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Prevalence, Morphological Types and Factors Associated With Anemia among Mothers Attending Antenatal Clinic at Mbarara Regional Referral Hospital, South Western Uganda

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ABSTRACT
Background: Anemia in pregnancy is defined as reduction in hemoglobin concentration of below 11.0g/dl in the first and third trimester, or hemoglobin below 10.5g/dl in the second trimester. Globally 50% of pregnant women have anemia, the prevalence is even higher in Uganda where 64% of pregnant women have anemia, and the factors that are associated with anemia in pregnancy include social demographic, nutritional and medical factors.
Objective: To determine the prevalence, morphological types, and factors associated with anemia in pregnancy among mothers attending antenatal clinic at Mbarara Regional Referral Hospital (MRRH).
Materials and methods: A total of 355 mothers were recruited in a cross-sectional study at MRRH, social demographic, obstetric and medical factors were collected. Samples of blood, urine and stool were examined for malaria parasitaemia, hemoglobin (Hb) estimate, features of urinary tract infection (UTI), hookworm infestation and fecal occult blood. Thin film was done for those with low Hb. The dependant variable was anemia. A univariate and multivariate analysis was done to determine factors associated with anemia in pregnancy. A p-value of 0.05 was considered significant.
Results: The prevalence of anemia among pregnant women attending antenatal clinic was 62.82%. Factors significantly associated with anemia were UTI (p=0.038), malaria parasitaemia (p=0.007), fecal occult blood (p=0.002) and use of hematinics (p=0.031). The common morphological type of anemia was found to be microcytic hypochromic anemia (76.68%).
Conclusion: Microcytic hypochromic anemia was the most common morphological type of anemia. There is need to strengthen policies on screening for anemia during pregnancy, hematinics supplementation and use of IPT (Intermittent Preventive Treatment) together with proper treatment of Malaria and UTIs.
Key words; Anemia, Pregnancy, Prevalence, factors, morphology, antenatal care

Background:

Anemia is defined as a reduction in the hemoglobin concentration below the expected normal range as adjusted for the person's age, gender and environment resulting into reduced oxygen carrying capacity (ACOG, 2008). During pregnancy there is more increase in the plasma volume than the red cell mass leading to hemodilution and the reduced hemoglobin concentration. This is physiological anemia of pregnancy, the peak of this effect being reached in the second trimester (Okeke et al., 2011). For this reason anemia in pregnancy is defined as reduction in hemoglobin concentration of below 11.0g/dl in the first trimester, or hemoglobin below 10.5g/dl in the second trimester or hemoglobin below 11.0g/dl in the third trimester (ACOG, 2008).

Anemia in pregnancy can be classified into three categories basing on the level of hemoglobin concentration: mild, moderate and severe anemia for hemoglobin ranging from 10.0-10.9g/dl (10.0-10.4g/dl for 2nd trimester), 7.0-9.9g/dl and below 7.0g/dl respectively (UDHS, 2011). Anemia can further be classified according to etiological causes. Commonly into three major categories which are: Nutritional anemia such as iron deficiency anemia, anaplastic anemia which is due to bone marrow diseases/defects and, hemolytic anemia which can be due to diseases like malaria.

Pregnant women who have anemia present with different symptoms which depend on the degree of anemia. These symptoms include general weakness, headaches, easy fatigability, palpitations and body swelling. Even in situations of mild forms of anemia it has been shown to influence the sense of well being, aggravate other disorders and affect work capacity (Haas et al., 2001). It also makes the body prone to infections (severe sepsis), and reduces the body's resistance to blood loss during delivery (Ross et al., 1996). The majority of the mothers in developing countries start pregnancy period with depleted iron stores; this makes their requirement even much higher in order to cater for the deficit. The total iron needed during pregnancy is around 1000mg. Fifty percent of this (500mg) is for the fetus and the placenta, 300mg is for the maternal use and 200mg is lost especially in the digestive system (Bruno de Benoist et al., 2001).

