**KWAME NKRUMAH UNIVERSITY OF SCIENCE AND TECHNOLOGY**

**KUMASI, GHANA**

**COLLEGE OF SCIENCE**

**DEPARTMENT OF THEORETICAL AND APPLIED BIOLOGY**



# DETERMINANTS OF ANAEMIA IN PREGNANT WOMEN IN PERI-URBAN KUMASI IN THE ASHANTI REGION

BY

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FEBRUARY 2011

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A THESIS SUBMITTED TO THE DEPARTMENT OF THEORETICAL AND APPLIED BIOLOGY, KWAME NKRUMAH UNIVERSITY OF SCIENCE AND TECHNOLOGY IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR THE AWARD OF MASTER OF SCIENCE (MSC) DEGREE IN ENVIRONMENTAL SCIENCE.

BY

**AGYIRI JOSEPH**

FEBRUARY 2011

**DECLARATION**

I hereby declare that this thesis is the result of my own work except for the references cited which have been duly acknowledged. It has never been submitted in substance for the award of any degree.

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**DEDICATION**

This work is dedicated to my wife, Isabella and children, Harriet and Angela.

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**ABSTRACT**

A field survey was conducted to investigate the causes of anaemia in pregnant women in peri-urban communities of North Suntreso and Pramso in the Ashanti Region of Ghana. Three hundred pregnant women attending antenatal were studied in both areas. Blood samples of participants were collected to determine serum ferritin levels, malaria parasitimia and haemoglobin concentration. Stool examination was also conducted to determine the presence or otherwise of parasitic intestinal worms. Questionnaires were administered to obtain information on the socio-demographic characteristics of participants. Majority of the participants (43.6% and 71.3% for Pramso and South respectively) were anaemic with Hb below 10g/dl. Mean ferritin levels ranged between zero and 275 microliters. Malaria parasitaemia was of a high prevalence (56.4% and 68.3%) among the participants in Pramso and North Suntreso respectively. There was a significant association between the knowledge of transmission and prevention and malaria parasitaemia. However no significant association was found between the low helminth parasitaemia recorded ($P < 0.005$) and anaemia.

There is the need for education on the contributory factors to anaemia especially, iron deficiency anaemia.

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# CHAPTER ONE

## 1.0 INTRODUCTION

Anaemia is generally referred to a condition or state in which blood is deficient in erythrocytes (red blood cells) [number of red cells less than 10 g/dl]or the red blood cells don't have enough haemoglobin. It is a widespread condition worldwide, with a high prevalence especially among pregnant women (56%). Anaemia as a major public health problem throughout the world is widely recognized by health authorities and policy makers alike (WHO, 2003), and about four hundred different kinds of anaemia have been identified each with its own cause, however, many of them are rare (WHO, 2003). Anaemia can be temporary or long-term and it can range from mild to severe.

The cause of anaemia may be blood loss, a chronic illness, a genetic or acquired defect, or disease; it may also be a side effect of a medication or an iron or vitamin deficiency (Dass *et al*., 1967). The symptoms of anaemia include weakness and sometimes being out of breath or, unable to work hard, paleness of the skin (WHO, 2003), less resistance to infections, impaired immune function and cardiac failure (Lemson *et al*, 1992).

The most common forms of anaemia include the following, iron deficiency anaemia, folic acid deficiency, vitamin B12 deficiency, haemolytic anaemia, haemolytic anaemia, and sickle cell anaemia.

Iron deficiency anaemia – the most common type of anaemia - is defined as a condition in which there are no mobilizable iron stores and in which signs of a compromised supply of iron to the tissues including the erytron are noted. Iron deficiency occurs when an insufficient amount of iron is absorbed and thus does not meet the body's iron

requirements (WHO, 2003). Iron deficiency result in a decrease in haemoglobin concentration and subsequent anaemia.

Iron is an essential mineral and an important component of proteins involved in oxygen transport and metabolism (Ursel, 2001). Almost two-thirds of the iron in our body is found in heamoglobin (an iron -rich protein in red blood cells that gives blood its red colour and carries oxygen from the lungs to our body's tissues) [WHO, 2003; Ursel, 2001].



Iron plays a crucial role in the processes of growth, cell division and in the transport of oxygen throughout the body. It is also important for the proliferation of cells of the immune system as well as for micro-organisms including the malaria parasites.

Anaemia results when haemoglobin level is below the normal range of 11g/dl for a person's age and sex. The heamoglobin level tends to decrease with age and is lower in females in reproductive age than the male in the same category - as a result of blood loss during menstruation (WHO 1990). Severe anaemia in pregnancy has been defined as < 7g/dL (WHO 1989). Smaller amounts of iron are found in myoglobin - a protein that helps supply oxygen to muscle, and in enzymes that assist biochemical reactions in cells (Ursel, 2001).

The onset of iron deficiency anaemia is gradual; the red cells die off faster than they can be made by the body. Iron deficiency can also be a nutritional problem that has far-reaching effects on productivity (WHO, 2003). The body gets its iron from food and the main food sources are meat and shellfish as well as iron-fortified foods (that is, foods that have iron added). A steady supply of iron is needed to form haemoglobin and healthy red blood cells (Matt, 2004). A person can have low iron levels for three main

reasons: Blood loss, either from disease or injury, nutritional deficiency, not being able to absorb the iron in the diet because of damage to the intestinal lining and inflammation leading to hepcidin anaemia. The more severe stages of iron deficiency are associated with anaemia. Iron deficiency, like most nutritional deficiencies of public health concern is mainly a consequence of poverty (WHO 2003).

According to WHO (2003), infants, pre-school children, adolescents particularly girls and women of childbearing age, particularly pregnant women, are at greatest risk of being anaemic.

There is increased risk of anaemia in pregnant women during delivery; during the perinatal period, their babies are at an increased risk of death. In pregnant women, the consequences of iron deficiency, and especially iron deficiency anaemia include increased maternal disability and death; increased foetal disability and death (Lemson *et al.*, 1992).

Due to the importance of anaemia, a National Nutritional Anaemia Prophylaxis Programme (NNAPP) was initiated in 1970 which had a five year plan with the aim to reduce the prevalence of anaemia to 25 percent by 2012 (WHO, 2003). Again, in the last decade the importance of iron deficiency anaemia as a public health problem has been recognized by health authorities and policy makers (Stolzfus and Dreyfuss 1988).

In pregnant women for example, iron-deficiency anaemia can increase the risk of premature deliveries and low-birth-weight babies (Stolzfus and Dreyfuss 1988). The causes of morbidity and mortality in pregnancy and childbirth can be direct (conditions occurring only in pregnancy), most often obstructed labour, ante partum and postpartum haemorrhage, puerperal infection, and hypertensive disorders, or indirect (diseases

aggravated by pregnancy) such as anaemia, malaria, heart disease, essential hypertension, *diabetes mellitus*, and haemoglobinopathies (Stolzfus and Dreyfuss, 1988).

Prevalence of anaemia among pregnant women in developing countries averages 56% and ranges between 35 and 100% among different regions of the world (WHO, 2003). Various studies from different regions of India have reported the prevalence of anaemia to be between 33 and 100% (Darbari, 1993). During pregnancy, at delivery and during the peri-natal periods, a total amount of about 700-850mg of iron is needed to meet the iron requirements of a mother and foetus. The average woman of reproductive age needs about 300-500mg additional iron to maintain iron balance during pregnancy. The difference has to come from the mothers store or from iron supplements. In developing countries 25-30% has no iron reserves at all, and because the situation is more serious among pregnant teenagers it is important to promote all measures that will improve iron reserves before and during pregnancy (Harrison, 1982).

In many areas of sub-Saharan Africa, the synergy between poor nutrition and infectious disease conspire to exacerbate some of these direct and indirect conditions. High fertility, child bearing at the extremes of the fertile years, and the poor social status of women (evidenced in part by low levels of education), increase the problem in a setting where often there is inadequate or improper health care. As indicated by the MMR, an excessive loss of life, and of disability, results (Stolzfus and Dreyfuss, 1988). In addition to the lack of quality foods, blood loss from parasites is the leading cause of iron-deficiency anaemia in most areas of the world. Inadequate dietary intakes of iron

are seen most often in premenopausal women, infants and children. Iron deficiency is another nutritional problem that has far-reaching effects on productivity

Globally, iron deficiency anaemia affects over two billion people. An estimated 39% of preschool children are anaemic as are 52% of the pregnant women. This number soon rises when children and adolescents are factored in, since about 10% of toddlers and 10% of adolescent girls are also iron deficient (Stolzfus and Dreyfuss, 1988). In Ghana, iron deficiency, is most common in young children and in women of childbearing. For example, anaemia during pregnancy is estimated in Ghana to affect close to 70% of pregnant women, and has implications for maternal mortality. Ghana has an unacceptably high maternal mortality rate of 214/100,000 and about 20% of this is due to anaemia (WHO, 2003). Data from national surveys indicate that, about 81% of children fewer than 5 years, 17% of non-pregnant women-and 69% pregnant women are anaemic (RCH, 2003). According to WHO (2001), there is at least a 1% reduction in productivity for each 1% drop in iron status. It was projected that between 1997 and 2001, 90 million dollars was lost in agricultural productivity as a consequence of iron deficiency anaemia in the female labour (WHO, 2001).

Although there is a relation between the incidence of malaria and anaemia, recent studies by Tutu *et al*., (2005) and Lena *et al*., (2006) on pregnant women in the Ashanti region of Ghana suggested that the levels of anaemia observed in pregnant women had no significant relationship with the level of malaria parasitaemia. In Ghana, although there are several programs, especially on malaria control that target pregnant women as a means of reducing anaemia among pregnant women, the rather high prevalence of anaemia calls for investigation into other factors that may contribute to anaemia. There

is therefore the need to find out the prevalence of anaemia in pregnant women and its contributory factors especially iron deficiency.

## 1.1 AIM AND OBJECTIVES OF THE STUDY

The main objective was to determine the prevalence of anaemia among pregnant women in peri-urban communities visiting antenatal clinics and the possible contributory factors.

The specific objectives were to determine,

i) Prevalence of anaemia among pregnant women

ii) Haemoglobin concentrations

iii) The iron status of the pregnant women

iv) Malaria parasitaemia and,

v) The presence of intestinal helminths

# CHAPTER TWO

## 2.0 LITERATURE REVIEW

### 2.1 ELEMENTAL IRON

Iron, one of the most abundant metals on Earth, is essential to most life forms including humans. It is an integral part of proteins and enzymes that maintain good health. In humans, iron is an essential component of proteins involved in oxygen transport. It is also essential for the regulation of cell growth and differentiation. A deficiency of iron limits oxygen delivery to cells, resulting in fatigue, poor work performance, and decreased immunity. On the other hand, excess amounts of iron can result in toxicity and even death (Matina, 2003).

Iron is also found in proteins such as ferritin that store iron for future needs and that transport iron in blood (Andrews, 1999). Iron has the longest and best-described history among all the micronutrients and it is a key element in the metabolism of almost all living creatures (Ursel, 2001).

### 2.2 ANAEMIA.

Anaemia may result from defects at any stage of red cell and haemoglobin production or when an increased rate of red cell destruction (haemolysis) exceeds the capacity of the bone marrow to mount a compensatory increase in production. Changes in the relationships between red cell and plasma volumes may also result in a reduced haemoglobin concentration, such changes occur physiologically in pregnancy, where red cell volume is increased less markedly than plasma volume. Generally anaemia occurs at a haemoglobin level below 11g/dl. Haemoglobin concentrations below which

anaemia is likely to be present at sea level are usually defined as follows: Children 6 months – 6 years: 11 g/dL; Children 6-14 years: 12 g/dL; Adult males: 13 g/dL; Non-pregnant females: 11 g/dL; pregnant females 12 g/dL. All anaemia sufferers manifest signs and symptoms attributable to tissue and organ hypoxia and the ensuing reduced metabolism. It also occurs when there are an inadequate number of red blood cells or an inadequate amount of haemoglobin for the body to function properly. Haemoglobin is a protein in red blood cells that carries oxygen to the brain, muscular system, immune system, and other parts of the body. Without adequate oxygen, the physical and mental capacities of individuals are reduced (Verhoef, 1999).

Most of the anaemic population lives in developing countries, where high anaemia prevalence is seen, particularly in pregnant women, young children, female adolescents, and women of childbearing age. (WHO. 2001). Overall, anaemia contributes to about 20 percent of maternal and peri natal deaths in developing countries. A recent WHO World Health Report noted that the risks of both maternal and peri natal mortality were reduced by 25 percent and 28 percent, respectively, for each gram increase in hemoglobin level between 50 and 120 g/L. This is contrary to the previous generally accepted understanding that only severe anaemia resulted in death. This finding is very important because the numbers of women and children with mild and moderate anaemia are vastly greater than the number with severe anaemia. It follows then that the great majority of anaemia-related maternal and peri -natal deaths are due to mild and moderate anaemia rather than severe anaemia. (WHO, 2003).

Anaemia's other serious negative consequences include poor pregnancy outcomes such as low birth weight and premature birth. Anaemia also has adverse implications for

social and economic development. There is now strong evidence that anaemia can reduce cognitive development and limit a child's learning in schools. This will lower the effectiveness of investments in education. Anaemia's role in reducing physical capacity and work productivity in adults has been long established. Nevertheless, anaemia continues to have a relatively low priority in health policies and programs, compared to other nutrition-related health problems with more obvious life-threatening implications (WHO, 2003)



These are major constraints for policy formulation and program development. A better understanding of the etiology or causes of anaemia and the identification of critical issues related to effective anaemia program design and implementation are key to developing more successful actions. Recent progress in understanding the nature of the problem and the achievements and limitations of existing programs provides a firm basis for designing effective strategies and interventions. MOST, the USAID Micronutrient Program has supported ministries of health in the Democratic Republic of the Congo, Ghana, Uganda, and Nicaragua to develop approaches to address anaemia.

