well done work
good work in the field
the investigation knows limitations
need more operational definitions

Masters Thesis

MALARIA IN PREGNANCY

IN THREE WOREDAS OF EAST SHOA

By

Negussie Taffa, MD

May, 1994

MALARIA IN PREGNANCY IN THREE WOREDAS OF EAST SHOA, ETHIOPIA

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Thesis submitted to the School of Graduate Studies of Addis Ababa University in partial fulfilment of the requirement for the Degree of Master of Public Health.

May, 1994

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School of Graduate Studies

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ABSTRACT

A cross-sectional, community based study of malaria was conducted from September-December 1993, in an unstable malaria endemic area in Eastern Shoa, Ethiopia, to determine the prevalence and document the clinical features and treatment responses in pregnant and non pregnant women.

A total of 1,767 women were studied. Ninety (11.3%) pregnant and 63(6.6%) non pregnant women were found to have malaria parasitemia (P=0.001). The rate in primigravidae was higher than in multigravidae, 212(12.24%) and 314(11.4%) respectively, though the difference was not statistically significant.

Overall Plasmodial prevalence showed 49.7% *Plasmodium vivax* and 50.3% *Plasmodium falciparum.* Mean gestational age of the pregnant women was 21.09± 10.2 weeks with Peak malaria parasitemia around mid-pregnancy. Majority of cases were symptomatic during diagnosis and symptoms were not different between pregnant and non pregnant.

Conjunctival pallor was more documented in pregnant than in non pregnant (OR = 3.04, 95% CI.1.47,6.33). Mean haematocrit in the pregnant was 30.3± 4.6% and 33.4± 6.8% in the non pregnant (P= 0.0001). Primigravidae showed lower haematocrit values and higher mean parasite counts when compared to multigravidae (P =0.03 and 0.06 respectively). Mean asexual malaria parasite count was 6,561± 5,259/micro litter of blood and 4,598± 4,275/micro litter of blood in pregnant and non pregnant respectively (P= 0.01).

Clinical responses in both groups were not significantly different. Findings are discussed and recommendations made regarding future studies.

I. Introduction

Malaria remains to be one of the most difficult Epidemiological,Pharmacological, and Immunological challenges in the developing world, particularly in Africa, Latin America, and Asian countries. Its influences on practically every aspect of life in many African countries has been profound and sustained over the centuries, and malaria remains a major cause of morbidity and mortality. Moreover,the Epidemiological situation appears to be changing for the worse in many countries. The vectoral capacity of *Anopheles gambiae* and *Anopheles funestus* continues to be intense, and resistance of these vectors to insecticides and of the parasites to drugs is on an increase(1), which poses a serious threat of increased severity of the disease and death. Quantitatively, the number of foci of intense malaria transmission is increasing because of changing environmental conditions in areas of vast economic developments and the spread of malaria to areas previously free of the disease, as millions of people move in to malarious areas to reclaim land particularly in nomadic life conditions, to seek survival or food in famine situations or escape civil disturbances and war. Malaria undermines the health and welfare of families, endangering the survival of children, debilitating the active population and straining both countries' and peoples' scarce resources by excessive public health costs, low productivity and impaired economic growth (2).

This, added on to the prevailing economic and social problems in implementing locally planned control strategies, puts African malariologists in difficulties when trying to forward lasting solutions.Yet, malaria is a curable disease, not an inevitable

burden. The vastly expanded knowledge of the disease and its control acquired over the years provides the basis for launching a new global initiative for malaria control. In most endemic countries, the goal of malaria control will be to prevent malaria mortality and to reduce morbidity and socio-economic losses provoked by this disease.

Success in achieving these goals depends on political support from the highest level. It also depends on a change in emphasis from highly prescriptive, centralized control programmes to flexible, cost- effective and sustainable programmes adapted to local conditions and responding to local capacities for assessing malaria situations and selecting appropriate control measures to reduce or prevent the disease problem in the community rather than to concentrate on reducing parasite rates in the population (2).

Malaria control is not the isolated concern of the health worker. It is every one's business, and everyone should contribute. It requires the partnership of community members and the involvement of those engaged in education, the environment in general, and water supply,sanitation and community development in particular. Malaria control must be an integral part of national health development and health concerns must be an integral part of national development.

Emphasis ought to be put on the vulnerable population sector (children and pregnant women) who are highly susceptible for the major health, economic, and social sequelae of malaria. The provision of prophylactic protection against this disease and the issue of vector control through community based actions must be investigated and applied with caution as both of these activities require careful evaluation of economic difficulties, complexity of the conditions and drug sensitivity problems.

In general, this community based activity must be sustained and supported by inter- sectoral collaboration at district, regional, national and international levels, by monitoring, training and evaluating through operational and basic research.

From the most recent information obtained by the World Health Organization (WHO 1990),it is estimated that 2,073 million people in the world (over 40% of the total population),living in more than 100 countries, are exposed to the risks of malaria (around 500 million of them live in Africa),and that some 270 million of these are infected with malaria parasites. While the number of cases reported to WHO has been of the order of 5 million annually for the past three years, the best estimate is that perhaps 110 million clinical cases occur every year, of which 90 million (>80% of the total) are in Tropical and sub- Saharan Africa. Global deaths are estimated at approximately 1.5-3 million a year and the majority are again from African countries (3).

In Ethiopia, around 2/3rd of the inhabitants live under malaria risks and the prevalence and incidence of malaria has greatly increased since the mid-1980s and early 1990s, partly due to the military campaigns and ecological changes during this time. The annual incidence of malaria in 1989-1990 was 21/1000 population based on the statistics reported by the health services to the Ministry of Health (MOH, Unpublished data). But this figure is undoubtedly a gross underestimate, since it includes only cases with laboratory confirmation. Inclusion of the additional cases diagnosed on clinical grounds alone would result in a three to four-fold increase in the estimated incidence (4).

Unpublished data for 1993 from the National Office for Control of Malaria and other Vector Borne Diseases (NOCMVD) suggest that an estimated 200,000 cases of malaria are known to occur every

year (5). The same data showed that the 1992 -1993 prevalence estimate of malaria infection (based on health institution reports) was 2.4%. It was also indicated that malaria accounted for 16.3% of all outpatient visits and 28.4% of all deaths in areas where malaria is endemic (5).

Many investigators have demonstrated an increased prevalence and severity of malarial infection during pregnancy (6-15).

Gilles et al (8), for example, followed a small group of primigravidae longitudinally before and after pregnancy at University College Hospital Ibadan, Nigeria and reported an increase in the rate of parasitemia from 13 (34%) before pregnancy to 30 (79%) during pregnancy.

In Bangui (C.A.R), a comparative survey regarding infection with malarial parasites was carried out in 250 pregnant and 250 non pregnant women. Both groups were matched by age, and none of them had taken any chemoprophylaxis or anti-malarial treatment for the month before the survey. Parasitic indices were significantly higher in pregnant women, and higher in primigravidae than multigravidae; the highest malarial indices were noted in the 15-20 age group (10). This increased prevalence and severity of malaria among pregnant women is more likely to be observed in areas of unstable malaria and among women who are less immune to the disease (6-15).

Among pregnant women residing in highly endemic areas who have acquired considerable immunity to malaria, age and parity appear to influence susceptibility to an important degree. Those women who are pregnant for the first time are most affected, showing an increased prevalence and density of parasitemia, increased frequency of clinical illness (but not mortality) and significantly

increased delivery of low birth weight children (9).

B.J.Brabin (6) in 1983, did a study of malaria during pregnancy in women living under stable malaria endemic area in western Kenya. Results showed that infection is more frequent and severe in primigravidae both during pregnancy and at the time of delivery. The peak prevalence of infection in primigravidae (85.7%) and multigravidae (51.7%) mainly occurred at 13-16 weeks of gestation.

In contrast,among partially immune pregnant women (in unstable malaria endemic regions predominantly), frequency and density of parasitemia increase progressively to peaks around mid-pregnancy, especially in primigravidae . An epidemiological survey in unstable malaria endemic area in rural Cameroon (11) assessed the presence of malaria infection in 225 pregnant and 75 non pregnant women. The results showed that *Plasmodium falciparum* infections occurred more frequently in pregnant (45%) than in non pregnant women (31%) in terms of parasite rates and density especially in primigravidae as compared to multigravidae matched for parity and age. One study (30), however, found no difference in parasite densities between primigravidae and multigravidae in unstable endemic area in India.

In women with little or no prior experience of the disease, *Plasmodium Falciparum* causes severe clinical illness, substantial mortality, increased rates of abortions, still birth and low birth weight of the offspring. Moreover, in such women, the clinical consequences seen are not modified by maternal parity. However, in pregnant women residing in highly endemic areas who have acquired considerable immunity through prolonged prior contact with malaria, parity appears to influence susceptibility to an important degree (9). In Zimbabwe, malaria was followed by abortion in 13 of 14

patients, in the first trimester and 8 of 19 in the second trimester and 50% of infected women in the third trimester went in to premature labour (Herd and Jordan, 1981).

A longitudinal study of splenomegaly in pregnancy was done in Madang province, Papua New Guinea (12), where malaria is transmitted all year round. Five hundred eighty two pregnant women attending monthly rural antenatal clinics were compared to 712 non pregnant women of child bearing age group who live in the same area. In non pregnant women who later became pregnant, spleen rates increased in early pregnancy. Peak spleen rates in early pregnancy occurred before 16 weeks of gestation for all gravida classes. There was a decrease in spleen rate and a fall in average enlarged spleen size with increasing gestational age for women attending on a first clinic visit and before receiving chloroquine prophylaxis. It is suggested that pregnancy induces a change in spleen function early in gestation, which in primigravidae results in increased frequency of *Plasmodium falciparum* parasitemia. In multigravidae, these early gestational changes did not predispose to parasitemia, which indicated that these women had developed enhanced malaria immunity.

