can be argued, therefore without improvement in de-worming and not applying helminthes control, may not result in a reduction of anemia in pregnant women in Gambella town (31).

Dietary habits of pregnant women, according to many studies have an influence on anemic status of pregnancy. In our finding the prevalence of anemia was higher among pregnant women who were consuming green leafy vegetables less (25%) than those who were consuming vegetables daily (7%). The present study revealed that a significant association occurred between green leafy vegetable consumption

daily and anemic status of pregnant women. This is consistent with the study done at westmore land, Jamaica showed that the risk of anemia was reduced among the women who consumed green leafy vegetables once or more in a week compared to those who did not(11). This indicates that frequent consumption of green leafy vegetables and other iron rich foods such meat and fish may enhance increased HBC thereby decreasing the probability of being anemic. Continuous awareness creation activity is required for pregnant women to increase the frequency of consumption of iron rich foods and avoid or decrease consumption of iron inhibiting foods (33).

The amount of iron absorbed from the diet is highly dependent on the composition of the diet, namely, the quantities of substances that enhance or inhibit dietary iron absorption. In this study it was expected that drinking tea or coffee after meal may have an association with anemia in pregnancy. But in the present study no association was found among those variables with anemia. This might be associated to less habit of drinking tea or coffee after meal by study participants.

Iron deficiency occurs if the amount of iron absorbed is too little to meet the body's demand(21). The present study found that a significant association between iron intake during pregnancy and anemia. From the total anemia 10% was attributed by lack of iron intake during pregnancy. Pregnant women who did not take iron tablet were 2.5 times more likely to be anemic compared to their counter parts. This is consistent with study conducted at Bushalo health center in southern Ethiopia (21) and similar study conducted in Gondar at Azezo health center(18). Iron deficiencies may develop during pregnancy because of the increased iron requirements to supply the expanding blood volume of the mother and the rapidly growing fetus and placenta. The high prevalence of anemia in pregnancy in sub-Saharan African women might be due to the fact that women may enter pregnancy with seriously depleted iron stores(34). The amount of iron required during the last half of pregnancy cannot easily be met from dietary sources alone, daily supplements of iron and folic acid has been universally recommended (17).

However not only lack of iron supplementation, but it is also agreed with other evidence that parasitic infection contributes significantly to the high prevalence of anemia in Africa and elsewhere(25). In our finding even though majority 71% of the pregnant women receives iron supplementation, treatment with iron alone is unlikely to be an effective strategy against anemia. In addition to iron supplementation parasitic infection control mechanisms play a great role in reducing magnitude of anemia (35) .

CHAPTER SEVEN: STRENGTH AND LIMITATION OF THE STUDY

Strength of the study

Study participants were examined for parasitic infection such as malaria and helminthes parasites.

Limitation of the study

There are, of course, limitations in this study. Even though this study tried to address some important factors that are associated with anemia, other factors such as other nutritional deficiencies (including folate, vitamin B12, Vitamin A), acute and chronic inflammation, and inherited or acquired disorders that affect hemoglobin synthesis, red blood cell production or red blood cell survival, which can all cause anaemia, were not addressed.

CHAPTER EIGHT: CONCLUSION

Based on the data presented in this study, the overall prevalence of anemia in the study area indicated that anemia is medium public health problem and we believe that it is a major health issue for pregnant women in Gambella town. It was found that parasitic infection such as malaria and helminthes, daily green leafy vegetable consumption and iron tablet supplementation were significantly associated with anemia among pregnant women.

The high physiological requirement for iron in pregnancy is difficult to meet with most diets. Pregnant women should routinely receive iron supplements in almost all contexts according to the iron supplementation guide lines. Since Gambella town is endemic for parasitic infection both malaria and helminthes infections were having a significant contribution to the anemic status of study participants. For this reason giving anti-helminthic or anti-malarial drugs along with iron supplements may increase the effectiveness of supplementation. Where appropriate, complementary parasite control measures are given along with the guidelines for iron supplementation.

CHAPTER NINE: RECOMMENDATION

Gambella hospital: since some of the parasitic infection like helminthes and malaria are asymptomatic, should start screening every pregnant woman when they came for ANC visit in addition to hemoglobin measurement which is currently done at the hospital. Provide appropriate treatment as per the guide line for those whose tests are positive for parasite infection.

Gambella regional health bureau and hospital: should work on improving the awareness of pregnant women by providing health education through the health extension package and when they appear for ANC visit to improve consumption of iron rich dietary food sources.

Gambella regional health bureau and its partners: have to work on controlling mechanisms of malaria and helminthes infection through providing ITN to pregnant women and encouraging them to use the bed net. Awareness creation activities should be done to pregnant women to avoid walking on bare foot and to keep washing hands with soap at critical times.