According to WHO (1997), approximately 30% of the world's population is affected by anaemia. In a meta-analysis of available data using WHO threshold criteria (De Maeyer and Adiels-Tegman, 1985), the problem was found predominantly in developing regions (especially south Asia and sub-Saharan Africa) where 36% of the total population were estimated to be anaemic compared to 8% in developed nations. Prevalence was particularly high in pre-school children (51% in less developed and 10% in more developed regions) and in adult females (50% and 13% respectively). In pregnancy, a WHO tabulation of available data averaged the prevalence to be 56% in

developing countries, ranging between 50-70% for Hb < 11g/dL and 5-15% for Hb < 7 g/dL in sub-Saharan Africa (WHO 1992).

Anaemia is a huge public health and nutrition problem with serious consequences (WHO, 2003). In Ghana the condition primarily affects young children and women. About 240,000 children between the ages of one and two have the condition. About 3.3 million women of child-bearing age have iron deficiency anaemia.

Although the classic symptoms of iron deficiency have long been known, another symptom is also becoming classic: a poor tolerance to cold. One way the body accelerates heat production when the environmental temperature falls involves the neurotransmitter norepinephrine and the thyroid hormones, which speed up the metabolic rate. Iron deficiency impairs temperature regulation in both animals and humans, probably by interfering with the normal production of these compounds (Andrews, 2001).

## 2.3 TYPES OF ANAEMIA

### 2.3.1 IRON DEFICIENCY ANAEMIA

Iron deficiency anaemia is a condition where there is a reduced amount of haemoglobin and decreased number of red blood cells in the body. Anaemia leads to less oxygen getting to the cells and tissues affecting their function. (Lanerolle and Atukorala, 2006; WHO, 2003) About 3.3 million women of child bearing age have iron deficiency anaemia. Iron deficiency with or without anemia reduces work productivity in adults and limits cognitive development in children, thus limiting their achievement in school and ultimately reducing investment benefits in education (WHO, 2002). It is estimated

that about 2,150 million people are iron deficient and that this deficiency is severe enough to cause anaemia in 1,200 million people globally. About 90% of all anaemia have an iron deficiency component (Fernando, 2008). In the developing world nearly half of the population is iron deficient. However, the industrial world is not free from it: 11 % of its population has iron deficiency (WHO, 2003; Fleming, 1970).

Roughly 47% of non-pregnant women and 60% of pregnant women have anaemia worldwide, and including iron deficiency without anaemia the figures may approach 60 and 90% respectively. In the industrial world as a whole, anaemia prevalence during pregnancy averages 18%, and over 30% of these populations suffer from iron deficiency (WHO, 2003; Fleming, 1970).

### 2.3.2 IRON DEFICIENCY IN PREGNANCY

The high risk of women of fertile age and pregnant women for incurring negative balance and iron deficiency is due to their increased iron needs because of menstruation and the substantial iron demands of pregnancy. Median requirements of absorbed iron are estimated to be 1.36 and 1.73 mg per day among adult and teen-age menstruating females. However, 15% of adult menstruating women require more than 2.0 mg per day, and 5% require as much as 2.84 mg per day. The superimposition of menstrual losses and growth in menstruating teenage girls increases the demands for absorbed iron; 30% need to absorb more than 2.0 mg of iron per day; 10% as much as 2.65 mg, and 5% 3.21 mg. These requirements are very difficult, if not impossible to satisfy even with good quality, iron-fortified diets (Fernando, 2008).

### 2.3.2.1 SYMPTOMS OF IRON DEFICIENCY

A mild case usually causes no symptoms or problems. However, a severe case can cause extreme fatigue (tiredness) and weakness. Severe iron-deficiency anaemia can lead to serious problems for young children and pregnant women, and it can affect the heart (Dass et al. 1967). This type produces red blood cells (RBCs) that are smaller than usual, and hence, the term microcytic is used when referring to them. RBCs are not only reduced in size and number, but contain a subnormal amount of hemoglobin, which causes cells to become pale. The term hypochromia describes cells that are pale in color because they do not contain their full complement of hemoglobin.



### 2.3.2.2 CAUSES OF IRON DEFICIENCY ANAEMIA

There are many causes for iron deficiency anaemia of which the major causes are as follows;

i. an inadequate dietary intake of iron (less than 1 to 2 mg/day) as a result of a poor diet; prolonged, unsupplemented breast- or bottle-feeding of infants; or during such periods of stress as rapid growth in children and adolescents. The elderly often develop anemia as their interest in food wanes. Dieters are another group that can become anaemic because they do not meet their iron need.

ii. iron malabsorption which prevents iron from being used and may occur because of chronic diarrhea, partial or total gastrectomy, or such other malabsorption syndromes as celiac disease (where the lining of the small intestine is damaged by the effects of gluten - the protein found in wheat and other cereal grains of the grass family)

iii. secondary blood loss resulting from drug-induced gastrointestinal bleeding (usually from anticoagulants, aspirin, or steroids); heavy menses; or hemorrhage from trauma, gastrointestinal ulcers, malignancy, or varices (enlarged veins, arteries, or lymphatic vessel)

iv. pregnancy, which diverts maternal iron to the fetus for erythropoiesis and then

v. mechanical erythrocyte trauma caused by a prosthetic heart valve or vena cava filters.

Such diseases as rheumatoid arthritis, connective tissue disorders, chronic infection, trauma or malignancy are commonly confused with mild iron deficiency anaemia (Ref). Instead, they are related to anaemia of chronic disease.

Other diseases that can lead to iron deficiency include the following:

Esophagitis is an inflammation of the esophagus and is the most common failure of the muscular valve between the esophagus and the stomach. This causes a failure to keep the highly acidic gastric contents from going back up (refluxing) into the esophagus. Since the esophageal lining is not adapted to withstand much acid, inflammation of the mucosa results and blood slowly leaks out, ultimately leading to anaemia.

Gastritis; an inflammation of the mucosa of the stomach. There are two major causes: infection from the bacterium *Helicobacter pylori*, and the other being various oral medications (mostly aspirin and NSAIDs [ibuprofen sold as Motrin, Advil, and Midol], naproxen [Aleve], and ketoprofen [Orudis, Acton]). These drugs may be excellent for

relieving pain, but their side effects are so common that some darkly refer to them as "gastroenterologist's little helpers".

Peptic ulcer is a localized area of inflammation in the lining of the duodenum or stomach. The inflammation is so intense that the mucosa and underlying tissues actually break down, resulting in a crater. Not until the 1990s did researchers know the cause, which is now mainly the result of the *Helicobacter bacterium*.



Benign tumors are fairly common in the lining of the gut. The most common are hyperplasic polyps which have no real medical ramification except when they become ulcerated, allowing blood to seep out and causing an iron deficiency. The common benign tumors of the colon and rectum are the "adenomas", of which there are two types: tubular (also called adenomatous polyps) and the villous adenoma. Aside from chronic bleeding and a resulting iron deficiency anemia, adenomoas have the potential of turning into frank cancers.

The body will naturally withhold iron from carcinogenic or infectious entities because of their need for iron. In these cases, iron supplementation can actually contribute to the spread of disease.

Cancer is the most serious cause of chronic iron loss and is the most common cancer of the gastrointestinal tract in North America. These cancers are generally slow-growing as they eat through the wall of the gut. They can also metastasize, causing daughter tumors in the lymph nodes draining the GI tract, as well as the liver. The second most important

cancer of the gut is adenocarcinoma of the stomach seen more commonly in Asia and developing countries. This type has a relatively poor prognosis.

### 2.3.2.3 STAGES OF DEVELOPMENT OF IRON DEFICIENCY ANAEMIA

Iron deficiency develops in stages, but blood symptoms do not develop until iron deficiency has progressed quite far to the point where brain and muscular activity may already be impaired. Unfortunately, low hemoglobin and hematocrit test results are often relied upon as the first indicators of iron deficiency.

The three overlapping stages of iron deficiency are as follows:

- *Stage One*: The body's iron stores for erythropoiesis are depleted; but, at this stage, erythropoiesis remains normal as does the hemoglobin content of the maturing erythrocytes.
- *Stage Two*: Not enough iron is transported to the marrow, and iron-deficient erythropoiesis begins.
- *Stage Three*: This stage begins when the small, hemoglobin-deficient cells enter the circulation in increasing numbers, replacing normal erythrocytes that have grown old and have been removed from the circulation. By this time, the development of iron deficiency anemia is associated with the depleted transport of iron stores and diminished hemoglobin production.

Iron is found in all plants foods but is more plentiful and bio-available in meat. Deficiency results from ineffective absorption of iron excess loss. Absorption is tightly regulated in the intestines depending on the iron status of the individual, the type of iron

and other nutritional requirements. Once iron is absorbed it is well conserved. Iron is depleted primary through blood loss, including from parasitic infections such as schistosomiasis and hookworm.

Mainly found in haemoglobin, iron is essential for the binding and the transport of Hoxygen as well as for cell growth and differentiation (Beard, 2001). Iron deficiency anaemia results in neurological impairment which may not be fully reversible (Grantham-McGregor and Ani, 1999). Iron deficiency is also known to decrease immune factors functions, but some investigators has also hypothesized that deficiency protects against infectious diseases or that iron supplementation increases infectious diseases (Caulfield *et al*., 2004). Stolzfus, Mullany and Black (2004) found that iron deficiency anaemia was the underlying factors in 841,000 deaths per year resulting from maternal and peri-natal causes and it directly causes the deaths of 134,000 young children.

Iron deficiency is the most principal source of anaemia worldwide (British Nutrition Foundation 1995). The principal cause is dietary iron deficiency (due to low iron intake or poor bioavailability caused by high intake of iron binders such asphytate or tannins common to cereal based diets). Physiological increases in iron losses (e.g. menstrual losses, iron losses to the foetus during pregnancy, iron lost in the milk during lactation), or pathological iron losses (e.g. gastrointestinal haemorrhage), increased tissue iron requirements (e.g. during periods of rapid growth in infants, children and adolescents) or, more rarely, malabsorption of iron due to intrinsic gastrointestinal disease, are all contributors to iron deficiency anaemia. The decline in body iron is first marked by the depletion of iron stores, which indicates the onset of iron deficient erythropoiesis.

Haemoglobin synthesis starts to become impaired and haemoglobin concentration falls. Iron deficiency anaemia is characterized by microcytic-hypochromia red blood cells.

Because of the gradual progression of the disorder, many patients are initially without symptoms and tend not to seek medical help until the anaemia becomes severe, that is, when haemoglobin levels have dropped to a certain level (about 7 or 8 g/dl). Long before the mass of the red blood cells is affected and anaemia is diagnosed, a developing iron deficiency affects behavior. Even at slightly lowered iron levels, the complete oxidation of pyruvate is impaired, reducing physical work capacity and productivity, resulting in common symptoms of fatigue, weakness, and shortness of breath.

In advanced stages, decreased hemoglobin and the consequent decrease in the blood's oxygen-carrying capacity cause the patient to develop dyspnea on exertion, fatigue, dizziness, listlessness, pale ear lobes, palms, conjunctiva, and mucous membranes, inability to concentrate, irritability, headaches, susceptibility to infection, and a tingling sensation in the fingers and toes. Decreased oxygen perfusion causes the heart to compensate with increased cardiac output and tachycardia. Other, less common manifestations of iron deficiency include glossitis (inflammation of the tongue), angular stomatitis (fissuring at the corners of the mouth), and koilonychia (concave or "spoon" fingernails).

Structurally or functionally altered epithelial tissue is often found in individuals with iron deficiency anaemia. The nails become brittle, thin, and "spoon-shaped" or concave as a result of impaired capillary circulation. The tongue may be sore, with redness and burning, caused by atrophy of the papillae. However, these changes can be reversed

within a week or two of iron replacement. Changes may also occur in the epithelium at the corners of the mouth, causing soreness, dryness, and cracking (angular stomatitis). The tongue turns smooth, and the patient complains of dysphasia (difficulty swallowing), or may develop pica. Dysphasia is often associated with a "web" of mucous and inflammatory cells at the juncture between the hypopharynx and esophagus. These lesions may become malignant. There may also be associated neuromuscular effects, including vasomotor disturbances, numbness and tingling of extremeties, and neuralgic pain. (Andrews, 1999).



### 2.3.3 FOLIC ACID DEFICIENCY ANAEMIA.

Folic acid is a member of the vitamin B family; it is used in the production of new red blood cells. Some people do not have enough folic acid in their normal diet, so their bodies are unable to produce enough red blood cells. In other cases, the body may not be able to properly use the folic acid ingested. Folic acid deficiency anaemia occurs most often in infants and teenagers. Some important sources of folic acid are cheese, eggs, fish, green vegetables, meat, milk, and yeast. Smoking can also interfere with the body's ability to use folic acid.

### 2.3.4 VITAMIN B12 DEFICIENCY ANAEMIA.

Like folic acid, vitamin $B_{12}$ is used to make red blood cells. The vitamin is found in meat and vegetables. Some symptoms of vitamin $B_{12}$ deficiency anaemia are loss of muscle control, loss of feeling in the arms and legs, soreness of the tongue, and weight

loss. Vitamin $B_{12}$'s primary functions are in the formation of red blood cells and the maintenance of a healthy nervous system. $B_{12}$ is necessary for the rapid synthesis of DNA during cell division. This is especially important in tissues where cells are dividing rapidly, particularly the bone marrow tissues responsible for red blood cell formation. If $B_{12}$ deficiency occurs, DNA production is disrupted and abnormal cells called megaloblasts occur. This results in anaemia. Symptoms include excessive tiredness, breathlessness, listlessness, pallor, and poor resistance to infection. Other symptoms can include a smooth, sore tongue and menstrual disorders. Anaemia may also be due to folic acid deficiency, folic acid also being necessary for DNA synthesis.

The most common form of vitamin $B_{12}$ deficiency anaemia is called pernicious anaemia. People between the ages of fifty and sixty are at highest risk for pernicious anaemia, which is a form of anaemia in which the body is unable to absorbed vitamin $B_{12,}$ Pernicious anaemia is characterized by decrease gastric production of hydrochloric acid and deficiency of intrinsic factor, a substance normally secreted by the parietal cell of the gastric mucosa that's essential for vitamin B12 absorption. Some conditions that can lead to pernicious anaemia are eating disorders, anorexia nervosa and bulimia entries, poor nutrition, diabetes mellitus stomach problems, and thyroid disease.