In five maternity centres in urban and rural Zaire, Steketee et al (13), evaluated the maternal prevalence of *Plasmodium falciparum* parasitemia and recorded fever, the frequency of abortions and still births, newborn birth weights and the feasibility of delivering anti-malarial prophylaxis. Women in their first and second pregnancy compared to others ($\geq$ 3 pregnancies), had a higher frequency of parasitemia, higher parasite densities, higher rates of still births and low birth weight babies. A more recent study that evaluated the efficacy of chloroquine prophylaxis in Madang, New Papua Guinea (14), found an increased susceptibility

of primigravidae to *Plasmodium falciparum* but not to other malaria species, with the peak prevalence occurring at 9-16 weeks of gestation. The incidence of *Plasmodium falciparum* infection per-person-month was only 14% among nulliparous women. In contrast, infection rates were 20% among primigravidae, 25% among those women in their second pregnancy, 17% for women in their third or later pregnancies.

In Ethiopia, Assefa and Awash (un published data presented to Ethiopian Medical Association Annual Conference,1988) documented pregnancy outcomes among 428 pregnant women who were resettled from non-malarious areas to the malaria endemic Pawi Resettlement. They found no significant association of unfavourable outcomes of pregnancy (abortions, still births, pre-term deliveries and maternal deaths) with malarial infections. A year later, Yohannis (15), analyzed 37 pregnant women admitted to Gondar Hospital with malaria. He described a maternal mortality rate of 12 (32.4%), 15 (40.5%) rate of spontaneous abortion, pre-term delivery with early neonatal death and still birth.

Though the exact reason for an increased prevalence and severity of malaria during pregnancy is not yet clearly defined, immunity acquired against this disease either with age or more definitely with frequent exposure to malarial episodes together with the general effect of pregnancy on the body immune system seem to be the most likely explanation (16-20).

Brabin (6),quoted Walton's assumption 30 years before his own work. Walton reported that pregnant women show the same pattern of malaria infection as school children. He stated, "during pregnancy the ability to limit the number of parasites appears to be lost and women revert to a "child type" reaction to infection". Brabin also noted that the mother seems to loose her age dependant

immunity only to re-acquire it after 9 months of pregnancy, and remarkably exhibit recovery at about the time of delivery.

An increase in the incidence of a number of infectious diseases has been reported in pregnant women in association with the depression of selected components of the immune system. The severity and risk of acquiring diseases which are normally controlled by the Cell-Mediated Immune System (CMI) (e.g Tuberculosis, Cytomegalo virus [CMV],and Toxoplasmosis) are enhanced during pregnancy. Since CMI mechanisms are believed to be most important in the development and maintenance of immunity to malaria, impairment of this immunity may in part explain the susceptibility of pregnant women to malaria. Of this system,particular insult is to the components such as: reduced lymphocyte transformation, suppression of T-cell dependant macrophage activation, an increase in serum hydrocortisone level and the appearance of pregnancy-associated new proteins such as Alpha-2 Globulins which are known to exert immuno-suppressant effect has been described widely by many researchers (16-19).

Although impairment of in-vitro cellular responses to various antigens has been reported in the pregnant women, some T-cell responses appear to be unaffected. E.M. Riley, et al (6), have shown that PHA-induced responses appear to be unchanged in pregnancy, which suggests that antigen handling and presentation at the macrophage level, rather than antigen recognition at T-cell level may be the crucial step. This data also demonstrated that malaria antigen-induced Gamma-Interferon production may also be depressed in pregnant women, which is assumed to be one factor that contributes to reduced antigen-presenting cell activity in pregnancy.

The concentration of both total (bound to corticoid-binding globulin plus free) and free cortisol was determined in sera from two groups of pregnant women in Tanzania (17). The study included 152 pregnant women exhibiting clinical malaria during pregnancy and 527 pregnant women who did not have a record of malaria during pregnancy. The serum concentration of total cortisol was significantly higher in women with clinical malaria than in women without recorded malaria. Free cortisol fractions did not differ significantly in these groups. Indications were obtained that higher total cortisol levels cause loss of malaria immunity rather than being concomitant with malaria infection only.

In a study of lymphocyte sub-population and Leucocyte Migration Inhibition Index (LMI) in 78 pregnant and 22 non pregnant women with malarial infection from Northern India, (19) T-cell percentage in controls (80 pregnant and 20 non pregnant women without infection) decreased to its lowest in first trimester and gradually rose to non pregnant levels by the end of puerperium. With malarial infection, there was further drop in T-cell percentage, more so in the third trimester. LMI in controls corresponded with T-cell percentage and was depressed in pregnant patients. Enhancement in LMI with malarial infection was depressed in pregnant patients maximally in the third trimester. Thus, cellular immunity status in malarial patients was found to be depressed during pregnancy especially in the third trimester.

Studies of humeral immunity in pregnancy,on the other hand, have shown that titters of malaria specific antibodies don't differ between pregnant and non pregnant women, and that there is no individual correlation between antibody titters and the susceptibility to infection during pregnancy (8,16,20).

Maternal morbidities correlate with the degree of anaemia, parasitemia and clinical presentation of the illness which range from few episodes of malaise to severe and complicated cerebral malaria. In the proceeding few paragraphs we shall see some of the important complications of malaria during pregnancy.

Anaemia :-Anaemia of malaria is primarily haemolytic and later as erythroid hyperplasia comes into the scene, folate deficiency complicates the problem (6,7). The pathophysiology of anaemia in malaria is also hypothesized further to involve many mechanisms that will include both increased red cell destruction and decreased red cell production. Potential causes of haemolysis include:-loss of infected cells by phagocytosis, removal of unaffected cells by rupture or phagocytosis, removal of unaffected cells due to antibody sensitization or other physio-chemical membrane changes, and increased reticuloendothelial activity, particularly in organs such as the spleen. Decreased production results from marrow hypoplasia seen in the acute infections, and dyserythropiesis, a morphological appearance which in functional terms is ineffective erythropoiesis. Nutritional deficiencies, particularly in women from low social classes, are involved as contributory factors in anaemia causation (7,8,21-26).

The commonness of the problem greatly varies with epidemiologic pattern of the disease. In unstable malaria areas anaemia occurs at all ages and often in both primi and multigravidae equally. In stable areas, although, anaemia is most common in young children and pregnant women, the interaction of other supplementary causes should also be taken into consideration.

Brabin (24), reported from women in Coastal New Papua Guinea that malaria reduces the mean haemoglobin by 0.3gm/dl which is less

than the effect on haemoglobin of malaria in children. The difference in haemoglobin values is greatest in primigravidae (0.7gm/dl)who are more susceptible to *Plasmodium falciparum* than are multigravidae. Comparable differences in haemoglobin levels associated with malaria in pregnancy have been reported by McGregor from the Gambia in 1984 (24).

Hercberg S. et al (27) studied Nutritional Anaemia in 126 pregnant Beninese women where they found anaemia in 55% of them. Interestingly enough, 99% of all patients (there were 95 menustrating women in the study) had *Plasmodium falciparum* infection. Fleming (7), studied 37 Zambian pregnant women with severe anaemia (Hgb.<7gm/dl); 34 (84%) had *Plasmodium falciparum* malaria, 23 (62%) were folate deficient and 13 (35%) were iron deficient. The anaemia of malaria and folate deficiency was both more common and more severe than anaemia due to iron deficiency. It occurred in younger women although primigravidae were not over-represented, it occurred earlier in pregnancy and was associated with low birth weight.

A study from rural Zaire (28) on anaemia of pregnancy in 2950 pregnant women attending antenatal clinics showed that 72% were suffering from moderate anaemia (Hgb.7-11gm/dl) and 3.7% from severe anaemia (Hgb.<7.0gm/dl) at their first visit, before receiving any hematinics or anti-malarial prophylaxis. Thick blood smears were examined from 379 women who presented in the first and second trimester. 70% of primigravidae had malaria parasitemia, compared with 13% of multigravidae. Haemoglobin levels rose with increasing parity and age.

Anaemia in pregnancy, particularly in those cases complicated by malaria infection, is associated with intrauterine growth retardation and low birth weight and hence,increased perinatal

mortality rates to the fetus; it would leave the mother with less reserves to withstand the stress of haemorrhage at delivery, and show a frequency of bacterial and viral infections because of malaria, iron and folate deficiency, as well as the depression of the immune mechanism by pregnancy itself. It also leads, in severe cases, to congestive heart failure and death. In Ibadan, Nigeria, A.Fleming (23) reported a fetal loss rate of 38% and low birth weight (<2000gm) rate of 50% in mothers with untreated severe anaemia (Hct<23%) (23,25,29) even though the proportion of these women with malaria is not stated.

Parasitemia:-Malarial parasite rate and mean parasite density is significantly higher in pregnant women as compared to non pregnant and more so in primigravidae than in multigravidae (6,10,13,31-33). However,some investigators from India (30), have noted different observations since their patients came from unstable malaria endemic areas as opposed to most African malaria and their results showed no significant difference in parasite densities between primigravidae and multigravidae.

References in the above paragraphs concerning high density parasitemia have indicated that peak mean parasite counts occur in the second trimester with slow rises during the first and slow decline after the second trimester. The Indian study (30) however, has observed maximum parasite densities in the third trimester for both primi and multigravidae, though findings were not statistically significant.

Women living in holoendemic malaria regions develop tolerance to high density of parasitemia and thus are asymptomatic most of the time. They also have lesser risks of hyperparasitemia and fatal *Plasmodium falciparum* infection with increasing age. This is

attributed to the active immunity developed following a repeated or sustained parasitemia. Among those women residing in unstable malaria areas or in regions where seasonal variation of malaria transmission is marked, pregnant women appear most of the time with malaria signs and symptoms even at lesser parasite densities that would have been tolerated by the immunes. They are also prone to having higher parasite densities and severe forms of malaria infection by the time they appear for investigations (6,22,29,34).