We recommend for further studies to be under taken on factors associated with anemia by including all other factors which are not included in this study can help in designing different interventional strategies which could be very important for the programme of ANC services and other public health programmes.

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ANNEXES

Annex I. Consent form

Code no: _____

Jimma university school of public health and medical sciences, department of Epidemiology

Hello. My name is _____ and I am member of the survey team. We are conducting a survey about factors associated with anemia among pregnant women who are attending ANC in this hospital. The survey usually takes about 30 to 45 minutes. All of the answers you give will be confidential and will not be shared with anyone other than members of our survey team. You don't have to be in the survey, but we hope you will agree to answer the questions since your views are important. If I ask you any question you don't want to answer, just let me know and I will go on to the next question or you can stop the interview at any time.

As part of this survey, after the interview we are asking pregnant mothers to take an anemia test. Anemia is a serious health problem that usually results from poor nutrition, infection, or chronic disease. We are also asking pregnant women to take malaria and helminthes parasite examination. For the anemia testing, we will need a few drops of venom blood. For malaria test we will need a few drops of blood from a finger and for helminthes test, we will need stool specimen. The equipment used to take the blood is clean and completely safe. It has never been used before and will be thrown away after each test. The blood will be tested for anemia and malaria immediately, and the stool will be tested for helminthes parasites and finally result will be told to you right away. The result will be kept strictly confidential and will not be shared with anyone other than members of our survey team. This survey will assist stake holders to develop programs to prevent and treat anemia, malaria and helminthes infection.

Do you have any questions?

You can say yes to the interview and the test, or you can say no. It is up to you to decide.

Will you take the entire test? May I begin the interview now?

1. Yes go to the next page
2. No stop.

Consent form Amharic version

የስምምነት መጠየቂያ ቅጽ

በጅም ዩኒቨርሲቲ የህክምናና ሕ/ሰብ ጤና ሳይንስ ኮሌጅ የኢፒዲምዮሎጂ ዲፓርትመንት

ጤና ይስጥልኝ። እኔ _____ እባላለሁ። ጥናት ከሚያካሂዱ ቡድን ውስጥ አባል ነኝ። ለቅድመ ወሊድ አገልግሎት ወደ ሆስፒታል የሚመጡ ነፍሰጡር እናቶች ላይ የደም ማነስ እና ተያያዥነት ያላቸው መንስኤዎች ጥናት እያካሄድን ነው። ጥናቱ ከ 30- 45 ደቂቃ የሚፈጅ ይሆናል። በዚህ ጥናት ለሚሰጡን ማናቸውም መልስ ጥናቱ ከሚያካሂዱ ሰዎች በቀር ሌላ ሰው እንዳያያው ይደረጋል። በዚህ ጥናት እንዲሳተፉ የሚያስገድድዎ አካል አይኖርም ነገር ግን የእርስዎ መልስ ለጥናቱ ጠቃሚ በመሆኑ እንደሚሳተፉ ተስፋ እናደርጋለን። እርስዎ እንዲጠየቁ የማይፈልጉት ጥያቄ ካለ በመሀል ይንገሩኝ ወደ ሚቀጥለው ጥያቄ እናልፋለን። ቃለ መጠይቁ በማንኛውም ሰዓት ማቋረጥ ሆነ ማቆም ይችላሉ።

ከቃለ መጠይቁ በኋላ የጥናቱ አንድ አካል የሆነውን የደም ማነስ ምርመራ ይካሄዳል። የደም ማነስ በሽታ በጣም አደገኛ የሆነ በተለያዩ መንስኤዎች ለምሳሌ ጥሩ ካልሆነ የአመጋገብ ስርዓት ፣ ተላላፊ በሽታዎችና እና ለብዙ ጊዜ (ዓመታት) ከሚቆዩ በሽታዎች አማካኝነት ሊከሰት የሚችል በሽታ ነው። በተጨማሪም በዚህ ጥናት የወባ በሽታ እና የሆድ ትላትል ምርመራ ይካሄዳል። ለደም ማነስ ምርመራ ትንሽ ደም ከእጅ እንወስዳለን። ለወባ በሽታ ደግሞ ከቀለበት ጣት የተወሰነ ደም የሚወሰድ ሲሆን ለሆድ ትላትል ምርመራ ግን ሰገራ በትንሽ ብልቃጥ እንድታመጡ ይደረጋል። ደም ለመውሰድ የምንጠቀምበት ዕቃ ንፅህናውን የጠበቀ እና ደህንነቱ የተጠበቀ ነው። ከዚህ በፊት ጥቅም ላይ ያልዋለ ሲሆን ከእያንዳንዱ ምርመራ በኋላ ይጣላል። የተወደሰው የደም ናሙና የደም ማነስ ምርመራ እና የወባ በሽታ ምርመራ ይካሄድበታል። የሰገራ ናሙና ደግሞ የሆድ ትላትል ምርመራ ይደረግበታል። ውጤቱ በአስቸኳይ እንዲደርስዎ ይደረጋል። ውጤቱም ጥናት ከሚያካሂዱ ሰዎች በቀር ሌላ ሰው እንዳያያው ይደረጋል። በመጨረሻም የዚህ ጥናት ውጤት አጋር ድርጅቶችና መንግስት የደም ማነስ ፣ የወባ በሽታ እንዲሁም የሆድ ትላትል በሽታዎች እና ሌሎች ተያያዥነት ያላቸው ችግሮች ፕሮግራም እንዲቀረፅና በቀጣይነት መፍትሄ እንዲያገኙ ይረዳል።

ጥያቄ አለዎት?