**2.3.5 HAEMOLYTIC ANAEMIA.**

Hemolytic anaemia occurs when red blood cells are destroyed faster than they are made. In some cases, an infection can cause this problem. In other cases, the bodies own

immune system destroys the red blood cells. Some symptoms of hemolytic anemia include pain, shock, gallstones, an enlarged spleen, and other serious health problems.

### 2.3.6 AUTOIMMUNE HAEMOLYTIC ANAEMIA

An autoimmune disorder is one in which a person's immune system attacks its own body. The normal function of the immune system is to protect the body against foreign invaders, such as bacteria and viruses. Autoimmune hemolytic anaemia is an uncommon group of disorders that can occur at any age. These disorders affect women more often than men. About half of the time, the cause of autoimmune hemolytic anaemia cannot be determined (idiopathic autoimmune hemolytic anaemia). Autoimmune hemolytic anemia can also be caused by or occur with another disorder, such as systemic lupus erythematosus (lupus), and rarely it follows the use of certain drugs, such as penicillin.

Destruction of red blood cells by autoantibodies may occur suddenly, or it may develop gradually. In some people, the destruction may stop after a period of time. In other people, red blood cell destruction persists and becomes chronic. There are two main types of autoimmune hemolytic anaemia: warm antibody hemolytic anemia and cold antibody hemolytic anaemia. In the warm antibody type, the autoantibodies attach to and destroy red blood cells at temperatures equal to or in excess of normal body temperature. In the cold antibody type, the autoantibodies become most active and attack red blood cells only at temperatures well below normal body temperature.

Some people with autoimmune hemolytic anaemia may have no symptoms, especially when the destruction of red blood cells is mild and develops gradually. Others have

symptoms similar to those that occur with other types of anaemia, especially when the destruction is more severe or rapid. When severe or rapid destruction of red blood cells occurs, mild jaundice may also develop. When destruction persists for a few months or longer, the spleen may enlarge, resulting in a sense of abdominal fullness and, occasionally, discomfort.

When the cause of autoimmune hemolytic anaemia is another disease, symptoms of the underlying disorder, such as swollen and tender lymph nodes and fever, may dominate. But the immune system can sometimes become confused. It thinks that parts of the body are a foreign invader. In the case of autoimmune hemolytic anaemias, the immune system attacks red blood cells, killing them just as it would destroy bacteria or viruses.



### 2.3.7 THALASSEMIA.

Thalassemia is an inherited blood disease resulting from defection production of haemoglobin. Haemoglobin is a protein made up of four chains of amino acid, two identical alpha chains and two identical beta chains. Thalassemias are caused by an imbalance in the production of alpha and beta chains, an imbalance that is caused by a mutation or change in the genes that direct their production. The severity of the disease in patients depends on the degree of imbalance between the alpha and beta chains production. Patients with thalassemia develop jaundice, poor growth, fatigue, enlarged spleen and irritability. The lack of the production of haemoglobin will eventually leads to less oxygen being delivered to the various vital organs. Lack of oxygen to these organs may lead to the death of the patient.

### 2.3.8 SICKLE CELL ANAEMIA.

Sickle cell anaemia is a genetic disorder. Cells receive genes that give them the wrong instructions for making red blood cells. Red blood cells are normally shaped like plump doughnuts. People with Sickle Cell Anaemia have Sickle haemoglobin (HbS) which is different from the normal haemoglobin (HbA). When sickle haemoglobin gives up its oxygen to the tissues, it sticks together to form long rods inside the red blood cells making these cells rigid and sickle-shaped whiles the normal red blood cells are flexible.

Because of their shape, sickled red blood cells can't squeeze through small blood vessels as easily as the almost donut-shaped normal cells. This can lead to this small blood vessel getting blocked which then stops the oxygen from getting through to where it is needed. This in turn can lead to severe pain and damage to organs. This condition can cause pain, weakness, and, in extreme cases, death.

### 2.3.9 APLASTIC ANAEMIA

Aplastic anaemia is a serious form of anaemia that can lead to death. The body makes too few of all kinds of blood cells: red blood cells, white blood cells, and platelets, because the bone marrow fails to produce them. One well known cause is the white cell attacking the bone marrow.(autoimmune). Platelets are blood cells that help blood to clot. Aplastic anaemia may be caused by a recent severe illness, long-term exposure to industrial chemicals, and the use of certain types of medication.

## 2.4 GENERAL CAUSES OF ANAEMIA

### 2.4.1 Inadequate intake of iron and other nutrients

Thisresults largely from consuming too little iron in a form that the body can absorbed. Other nutrient deficiencies that contribute to anemia include vitamins A and C, folate, riboflavin, and B12.

### 2.4.2 Poor absorption of iron

Poor absorption of iron occurs because dietary components such as phytates in cereal foods bind to the little iron present in plant foods making much of it unavailable for absorption. As a result, iron taken into the body cannot be readily absorbed and used.

In the Western world, however, high sugar, fat, and cow's milk intakes are often responsible for low iron levels since all of them interfere with iron absorption. The American Academy of Pediatrics (AAP) recommends that children under the age of one year should not drink cow's milk because it interferes with iron absorption, causing anemia.(WHO, 2001) The AAP published the results of a University of Iowa study that found the blood content in the stool of infants fed cow's milk was five times higher than children fed other kinds of infant formula. Researchers also concluded that the amount of lost iron was "nutritionally important".

### 2.4.3 Malaria

Anaemia in malaria is caused by the destruction of red cell (haemolytic anaemia) in the body or the depression of red blood cell production in the bone marrow (WHO, 2001). As the red blood cell becomes less the patient weakens. In severe cases the patient is unable to deal with basic life essential task and may die.

### 2.4.4 PARASITIC WORMS

Most parasites require some host to complete their life cycle. These will vary in size from one-thousandth of a micron to whale tapeworm which is one hundred feet long. Parasite and worms can invade the bodies through food and water intake or through transmitting agents like mosquitoes. Once established they will eat the same food you eat or they will eat you. They are responsible for many health problems, they secret toxins and "steal" the vital nutrients from our bodies. Hookworms use their teeth to burrow through the intestinal walls and feed on the blood resulting in anaemia.

### 2.4.5 Infections, - both chronic and systemic (e.g., HIV/AIDS).

Multiple factors both diseases related and treatment related can cause anaemia during HIV infection. In this instance there is an attack on the cells and these caused immunosupression. There is both haemolytic and aplastic anaemia

### 2.4.6. MENSTRUATION AND BLEEDING.

In women, profuse menstruation (menorrhagia) and pregnancy are reasons for primary iron deficiency anemia. In males, the condition is uncommon and occurs mainly as a result of gastrointestinal bleeding, which may be the result of gastric or duodenal ulcers, hiatal hernia, esophageal varices, cirrhosis, haemorrhoids, ulcerative colitis, or carcinoma. A daily blood loss of two-four ml is enough to cause iron deficiency anemia. Other causes in both sexes include the use of medications that cause gastrointestinal bleeding; surgical procedures that decrease stomach acidity, intestinal transit time, and

absorption; insufficient dietary intake of iron; and such eating disorders as pica (the craving and eating of non-nutritional substances).

The relative contributions of these causes of anaemia vary in different countries. To reduce anemia prevalence in a population, it is important to select interventions that address the primary causes in that population, as listed above. Strong evidence links anaemia to health and development problems. Overall, about 20 percent of maternal and peri- natal mortality in developing countries can be attributed to anaemia (WHO, 2002). Recent work has shown that most of this impact is in the mild and moderate grades of anaemia, rather than being limited to severe anaemia. Anaemia in pregnant women results in lower birth weight babies who have higher risk of death. (Chestman *et al*.,1991).

## 2.5 EPIDEMIOLOGY

The World Health Organization (WHO) estimates that anaemia affects between one-quarter to one-third of the world's population or up to 2 billion people. Iron deficiency affects many more people. Most of the anaemic population lives in developing countries, where high anaemia prevalence is seen, particularly in pregnant women, young children, female adolescents, and women of childbearing age.

Overall, anaemia contributes to about 20 percent of maternal and peri natal deaths in developing countries. A recent WHO World Health Report noted that the risks of both maternal and peri natal mortality were reduced by 25% and 28%, respectively, for each gram increase in haemoglobin level between 50 and 120 g/L. This is contrary to the

previous generally accepted understanding that only severe anaemia resulted in death. This finding is very important because the numbers of women and children with mild and moderate anaemia are vastly greater than the number with severe anaemia. It follows then that the great majority of anaemia-related maternal and peri -natal deaths are due to mild and moderate anaemia rather than severe anaemia. Anaemia's other serious negative consequences include poor pregnancy out comes such as low birth weight and premature birth. Anaemia also has adverse implications for social and economic development. There is now strong evidence that anaemia can reduce cognitive development and limit a child's learning in schools. This will lower the effectiveness of investments in education. Anaemia's role in reducing physical capacity and work productivity in adults has been long established. Nevertheless, anaemia continues to have a relatively low priority in health policies and programs, compared to other nutrition-related health problems with more obvious life-threatening implications. Difficulties are posed by Anaemias multiple causes and by the still-limited programmatic experience in and insufficient evidence on effective intervention approaches and best practices to control anaemia.

These are major constraints for policy formulation and program development. A better understanding of the etiology or causes of anaemia and the identification of critical issues related to effective anaemia program design and implementation are key to developing more successful actions. Recent progress in understanding the nature of the problem and the achievements and limitations of existing programs provides a firm basis for designing effective Strategies and interventions. MOST, the USAID

Micronutrient Program has supported ministries of health in the Democratic Republic of the Congo, Ghana, Uganda, and Nicaragua to develop approaches to address anaemia. Anaemia may result from defects at any stage of red cell and haemoglobin production or when an increased rate of red cell destruction (haemolysis) exceeds the capacity of the bone marrow to mount a compensatory increase in production. Changes in the relationships between red cell and plasma volumes may also result in a reduced haemoglobin concentration: such changes occur physiologically in pregnancy where red cell volume is increased less markedly than plasma volume. All anaemia sufferers manifest signs and symptoms attributable to tissue and organ hypoxia and the ensuing reduced metabolism.



## 2.6 FOOD SOURCES RICH IN IRON.

There are two forms of dietary iron: heame and non-heame. Iron in meat, fish, and poultry is found in a chemical structure known as heame. Heame iron is absorbed very efficiently by the body. Iron in plants such as lentils and beans are arranged in a different chemical structure called non-heame iron. Non-heame iron is not as well absorbed as heame iron. Flours, cereals, and grain products that are enriched or fortified with iron are good dietary sources of non-heame iron the addition of iron to infant formulas, cereals, and grain products has been credited with improving the iron status of millions of infants, children, and women. The tables of selected food sources of heame and non-heame iron suggest many dietary sources of iron. Foods rich in iron and the highest source of iron usually come from animal origin for example liver, kidneys and meats.

The amount of iron in food that is absorbed and used by the body is influenced by the iron nutritional status of the individual and whether or not the iron is in the form of haeme. Haeme iron is absorbed better than nonhaeme iron, but most dietary iron is nonhaeme. Haeme iron comes mainly from haemoglobin and myoglobin in meat, poultry, and fish.



**2.7 HUMAN GROWTH**

The European Union RDA for the general population is set at 15 mg/day, however, some people need more iron than others, children between the ages of six months and six years need more. A full-term infant's iron stores are usually sufficient to last for six months. High iron requirements are due to the rapid growth rates sustained during this period.

Early adolescence is another period of rapid growth and in females, the blood loss that occurs with menstruation adds to the increased iron requirement of adolescence. In pregnant women increased iron utilization by the developing foetus and placenta, as well as blood volume expansion significantly, increases the iron requirement during pregnancy.

Individuals with chronic blood loss -Chronic bleeding or acute blood loss may result in iron deficiency. One millilitre (ml) of blood with a haemoglobin concentration of 150 grams/litre contains 0.5 mg of iron. Thus, chronic loss of very small amounts of blood may result in iron deficiency. A common cause of chronic blood loss and iron deficiency in developing countries is intestinal parasitic infection. Individuals who donate blood frequently, especially menstruating women, may need to increase their

iron intake to prevent deficiency because each 500 ml of blood donated contains between 200 and 250 mg of iron (Clarke, 1976).

## 2.8 IRON ABSORPTION

Iron absorption refers to the amount of dietary iron that your body obtains from food. Healthy adults absorb about 15% of the iron in their diet. The greatest influence on iron absorption is the amount stored in your body. Iron absorption significantly increases when body stores are low. When iron stores are high, absorption decreases to help protect against iron overload. Iron is mainly absorbed in the duodenum and upper jejunum (Novartis, 2008) Iron absorption is regulated by three mechanisms:

(i) Dietary regulator: a short-term increase in dietary iron is not absorbed as the mucosal cells have accumulated iron and "block" additional uptake.

(ii) Stores regulator: as body iron stores fall, the mucosa is signaled to moderately increase absorption.

(iii) Erythropoietic regulator: in response to anaemia the erythroid cells will signal the mucosa to increase iron absorption more significantly.

The absorption of iron is a complex process influenced by the intestinal mucosa and the amount of iron present in the diet. Vitamin C (ascorbic acid) enhances the absorption of irons while iron binding phenol compounds in tea and coffee inhibits its absorption. Not

all forms of iron are available for absorption. Generally, the animal sources appear superior to those of plant sources.

Since the average diet of a young woman seldom provide more than 6 mg-10 mg of iron daily and absorption is not more than (10-20) % of the food iron, many women enter pregnancies with inadequate stores (Clark, 1976).

To be absorbed, dietary iron must be in its ferrous $Fe^{2+}$ form. A ferric reductase enzyme on the enterocytes reduces ferric $Fe^{3+}$ to $Fe^{2+}$. A protein called divalent metal transporter 1 DMT1, which transports all kinds of divalent metals into the body, then transports the iron across the enterocyte's cell membrane and into the cell. These intestinal lining cells can then either store the iron as ferritin (in which case the iron will leave the body when the cell dies and is sloughed off into feces) or the cell can move it into the body, using a protein called ferroportin. The body regulates iron levels by regulating each of these steps. Our bodies' rates of iron absorption appear to respond to a variety of interdependent factors, including total iron stores, the extent to which the bone marrow is producing new red blood cells, the concentration of haemoglobin in the blood, and the oxygen content of the blood. We also absorb less iron during times of inflammation. Recent discoveries demonstrate that hepcidin regulation of ferroportin is responsible for the syndrome of anaemia of chronic disease.