High parasitemias often result in morbidities consequent to anaemia in the mother and parasitization of the placenta, though often, the rate and intensity of which may not correspond with the rate of peripheral blood parasitemia.

Low birth weight: Placental infection by malarial parasite will result in fetal growth retardation and hence low birth weight deliveries (<2,500gm). Frequently, primigravidae suffer the greatest burnt than multigravidae besides the effect of first pregnancy on birth weight of the newborn. However, there are different views as to whether malaria perse can be the cause of low birth weight rather than being a single factor among the many which can have influence on birth weight (6,8,34-37). Randomized trials of chloroquine prophylaxis (35), were not able to show any significant result of birth weight differences of newborns between the protected and the exposed group of pregnant women though prophylaxis was able to prevent placental infection in the protected group.

In view of the lack of data regarding the importance of malaria in pregnancy in Ethiopia, this study was conducted with the following objectives.

Objectives of the study

General Objective - To describe the prevalence of malaria in pregnancy, and compare its clinical features and treatment response to that of non- pregnant controls.

Specific Objectives - 1. To assess the relative prevalence of malaria in pregnant women as compared to non-pregnant ones.

2. To compare the clinical features and severity of malaria in pregnant women with that of non pregnant women.

3. To document differences in treatment response between the pregnant and non - pregnant cases.

4. To compare the features of malaria prevalence, severity, and treatment response between the primigravid and multigravid women.

II. Subjects and Methods

A. Overall Design This is a cross sectional, community based, comparative study of malaria in pregnancy.

B. Study area and population : The study was carried out from September 15th to December 30th 1993 in order to cover the peak malaria transmission period that follows the heavy rain-fall of the summer. It included 30 rural and urban Kebeles (communities) in Akaki, Debrezeit and Mojo woredas within 75 Kms of Addis Ababa (Figure I).

About 145,000-150,000 people are estimated to live under risk of malaria in the study area (Health profile from the three woredas, Un published data). Approximately 60-80% of the study population in the rural areas live within 10 Km of health institutes. Malaria transmission is mostly water based which actually exists during the rainy season and shortly afterwards. In Debrezeit alone, there are 5-6 large lakes that serve as breeding sites for mosquitoes. The malaria control unit responsible for these areas (extending from Akaki to Mojo) documented 18,964 patients presented with malaria signs and symptoms at out-patient departments in health facilities in the above areas (September to February, 1992-1993). Among these, 9,353 were found to be blood smear positive, where 4,614 (49.33%) were *Plasmodium falciparum* and 4,809 (51.4%) *Plasmodium vivax* cases. The 1983-84 E.C fiscal year morbidity report from Debrezeit and Akakai woreda health units indicated that malaria was one of the top three reasons for out patient visits.

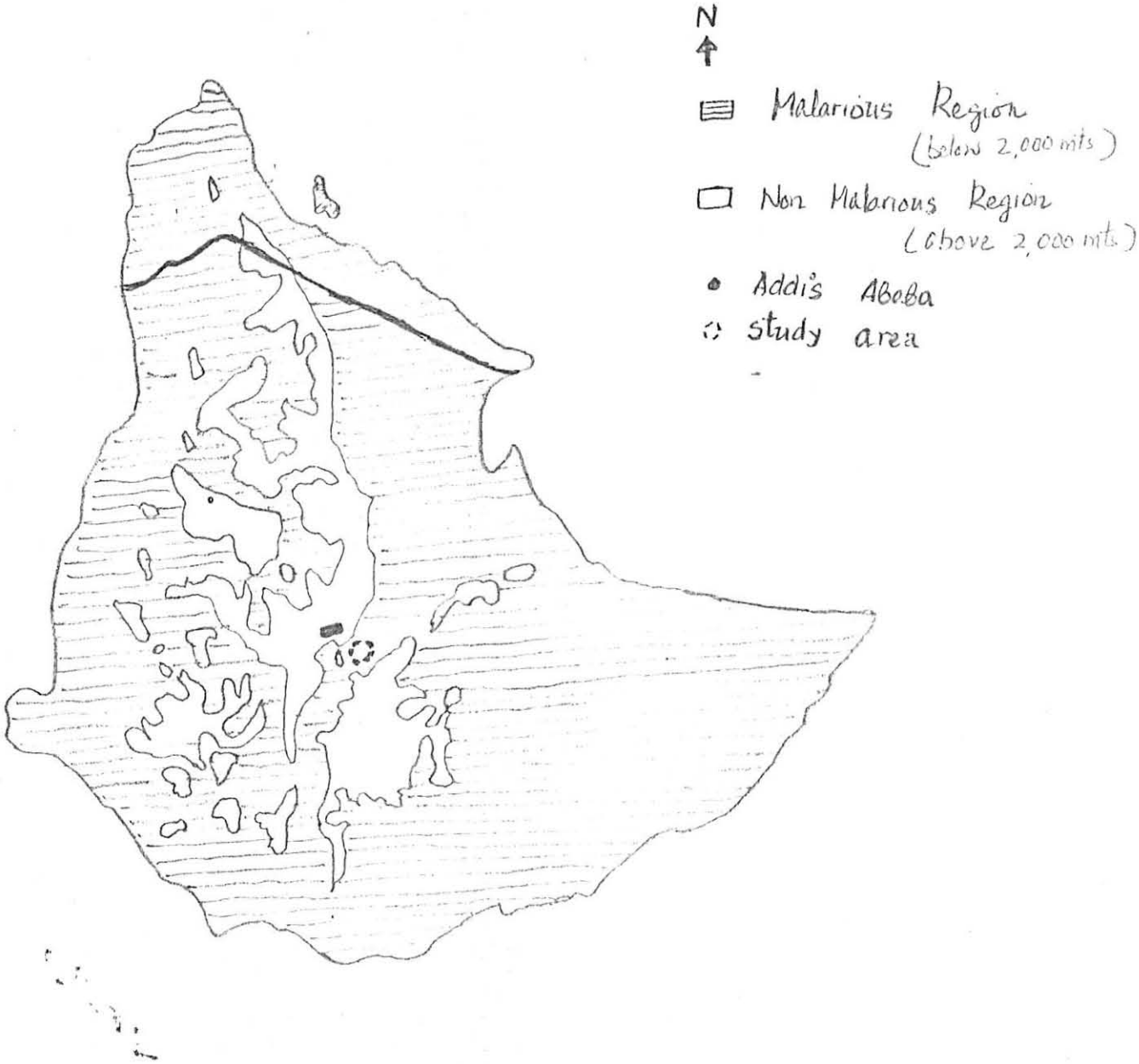


Figure I Malarious and non-malarious Regions of Ethiopia and Location of the Study Area

(Adapted from Gebre Mariam 1988)

Based on the selection criteria, list of malarious localities was obtained from the local malaria control program office. The study localities were selected based on the blood film study results during the preceding years and day to day observations by the office as they had the highest prevalence of smear positivity.

1. Selection Criteria for the study communities.

1-1 Accessibility to 4-Wheel Drive Vehicles and to health service units within 10 Kms diameter.

1-2 The presence of a malaria control office for active surveillance, case detection and control of epidemics.

1-3 The homogeneity of the people and similarities of the ecologic set up as far as malaria transmission is concerned.

2. Inclusion criteria for selection of study population

A house to house visit was made in search of all pregnant women in a locality. Any woman who is pregnant by history which was supplemented by physical examination of a gravid uterus (abdominal fundal height measurement), was made a candidate for participation in the study. Early pregnancies and non pregnant states were differentiated using a urine pregnancy test (Omega-Betatex direct plus agglutination test with sensitivity at 0.2 iu hCG/ml- a level usually achieved 2-3 days after the first missed menustral period).

Any candidate who reported taking antimalarial medication within the past week of study date was excluded from the study. A non pregnant neighbourhood "control"was selected for each case matched for age and parity. The non pregnant status was confirmed by urine pregnancy test. Any problem regarding patient compliance in taking medications appropriately was relied exclusively on histories given on subsequent days and any vomiting or diarrhoea reported on day 1 or 2 were criteria for excluding patient from the

study.

C. Sample Size

As there are no community-based studies documenting any prevalence of parasitemia among pregnant women in Ethiopia, other African country study results were used to estimate the expected proportion of pregnant women with malaria. However, most study areas did have an epidemiologic feature of stable malaria endemicity as opposed to the seasonal variation characteristic to the study area which has got features of unstable malaria.
The following data is taken from some studies conducted in African countries :

Country	Rates of malaria with pregnancy	Year
Gabon	35%	1981
Tanzania	41%	1985
Kenya	42%	1988
‡Cammeroon	45%	1992

‡ - Unstable malaria endemic area.

Based on the above fact, a conservative estimate of 20-25% prevalence of malaria in pregnancy in the study area with 3-5% marginal error (ME), at 95% Confidence Interval, allowed the following sample size calculation.

$$n=\frac{Z^2(\Pi)(1-\Pi)}{M.E^2}$$

Where, n = the estimated sample size

M.E = the degree of certainty for the result to be within this discrepancy.

Z = the critical value corresponding to a 95% confidence level. (two tailed)

Therefore, the above calculation gave a sample estimate of 682.95

in each group (pregnant and non pregnant), that is, 1366 overall. With 30% assurance of loss to follow up and withdrawal from the study, a total of 1,775 (886 per group) were estimated to be the sample population.