ምርመራው ያካሂዳሉ? ጥያቄውን ልቀጥል?

- 1. እሺ ካሉ ወደ ሚቀጥለው ገፅ ቀጥል
- 2. አይሆንም በቃ አቁም.

ANNEX II. Questionnaires for associated factors of anemia among pregnant women

This questionnaire is developed to assess factors related to anemia among pregnant women attending ANC in Gambella hospital. To be filled after obtaining informed consent.

Code no _____

I. socio-demographic and economic back ground			remarks
101	How old are you (in years)?	_____	
102	What is your occupation?	1. House wife 2. Merchant 3. Civil servant 4. Farmer 5. Others(specify)_____	
103	What is the total number of family members in your house hold?	_____	
104	What is your family monthly average income?	_____	
105	Have you ever attended school?	1. Yes 2. No	
106	If yes for Q. 105, What is the highest level of education/school you attended?	_____	
107	If No for Q.105, Do you read and write?	1. Yes 2. No	
108	What is your marital status?	1. Married 2. Single 3. Widowed 4. Divorced	
109	What is your religion?	1. Orthodox 2. Muslim 3. Protestant 4. Others specify_____	
110	What is your ethnicity?	1. Agnuwak	

		2. Nuer 3. Oromo 4. Kembata 5. Wolayta 6. Tigre 7. Amara 8. Others specify_____	
111	Where is your residence?	1. Urban 2. Rural	
II. parasitic infection and chronic illness related factors			
201	Have you had malaria infection in the last 3 months?	1. Yes 2. No	
202	Do you have ITN in your home?	1. Yes 2. No	
203	If yes for Q.202, do you use it always/regularly?	1. Yes 2. No	
204	Have you had helminthes disease in the last 3 months?	1. Yes 2. No	
205	Do you walk on bare foot?	1. Most of the times 2. Some times 3. Never	
206	Do you have latrine?	1. Yes 2. No	
207	Do you wash your hand with soap after visiting the toilet?	1. Yes 2. No	
208	Do you wash your hand with soap before eating meal?	1. Yes 2. No	
209	What is the principal source of drinking water for your home?{check one}	1. Piped water into dwelling 2. Piped water to yard/plot 3. Public tab/standpipe 4. Protected dug well/spring	

		5. Unprotected dug well/spring 6. Rain water collection 7. Surface water	
210	Have you had a disease called chronic kidney disease?	1. Yes 2. No	
III. nutrition and nutrition related factors			
301	Do you eat meat and animal products?	1. Yes 2. No	
302	If yes for Q.301, how frequently do you eat?	1. Every day 2. Every other day 3. Once a week 4. Once a month 5. Others specify.....	
303	Did you eat meat or animal products in the last 24 hours?	1. Yes 2. No	
304	Do you eat green leafy vegetables?	1. Yes 2. No	
305	If yes for Q.304, how frequently do you eat?	1. Every day 2. Every other day 3. Once a week 4. Once a month 5. Others specify.....	
306	Did you eat green leafy vegetables in the last 24 hours?	1. Yes 2. No	
307	Do you eat fruits?	1. Yes 2. No	
308	If yes for Q.307, how frequently do you eat?	1. Every day 2. Every other day 3. Once a week 4. Once a month 5. Others specify.....	

309	Did you eat fruits in the last 24 hours?	1. Yes 2. No	
310	What is your staple diet? (Encircle all response)	1. Injerra 2. Pourage 3. Fish 4. Others specify_____	
311	Do you take tea after meal?	1. Yes 2. No	
312	If yes for Q.311, how often do you take tea?	1. After every meal 2. Once a day 3. Every other day 4. Occasionally	
313	Do you take coffee after meal?	1. Yes 2. No	
314	If yes for Q.313, how often do you take coffee?	1. After every meal 2. Once a day 3. Every other day 4. Occasionally	
315	Now I would like to take your MUAC measurement?	_____	
316	Now I would like to take your BMI measurement?	_____	
IV. obstetric and reproductive related factors			
401	How many months/weeks pregnant are you? (probe and write in weeks)	_____	
402	Was there any blood loss during this pregnancy?	1. Yes 2. No	
403	Have you ever given birth?	1. Yes 2. No	
404	If yes for Q.402, how many total births have you had during your life?	_____	