While DMT1 is unique to iron transport across the duodenum, ferroportin is distributed throughout the body on all cells which store iron. Thus, regulation of ferroportin is the body's main way of regulating the amount of iron in circulation.

Hephaestin, a ferroxidase that which can oxidize $Fe^{2+}$ to $Fe^{3+}$ and is found mainly in the small intestine, helps ferroportin transfer iron across the basolateral end of the intestine cells.

The composition of the diet may also influence iron absorption. Citrate and ascorbate (in citrus fruits, for example) can form complexes with iron that increase absorption, while tannates in tea can decrease absorption. The iron in heame found in meat is more readily absorbed than inorganic iron by an unknown mechanism. The ferrous form of iron is more readily absorbed than ferric iron. Duodenal microvilli contain ferric reductase to promote absorption of ferrous iron. Only a small fraction of the body's iron is gained or lost each day. Most of the iron in the body is recycled when old red blood cells are taken out of circulation and destroyed, with their iron scavenged by macrophages in the mononuclear phagocyte system, and returned to the storage pool for re-use. Iron homeostasis is closely regulated via intestinal absorption. Increased absorption is signalled by decreasing iron stores, hypoxia, and erythropoietic activity. (Baksh *et al*., 1999).

## 2.9 STIMULATORS OF IRON ABSORPTION

The following food components have been found to stimulate the absorption of iron**:**

Vitamin A – vitamin A deficiency may increase iron deficiency anaemia. The combination of vitamin a and iron seems to improve anaemia more effectively than either iron or vitamin a alone.

Copper – adequate copper nutritional status appears to be necessary for normal iron metabolism and red blood cell formation. Anaemia is a clinical sign of copper deficiency.

Meat proteins – meat proteins improves the absorption of nonhaeme iron.

Vitamin C – vitamin c improves the absorption of nonhaeme iron.



## 2.10 INHIBITORS OF IRON ABSORBTION

Tea, cocoa, herbal infusion and some spices example oregano are some of the inhibitors of iron absorption, calcium particularly from milk and milk products (Lemson, 1992). In addition to the lack of quality foods, blood loss from parasites is the leading cause of iron-deficiency anemia in most areas of the world. Inadequate dietary intakes of iron are seen most often in premenopausal women, infants and children (Lemson, 1992). The heart is affected when there is a lack of oxygen in the body. The heart has to work harder to get enough oxygen throughout the body. Over time, this stress on the heart can lead to a fast or irregular heartbeat, chest pain, an enlarged heart, and even heart failure.

## 2.11 Storage of iron

**Table 1.2 Table of Total Iron found in Adults**

| | Men(mg) | Women(mg) |
|---|---|---|
| **Haemoglobin** | 2100 | 1750 |
| **Myoglobin** | 300 | 250 |
| **Enzymes** | 50 | 50 |
| **Storage Ferritin and Haemosiderin** | 1000 | 400 |
| **Total** | 3450 | 2450 |

Source: Contran *et al*. (1999).

Ferritin and Haemosiderin are specially designed protein for the reversible storage of iron Haemosiderin is a kind of polymer of ferritin micelles which is metabolically less active but serves the same purpose as ferritin in the storage of iron. Ferritin stores about 4500 Fe atoms (Garrow, 1993). Iron is initially stored as ferritin, but ferritin can be incorporated by phagolysosomes to heamosiderin. There are about 2 gm of iron in the adult female and up to 6 gm iron in the adult male. About 1.5 to 2 gm of this total is found in red blood cells as heame in heamoglobin and 0.5 to 1 gm occur as storage iron, with the remainder in myoglobin and in enzymes that require iron. A total of 4g of iron is found in the normal human body of which 70% is found in the haemoglobin and the remainder bound to a protein called ferritin found in the reticuloendothelial cells (mainly in the liver), (Garrow, 1993).

In the blood plasma, iron is transported in the form of ferric iron ($Fe^{3+}$) bound to its carrier protein, transferrin (a globulin). The plasma constitutes a total of 5mg iron being

iron in transient from the reticuloendothelial cells where erythrocyte and the haemoglobin are broken down to the blood forming tissue of the bone marrow, where the ion is used again for new haemoglobin synthesis. In all 25mg of iron is produced per day by haemoglobin breakdown and re-used for erythropoiesis.

Transferrin is determined as the Total Iron-Binding Capacity (TIBC) of the plasma expressed µmol iron/l, where the degree of saturation of the transferring with iron is calculated by dividing iron concentration by TIBC. Approximately 3 milligrams or 0.1 % of about 3g iron in the adult male circulates in the plasma as an exchangeable pool. Essentially all circulating plasma iron normally is bound to transferrin. This chellation serves three purposes: it renders iron soluble under physiologic conditions, it prevents iron-mediated free radical toxicity, and it facilitates transport of iron into cells. Transferrin is the most important physiological source of iron for red cells (Chesterman *et al.,* 1990). The liver synthesizes transferrin and secretes it into the plasma. Transferrin is produced locally in the testes and the Central Nervous System. These two sites are relatively inaccessible to proteins in the general circulation (blood: testis barrier, blood brain barrier). The locally synthesized transferrin could play a role in iron metabolism in these tissues. Plasma transferrin is an 80 kDa glycoprotein with homologous N-terminal and C-terminal iron-binding domains.

The simplest tests that indirectly give an indication of iron stores are the serum iron and iron binding capacity, with calculation of the percent transferrin saturation. The serum ferritin correlates well with iron stores, but it can also be elevated with liver disease, inflammatory conditions, and malignant neoplasms. The CBC will also give an indirect measure of iron stores, because the mean corpuscular volume (MCV) can be decreased

with iron deficiency. Iron administered into the body is absorbed into the plasma. This iron is taken from the plasma by transferrin into iron stores. In the process of red cell formation, transferrin carries iron from the stores into the bone marrow for the synthesis of red blood cells.

## 2.12 IRON DEFICIENCY AND MATERNAL ANAEMIA

Pregnancy is a physiological state during which a lot of physical and physiological adaptive changes take place within the female body, as a result of the developing foetus. The most important of the changes is the change in blood picture. As a result of the anaemia tends to be the commonest medical disorder in pregnancy especially in developing countries with iron deficiency anaemia accounting for the majority of the cases. In pregnant women, iron-deficiency anaemia can increase the risk of a premature delivery and a low-birth-weight baby (Pelletier, 1993).

A woman who is not pregnant needs 15 milligrams of iron a day while a pregnant woman needs 30 milligrams of iron a day. Women need twice as much iron as normal during pregnancy. In order to supply this, about 50 per cent of all women need an iron supplement during their pregnancy. This is usually taken with food from the 20th week of pregnancy, and can take the form of pills or a natural herbal product. Iron supplements can cause stomach pains, nausea or vomiting and slow down digestion. This can be avoided by taking iron supplements just before going to bed or eating a fibre-rich diet.

During pregnancy, the needs of the growing fetus and placenta, as well as the increasing maternal blood volume and red cell mass, impose such a demand on maternal iron stores that iron supplementation at daily doses between 18 and 100 mg from 16 weeks gestation onwards could not completely prevent the depletion of maternal iron stores at term (Thomsen *et al.*, 1993). The development of iron deficiency anaemia is associated with increased risk of preterm births and low birth weight infants (Ulmer *et al.*, 1992). Furthermore, mothers given iron supplementation had decreased risk of preterm delivery compared with mothers without supplements (Ulmer. *et al.*, 1992). Thus the relationship between maternal iron deficiency and preterm birth and fetal growth restriction seems to be well established. In the developing world, rates are as high as 700 per100, 000 live births in many parts of Africa and in some countries in south Asia. These large differences in risk are related primarily to differences in available obstetric care for women living in areas with inadequate antenatal and delivery care facilities.

Harrison (1989) has championed the arguments for developing improved pregnancy care to reduce maternal mortality in developing countries. In reports from Nigeria, he has highlighted the importance of maternal anaemia as a contributory factor to maternal death (Harrison, 1975; Harrison and Rossiter, 1985). In 1987, international agencies and leaders from 45 countries established the Safe Motherhood initiative with the goal of reducing half of maternal deaths by the year 2000 (World Bank, 1993).

A key component of Safe Motherhood is the eradication of anaemia during pregnancy. The relationship of anaemia as a risk factor for mortality is derived mainly from cross-sectional studies and can be confounded for several reasons. Most studies report hospital data, often for moribund women, and there is limited attention to factors such

as pregnancy hemodilution, hemoglobin rise in late pregnancy, concurrent infection, hemorrhage, prior treatment or poor maternal nutritional status.

In young women living under endemic malaria conditions, especially in urban areas in which adults may have poor malaria immunity, severe malaria anaemia and cerebral malaria may occur and can rapidly lead to death (Granje *et al*., 1998). For these reasons, most studies form an inadequate basis for determining how anaemia relates causally to maternal survival in communities, and extrapolation from hospital delivery data must be considered an approximation that may be misleading. Intervention studies with maternal mortality as an outcome measure are required to determine causality, but these are very difficult to conduct for both ethical and logistic reasons. For example, there are very few studies that did not use transfusion as an emergency procedure in severely anemic women at term (Fullerton and Turner, 1962).

Iron deficiency and subsequent iron deficiency anaemia are the most prevalent nutrient deficiency problems afflicting pregnant women (Allen, 1997). Iron deficiency may develop during pregnancy because of the increased iron requirements to supply the expanding blood volume of the mother and the rapidly growing foetus and placenta. The net additional iron requirements during pregnancy are estimated to be 1000 mg (British Nutrition Foundation, 1995).

There are generally no changes in dietary intakes of iron during pregnancy yet there are extensive changes in maternal iron metabolism to ensure effective uptake from food and delivery to the foetus. Also, the use of iron stores at this time is suggested by lower serum ferritin levels in pre-menopausal multiparous women compared to nullipara and postmenopausal women (Milman *et al.*, 1992; White *et al.*, 1993). However, in the

context of sub-Saharan Africa women may enter pregnancy with seriously depleted iron stores (Lamparelli *et al.*, 1988).

When pre-pregnancy iron stores are low the amount of iron required during the last half of pregnancy cannot easily be met by diet and the risk of iron deficiency anaemia will be high, especially toward the end of pregnancy (Taylor *et al.*, 1982). The extent to which iron deficiency alone affects maternal and neonatal health is uncertain.

Interpretation of studies on the effect of iron deficiency on the mother, foetus or child is difficult because many do not consider gestation, parity, physiology of the woman, or socio-economic and nutritional confounders (British Nutrition Foundation, 1995; Allen, 1997). However, existing data suggest that maternal iron deficiency anaemia is associated with adverse outcomes, including abortion (Sirota *et al.*, 1989), pre-term delivery and maternal mortality (Allen, 1997). Currently iron-deficiency anaemia is ranked as the third leading cause of loss of disability-adjusted life years (DALYs) for women aged 15-44 worldwide (Murray and Lopez, 1996). Iron absorption during pregnancy is determined by factors such as the amount of iron in the diet, the bioavailability of the iron and the changes in iron absorption.

In the first trimester (the first 3months of pregnancy), the rate of absorption of iron is lower and this is related to the reduction in iron requirement during this period compared to the non pregnant woman (Clark and Affonso, 1976). The increase in iron requirement during pregnancy is directly proportional to the increase need for oxygen transport during pregnancy. There is an exponential growth of the foetus at the early stages implying that iron need is almost negligible in the first trimester.

In the second trimester, iron absorption is increased by about 50 % (Clark and Affonso, 1976). During the last trimester, it may increase by up to four times. The concentration of iron in the blood usually decreases from the normal level of about 150 μg% to about (80-90) % μg during the second half of pregnancy (Garrow and James, 1993).

## 2.13 MALARIA INFECTION

Malaria is a mosquito-borne protozoan disease, caused in Man by four species of the genus *Plasmodium: P. falciparum* (the most pathogenic and accounting for the majority of infections), *P. malariae, P. ovale* and *P. vivax.* More than 2000 million people live in areas where malaria transmission occurs and mainly children and pregnant women are affected. Malaria is widely distributed in the tropical and subtropical zones being endemic throughout South and South East Asia, Africa, areas of the Middle East and south and Central America (Cowan, 1993). In Africa alone it is estimated that 300-500 million clinical cases of malaria occur annually resulting in 1-2 million deaths (Breman, 2001), the majority occurring in African children – approximately 1 million annually (Snow *et al.*, 1999).

### 2.13.1 MALARIA IN PREGNANT WOMEN

Malaria infection during pregnancy is a major public health problem in tropical and subtropical regions through the world (Sanchez *et al.*, 2003). It presents a special risk for both the pregnant woman and her baby. In non immune women clinical attacks may be severe and the woman may be particularly prone to pulmonary oedema and hypoglycemia (Whitfield, 2004). In most endemic areas especially Africa, pregnant

women are the main adult risk group (Caroline, 2004; Sanchez, 2003). The effect of malaria on pregnancy is dependent on the malaria epidemiology and the immunity of the women. In areas of low malaria transmission where women have little immunity, the women will have acute often more severe symptomatic malaria especially during epidemics. In high of high transmission, pregnant women especially those with first pregnancy and teenage pregnancy and to a lower extent, second pregnancy are more likely to be infected by malaria than non pregnant women. They are also more likely to be anaemic (Bell, 1990).

Compared with non-pregnant women, pregnant women are at increased risk of malaria infection and its disease consequences in settings of both low and high transmission of malaria (Brabin 1991; Menendez 1995). This is probably due to hormonal modulation of the immune response during pregnancy and is heavily compounded by increased blood volume and sequestration of the parasites in the placenta (Riley *et al.*, 1989; Rasheed *et al*,. 1993; Menendez, 1995).

The Increased risk of malaria is not evenly distributed throughout parities, affecting primarily primigravidae in sub-Saharan Africa, not throughout each pregnancy as the prevalence of infection and parasite density is generally highest in the first half of the pregnancy and decreases progressively until delivery (Nosten, 1991; Brabin, 1991).