D. Data Collection

A standardized, pre-tested questionnaire in the local language (Oromyna) was administered to each participant by trained local interviewers to obtain data on age, address, gravidity/parity, last menustral period (month of gestation), history of fever, headache, malaise, chills/rigor, etc in the last 48-72 hours, and any medication for malaria symptoms in the last 7 days. The clinical status of women with parasitemia was assessed by a Nurse or Physician prior to and after treatment (7 days later) and findings such as fever, diaphoresis, conjunctival pallor, spleen size, fundal height and status of fetal heart-beat were recorded.

Blood was taken by a finger prick with a sterile blood lancet from the tip of the middle finger on day 0 (the first day of examination) from all participants (pregnant women and their matched "controls") for a thin and thick blood smear on a single slide. The blood films were stained according to the procedures described by Bruce (62). Microscopic examination of a thick blood smear, which has been dehaemoglobinized and stained with Giemsa was used for species differentiation and asexual parasite count. Thin smears fixed with 95% ethylene alcohol and stained with Giemsa allowed better species differentiation.

Asexual parasites were counted using an indirect method of malaria parasite density estimation (Trape, 38), whereby, the number of parasites per white blood cell (WBC) in a thick blood film was recorded using a high power microscopic field, (x 100 oil

immersion objective and a x-6 eye piece) and parasite density was determined by multiplying this figure by 8000, an average WBC count per micro-litter of blood.

Blood smear positive women also underwent an interview for a history of malaria signs and symptoms 48-72 hours before blood was taken for examination. Some signs related to malaria infections were looked for after history was taken (annex 1). Blood was also withdrawn in a haematocrit capillary tube for determination of haematocrit.

Response to treatment was monitored both clinically and haematologically. The clinical assessment was done on day 7 using the history and physical examination format used on day 0. The haematologic response monitoring was done using a simplified in-vivo test system developed by K.H.Reckmann (40), a modified three days (day 0,2, and 7) assessment of parasite density to establish partial facts regarding the areal parasite resistance pattern. This method involves administration of 600mg Cq on day 0 and day 1, and the remaining 300mg Cq (5mg/kg) is taken on day 2. Follow up blood smears on days 2 and 7 involve examination for smear positivity and asexual malaria parasite count. Counts on day 2 were interpreted based on the finding made on day 0 which was taken as 100%. If the level of parasitemia was more than 25% of that observed on day 0, the infection was considered to be resistant at the R-III level. These patients received Pyrimethamine/Sulfadoxine (fansidar ^{R} 1575mg po stat). If patients became negative for malarial parasite on day 2, they were considered S/R-I. When patients turned blood smear negative on day 7, they were labelled as S/R-I late and any positivity on day 7 is recorded as R-I early /R-II and provided with fansidar (40,41).

Operational Definition

1. Positive blood smear :- The presence of plasmodial species in a thick blood smear examined in a 100 microscopic fields under high power (x 100 oil emersion objective and a x-6 eye piece).
2. Parasitemia :- Is the number of asexual malaria parasites per 100 Leucocytes extrapolated for an average total white blood cell count in a micro litter of blood (8000). Parasite Loads of < 10,000/micro litter of blood was taken as moderate and >10,000 as severe parasitemia.
3. Anaemia :- Haematocrit values corresponding to haemoglobin level of 11.8gm/dl (altitude adjusted anaemia level in pregnant women - WHO 1975), That is, Hct = 3.2 x Haemoglobin, which is equal to 37.7% were taken as a cut-off point for anaemia. Severe anaemia was taken as Hct <22.4% or Haemoglobin level of 7.0gm/dl.
4. Multigravidae :- Those pregnant women with gravidities of 4 or more.

is this a standard def?

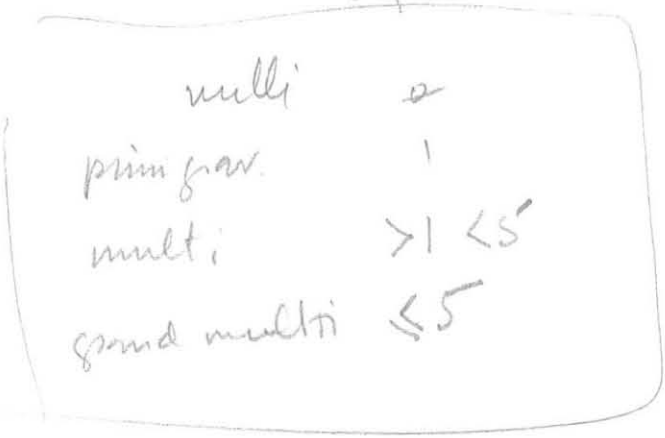
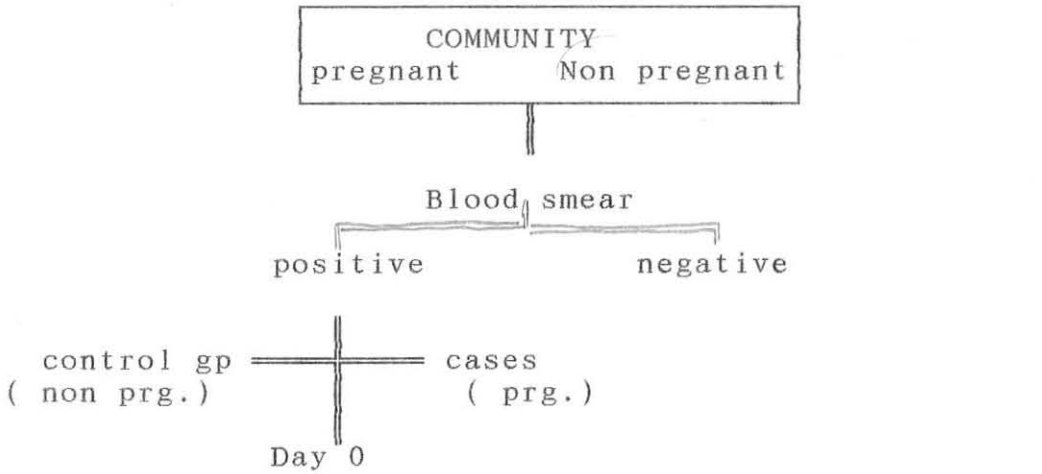


Figure II. Schematic Diagram for Administration of Questionnaire and Management of Study Participants



. Administration of questionnaire

. Hx. P/E

. hct determination

. Treatment

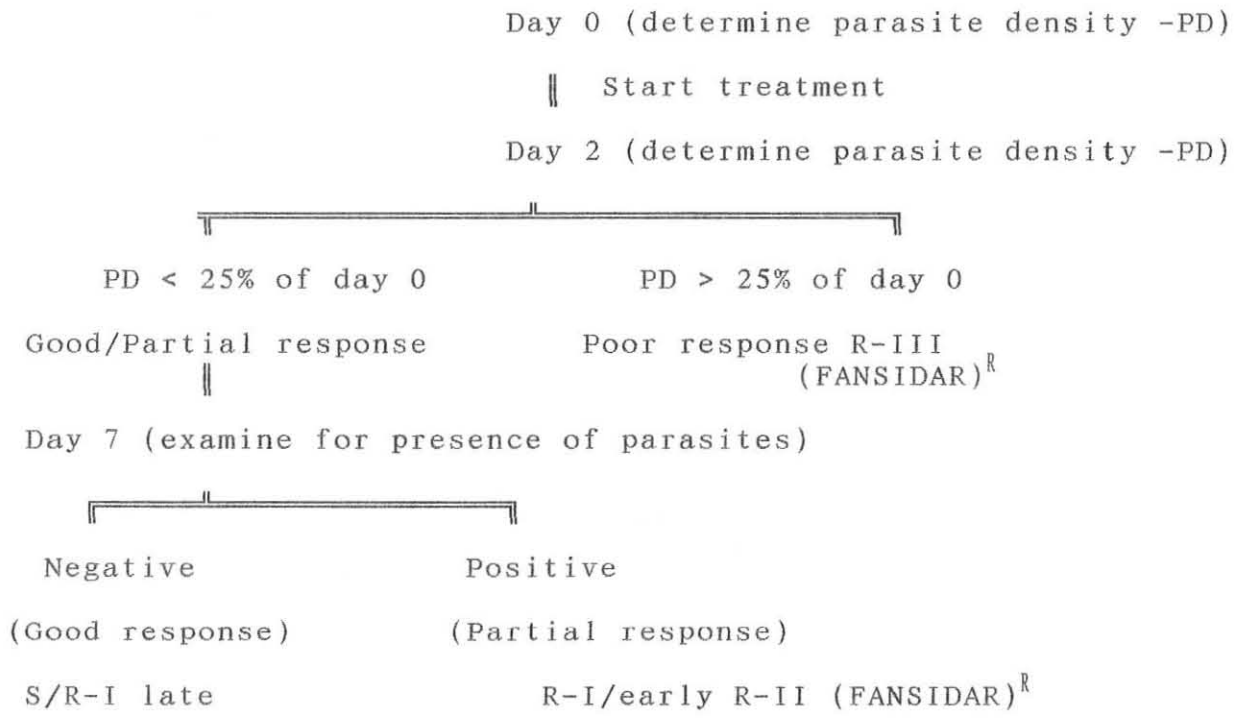
Day 2 . Blood smear taken

. 3rd dose of Cq administered

Day 7 . Blood smear taken

. Hx. and P/E

Figure III. Schema for Monitoring the Response of Malaria Infections to Treatment



Ethical Consideration

Verbally informed consent was obtained from the study population. All procedures, including the blood drawing, physical examination, and drug administration were explained in details before, at the beginning and during the investigation. The side effects of the procedures, if there were any (e.g. abdominal discomfort in pregnant women after fundal manipulation, Chloroquine related pruritus and side-effects of ferrous sulphate - such as nausea, vomiting and some dyspeptic symptoms), were explained fully together with some methods to relieve them.