405	Where did you give birth in your last delivery?	1. Health facility 2. Home 3. Others specify _____	
406	What is the birth interval between your last birth and your current pregnancy (in years)?	_____	
407	Have you ever had a pregnancy that miscarried or aborted?	1. Yes 2. No	
408	If yes for Q.407, how many of such pregnancies have you ever had?	_____	
409	If yes for Q.407, when did the last such pregnancy end?	_____	
410	Have you used any kind of contraceptive methods prior to your current pregnancy?	1. Yes 2. No	
411	Have you faced excess menstrual bleeding prior to your current pregnancy?	1. Yes 2. No	
412	Was there any irregularity in your menstrual cycle prior to this pregnancy?	1. Yes 2. No	
V. health service related factors			
501	Did you attend ANC services in your last pregnancy?	1. Yes 2. No	
502	If yes for Q.146, How many times did you visited for ANC services?	_____	
503	During this pregnancy, were you given or did you buy iron tablets?	1. Yes 2. No	
504	During this pregnancy, did you take any drug for intestinal worms?	1. Yes 2. No	

AMHARIC VERSION OF QUESTIONNAIRES

ይህ መጠየቂያ በጋምቤላ ሆስፒታል ለቅድመ ወሊድ ክትትል የሚመጡ ነፍሰጡር እናቶች ላይ የሚከሰተውን የደም ማነስ በሽታ እና ተያያዥነት ያላቸው መንስኤዎች ለማጥናት የተዘጋጀ መጠየቂያ ነው። መጠየቂያው የሚሞላው የተሳታፊዎችን ፍቃደኝነት ከተረጋገጠ በኋላ ነው።

የተሳታፊ መለያ ቁጥር.....

II. ስነ ማህበራዊና ኢኮኖሚያዊ			ምርመራ
101	ዕድሜዎ ስንት ነው(በዓመት)?	_____	
102	ስራዎት ምንድነው?	1. የቤት እመቤት 2. ነጋዴ 3. የመንግስት ሰራተኛ 4. አርሶ/አርብቶ አደር 5. ሌላ ካለ (ይጠቀስ)_____	
103	በቤትዎ ውስጥ ስንት የቤተሰብ አባላት አሉ?	_____	
104	በአማካይ የወር ገቢዎ ስንት ነው ?	_____	
105	የትምህርት ደረጃዎ ስንት ነው?	1. መፃፍና ማንበብ የማይችል 2. የተማሩ ከሆነ የደረሱበትን የትምህርት ደረጃ ይጠቀስ _____	
106	የጋብቻ ሁኔታ?	1. ያገባች 2. ያላገባች 3. ባል የሞተባት 4. አግብታ የፈታች	
107	እምነትዎ ምንድነው?	1. ኦርቶዶክስ 2. እስልምና 3. ጴጌነት 4. ሌላ ካለ ይጠቀስ_____	

108	የእርስዎ ብሄር ምንድነው?	1. አኙዋ 2. ኑዌር 3. ኦሮሞ 4. ከምባታ 5. ሌላ ካለ ይጠቀስ_____	
109	የመኖሪያ ቤትዎ የት ነው?	1. ከተማ 2. ገጠር	
II. ተላላፊ እና ሌሎች በሽታዎች ተያያዥነት ያላቸው ጥያቄዎች			
201	ባለፉት ሶስት ወራት በወባ በሽታ ታመው ያውቃሉ?	1. አዎ 2. አይደለም	
202	በቤትዎ ውስጥ የአልጋ አጎበር አለ?	1. አዎ 2. የለም	
203	የጥያቄ ቁ.202 መልስ አዎ ከሆነ፤ ሁሉ ጊዜ ይጠቀሙበታል?	1. አዎ 2. አይደለም	
204	ባለፉት ሶስት ወራት በሆድ ትላትል በሽታ ታመው ያውቃሉ?	1. አዎ 2. አይደለም	
205	በባዶ እግር (ጫማ ሳይለብሱ) ሂደው ያውቃሉ ?	1. አብዛኛው ጊዜ 2. አልፎ አልፎ 3. በፍፁም	
206	መፀዳጃ ቤት አለዎት?	1. አዎ 2. የለም	
207	ከመፀዳጃ ቤት መልስ እጅዎትን በውኃና በሳሙና ይታጠባሉ?	1. አዎ 2. አይደለም	
208	ምግብ ከመመገብዎ በፊት እጅዎትን በሳሙና እና በውኃ ይታጠባሉ?	1. አዎ 2. አይደለም	
209	ምግብ ከማዘጋጀትዎ በፊት እጅዎትን በውኃና በሳሙና ይታጠባሉ?	1. አዎ 2. አይደለም	
210	በዋናነት ለመጠጥ አገልግሎት የሚውል ውኃ ከየት ነው የምታገኙት?{አንዱን	1. ግቢ/ቤት ውስጥ ከተዘረጋው ቧንቧ	