In hypo-endemic settings with low or unstable levels of malaria transmission, adults have relatively little acquired immunity, and people at all ages are at risk of severe disease if infected. Here, pregnant women of all parities are at 2-3 times greater risk of developing severe disease than non-pregnant women and at approximately 3 times greater risk of dying if they do develop severe disease (Luxemburger *et al*,. 1997). The

symptoms and the complications of malaria during pregnancy differ with the intensity of malaria infection and the level of immunity acquired by the pregnant women (Caroline *et al*., 2004)

Reports of adverse pregnancy outcomes associated with *P. falciparum* infection have included cerebral malaria, severe haemolytic anaemia, and a high risk of miscarriage, premature delivery or neonatal death (Menon, 1972; Herd and Jordan, 1981; Sholapurkar *et al.*, 1988; Meek 1988). In contrast, in much of sub-Saharan Africa, stable transmission of *P. falciparum* malaria is the rule. In these areas, women of childbearing age have a relatively high level of acquired ant malarial immunity. When pregnant, these women who are anaemic also demonstrate an increased susceptibility to *P. falciparum* manifested by a higher frequency and density of parasitaemia compared with non-pregnant women (Brabin, 1983; McGregor, 1984). This susceptibility appears to wane with subsequent pregnancies (McGregor, 1984; Brabin, 1991), although some studies from highly endemic areas have reported an increased susceptibility to malaria in grand multigravidae, that is, women with more than 5-7 previous pregnancies, compared to other multigravidae (Watkinson and Rushton, 1983).

Further, research from Senegal has suggested that, after controlling for use of anti malarial drugs, the pregnancy-associated increase in susceptibility to malaria persists for 60 days after delivery among women who live in areas where malaria is highly endemic (Diagne *et al.*, 2000). Despite severe disease being uncommon in pregnancy in endemic areas, *P.falciparum* infection during pregnancy is a major public health problem and the increased risk of parasitaemia has adverse effects for both mother and child. It leads to parasite sequestration in the maternal placental vascular space, with

consequent infant low birth weight due to both prematurity and intra-uterine growth retardation (McGregor, 1984; Brabin, 1991).

It has been estimated that the fatality rate of newborns with malaria-related low birth weight is 37.5% which, when adjusting for the current birth rate in sub-Saharan Africa translates as 3-17 deaths per 1,000 live births from malaria-induced LBW annually (Murphy and Breman, 2001). In addition, *P. falciparum* is an important, and often underestimated, cause of severe maternal anaemia (Gilles *et al.*, 1969; Shulman *et al.*, 1996). It has been estimated that as many as 400,000 cases of severe anaemia in pregnancy were caused by malaria infection in sub-Saharan Africa in 1995 (Guyatt and Snow, 2001).

Maternal death may result either directly or indirectly from malaria related severe anaemia. In addition, malaria may result in a range of outcomes including low birth weight, spontaneous abortion and neonatal death (Shulman, 2003) Maternal mortality continues to be a major health problem in the developing world. Nearly 600,000 women die each year as a result of complications of pregnancy and childbirth; most of these deaths could be prevented with attainable resources and skills (WHO, 1996). The worldwide maternal mortality ratio (annual number of deaths of women from pregnancy related causes per 100,000 live births) is estimated to be 390 per 100,000 live births (Abousahr and Royston, 1991). Most of these occur in developing countries, where women have a risk of dying in pregnancy and childbirth that is 50–100 times greater than that of women in the developed world (Starrs, 1987). The malaria parasites in the anaemic condition of the pregnant women increases the risk of low birth which is a factor related to poor health of the new born infant (Robb, 2000).

## 2.14 HAEMATOLOGICAL STATUS AND IRON NUTRITION.

Mild maternal iron deficiency and anemia have little significant repercussions on the hematological status of newborns. It has been suggested that placental transferring receptors protect the fetus. However, it appears that the capacity of iron to transfer from placenta to foetus is limited by a threshold mechanism so that fetal iron deficiency exists in severe maternal iron deficiency and anaemia. Also, the fetus of iron deficient mothers accumulates less iron reserves, and has smaller hemoglobin mass than their normal counterparts. This has been termed "hidden iron deficit" and is further magnified by low birth weight, mainly due to preterm delivery (Beard, 1994). Further evidence of "hidden iron deficiency" at birth comes from studies that showed that maternal and cord serum ferritin levels were lower in the presence of maternal iron deficiency and that this difference with children from non-anaemic mothers was magnified when the children were again studied at 2 months of age. This may account for the well documented higher prevalence of iron deficiency and anaemia in late infancy among populations where anaemia of pregnancy is highly prevalent. This situation increases the risk of long term and even permanent impairments in mental and physical development among such children (Beard, 1994).

Over half of the world's population is 25 years old and .80% of the world's youth live in developing countries. In the mid-1990s, the global teenage population was estimated at 513 million. In this group of adolescents (10–19 y), the WHO has estimated that anaemia prevalence (Hb ,110 g/L) is 16% in less-developed countries but 45% in Africa (DeMaeyer and Adiels-Tegman, 1985). The risk of anaemia is high in teenage primigravidae in developing (Arkutu 1979, Barr *et al.*, 1998, Fazio-Tirrozo *et al*., 1998)

and developed countries (Beard 1994, Osbourne *et al*., 1981). Maternal deaths in a community study using verbal autopsy in Tanzania showed no association with maternal age (Macleod and Rhode, 1998). These authors did not examine whether maternal deaths related to anaemia were more common in adolescents. In a large hospital-based study in Northern Nigeria, a higher maternal mortality from severe anaemia (43%) was compared in very young (15years) adolescent, older adolescent and non adolescent pregnant women (10%) (Harrison, 1989). Lawson and Lister (1954) in an early Nigerian study of 188 moderately anemic women (Hb 70 g/L) observed a case fatality of 1.89% in adolescent pregnancies compared with 8.89% in non adolescent women (x2 5 2.9, *P* , 0.1). Only 3 of the 53 adolescents were 16 years old.

**2.15 Role of Hookworm in anaemia during pregnancy.**

Hookworm infection as a recognized factor in iron deficiency anaemia is shown as microcytic hypochromia (Schade and Warren, 1990). Fleming (1989) did a study on the etiology of severe anaemia in pregnancy in Ndola Zambia, and found that iron deficiency anaemia was mainly nutritional problem but hookworm infection was a contributory factor in about one third of the patients. Hookworm as a risk in anaemia in pregnancy was studied by Shulman *et al*. (1996) in Kenya, they found out that in multigravidae iron deficiency and hookworm infections were important factors to consider in anaemia in pregnancy

Anathakishnan *et al*. (1997) in a study in India also found that 90% of the pregnant were anaemic as they lived in hookworm endemic areas. To show the effect of hookworm infection on iron metabolism, Compton and Whitehead (1993) stated that

hookworm contributes significantly to iron deficiency anaemia, and that anaemia may be precipitated by even a small burden of hookworm. This is supported by Roche and Lagrisse (1996) that hookworm burden was related to the risk of iron deficiency anaemia however the precise function of this relation is unknown and depends on host status particularly iron balance in terms of diet, state of iron resources and the intensity and duration infection (Cromption and Stephenson, 1990).

In hookworm infection the loss of red cell in the gut in proportional to the worm burden as indicated above, and it is said that N americanus caused a loss between 0.03-0.05mls of blood per worm per day and between 0.16 -0-34 mls in the case of A duodenale (WHO, 1981). The relation between hookworm related blood loss and iron deficiency anaemia was also proven by Stolzfus *et al*. (1996) in which they found out that the intestinal blood loss was strongly related and linearly related to the hookworm egg count and the degree of degradation of the faecal haem, indicating that blood loss has occurred in the upper gastro intestinal tract. Hookworm infection has long been recognized among the major causes of anaemia in poor communities but understanding of the benefits of the management of hookworm infection in pregnancy has lagged behind the other major causes of maternal anaemia.

In a study by Bruce and Tagoe (1997) they found out that hookworm infection and high parity had little effect on the haemoglobin levels. They also found lower haemoglobin levels in women with lower parity and they attributed this to the fact that women of high parity are likely to feed well.

An epidemiological study in 1995 by the WHO highlighted the paradox presented to public health workers that an estimated one-third of all pregnant women in developing

countries were infected with hookworm and yet, in the absence of safety data, the appropriate advice then was to avoid the use of anthelmintics in pregnancy. Furthermore, the lack of an acceptable intervention constrained the development of evidence-based understanding of the impact of hookworm infection on maternal anaemia. These issues were addressed directly by de Silva and colleagues, who analyzed the safety profile of some 20 years of mebendazole use in antenatal clinics in Sri Lanka.



In 2002, WHO published new guidance indicating that pregnant women should be treated for hookworm infection, ideally after the first trimester. This immediately provided the opportunity for improved service delivery, and also encouraged studies to assess the contribution of hookworm to anaemia in pregnancy and the impact of treatment, some of which have been undertaken since 2002. These provide a rich new source of data to help inform public health decision making, and a systematic review of hookworm as a risk factor for anaemia among pregnant women.

Prevalence of infection increases with age in children, typically reaching maximum levels at 15-20 years and leveling off in adulthood (Bundy, 1990). Infection intensity follows a similar pattern of increasing with age, but does not necessarily level off during adulthood, often continuing to increase in older adults. Treatment with mebendazole in a single 500-mg dose is effective in substantially reducing the intensity and prevalence of infection, is safe in pregnancy (beyond the first trimester) and during lactation (WHO, 1995). Where anaemia is prevalent, and hookworm prevalence is endemic (>20-30%) WHO recommend that “hookworm control be included in strategies designed to improve the health, development and nutritional status of girls

and women." Hookworm disease as a major cause of iron deficiency anaemia is related to the worm burden and faecal egg count (Roche and Layrisse, 1966; Stoltzfus *et al.*, 1997a) with a wide range of intensities, equivalent to burdens of 40 to 160 worms, being associated with haemoglobin levels below 11g/dL in pregnant women (Lwambo *et al.*, 1992).



## 2.16 SICKLE CELL DISEASE AND IRON DEFICIENCY ANAEMIA

Sickle cell anaemia (SCA) is an inherited disorder of hemoglobin synthesis that is characterized by life-long severe hemolytic anaemia, attacks of pain crisis, and chronic organ system damage. A third of the haemolysis in SCA is intravascular and the resulting urinary losses of iron may lead to iron deficiency. There is no evidence of iron overload in SCA and iron deficiency may be more common than suspected, especially in men. Absence of bone marrow iron remains a gold standard for the diagnosis of iron deficiency in these patients. Although low serum ferritin is highly specific for the diagnosis of iron deficiency, its sensitivity is quite low in SCA because of non-specific elevation due to increased red cell turnover. The kinetics of sickling is strongly concentration dependent such that small decreases in the mean corpuscular deoxyhemoglobin-S concentration (MCHC-S) cause a substantial delay in sickle hemoglobin polymerization. Prolongation of the "delay time of gelation" in excess of the capillary transit time may allow the erythrocyte to traverse the capillary bed to escape to the arterial side before there is rheologic impairment of the erythrocyte from polymerization of sickle hemoglobin. Overt iron deficiency lowers the MCHC-S and thereby decreases the sickling tendency and the severity of haemolysis. The clinical

improvement in SCA following the induction of iron deficient erythropoiesis by repeated phlebotomies or by erythrocytapheresis has been reported. Prospective controlled studies are needed to evaluate further, the therapeutic strategy of inducing controlled iron deficient erythropoiesis in selected patients with SCA (Wile, 2003).

# CHAPTER THREE

## 3.0 MATERIALS AND METHODOLOGY

### 3.1 STUDY AREA

The study was conducted in Peri-urban communities (Pramso and South Suntreso) of Kumasi, located in the Ashanti region. The region has a tropical climate characterized by moderate temperatures all year round. The average minimum temperature is about 21$^{o}$C and a maximum average temperature of 30.7$^{o}$C. The average humidity is about 84.16% in the morning and 60% in the evening. There is a maximum rainfall regime 214.3mm in June and 165.2mm in September.

The Bosomtwe District, located at the central portion of the region is one of the newly created 26 districts in the Ashanti Region, having been carved out of the former Ejisu-Juaben -Bosomtwe District. The District is bounded on the North by Atwima Nwabiagya and Kumasi Metropolis and on the East by Ejisu-Juaben District. The southern section is bounded by Amansie West and East Districts. Kuntanase is the District Capital.

Slightly over half, 51.5 per cent, of the population of the region reside in four districts. While more than half of the population in the region resides in urban areas, in 15 of the 18 districts, over half of the population lives in the rural areas of the region. The high level of urbanization in the region is due mainly to the high concentration of the population in the Kumasi metropolis (which has almost about a third of the region's population).

The Kumasi metropolis is the most populous district in the Ashanti Region. During the 2000 Population Census it recorded a figure of 1,170,270. It has been projected to have a population of 1,625,180 in 2006 based on a growth rate of 5.4% per –annum ( p.a) and this accounts for just under a third (32.4%) of the region's population.

### 3.2 SAMPLING SITES

The sampling points from where the participants were recruited, were the Suntreso South Government hospital and Michael's hospital. The St. Michaels hospital is a mission hospital, located in Pramso within the Bosomtwe District. This hospital serves several communities around it including Aboatam, Feyiesi, Esreso, Atiwa etc. the majority of people living in this area are petty traders and subsistence farmers.

The Suntreso Government hospital is in the Kumasi metropolis under the Bantama sub-metro. It is found close to the boundary between Bantama and North Suntreso. The Suntreso Government hospital is the bigger of the two hospitals. This hospital serves the local populace as well as patients from neighboring suburbs such as Kwadaso, Patasi Estate, and Asuoyeboa, Abrepo, Suame among others. The residents are mostly public servants, traders and artisans.

### 3.3 SELECTION OF PARTICIPANTS

The study design was a cross sectional study restricted to pregnant women attending antenatal clinic at the hospitals. Pregnant women attending antenatal clinics in both hospitals for routine check-up (which includes, body temperature, pressure and urine examination) were recruited to participate in this project work. Informed consent was

sought from the participants. All the participants were registered members of the Department of Community Medicine in the two hospitals.