Those who consented to participate were also informed that they may withdraw from the study at any time without fear of reprisal. Privacy of participants during interviews and physical examinations was fully safeguarded. Confidentiality of the data was protected by coding and other appropriate care of nominative study records.

All women who were found to be positive for malaria parasites were treated using chloroquine (cq) 25 mg/kg body weight over 3 days of which the first dose of 10 mg/kg was taken at the spot and the patient was observed for 60 minutes for any vomiting on day 0. An addition of analgesics such as ASA 600mg and Paracetamol 1000mg tablets were given for any fever and headaches accordingly. Anaemia of haematocrit $\leq$ 30% was treated by FeFol (Ferrous Sulphate 60mg + Folic Acid 200mcg) po tid for 10 days on day 7 for fear of recrudescence when given earlier.

Statistical Methods

Data entry and analysis was made using Epi- Info (version 5),and SASS statistical package, using conditional logistic

regression for matched set of samples (A one to one match and conditional logistic regression was done for 60 paired sets of pregnant and non pregnant malaria positive women matched for age, parity and place of residence during data collection period). Cross tabulations and tests of significance were made using chi square to analyze the difference in proportions. Student's t - test and analysis of variance were used to compare the difference between sample means. A multivariate analysis was carried out for different variables to determine their respective associations with parasite densities, clearance after treatment and haematocrit level.

III RESULTS

Out of the total 1,767 women who took part in the study (816 pregnant and 948 non pregnant), Ninety (11.03%) of the pregnant and 63 (6.64%) of the non pregnant women were found to have malaria parasitemia during the study period. The difference in prevalence was statistically significant (P = 0.001 by student's t -test). The rate in primigravidae was 212 (12.24%) and 314 (11.4%) in multigravidae. The overall plasmodial infection indicated 49.7% *Plasmodium vivax* and 50.3% *Plasmodium falciparum* occurrence. When primigravidae and multigravidae were compared in terms of their plasmodial infection, the former group had more infection with *Plasmodium falciparum* 57.7% vs.42.2% for *Plasmodium vivax*, However, the difference was not statistically significant. Mean age of the pregnant women was 24.08 $\pm$ 5.4 years while that of the non pregnant was 25.6$\pm$ 6.04 years (Figure IV). Mean parity of the pregnant women was 2.03$\pm$ 1.9 while that of their non pregnant counter part was 2.5$\pm$2.3. The difference in the above two variables was not significant (P= 0.09 and 0.19 by student's t - test respectively) and this shows that the two groups were comparable in terms of their ages and parity.

Mean gestational age of the pregnant women was 21.09$\pm$ 10.2 weeks with more or less comparable distribution over the three trimesters, that is, 28 (31.1%) in their 1st trimester, 29 (32.2%) in their 2nd trimester and 33 (39.7%) in their 3rd trimester. Eighty (88.9%) of the pregnant women were in their 32nd week of gestation or below it. Peak parasitemia occurred between gestational ages ranging from 12 - 24 weeks which correlates with usual second trimester severity of malaria parasitemia in pregnancy. There were

also few cases of high parasitemia between gestational ages of 28 - 32 weeks. In general, peak parasite loads were observed before 32 weeks of gestation.

Malaria Symptoms and Signs

Headache, muscle and joint pain,malaise, fever, and chills/ rigors were the most frequently observed clinical symptoms in both groups of women. There was no significant difference between both groups in terms of malaria related symptoms (Table I). Thirty one cases (20.2%) complained of minor symptoms such as dizziness, cough and jaundice. One pregnant woman presented in coma and died on the 3rd day of follow up. Two of the pregnant malaria positive women delivered normal babies weighing 2.75 Kg and 3.0Kg during the seven days of follow up.

Clinical signs at the time of examination on day 0 showed some differences between pregnant and non pregnant women (Table II). However, this difference was not statistically significant except for the conjunctival pallor.

Malaria and Haematologic Indices

The mean haematocrit value for pregnant women was 30.3± 4.67% and it was 33.4±6.18% in non pregnant women. This finding was significantly different between the two groups (P= 0.0001 by student's t - test). When a cut-off point of 37.7% HCT (which corresponds to haemoglobin level of 11.8 gm/dl, WHO's definition for altitude adjusted pregnancy anaemia; WHO - 1975) was taken to label a woman with malaria infection as anaemic or not, 83 (92.2%) of the pregnant and 47 (74.6%) of the non pregnant women were found to be anaemic and this difference was also statistically significant (P = 0.005) (Table III). Eighty (96.4%) of the pregnant and 44 (93.6%) of the non pregnant anaemic women were

found to have mild to moderate anaemia (Haemoglobin values corresponding to 11gm/dl - 9gm/dl). Six cases, (3.6% and 6.4% of the anaemic women from the respective group), were found to have severe anaemia (hct $\leq$23%).

The haematocrit level difference between the primigravid and multigravid women also showed the same pattern. Mean value in the former group was 28.77$\pm$3.2% and it was 31.86$\pm$5.4% in the latter group (P = 0.008) (Table IV). Among the pregnant women, anaemia was also common in the younger ages and lower gravidities. Comparison of these women by categorizing them in to gravidities $\leq$ 3 and $\geq$ 4, & HCT level $\leq$ 37.7% & $\geq$ 38%, showed the results depicted in (table V) with significant difference between the two groups (Fisher's exact P-value = 0.01, 2 - tailed; OR =10.60, 95% CI = 1.16,245).

Malaria Parasite Count

Pregnant women had a mean parasite count of 6,561$\pm$5259/micro litters of blood on day 0 and non pregnant women had a mean count of 4,598$\pm$4275/micro litters of blood on the same day. This difference is statistically significant (P = 0.01 by student's t - test). An arbitrary cut-off point of 10,000 asexual malaria parasites per micro litters of blood was taken to classify cases in to moderate and heavy parasitemia. Accordingly, 24 (26.7%) of the pregnant and 7 (11.1%) of the non pregnant women had heavy parasitemia (P = 0.03). There was also a difference, though it's statistically marginal, in asexual blood parasite load on day 0 between primigravid and multigravid women. The mean count in the former group was 8,290$\pm$5202/micro litter of blood and 5906$\pm$4782/micro litter of blood in the later group (P = 0.06 by student's t- test).

Response to treatment

The clinical assessment of response to treatment consisted of history-taking and physical examination on day 7. Accordingly, almost all of the women in both groups who had malaria symptoms on day 0 responded that they did not have them on day 7 (table VI). Among the objective signs, fever ($T^0 \geq 37.5^0c$) for example, was not recorded in any of those women with previous records of fever in both groups. As Iron therapy was provided on day 7, conjunctival pallor was dropped out of the evaluatory signs for response monitoring on day 7. The difference in spleen regression rate between the pregnant and non pregnant was also not significant (table VII).

The rate of asexual malaria parasite clearance on day 2 (S/R-I) in non pregnant women was faster. But this difference was statistically marginal (P = 0.07) (table VIII). The mean parasite count on day 2, however, indicted that the two groups were different significantly. The mean asexual malaria parasite count in the pregnant was 549/micro litter of blood while that of the non pregnant was 165/micro litter of blood (P = 0.01), which shows that pregnant women with parasitemia, as a group, were slower to clear their parasitemia than do their non pregnant controls following treatment with Chloroquine (Figure V). Twenty one (23.6%) of the pregnant and 9 (14.28%) of the non pregnant women had S/R-I late response pattern (Blood smear negative for malaria on day 7) according to Reckmann's treatment response classification. There were 4 (4.5%) pregnant women and one (1.58%) non pregnant woman who showed R-III (day 2 asexual parasite count > 25% of day 0) response pattern. One woman from each group proved to have a positive blood smear on day 7 (R-I/R-IIearly). One pregnant woman

with *Plasmodium falciparum* malaria on day 0 presented with coma and died on day 3 of the follow up.

Among the pregnant women who became blood smear negative on day 7, 9(42.8%) were primigravid, 7(33.3%) were multi gravid and 5 (23.9%) were women in gravida 2 or 3. The only R-I/R-II early response pattern documented among the pregnant group was by a primigravid woman. The R-III pattern was more or less distributed equally among the primigravid, gravida 2 and 3 and the multigravid.

Matched Statistical Analysis

A one to one matched logistic regression using SAS was worked out for 60 pairs of cases (pregnant), and controls (non pregnant), with malaria parasitemia. Variables that included clinical symptoms (Fever,Chills/Rigor, Malaise and Fatigue and Headache), and signs (Conjunctival pallor and Splenomegaly), with some laboratory findings (Haematocrit and asexual malaria parasitemia on day 0)(annex 2) were used to calculate the conditional logistic regression. Accordingly, our finding confirmed the crude analysis result done in Epi-Info for the whole group of malaria positive study subjects (153 women) except that the Odds Ratio and significance tests seemed to shift away from the Null value in some variables (table IX and X).

A Multivariate analysis was carried out to remove any confounding effect by controlling the species difference between matched sets of malaria cases and controls. There was a difference in the calculated Odds Ratio and 95% Confidence Intervals for majority of signs, symptoms and laboratory findings. Hence, clearly indicating that difference in plasmodial species infection results in varied degrees of clinical manifestation in pregnant and non pregnant women (table XI).