	ምረጥ}	2. የጋራ ቧንቧ/ቦኖ 3. የተጠበቀ ምንጭ/ጉድጓድ ውኃ 4. ያልተጠበቀ የምንጭ/ጉድጓድ ውኃ 5. ከዝናብ ከሚጠራቀም ውኃ 6. ከወንዝ 7. ሌላ ካለ ይጠቀስ.....	
211	ለረዥም ጊዜ የቆየ የኩላሊት በሽታ አለብዎት?	1. አዎ 2. አይደለም	
III. ከስነ ምግብ እና የአመጋገብ ስርዓት ተያያዥነት ያላቸው ጥያቄዎች			
301	ስጋ ወይም ዓሳ ይመገባሉ?	3. አዎ 4. አይደለም	
302	የጥያቄ ቁ.301 መልስ አዎ ከሆነ፣ በምን ያህል ጊዜ ስጋ ወይም ዓሳ ይመገባሉ?	1. በየቀኑ 2. በሁለት ቀን አንድ ጊዜ 3. በሳምንት አንድ ጊዜ 4. በሁለት ሳምንት አንድ ጊዜ 5. በወር አንድ ጊዜ 6. ሌላ ካለ ይጠቀስ.....	
303	የጥያቄ ቁ.301 መልስ አዎ ከሆነ፣ ባለፈው 24 ሰዓት ውስጥ ስጋ ወይም ዓሳ ተመግበዋል?	1. አዎ 2. አይደለም	
304	የእንስሳት ተዋፅኦ ለምሳሌ እንቁላል ይመገባሉ?	1. አዎ 2. አይደለም	
305	የጥያቄ ቁ.304 መልስ አዎ ከሆነ፣ በምን ያህል ጊዜ እንቁላል ይመገባሉ?	1. በየቀኑ 2. በሁለት ቀን አንድ ጊዜ 3. በሳምንት አንድ ጊዜ	

		<ul style="list-style-type: none"> 4. በሁለት ሳምንት አንድ ጊዜ 5. በወር አንድ ጊዜ 6. ሌላ ካለ ይጠቀስ..... 	
306	የጥያቄ ቁ.304 መልስ አዎ ከሆነ፤ ባለፈው 24 ሰዓት ውስጥ እንቁላል ተመግቦታል?	<ul style="list-style-type: none"> 1. አዎ 2. አይደለም 	
307	አረንጓዴ ቅጠላ ቅጠሎችን ይመገባሉ?	<ul style="list-style-type: none"> 1. አዎ 2. አይደለም 	
308	የጥያቄ ቁ.307 መልስ አዎ ከሆነ፤ በምን ያህል ጊዜ አረንጓዴ ቅጠላ ቅጠሎችን ይመገባሉ?	<ul style="list-style-type: none"> 1. በየቀኑ 2. በሁለት ቀን አንድ ጊዜ 3. በሳምንት አንድ ጊዜ 4. በሁለት ሳምንት አንድ ጊዜ 5. በወር አንድ ጊዜ 6. ሌላ ካለ ይጠቀስ..... 	
309	የጥያቄ ቁ.307 መልስ አዎ ከሆነ፤ ባለፈው 24 ሰዓት ውስጥ አረንጓዴ ቅጠላ ቅጠሎችን ተመግቦታል?	<ul style="list-style-type: none"> 1. አዎ 2. አይደለም 	
310	የአትክልት ፍራፍሬ ይመገባሉ?	<ul style="list-style-type: none"> 1. አዎ 2. አይደለም 	
311	የጥያቄ ቁ.310 መልስ አዎ ከሆነ፤ የአትክልት ፍራፍሬዎች በምን ያህል ጊዜ ይመገባሉ?	<ul style="list-style-type: none"> 1. በየቀኑ 2. በሁለት ቀን አንድ ጊዜ 3. በሳምንት አንድ ጊዜ 4. በሁለት ሳምንት አንድ ጊዜ 5. በወር አንድ ጊዜ 6. ሌላ ካለ ይጠቀስ..... 	
312	የጥያቄ ቁ.310 መልስ አዎ ከሆነ፤ ባለፈው 24 ሰዓት ውስጥ የአትክልት ፍራፍሬዎች ተመግቦታል?	<ul style="list-style-type: none"> 1. አዎ 2. አይደለም 	
313	አዘውተረው የሚመገቡት የምግብ ዓይነት	<ul style="list-style-type: none"> 1. እንጀራ በወጥ 	