### 3.4 ETHICS.

Ethical approval was received from the appropriate authorities before the study was undertaken. Consent was sought from pregnant women attending antenatal at the selected hospitals by explaining the concept of study to them. Those who gave their consent were recruited to participate in the study.

### 3.5 SAMPLE COLLECTION

Stool and blood samples were collected for examination. Stool samples of the pregnant women were collected into small sterile sample containers and sent to the laboratory for analysis.

Venous blood (5 ml) was taken from the pregnant women into pre-labeled tubes based on patient's identification number. A portion of the blood was put into a tube containing EDTA and the rest into a test tube to be centrifuged and analyzed for serum ferritin in the lab. Part of the sample that was to be used the ferritin analysis was stored in the hospital deep freezer at -20$^{o}$C.

Samples taken were investigated for malaria parasitaemia, ferritin levels and haemoglobin concentrations.

### 3.6 QUESTIONNAIRE ADMINISTRATION

The pregnant women were interviewed using pre-structured, pre-tested questionnaires. The questionnaire was administered in either the local dialect (Twi) or English to those who gave their consent. The questionnaire were generally aimed at addressing various parameters such as age, education background, knowledge of iron deficiency anaemia, type of food normally eaten, usage of bed net and other parameters.



### 3.7 LABORATORY ANALYSIS.

Laboratory investigations were carried out to determine malaria parasitaemia, sickling status haemoglobin, ferritin levels in the sera of the participants. Stool examination was also carried out for the determination of the type of intestinal parasites.

#### 3.7.1 MEASUREMENT OF HAEMOGLOBIN

Heamoglobin measurement was carried out using a photometer. The photometer was turned on and allowed to warm up for 10 minutes.

A previously taken blood sample of 20 µl (well mixed with EDTA) was pipetted into a 4 ml Drabkins reagent (coral clinical), well mixed and allowed to stand for at least or at least 10 minutes. This was transferred into the cuvette of the photometer (colorlite 850). The absorbance was read at 540 nm against a reagent blank.

### 3.7.2 PREPARATION OF BLOOD FILM/ STAINING FOR MICROSCOPY.

Microscopy is the main diagnostic tool by which the various parasitic stages of *plasmodium* in blood films are identified. Drop of the blood collected is placed at the centre of a sterilized glass slide and evenly spread using a spreader. The film is then air dry. Three percent Giemsa stain was used in staining the blood film for a period between 10 to 20 minutes. The slides were then washed with distilled water and left to dry at room temperature. Drops of immersion oil was placed on the blood film and observed under the oil immersion lens of the microscope for the absence or presence of diagnostic stages of the malaria parasite.

### 3.7.4 FERRITIN MEASUREMENT

The test was conducted using a Microparticle Enzyme Immunoassay (MEIA) technology which uses a solution of suspended, submicron sized latex particles to measure analytes – in this case, Ferritin. The particles are coated with a capture molecule that is specific for Ferritin. Before the start of the automated procedure, each sample was firstly centrifuged (BS400 Denly Centrifuge) at 1000 rpm for five minutes. The purpose of the centrifugation was to separate the serum from the plasma, after which the serum was pipetted into a 5mL sample tube, and lined up at the sampling center of the machine (Abbott AxSYM system). The rest of the procedure is automated, and it is described below as it is delineated in the Abbott AxSYM System operations manual (1996):The sample and microparticles were combined and incubated to reaction temperature. During this incubation period, the analytes (Ferritin) bound to the microparticles creating an immune complex.

The processing probe then aspirated the reaction mixture from the incubation well of the reaction vessel and dispensed it onto the matrix cell. The immune complex then bound irreversibly to the glass fibers matrix. After a matrix cell wash removed the unbound materials, the immune complex was retained by the glass fibers while the excess reaction mixture flowed rapidly through the large pores in the matrix.

The processing probe then transferred alkaline phosphatase-labeled conjugate from a reagent well in the reaction vessel to the matrix cell. The conjugate then bound to the immune complex thereby completing the antibody-ferritin-conjugate complex, after which the matrix was washed again.

The bulk solution 1 dispenser then added the substrate 4-methylumbelliferyl phosphate (MUP) to the matrix cell. The alkaline phosphatase conjugate then catalyzed the hydrolysis of MUP to 4-methylumbelliferone (MU).

The MEIA optic then measured the rate at which MU, which is a fluorescent product, was generated on the glass fiber matrix. The rate at which MU is generated on the matrix is proportional to the concentration of the Serum Ferritin in the test sample. After internalized calculations, the Serum Ferritin level of each sample was then displayed on an attached monitor, and printed out. The results were then compared with the age-adjusted standards reported by Jacobs *et al* (1989).

### 3.7.5 STOOL EXAMINATION

About 5mls of normal saline solution was added to a small portion of the stool (about 0.2g) and mixed well. A drop of the mixture was then put on a glass slide and covered with a cover slip. It was then observed under the microscope (X10) for parasitic ova or larvae in the preparation using the WHO bench aid (2003) as a reference.



### 3.8 SURVEY OF THE ENVIRONMENT

The communities were surveyed for possible mosquito breeding sites such as refuse dump, choked gutters, pools, puddles, and for receptacles such as cans and vehicle tyres etc. Water samples were taken from these sites in February 2011 and examined in the laboratory for the presence of mosquito larvae and pupae. The larvae and pupae were held in cages until adults emerged. The adult were identified to determine the exact types and species of mosquitoes present.

### 3.9 IDENTIFICATION OF MOSQUITOES

Mosquitoes that emerged from the water samples collected from the study communities were viewed under a light dissecting microscope and identified using special identification keys (WHO, 2004).

# CHAPTER FOUR

## 4.0 RESULTS

A total of three hundred pregnant women at different stages of pregnancy attending antenatal clinics at the South Suntreso and St Michael's Hospitals were randomly selected for the cross sectional study. Of the 300 selected women, 55 (18.3%) were in their first trimester of their pregnancy, one hundred and sixty six (55.4%) were in the second trimester, while 79 (26.3%) were in the third or last trimester of their pregnancy.



## 4.1DERMOGRAFIC INFORMATION OF THE STUDY POPULATION

### 4.1.1 Age distribution of the pregnant women sampled

The wome are distributed in the various age categories as shown on Table 4.1 The age group 25-29 recorded the highest with 102 and this was followed by 20-24 with 76. The least was number was recorded by 40-44. The rest can be seen on the table.

### 4.1.2 Educational background of pregnant women:

With respect to education, 8.4% of the pregnant women sampled had no formal education, while 71.3% had acquired basic education. Forty four of them (14.8%) had acquired secondary education whereas 5.5% had tertiary education. Irrespective of level of education, 4.4% of the women surveyed had an idea about Iron deficiency anaemia. Most pregnant women surveyed had some knowledge about the drugs supplied to them, when they visit the antenatal clinic in the hospitals (Table 4.1).

**Table 4.1 Background information of pregnant women**

| CHARACTERISTICS | CATEGORY | SOUTH-SUNTRESO (%) | PRAMSO (%) | TOTAL | TOTAL PERCENT-AGE (%) |
|---|---|---|---|---|---|
| Stage of pregnancy | 1st trimester | 25 | 30 | 55 | 18.3 |
| | 2nd trimester | 82 | 84 | 166 | 55.4 |
| | 3rd trimester | 43 | 36 | 79 | 26.3 |
| Age group | 15-19 | 9 | 9 | 18 | 5.9 |
| | 20-24 | 34 | 42 | 76 | 25.3 |
| | 25-29 | 56 | 46 | 102 | 34.1 |
| | 30-34 | 30 | 26 | 56 | 18.7 |
| | 35-39 | 19 | 22 | 41 | 13.6 |
| | 40-44 | 5 | 2 | 7 | 2.4 |
| Educational level | Illiterate | 15 | 10 | 25 | 8.4 |
| | Basic | 89 | 125 | 214 | 71.3 |
| | Secondary | 30 | 14 | 44 | 14.8 |
| | Tertiary | 13 | 4 | 17 | 5.5 |
| | **Total** | **150** | **150** | **300** | **100** |



## 4.2 HAEMOGLOBIN (Hb) AND FERRITIN LEVELS

The haemoglobin levels of the participants ranged from 6.3 to 13.5g/dl with an overall mean of 10.95 g/dl. The distribution was approximately normal. The percentage of pregnant women with Hb less than11.0g /dl was 43.6% and 73.4% for pregnant women

living in Pramso and South Suntreso respectively. Peak values were between 10 and 12 g/dl (Figure 4.1 and 4.2) respectively.

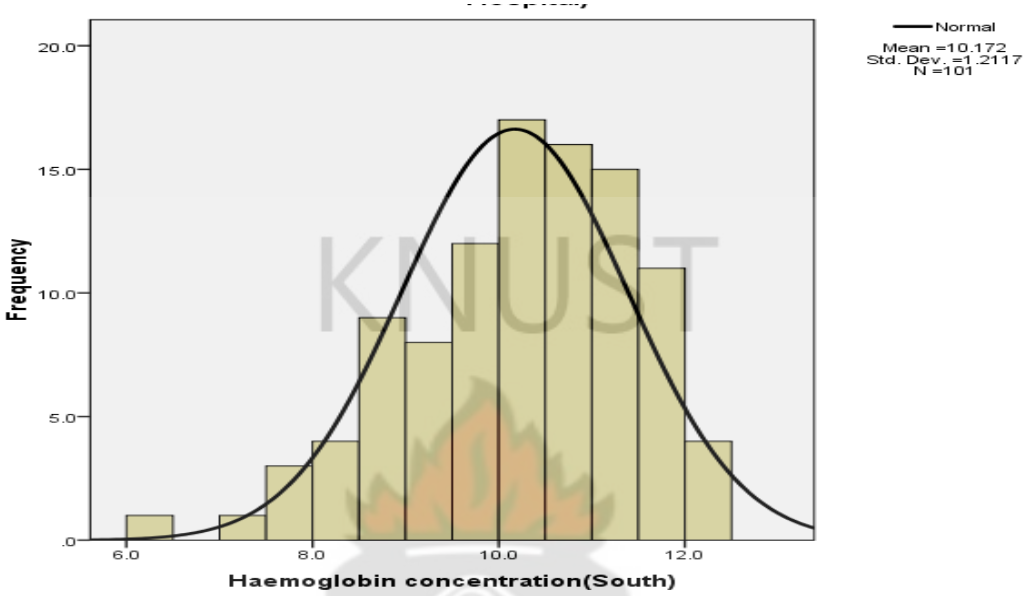


**Figure 4.1Mean haemoglobin concentration (g/dl) in South Suntreso participants**

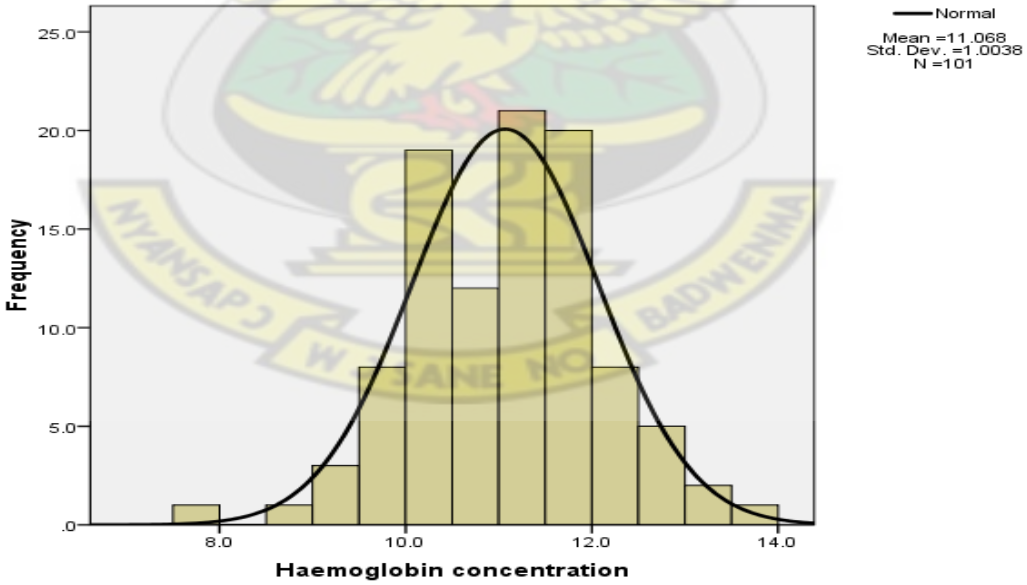


**Figure 4.2 Mean haemoglobin concentrations(g/dl) in Pramso participants**

The concentration of serum ferritin levels measured in the pregnant women was found to be between 0 and 275 microliters. However, most of the participants (Pramso) had their serum ferritin between 0 and 150 microliters with the peak mean concentration being 25 microliters indicating that serum ferritin levels for most of the participants was very low (Figure 4.3).

The mean ferritin levels observed in participants from South Suntreso, the ranged between 0 and about 175 microliters. The concentration of 50 microliters was, however, found to be more frequent followed by that of 75 microliters. A normal distribution for the mean ferritin levels was, however, found in the participants of South Suntreso (Figure 4.4).

Ferritin levels, were found to be dependent on haemoglobin concentration thus the higher the haemoglobin concentrations the higher the ferritin levels.

Mean haemoglobin concentrations were normally distributed between six microliters and 13 microliters. Peak levels were found to be at 10 microliters followed by 11.5 microliters. Levels of 6 microliters and 12.5 microliters were however found to be the least occurring in the pregnant women of South Suntreso with pregnant women from Pramso the mean haemoglobin levels was normally distributed between 8 and 14 microliters with peak values been between 10 and 12 microliters.