Malaria Prevalence by age in pregnant & non pregnant women, East Shoa, Ethiopia, 1993

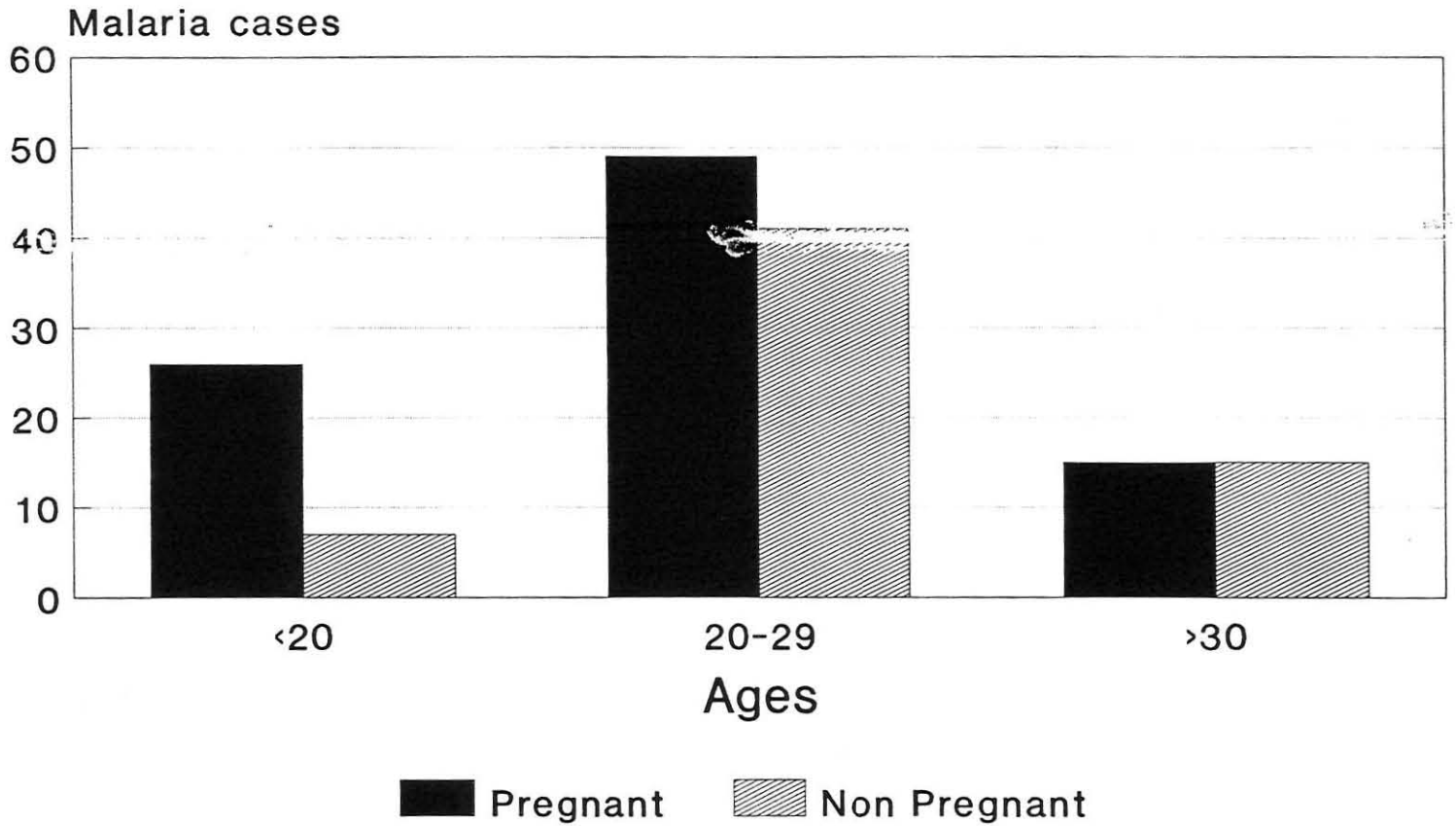


Figure IV

Table - I. Comparison of malaria related symptoms on the first day of examination by pregnant and non pregnant women. (East Shoa, Ethiopia 1993).

Symptoms	Frequency of complaint of each symptom		Odds Ratio	95% Confidence Interval
	Pregnant N = 90	Non pregnant N = 63		
Fever	73(81.1)	48(76.2)	1.34	0.57, 3.18
Malaise	79(87.8)	59(93.7)	0.49	60.12, 1.79
Chills/Rigor	52(57.8)	39(61.9)	0.84	0.41, 1.72
Headache	88(97.8)	60(95.2)	2.20	0.28, 19.77
Muscle/Join.pn	82(91.1)	58(92.1)	0.88	0.23, 3.22
Abdominal pain	20(22.2)	16(25.4)	0.84	0.37, 1.92
Nausea	30(33.3)	20(31.7)	1.07	0.51, 2.28
Vomiting	14(15.6)	9(14.3)	1.11	0.41, 3.04
Diarrhoea	5(5.6)	4(6.3)	0.87	0.19, 1.71

( ) = Percentages

Explain why such a table of symptoms in view of what are we looking for: prevalence of malaria (if it is severity of the disease, we should know)

Table II. Comparison of malaria related signs detected on day 0 in pregnant and non pregnant women. (East Shoa, Ethiopia 1993)

Signs	Frequency of signs		Odds Ratio	95% Confidence Interval
	Pregnant N = 90	Non pregnant N = 63		
1. Recordable Fever ($T^0 \geq 37.5^0 c$)	47(52.2)*	32(50.8)	1.06	0.52, 2.14
2. Conjunctival Pallor	60(66.7)	25(39.7)	3.04**	1.47, 6.33
3. Splenomegaly	27(30.0)	16(25.4)	1.26	0.58, 2.77

* Percentages
** Yates corrected P = 0.001 (2-tailed)

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Table III. Comparison of pregnant and non pregnant malaria positive women on their Haematocrit status (East Shoa, Ethiopia 1993).

	Pregnant	Non pregnant	Total
HCT ≤ 37.7%	83(92.2)	47(74.6)	130
HCT ≥ 38%	07(7.8)	16(25.4)	23
Total	90(100)	63(100)	153

OR = 4.04 ; 95% CI = 1.42,11.84 ; P = 0.005 (Yates corrected)

Table IV. Comparison of primigravid and multigravid malaria positive women in terms of their Haematocrit status. (East Shoa, Ethiopia 1993)

	Primigravid	Multigravid	Total
Hct ≤ 37.7%	26(100)	36(85.7)	62
Hct ≥ 38%	0	06(14.3)	06
Total	26(100)	42(100)	68

Fisher's exact P value = 0.03

Table V. Comparison of pregnant women by their gravidities for anaemia when there is malaria parasitemia (East Shoa, Ethiopia 1993).

	Gravida ≤ 3	Gravida ≥ 4	Total
Hct ≤ 33%	47(87.03)	22(61.1)	69
Hct ≥ 34%	07(12.97)	14(38.9)	21
Total	54(100)	36(100)	90

OR = 4.18 ; 95% CI = 1.33,13.53 ; P = 0.01

(Anaemia here was taken at haemoglobin level of 11.0 gm/dl without correcting for Altitude).

Table VI. Comparison of response to treatment between pregnant and non pregnant as documented using malaria related sign and symptoms (East Shoa, Ethiopia 1993).

Sign and Symptoms on day 7	Frequency of women without malaria sign and symptoms		Odds Ratio	95% Confidence Interval
	Pregnant N=88	Non pregnant N= 63		
Fever	88(100)	63(100)	--	---
Malaise	87(98.8)	61(96.8)	0.35	0.01, 5.16
Chills/Rigor	87(98.8)	61(96.8)	0.35	0.01, 5.16
Headache	88(100)	61(96.8)	0.00	0.00, 2.97
Muscle/Joint pain	88(100)	60(95.2)	0.00	0.00, 1.61
Abdominal pain	85(96.6)	62(98.4)	*2.19	0.19,56.91
Nausea	85(96.6)	59(93.6)	0.52	0.09, 2.92
Vomiting	88(100)	62(98.4)	0.00	0.00,12.73
Diarrhoea	87(98.8)	61(96.8)	0.70	0.02,26.69
Rec. T^0 ($\geq 37.5^0$c)	88(100)	63(100)	---	-----
Diaphoresis	88(100)	61(96.8)	0.00	0.00, 2.97
Splenomegaly (absent)	83(94.3)	60(95.2)		

( ) = Percentages

* Fisher's exact P- value = 0.6 (2 - tailed)

Table VII. Comparison of pregnant and non pregnant women on the basis of their splenomegaly on day 7. (East Shoa, Ethiopia 1993)

	Pregnant	Non pregnant	Total
Spleen Regressed	23(85.2)	14(87.5)	37
Spleen not Regressed	4(14.8)	2(12.5)	6
Total	27(100)	16(100)	43

OR = 0.82 ; 95% CI = 0.09,6.39 Fisher's P-value = 1.0

Table VIII. Comparison of malaria parasite clearance on day 2 in pregnant and non pregnant. (East Shoa, Ethiopia 1993)

	Pregnant	Non pregnant	Total
Parasitemia on day 2 cleared	62(69.7)	52(82.5)	114
Not cleared	27(30.3)	11(17.5)	38
Total	89(100)	63(100)	152

OR = 0.49 ; 95% CI = 0.2, 1.14 ; P = 0.07 (Un corrected)