	ምንድነው? (ሁሉንም መልሶች ማክበብ ይቻላል)	2. ገንፎ 3. ዓሳ 4. ሌላ ካለ ይጠቀስ_____	
314	ከምግብ በኋላ ሻይ ይጠጣሉ?	1. አዎ 2. አይደለም	
315	የጥያቄ ቁ.314 መልስ አዎ ከሆነ፣ በምን ያህል ጊዜ ሻይ ይጠጣሉ?	1. ሁሉ ጊዜ ከምግብ በኋላ 2. በቀን አንድ ጊዜ ብቻ 3. በሁለት ቀን አንድ ጊዜ ብቻ 4. አልፎ አልፎ	
316	ከምግብ በኋላ ቡና ይጠጣሉ?	1. አዎ 2. አይደለም	
317	የጥያቄ ቁ.316 መልስ አዎ ከሆነ፣ በምን ያህል ጊዜ ቡና ይጠጣሉ?	1. ሁሉ ጊዜ ከምግብ በኋላ 2. በቀን አንድ ጊዜ ብቻ 3. በሁለት ቀን አንድ ጊዜ ብቻ 4. አልፎ አልፎ	
318	የግራ እጅ የላይኛው የመሀል ክንድ ዙሪያ መጠን? (መጠኑን ለክትህ/ሽ ውሰድ/ጂ)	_____	
319	ቦዲ ማስ ኢንዴክስ (BMI) መጠን? (ቁመትና ክብደት ለክትህ/ሽ ውሰድ/ጂ)	_____	
IV. እርግዝናና ሌሎች ተያያዥነት ያላቸው ጥያቄዎች			
401	የአሁኑ ጽንሰ ምን ያህል ወር/ሳምንት ይሆነዋል? (በሳምንት ዓፍ/ፊ)	_____	
402	በአሁኑ እርግዝና የደም መፍሰስ ችግር አጋጥሞዎት ያውቃል?	1. አዎ 2. አይደለም	
403	ከአሁኑ እርግዝና በፊት ወልደው ያውቃሉ?	1. አዎ 2. አይደለም	
404	የጥያቄ ቁ.403 መልስ አዎ ከሆነ፣ በአጠቃላይ በሀይወት ዘመንዎ ምን ያህል	_____	

	ልጅች ወልደዋል?		
405	የጥያቄ ቁ.403 መልስ አዎ ከሆነ፤ የመጨረሻ ልጅዎን የተገላገሉት/የወለዱት የት ነበር?	1. በጤና ተቋም ውስጥ 2. በቤት ውስጥ 3. ሌላ ካለ ይጠቀስ_____	
406	የጥያቄ ቁ.403 መልስ አዎ ከሆነ፤ የመጨረሻ ልጅዎን ከወለዱ በኋላ አስከሁኑ እርግዝና ያለው ጊዜ ምን ያህል ዓመት ይሆናል?	_____	
407	በህይወት ዘመንዎ በተለያዩ አጋጣሚዎች ጽንሰ የማስወረድ ችግር አጋጥመዎት ያውቃል?	1. አዎ 2. አይደለም	
408	የጥያቄ ቁ. 407 መልስ አዎ ከሆነ፤ እንደዚህ ዓይነት የጽንሰ ችግር ለምን ያህል ጊዜ አጋጥመዎታል?	_____	
409	የጥያቄ ቁ.407 መልስ አዎ ከሆነ፤ እንደዚህ ዓይነት የጽንሰ ችግር ለመጨረሻ ጊዜ ያጋጠምዎት ከሰንት ዓመት በፊት ነበር?	_____	
410	ከአሁኑ እርግዝና በፊት ማንኛውም ዓይነት የወሊድ መከላከያ እንክብል/መድሃኒት ወስደው ያውቃሉ?	1. አዎ 2. አይደለም	
411	ከአሁኑ እርግዝና በፊት የወር አባባ የሚመጣበት ጊዜ የመዛባት ሁኔታ ተፈጥሮ ያውቃል?	1. አዎ 2. አይደለም	
412	ከአሁኑ እርግዝና በፊት በወር አባባ ጊዜ ከወትሮው የተለየ ብዙ የደም መፍሰስ ችግር አጋጥመዎት ያውቃል?	1. አዎ 2. አይደለም	
V. ከጤና አገልግሎት ጋር ተያያዥነት ያላቸው ጥያቄዎች			
501	ከዚህ በፊት አርግዘው ያውቃሉ?	1. አዎ	

		2. አይደለም	
502	የጥያቄ ቁ.501 መልስ አዎ ከሆነ፤ ከዚህ በፊት በነበረው እርግዝና የቅድመ ወሊድ ክትትል አድርገዋል?	1. አዎ 2. አይደለም	
503	የጥያቄ ቁ. 502 መልስ አዎ ከሆነ፤ ምን ያህል ጊዜ የቅድመ ወሊድ ክትትል አድርገዋል?	_____	
504	በአሁኑ እርግዝና አይረገገ(iron tablet) ከጤና ተቋም ተሳታፊነት ወይም ከሱቅ ገዝተው ተጠቅመዋል?	1. አዎ 2. አይደለም	
505	በአሁኑ እርግዝና የፀረ ሆድ ትላትል መድኃኒት ወስደዋል?	1. አዎ 2. አይደለም	

Laboratory Requesting and recording format

This format is developed for the study purpose to record laboratory examination results of pregnant women. To be filled and completed carefully and timely by the laboratory technicians working in the laboratory department. This format is applicable only for this study.