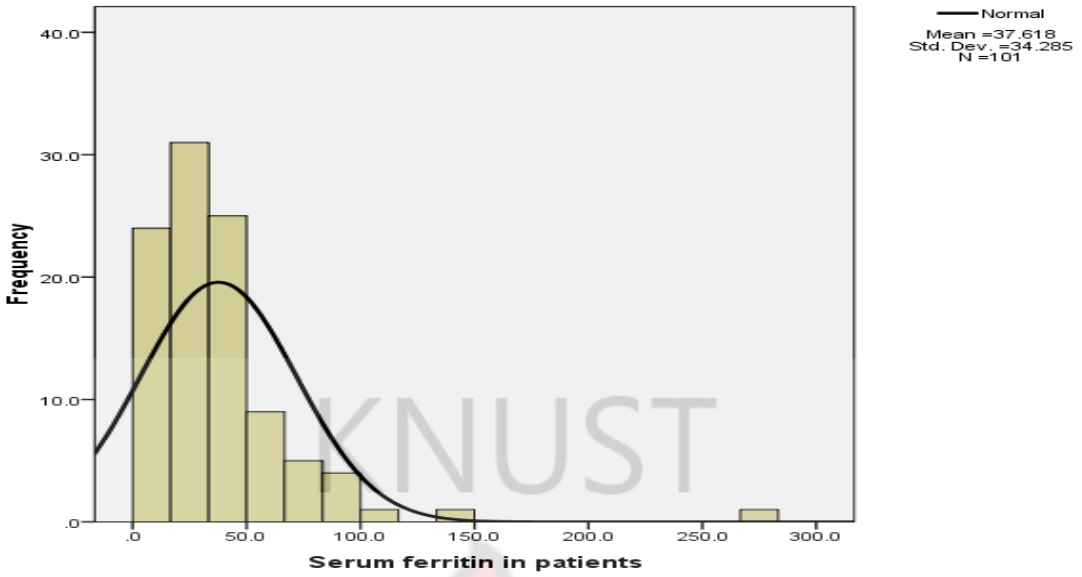


**Figure 4.3 Mean concentration of Ferritin (ul/L) in participants of Pramso**

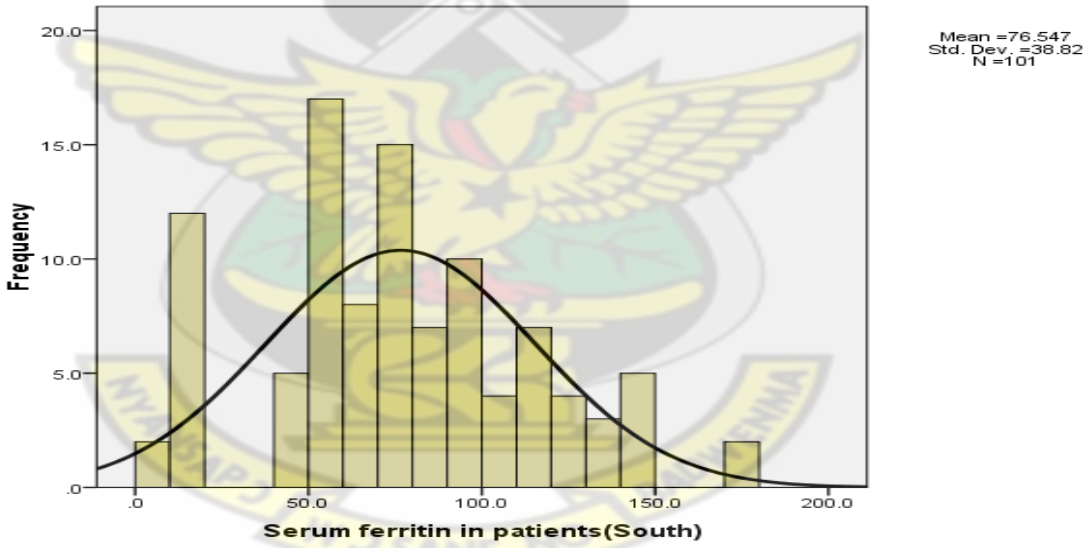


**Figure 4.4 Mean concentration of Ferritin(ul/L) in participants of South Suntreso**

## 4.3 PARASITAEMIA

### 4.3.1 MALARIA PARASITAEMIA

Presence of malaria parasites mainly *Plasmodium falciparum* was observed in 56.4% of the pregnant women in Pramso whiles 68.3% of the pregnant women in South Suntreso also had malaria parasites present in their blood (Figure 4.5).

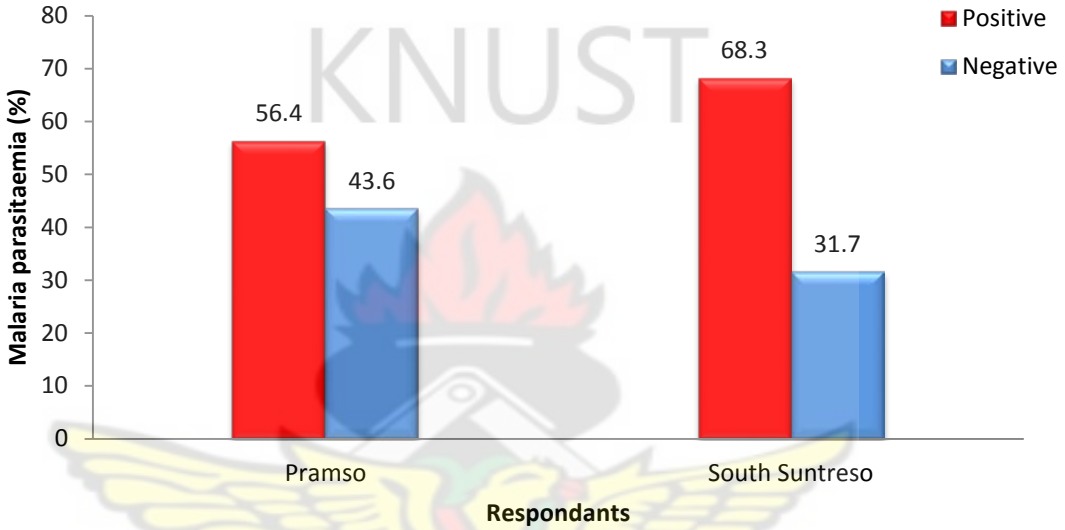


**Figure 5 Malaria parasitaemia**

Of the women in the first trimester of pregnancy, 2.8% had parasitaemia, however, the mean parasite density was low (++). Parasite density in those in the second trimester averaged +++, however, the differences in parasiteamia and parasite density among the various pregnancy groups were not significant at $P<0.05$

#### 4.3.1.1 MALARIA PARASITAEMA AND HB

The relationship betwwen parasiteamia and haemoglobin fellows a normal trend where the correlation is positive as exhibit by the participants in Pramso(figure 4.6), however

that of South deviated from that, this may be due to other factors like their diet and the drugs that they have been taken which may help or inhibit iron absorption.

Most of the anaemic participants had no malaria parasitaemia, compared to their normal Hb participants (Figure 4.7) the differences were not significant.

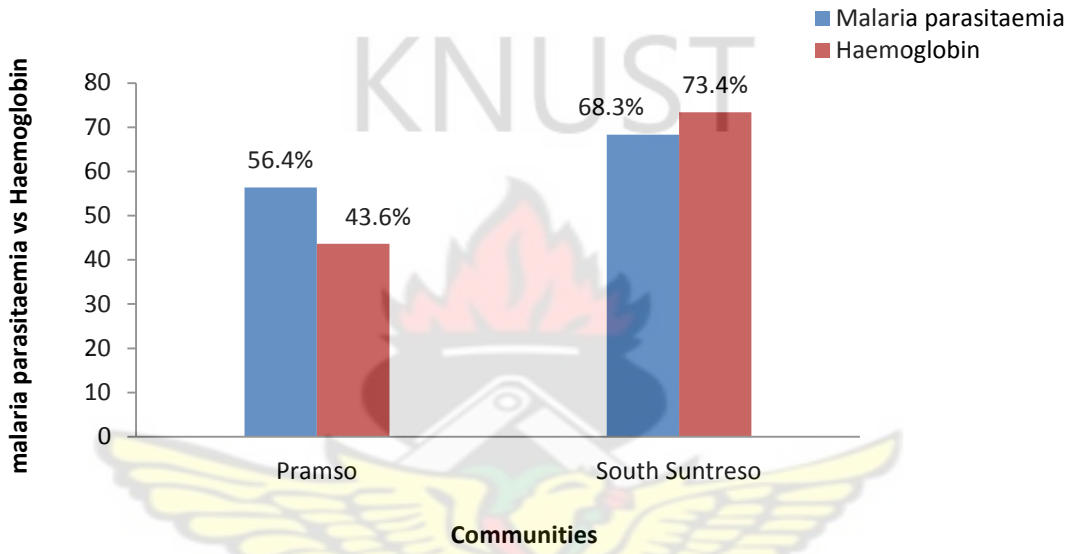


**Figure 6 Relationship of malaria parasitaemia and haemoglobin**

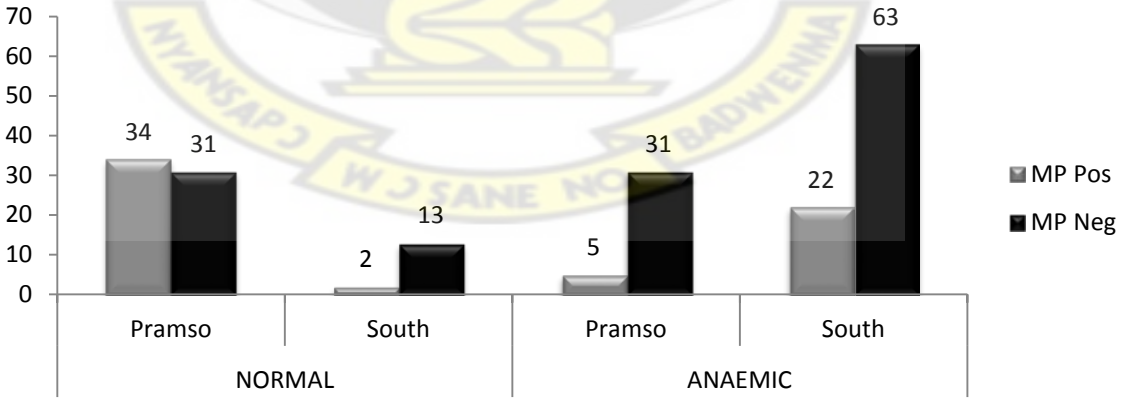


**Figure 4.7 Presence of malaria parasitimia in anaemic and non-anaemic participants of both communities**

#### 4.3.1.1 HELMINTH INFECTION AND HB

Helminth infections were generally absent in the participants who were in their second trimester of pregnancy (Figure 4.6). However, anaemic participants were found associated with low levels of Helminth infestation (Figure 4.7). The parasites mainly encountered were Ascaris lumbricoides and hookworms however they were of very low intensities and their presence was not found to significantly impact Hb levels.

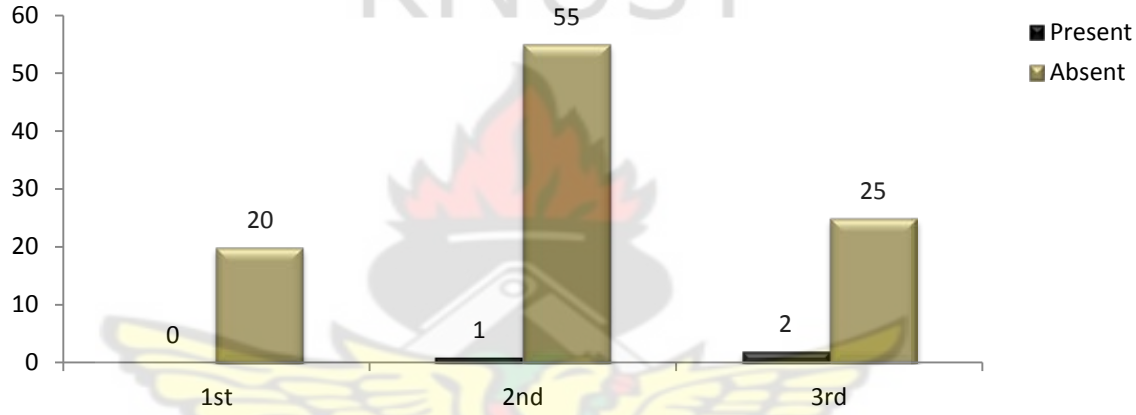


**Figure 4.8 Helmith infection in relation to the stage of pregnancy**

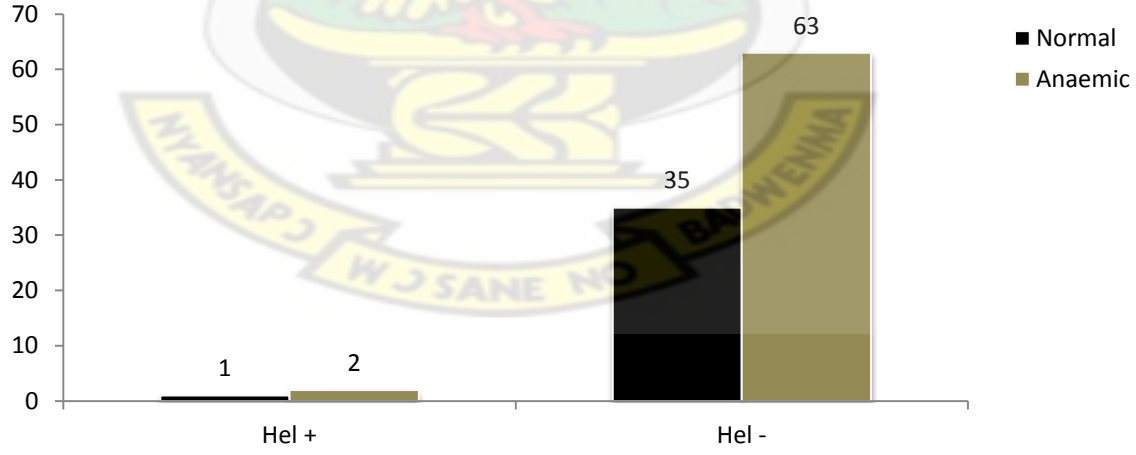


**Figure 4.9 Helmith infection in relation to anaemic status**

## 4.4 KNOWLEDGE OF MALARIA

Asked of their knowledge about malaria disease, all pregnant women gave an affirmative response. Two hundred and thirty eight (79.4%) of the pregnant women had experienced no malaria incidence during their pregnancy, while 20.6% recall at least a malaria incidence during the current pregnancy. The frequency of the malaria attacks categorized as quite often, not often and very often had respective percentage 10.9%, 86.2% and 2.9%.