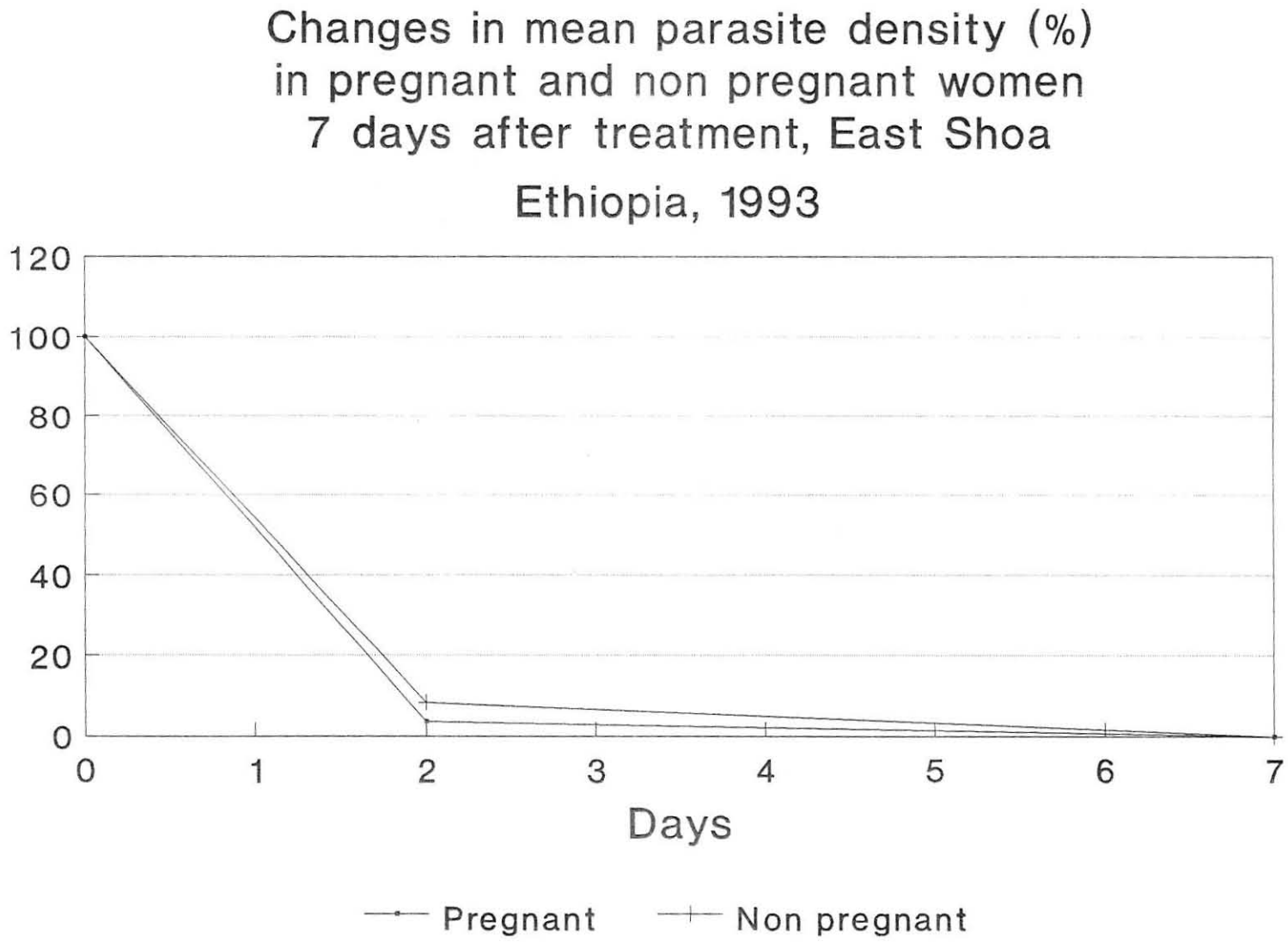
Changes in mean parasite density (%)
in pregnant and non pregnant women
7 days after treatment, East Shoa
Ethiopia, 1993
120
100
80
60
40
20
0
0
1
2
3
4
5
6
7
Days
Pregnant
Non pregnant

Figure V

Table IX. Estimated Coefficients of malaria related Signs and Symptoms for 60 matched sets of pregnant and non pregnant women on day 0. (East Shoa, Ethiopia 1993)

Variables	Parameter Estimate	Standard Error	Odds Ratio	95% CI
Fever	0.6190	0.4688	1.85	0.74,4.65
Headache	0.00	1.4142	1.00	0.06,15.9
Muscle/Joint pain	-0.9163	0.8367	0.40	0.07,2.06
Malaise	-1.2528	0.8018	1.85	0.74,4.65
Chills/Rigor	-0.2231	0.3354	0.28	0.06,1.37
Conjunctival Pallor	1.0986	0.4364	0.80	0.41,1.54
Splenomegaly	0.2412	0.4029	2.99*	1.27,7.05

* P = 0.5

Table X. Estimated Coefficients of Haematocrit and Parasite counts on day 0 for 60 matched sets of pregnant and non pregnant women. (East Shoa, Ethiopia 1993)

Variables	Parameter Estimate	Standard Error	Odds Ratio	95% CI
Hct Group‡	-1.5261	0.4934	0.22	0.08,0.57
Day 0 Parasite count Group‡‡	1.5041	0.5528	4.5	1.52,13.29

* = Grouping was made using a cut-off point of 0 -33% Hct and 34 - 47% Hct; P = 0.002

* = Grouping was made using a cut-off point of 0-10000/mic.lit. of blood and 10001 -21,276/mic.lit of blood malaria parasite count ; P = 0.006.

Table XI. Comparison of Adjusted (for species difference) and non adjusted Odds Ratios for malaria related signs,symptoms and lab. findings on day 0 for 60 matched sets of pregnant and non pregnant women
(East Shoa, Ethiopia 1993).

Variables	Crude Estimates		Adjusted Estimates	
	Odds Ratio	95% CI	Odds Ratio	95% CI
Fever	1.85	0.74,4.65	2.39	0.87,6.53
Headache	1.00	0.06,15.98	1.35	0.08,22.93
Muscle/Joint pain	0.40	0.07,2.06	0.38	0.07,2.04
Malaise/Fatigue	1.85	0.74,4.65	0.19	0.03,1.04
Chills/rigor	0.28	0.06,1.37	0.82	0.41,1.60
Conj.pallor	0.80	0.41,1.54	2.78	1.17,6.63
Splenomegaly	2.99	1.27,7.05	1.35	0.60,3.04
Hct group	0.22	0.08,0.57	0.20	0.07,0.55
Day 0 count gp.	4.50	1.52,13.29	7.00	2.03,24.15

IV DISCUSSION

This study was able to show that pregnant women in these localities during high malaria transmission season would have more malaria parasite prevalence (11%) as compared to the non pregnant women (6.6%). This result agrees with previous findings from Africa and South east Asian countries (6-8,11,13,30.34).

The prevalence rate of malaria in our study area is lower than the rate in other areas and does not correspond to the usual African malaria situation - holo endemicity and moreover, the study area even in the national endemicity categorization, is categorized under the high-land fringe type with usual low prevalence, but known for its catastrophic epidemics (Un published data, NOCMVD 1984 E.C.). Secondly, the 1986 E.C. (1993-94) experience of the National Office for Control of Malaria and other Vector Borne Diseases has been one of the few years when the problem is very minimal relative to the past few years experience, one reason being the less amount of rain fall in the year the study was conducted (Personal observation). Thirdly, the fact that the study area is one of the regions under surveillance by the local malaria control office, regular monitoring of epidemics and environmental malaria control activities might have offered better protection against Mosquito bites to the general population. Lastly, as the study was a community based and representative of the situation in its true sense, the prevalence rate might be lower than that observed by a passive case detection system based on self presenting cases.

Our finding correlates with earlier reports by B.J.Brabin (6), where he analyzed previous findings from Uganda, 1972 (4.8%)

and Gambia, 1978 (12%); a study from Zaire (13), reported 22% prevalence of malaria parasitemia in pregnant and 6.1% in non pregnant; S.L. Shola Purkar et al (30), found a rate of 14 malaria parasitemias per 1000 deliveries in their hospital and Liljestrand et al (21), from Mozambique, found 17% prevalence of malaria in pregnant women. This study has also shown that primigravid (12.24%) had more prevalence of malaria parasitemia as compared to either pregnant multiparae (11.47%) and non pregnant women although the difference from multigravidae was not significant. The above finding agrees with the works of different investigators from Africa and South east Asian countries (6,8,10,13,16,31-34).

The rate of *Plasmodium falciparum* (50.3%) in our study was not as high as in the case of most sub-Saharan and Tropical African countries which repot a rate of 80% or more infection (from Burkina Faso,(31); Thailand (34); Papua New Guinea,(24); and Cameroon (30). But still there are some reports like the one by S.L. Shola Purkar, 1989 India (30), who actually did studies in unstable malaria endemic regions of the country and found that the prevalence of *Plasmodium vivax* in the pregnant women was 60% and that of *P.falciparum* was 40%. Our finding also agrees with the usual proportion of plasmodial species distribution in the country or study area reported by areal malaria control office [Un published data MOH, (4)] which showed a proportion of 49.3% of *Plasmodium falciparum* and 51.4% *Plasmodium vivax* in a general population.

The pregnant women in the current study had a mean gestational age of 21.09 weeks with more or less proportional distribution of pregnant women over the three trimesters. This has enabled us to see the clinical features and response to treatment of pregnant women with malaria at different gestational periods as opposed to many studies that were unable to report on early pregnancies.

Nonetheless, our study had the limitations of being cross sectional since it failed to follow cases until delivery and document on pregnancy outcomes. The fact that 33(36.7%) of the pregnant women with malaria parasitemia were aged ≤ 20 years and 54(60%) of them were aged ≤ 25 years seemed to show that age influences the prevalence of malaria parasitemia (Figure IV). This finding is similar to other studies conducted elsewhere (6,8,10,31). Our finding of peak parasitemia in the 2nd trimester agrees with earlier study results by B.J. Brabin (6,12).

Majority of our study subjects complained of malaria symptomatology when diagnosis of parasitemia was made and this was more or less equally true in both pregnant and non pregnant women (table I). This indicates that our study subjects were less immune to the disease owing to their fewer exposure experiences. Whereas, most studies from stable malaria endemic regions reported the majority of malaria parasitemias to be asymptomatic (6,8-9,11 34,29).