Personal information and laboratory examination result

1. Personal data

1.1 Name _____

1.2 Code no. _____

1.3 Date of sample collection _____

2. Stool specimen examination

2.1 Physical examinations

I. Consistency of the stool

1. Formed

2. Semi formed

3. Soft

4. Watery diarrhea

5. Bloody diarrhea

II. Appearance of the stool

1. Blood stained

2. Mucus

3. Normal

2.2 Direct microscopy examination

I. No ova parasite

II. Type of ova parasite seen

1. Hook worm

2. A. lumbricoids

3. T. trichuria

4. Others specify _____

3. Blood film examination

3.1 Blood film examination of malaria

I. No malaria parasite seen

II. Type of malaria parasite seen

1. PF 2. PV 3. Others specify _____

III. Parasite load grading using plus sign scheme

1. + 2. ++ 3. +++ 4. ++++

3.2 Hemoglobin concentration (HBC) _____

1. Normal 2. Mild anemia 3. Moderate

4. Severe 5. Very severe

Name of lab. Technician _____ Signature _____ Date _____

Annex V. Laboratory examination procedures

I. Stool specimen examination

Bench-level Standard Operating Procedure
PARASITES IN STOOL - DIRECT SMEAR MICROSCOPIC EXAMINATION
Staff able to perform test: Laboratory Assistant and higher
<p>Principle of the Test Method:</p> <p>Many parasites cause disease in man. Some of these parasites are excreted in stool; they are called intestinal parasites. Intestinal parasites can be identified by examination of fresh stool samples. In stool samples we can find worms (eg. <i>Ascaris lumbricoides</i>) and segments of worms (e.g. <i>Taenia</i> species) visible to the eye. By microscopic examination of fresh stool samples, we can find eggs (e.g. Hookworm) and larvae of worms (e.g. <i>Strongyloides stercoralis</i>). We also find protozoa trophozoites (e.g. <i>Amoeba</i>) and cysts (e.g. <i>Cyclospora cayetanensis</i>). In heavy and moderate infection, a direct smear examination with normal saline and/or iodine to stain cysts, is usually sufficient. For light infections, a concentration of the stool sample might be required to find helminth (worm) eggs and protozoa by microscopic examination</p>
<p>Clinical Significance of the Test:</p> <p>Many pathogenic parasites are excreted in stool. Often, when a person is infected with intestinal parasites, other symptom such as anaemia. However diagnosis by physical examination is not sufficient to identify intestinal parasitic infection. Stool examination is essential to identify parasites that cause the disease.</p>
<p>Specimen:</p> <p>Fresh stool samples.</p>
<p>Equipment Requirements:</p> <ul style="list-style-type: none"> <input type="checkbox"/> Glass slides <input type="checkbox"/> Cover slip (20 x 20 mm) <input type="checkbox"/> Wooden applicator <input type="checkbox"/> Grease pencil. <input type="checkbox"/> Microscope.
<p>Reagents & Stain Requirements:</p> <ul style="list-style-type: none"> ○ Normal Saline (0.9% Sodium chloride solution) ○ Lugol's iodine.
<p>How to prepare 0.9% Sodium chloride solution –Normal Saline</p> <p>Sodium chloride NaCl 9 g Distilled water.....1000 ml</p>

How to prepare Lugol's iodine:

Potassium iodide..... 2 g

Iodine..... 1 g

Distilled water.....up to 100 ml

Mix the potassium iodide with about 200 ml of water until it is completely dissolved. Add the iodine to the potassium iodide solution, mix until dissolved. Add water until the 1000 ml mark. Store in dark brown bottle. Use a small amount in a brown dropper bottle. Prepare a new solution if colour fades.

Test Procedure Instructions:

- Always first examine the stool sample macroscopically (with your eyes).
- Note the colour, consistency and look for mucus, blood stains and worms.

- Label a glass slide with the patient name and/or lab number.
- Put one drop of Normal Saline in the middle of the left half of the slide.
- Put one drop of Lugol's iodine in the middle of the right half of the slide.
- You can examine the slide using Normal Saline only but you would not be able to see cysts well. Therefore it is advisable to examine each stool sample with Lugol's iodine and Normal Saline.