### 4.4.1 KNOWLEDGE OF TRANSMISSION AND PARASITAEMIA

Regarding knowledge of malaria transmission 82% of the women indicated that malaria is transmitted by mosquitoes, incidence of infection in this group was 15%. Ten percent indicated that transmission is through dirt (20% incidence of infection), whilst 8% had no knowledge of the transmission (33% incidence of infection) (Figure 4.8).

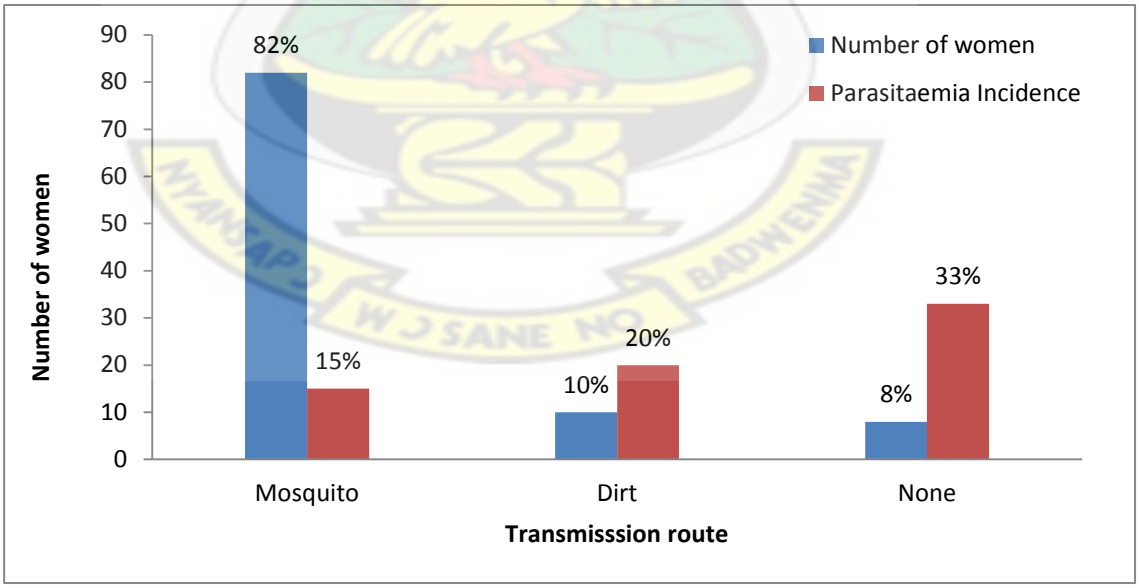


**Figure 4.10 Parasitaemia in relation to knowledge of transmission**

### 4.4.2 KNOWLEDGE OF MALARIA PREVENTION AND PREVENTIVE METHOD USED

Ninety six percent (288) of the women had knowledge regarding ways of preventing malaria. Preventive methods adopted by the pregnant women for malaria included the use of insecticide treated bed nets, mosquito coil, insecticide spray and those who do not use any of the options mentioned. In South Suntreso, 21.8%, 5%, 4% and 69.3% of the pregnant women used the preventive methods mentioned respectively, whereas in Pramso 21.8%, 4%, 5.9% and 68.35% used the preventive methods mentioned respectively (Table 4.2).

Apart from the options in the questionnaire some women made mention of other measures like desilting of gutters, getting rid of dirt as additional preventive methods used.

**Table 2.2 Knowledge of Malaria Preventive methods used by pregnant women in both communities**

| | Preventive methods for malaria control (**South Suntreso**) | | | | | |
|---|---|---|---|---|---|---|
| | | None | net | coil | insecticide | Total |
| Educational Status(**South Suntreso**) | Illiterate | 11 | 2 | 2 | 0 | 15(9.9%) |
| | Basic | 63 | 21 | 0 | 5 | 89(59.4%) |
| | Secondary | 18 | 8 | 3 | 1 | 30(19.8%) |
| | Tertiary | 12 | 2 | 2 | 0 | 16(10.9%) |
| | Total | 104(69.3%) | 33(21.8%) | 7(5%) | 6(4%) | 150 |

| | Preventive methods for malaria control(**Pramso**) | | | | | |
|---|---|---|---|---|---|---|
| | | None | net | coil | insecticide | Total |
| | Illiterate | 4 | 2 | 0 | 4 | 10(6.9%) |
| Educational | Basic | 94 | 23 | 3 | 5 | 125(83.2%) |
| Status(**Pramso**) | Secondary | 4 | 3 | 2 | 0 | 9(5.9%) |
| | Tertiary | 1 | 4 | 1 | 0 | 6(4.0%) |
| | Total | 103(68.3%) | 32(21.8%) | 6(4%) | 9(5.9%) | 150 |



### 4.4 Study communities and mosquito breeding sites

All study communities sites contained both residential and slum (Zongo) settlements. Both had poor drainage facilities (especially North Suntreso). Those with quite good drainages were choked creating room for mosquito breeding. A few meters from the Pramso hospital an old refuse dump had some old buckets and other containers holding water in which mosquitoes were breeding. Some old lorry tyres were found in open places breeding mosquitoes. North Suntreso also had a huge refuse dump in the middle of a residential area with so many receptacles and puddles where mosquitoes bred (Plates 1&2, Appendix 2).

### 4.5 Mosquito species identified

From the water sample collected at various points in the study communities, adult mosquitoes emerged over a period of between one and two weeks from each sample

collected. The Anopheles mosquito was the dominant mosquito species in the samples collected. South Suntreso, however, recorded the higher number of mosquitoes compared to Pramso. Choked gutters also recorded the highest number of mosquitoes in all samples collected.

A total of 78 adult mosquitoes emerged in the laboratory of which 63.4% were Anopheles mosquitoes, 32.0% were culex while 4.6% were Aedes. The highest number of mosquitoes (70) emerged from the samples collected from North Suntreso representing 87.5%. The samples from Pramso recorded 8 (12.5%) mosquitoes (Fig. 4.9).

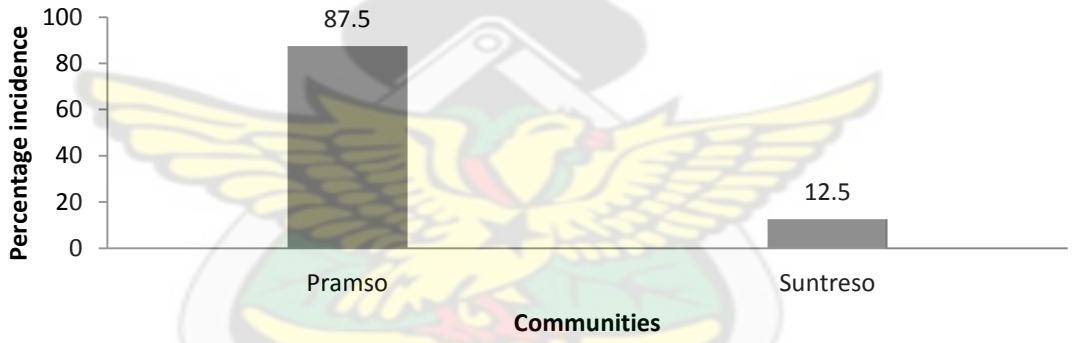


**Figure 4.9 Percentage occurrences of mosquitoes in the study areas**

For the purpose of this study, further analysis of the Anopheles mosquitoes were done to ascertain the exact number of female and male Anopheles mosquitoes that merged as well as the species of the Anopheles mosquitoes.

The 50 Anopheles mosquitoes recorded were of different species. The species were Anopheles gambiae (76%) and Anopheles funestus (24%). Of the 50 Anopheles

mosquitoes recorded, 32 were females whiles 18 were males representing 64.0% and 36.0% of the total number of Anopheles mosquitoes respectively (Fig 4.10)


**Figure 4.10 Percentage occurrences of male and female Anopheles gambiae mosquitoes**

Pramso and its environs where St Michael hospital is sited has less frequency of breeding sites. The main breeding site observed in this area is overgrown vegetation which occurs about 10m-15m. Suntreso and its vicinities (Bantama, North Suntreso, Abrepo etc) has more breeding sites, these breeding sites include overgrown vegetation, choked drainage systems, plastic bags and abandoned used car tyres. These were encountered at a distance of about 5m-10m.

# CHAPTER FIVE

## 5.0 DISCUSSION

The present study indicates that the prevalence of anaemia (Hb<11.0g dl) among pregnant women living in Pramso and South were 43.6% and 73.4% respectively indicating that IDA remains a major health problem in these communities.

The cause of anaemias stated by other studies (Aikawa*et al*., 2005; Brooker*et al*., 2008) is multifactorial, with risk factors including lack of iron intake, hookworm infestation, malaria parasitaemia (Aikawa*et al*., 2005), diet, infection and genetics,and for some of the commonest causes of anaemia there is goodevidence of the effectiveness of simple interventions: for example iron supplementation (Reveiz*et al*., 2007), long-lasting insecticide nets and intermittentpreventive treatment for malaria (Gamble *et al*., 2007; Garner and Gulmezoglu).

Though none of the contributing factors as stated by other studies (Aikawa*et al*., 2005; Brooker *et al*., 2008) was solely responsible for the observed anaemia in the respondants in the present study, malaria parasitaemia, as earlier mentioned have the ability to cause anaemia, the malaria parasite have the ability to cause haemolytic anaemia due to the destruction of the red blood cells by haemolysis. Although majority (82%) of the repondants knew that malaria was caused by mosquitoes, most of them (69.3% in Pramso and 68.3% in South) used no preventive method against malaria infection hence a high incidence of parasitaemia.

Significance level was established between haemoglobin and malaria in both communities (p = 0.000). However, incidence of malaria was high in South suntreso compared to that of Pramso.

This observation is due to anthropogenic activities (such as throwing of solid waste into constructed drains clogging them in the process). This leads to stagnation of waste water consequently creating suitable breeding sites for mosquitoes. The environmental quality of Pramso is much better than South Suntreso where there is few incidence of stagnant pools and overgrown weeds serving as breeding sites for disease causing vectors.

Serum ferritin levels as determined in all pregnant women was found to below as reported in the results (Figures 4.3 and 4.4),with the peak levels for Pramso at 25ul and South Suntreso at 50ul. The transportation of iron is enhanced by ferritin therefore a low level of ferritin will then automatically lead to low levels of haemoglobin which will consequently result in anaemia.

During pregnancy women normally have the craving for all kinds of food and non-food substances like red sand or clay (locally referred to as shile or ayillo). These foods and non-food substances might be low in iron or may contain substances that that would interfere with the absorption of iron in the diet, for instance as mentioned earlier, tea, cocoa, some herbal preparations contain iron binding phenol compounds. These may interfere with iron absorption.

Though hookworm infestation has been found to be associated with IDA (Merchant *et al*., 2002), its contribution to the present study was virtually absent – with significance level $p > 0.05$ for both communities, but that notwithstanding a few of the women sampled had helmithes infection which have the potential of causing anaemia due to sucking of blood.

The supply of iron tablets to women of childbearing age is one of the national nutrition policies of which Pramso and South are included. Previous studies have reported difficulties in compliance with taking iron tablets because of their side-effects and the low awareness of anaemia.

In this survey, it was found that the presence of hookworm and *Ascaris lumbricoides* ova in stool samples were not associated with a significantly increased risk of IDA. According to WHO guidelines, a de-worming tablet is recommended during the second and third trimesters of pregnancy and this may have accounted for the low levels of worm infestation.

In addition to anthelminthic therapy, emphasis should also be given to other preventive strategies, including sanitation control programmes and use of footwear to protect the feet from contact with contaminated soil.

Preference for traditional medicine was identified as one of the independent risk factors for IDA in the present study. Thus, emphasis should be given to the counseling of pregnant women who prefer traditional medicine to ensure that adequate iron status is achieved.

Geographic location is considered to be associated with IDA, as IDA prevalence is often higher in rural areas than in urban areas (Preziosi*et al*., 1997). As the current survey was conducted entirely within a rural district, this difference was not assessed. Also, social services, such as education and health care, were evenly distributed throughout the study area. The overall equality in social development and opportunity limited the comparison of our participants' social backgrounds.

# CHAPTER SIX

## 6.0 CONCLUSION AND RECOMMENDATION

### 6.1 CONCLUSIONS

The results of the present survey suggest that, intake of iron tablets, hookworm infestation, low frequency of egg consumption, malaria parasitimia during pregnancy, foetal iron demand and preference for traditional medicine are important risk factors of IDA among pregnant women living in urban and peri-urban settlements.

In this survey, we found that the presence of hookworm and *Ascaris lumbricoides* ova in stool samples was not associated with a significantly increased risk of IDA. According to WHO guidelines, a de-worming tablet is recommended during the second and third trimesters of pregnancy and this may have accounted for the low levels of worm infestation.

Preference for traditional medicine was identified as one of the independent risk factors for IDA in the present study. Thus, emphasis should be given to the counseling of pregnant women who prefer traditional medicine to ensure that adequate iron status is achieved.

In addition to anthelminthic therapy, emphasis should also be given to other preventive strategies, including sanitation control programmes and use of footwear to protect the feet from contact with contaminated soil.

## 6.2 RECOMMENDATION

Further research should be carried out in other communities so as to obtain an iron deficiency data base for effective planning and implementation of control programmes.

A stratified research of high income earning and low income earning pregnant participants should be carried out to determine the level of risk factors between the two groups.

A case control study should be investigated to determine the level of contribution of nutrition to iron deficiency in remote rural communities and urban communities.



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**APPENDIX 1**

**Table 1 Types of non-food substances and fruits taken by pregnant women**

| | Non-food substance | Pramso | Suntreso |
|---|---|---|---|
| Type of non food substance | Ice block | 3 | 1 |
| | White substance | 3 | 0 |
| | Red substance | 1 | 0 |
| | **Total** | **7** | **1** |

APPENDIX 2

PLATE 1Gutter choked with solid and liquid waste (Suntreso)

PLATE 2 Waste water in streets (Suntreso)