Most malaria symptomatologies seen in our patients were more or less found in Yohannis's study result of severe malaria cases in Gondar Hospital (15). A majority of the complaints in the latter study included fever, chills,vomiting, headache, arthralgia and failure to communicate. In this study too, these symptoms were among the top five complaints of pregnant women with malaria. Malaria related signs seen on day 0 seemed to be different between the pregnant and non pregnant. As these findings were among the severity measures of malaria infection, pregnant women with conjunctival pallor and splenomegaly appeared to be higher in proportion than the non pregnant (table II). Conjunctival pallor could result from many causes leading to anaemia and there can also be inter- observer variations in interpreting this sign because of

its subjective nature. Therefore,the interpretation of its Odds Ratio between the two groups must be viewed with caution.

Splenomegaly was more detected in the pregnant women, though the difference was not statistically significant. The non significance of the test could be explained by the fact that gravid uteruses would mechanically make the abdomen tense that it becomes difficult to palpate moderate to mild splenic enlargements in mid to late pregnancies hence making the number of splenomegalies detected to be lower than expected in the pregnant women. Therefore, it's probably true that we missed these splenomegalies in the pregnant women. In areas where malaria transmission is stable and intense, splenic enlargements occur in the early gestations peaking at around 16 weeks (12) but these observations were made in a longitudinal study.

Our findings of malaria related anaemia rates (92.2%) and (74.6%) in pregnant and non pregnant women respectively, correspond with findings from Zambia and Zaire (7,28). The inflated rate of anaemia could possibly be due to the altitude adjusted anaemia cut-off point which is higher by 0.8gm/dl than the usual WHO definition of anaemia at sea level. Earlier study results on anaemia and pregnancy in Ethiopia (50) have however, indicated that pregnancy related anaemia is rare. Nonetheless, there were fewer reports on malaria related pregnancy anaemia and further investigation is recommended to confirm or nullify our results. Altitude correction was made due to the High-Land fringe ecology of the study area with altitudes ranging from 1800 - 2500 mts above sea level.

Primigravidae had more reduction in haemoglobin level than multigravidae and non pregnant women and this also agrees with other study results (8,28,34,24). Among the pregnant women, the younger age groups and those with fewer parities were at an

increased risk of developing malaria related anaemia compared to the multigravidae and older age groups. This would show that younger gravidities (most probably younger in age also) predispose to severe clinical forms of malaria infection. Our finding in this respect agrees with a study from rural Zaire (28).

Parasitemia was heavier in pregnant and primigravidae than non pregnant women as indicated also by other studies (6,8,11,13). The findings of some investigators from India and Burma differ a bit from our study result as they reported that the parasite density of pregnant women was significantly higher than those in the non pregnant,but this difference was not true when primis and multigravidae were compared (30,33).

As in most unstable malaria endemic regions, malaria symptoms were not differently complained by both groups and there was no difference in their response to treatment with Chloroquine. This finding could not be compared to other works owing to the lack of similar study in Africa or South east Asian countries.

The fact that majority of our patients cleared their parasitemia on day 2 bears a similarity with the findings from Cameroon (11), although their methodology was different in that they followed their patients for 4 weeks with Chloroquine prophylaxis after initial treatment of parasitemia. Thet Naing et al,(33), conducted a study of Falciparum Malaria and Pregnancy in 1985-86 in Burma (a hospital based study) in which they assessed response to treatment using Quinine and Amodiaquine. They found that although the pregnant and non pregnant cleared their parasitemia (especially in the Amodiaquine group) differently, this was not statistically significant. Hence, having a more or less similar epidemiologic situation in between the two countries (unstable malaria situation), our findings are somehow comparable.

Although mean parasite count on day 2, for pregnant and non pregnant women showed a significant difference, we felt that this was not the ideal variable to show how much of the initial parasitemia (day 0) have been cleared following treatment with Chloroquine in each case and what difference lies between the pregnant and non pregnant. The most important finding was the fact that there were 4 (4.5%) of the pregnant and one (1.58%) of the non pregnant who showed R-III pattern (day 2 malaria parasitemia > 25% of day 0) (OR = 2.88, 95% Confidence Interval = 0.29,69.44). As the sample size of our study was minimal, this Odds Ratio could not have been significant.

Two women (1.3%) of the total (pregnant and non pregnant) were slide positive on day 7 following treatment with Chloroquine and this finding was similar to the finding by F.Nosten et al from Thailand (34). *Plasmodium falciparum* infection has resulted in one death (Case Fatality Rate of 1.31%), in the pregnant group indicating its severity and catastrophic clinical outcomes. Primigravidae among the pregnant have exhibited severe forms of clinical infection, and accounted for the majority of slow response to treatment, though this was not statistically significant (OR =1.85, 95% CI =0.73,4.79), which we think might have been accounted for by the smaller sample size. Thet Naing et al, (33) found same Fever and Parasite clearance pattern among pregnant patients with malaria whether it was the first or the subsequent pregnancy. However, most studies from stable malaria endemic areas report that primigravidae clear their parasitemias at a slower rate than multigravidae following treatment with chloroquine (6,11).

V. Conclusion and Recommendations

Despite its limitations, this study was able to document that pregnant women are differentially affected by malaria infection than non pregnant. Primigravidae were at a higher risk among the study groups. The overall rate of *Plasmodium falciparum* in the pregnant and the non pregnant women did not appear to be as high as in many sub- Saharan African countries.

Many investigators have earlier proposed that the course of malaria in pregnancy differs in areas of stable and unstable malaria endemicity. This fact has been observed in our study subjects in that both groups were, in the majority of cases, symptomatic when malaria diagnosis was made and complaints were similar in pregnant and non pregnant cases.

Only some malaria related signs and symptoms such as conjunctival pallor, splenomegaly, mean asexual malaria parasite count and haematocrit level could show that pregnant women had more severe forms of plasmodial infection than non pregnant women. Age and parity were found to influence the clinical severity of malaria in pregnant women. Peak parasitemias were observed in the second trimester of pregnancy. Primigravidae had higher parasite rates, lower haematocrits, higher mean parasite counts and were slower to clear their parasitemias following treatment with Chloroquine.

Response to treatment also showed some difference between pregnant and non pregnant, the former group being slower to clear their parasitemias after treatment. However, the method used for monitoring response to treatment was not clearly an in-vivo test

system and resistance patterns mentioned here can not be taken as a reference or otherwise, as they lacked serious compliance monitoring system such as urine chloroquine metabolite tests.

Pregnant women and children are the major victims of malaria morbidity and mortality. Hence, the National Office for Control of Malaria and other Vector Borne Diseases must draft a national policy as to how to protect this group of population. However, as our study is the first of its kind, adequate information is not yet fully available. Therefore, we recommend a more longitudinal study with larger sample size conducted in highly endemic areas of the country.

Many African countries, having had adequate information about the magnitude of the problem locally, are currently even questioning the importance and cost-effectiveness of malaria chemoprophylaxis in pregnant women, particularly primigravidae, for fear of drug resistance and due to the low compliance observed in many studies. However, there is a lot to be done in this regard in our country and we recommend that the ministry, particularly the NOCMVD carry out such studies for improved quality and coverage of malaria control activities.

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Annex 1

Malaria in Pregnancy Data Collection Format

I IDENTIFICATION

Code Number ----- Address (kebele, keftegna, house no.--------

------. Age ----.Gravidity-----. Parity------.

Are you pregnant Yes---- No----. If no, urine pregnancy test result ------; If yes, what gestational age (using Last menustral period or approximate month of pregnancy--------.

Note : Urine for hCG will be testedne for those pregnant women in their 1st trimester.

II MEDICAL HISTORY

Did you take any medication in the last two weeks : Yes --- No ---, If yes, specify what type and for what ? (detailed information must be collected to rule-out medication for malaria).

Symptoms within the last 48 - 72 hours	Day 0		Day 7	
	Yes	No	Yes	No
Fever Malaise Chills/Rigor Headache Muscle/Joint pain Abdominal pain Nausea Vomiting Diarrhoea				

Others, specify ----------

III.PHYSICAL EXAMINATION

Signs	Day 0		Day 7	
	Yes	No	Yes	No
TO Diaphoresis Conjunctival Pallor Splenomegaly Fetal heart (+ve/-ve)				

Others, specify ----------.

IV. LABORATORY FINDINGS

Blood film result *Plasmodium falciparum* ( ) *Plasmodium vivax* ( ) Mixed Species ( )

Haematocrit value ---

V. TREATMENT AND FOLLOW UP

Days	Parastemia/mic. liter of blood.	Treatment			
		Chloroquine	ASA	FeFol	Fancidar
0	----------				
2	----------				
7	----------				

Note:-1. Parasite count is recorded after conversion for the average White Blood Cell count per micro liter of blood is done.

$$\text{Count} = \frac{\text{(No. of asexual parasite found x 8,000)}}{100}$$

2. Chloroquine is given four tablets on day 0, two each on day 2, and day 7.

3. Fancidar is provided by the principal investigator or when it is understood that patient needs it.

Annex 2

Variables used for Matched Statistical Analysis

Paired Code No.-------

Medical History Day 0

fever (Yes) (No)

malaise/fatigue (Yes) (No)

chills/rigor (Yes) (No)

headache (Yes) (No)

muscle/joint pain (Yes) (No)

Physical Examination Day 0

conjunctival pallor (Yes) (No)

splenomegaly (Yes) (No)

Laboratory Finding day 0

species of plasmodium

P. falciparum -----

P.vivax ------

mixed ------

haematocrit -------

Parasite Count

day 0 -----

day 2 -----

day 7 -----

Note : Same data was filled for both pregnant and non pregnant women simultaneously.

DECLARATION

I the undersigned, declare that this is my work and that all sources of material used for this thesis have been duly acknowledged.

Name ... Negassie Tafese ...

Signature .....

Place ... AAU/IE, CTD .....

Date of submission ... May 16th, 1994 .....

This thesis has been submitted for examination with my approval as University Advisor.

Dr. Ahmed Ali, ...

Advisor