- Take a small piece of stool with the wooden applicator. (About this size).
- If the stool is formed take the piece from inside and the surface of the sample.
- If the stool is liquid take a drop. Any part is o.k.
- N.B. If the specimen is very liquid place one or two drops of stool directly onto the slide and cover it with the cover glass, do not add the saline as this would further dilute the specimen.*

- Mix the sample first with the drop of Normal Saline on the left half of the slide.
- If the stool contains mucus or bloodstained parts, prepare a second slide with a drop of Normal Saline and take the piece from the mucus or blood-stained part.

N.B. The Iodine preparation is useful to identify cysts but kills all living organisms in the specimen.

- Take a small piece again and mix it with the drop of Lugol's iodine on the right half of the slide.
- The iodine preparation is useful to identify protozoa cysts.

- Place a cover slip over each drop.
- Put the cover slip slowly letting it move down from the side to avoid air bubbles.

- Examine the entire cover slip systematically.
- For the saline preparation use the 10x objective and the 40x objective.
- For the Lugol's iodine preparations use the 40x objective, as there you might find cysts.

II. Blood film examination for malaria infection

SOP 1 PREPARATION OF MATERIALS FOR SPECIMEN COLLECTION

- Safety precautions must be considered, e.g. gloves
- All collection materials must be ready and arranged in order of use:
- Cotton wool/spirit swabs
- Slides/blood containers – clean for proper diagnosis
- Needles/syringes/lancets
- Sharps disposal box

SOP 2. METHODS OF SPECIMEN COLLECTION: CAPILLARY & VENOUS BLOOD

- Emphasize precautions to be taken to avoid contamination when blood films are made: cleaning slides, avoiding dust and flies
- Wearing protective gloves whenever handling blood or taking blood samples
- Avoiding fingers or hands getting into contact with blood
- Covering any cuts or abrasions on the hands with adhesive dressings
- Taking care not to prick others with contaminated sharp objects
- Not to use disposable lancets more than once
- Washing hands with soap and water after handling blood specimens
- Wiping the blood immediately it gets into contact with the skin using cotton wool soaked in alcohol and then washing the affected area with soap and water ²³

SOP 3. SPECIMEN PREPARATION

Thick and thin blood films:

- obtaining the blood specimen through finger prick
- making thick and thin films on the same microscope slide in duplicate
- labeling the dry thin film using a pencil

SOP 4. Preparation of equipment & reagents

Equipment

The microscope:

- Demonstrate the major components of the microscope
- Setting of the microscope
- Examine the specimen using the microscope
- Examine specimens using low power objectives (x10 & x40) before using the oil immersion objective

Weighing balance:

- Demonstrate the chemical balance
- Operate the chemical balance

Reagents

- Demonstrate the preparation of buffered water
- Checking and adjustment of pH of buffered water
- Preparation of stains (Giemsa stain, Field stain A & B)
- Identify the absolute methyl alcohol (methanol)
- Identify the anti-coagulant of choice for venous blood

SOP 5. PROCESSING OF SPECIMENS: THICK & THIN FILMS, RDTs

Giemsa staining technique

- Transfer the slide into a staining jar
- Two films can be arranged such that each film faces away from the other, while being slotted into the grooves in a staining jar
- Most of the jars can carry 5 – 20 slides, depending on the capacity and arrangement
- The jars are then filled with Giemsa stain diluted 3% and 10% with buffer solution at pH 7.0 – 7.2 and left to stain for 45 min (3%) and 45 min (10%).
- Washing of the films involves adding more buffered water or tap water to float off the scum forming on the surface. This is followed by thorough rinsing until no more stain can be detected in the washing water
- The back of every film is wiped dry with a piece of dry gauze and the films are then air dried. This can be achieved by the use of a drying rack or spreading them on a clean bench.

SOP STAINING OF SLIDES

SOP 6. EXAMINATION OF SPECIMENS AND REPORTING OF RESULTS

i) Examination of specimen

- Immersion oil is applied onto thick and thin films to be examined under the microscope
- The films are then set on the mechanical stage
- The x100 objective is put in place
- Focusing is achieved through the use of the coarse and fine adjustments
- When the film is in focus a thorough search is made for parasites that could be in blood. The whole thick film should be examined before the film is declared negative.

ii) Reporting of results

- Enter the following in the laboratory request form and the daily register:
- Results including parasite species and count
- Remarks
- Initials of the person performing the test

SOP 7. QUALITY CONTROL

This can be observed through:

- Inclusion of known positive samples in the procedure at regular intervals
- Every new stock of Giemsa and Field stain should be tested with known positive slides before being used for routine work
- The in-charge can occasionally examine the films before they are discarded
- Develop Standard Operating Procedures
- Establish quality assurance protocols
- Establish record keeping procedures and regularly review the accuracy of record keeping
- Monitor the diagnostic systems regularly