Rita Marahatta in 2007 reported poor pregnancy outcome in the anemic mothers than the non anemic ones. These poor outcomes were poor APGAR scores, low birth weight, preterm labor and intrauterine fetal demise

Globally 50% of the pregnant women are estimated to be anemic, prevalence of anemia in pregnancy is much higher in developing countries in the ranges of 56%-61% and at least 50% of these anemic mothers suffer iron deficiency anemia (WHO, 2002). According to world statistics in the Health Nutrition and Population statistics, Uganda is ranked the eighth with a 64.4% prevalence of anemia among pregnant women. UDHS 2006 estimates that the prevalence of anemia in pregnancy is 64%. The prevalence is much higher in malaria endemic areas and areas much affected by helminthic infestations, (Mukaya et al., 2009). The commonest type of anemia in pregnancy is iron deficiency anemia, it accounts for 76% of the anemia in pregnancy (Sifakisi et al, 2000).

In a matched case control study done in Uganda at Mbarara Regional Referral Hospital, the proportion of anemic pregnant mothers was found to be 84.4% and low serum iron levels correlated well with the low hemoglobin levels (Kiwanuka et al., 2000).

Anemia is common in prime gravidae than in the parous women (Anorlu R.I et al., 2006), especially in malaria endemic areas because the prime gravidae do not have immunity against malaria. Staying in rural areas and high parity with poor child spacing, poor employment, ethnic groups and Gestational age, late antenatal care booking, Low education level are associated with anemia (Desalegn, 1993, Yuan Xing et al., 2009, Anorlu RI et al., 2006 and Omoniyi et al., 2005).

Association between infections and anemia in pregnant women: Malaria is important factor for developing anemia most especially moderate anemia (Ouma et al., 2007). The use of insecticide-treated mosquito net (ITN) and fansidar prophylaxis for malaria among pregnant women is protective against anemia (Ndibaza et al., 2009). Anemia is still the commonest hematological problem in HIV infected population (Ramezani et al., 2008). This mainly is due to the effect of opportunistic infections, autoimmune states related to HIV, drugs like Azidothymidine (AZT) used in HIV/AIDS management and HIV itself, (Akimbo et al., 2009). UTIs are also associated with anemia in pregnancy (Geelhoed et al., 2006). Anemia is more in those with hookworm heavy infestation (Ndibaza et al., 2009, Mukaya et al., 2010).

Study rationale: Mbarara Regional Referral Hospital (MRRH) registers a high turn up of women attending ANC with an average daily ANC attendance of fifty mothers per day but the proportion that is anemic is not determined. Despite the fact that World Health Organization (WHO) and Ministry of Health (MoH) of Uganda recommend routine hemoglobin estimation to screen for anemia, this is not the practice at MRRH. According to the Hospital Audit Report of 2010, only 3% of all transfused patients had their Hb estimation done. This might be due to inadequate investigative practices by the hospital.

A supplementation dose of hematinics is given to all pregnant women attending antenatal care at MRRH even when they needed treatment doses because some of the anemic pregnant mothers are not diagnosed.
Despite the fact that iron supplementation for all pregnant women is routine, anemia still contributes significantly to morbidity and mortality at MRRH. According to the obstetrics and gynecology department report of 2010, 32 maternal deaths were registered in 2010 and four (12.5%) of which had presented with severe anemia in pregnancy at MRRH. Although anemia contributes to the causes of morbidity and mortality in our department, the prevalence and factors that are associated with anemia in pregnancy among women attending ANC at MRRH are not yet documented and this research intended to determine the prevalence, morphological types and factors associated with anemia among pregnant women attending ANC at MRRH. In low resource settings it will also help refocus resources on mandatory screening of pregnant women who are at higher risk of developing anemia.

The aims of the study were to determine the proportion of pregnant women with anemia in pregnancy; social demographic, obstetric and medical factors associated with anemia in pregnancy and the morphological types of anemia in pregnancy among mothers attending Antenatal Care at Mbarara Regional Referral Hospital.

Methods

Study Site: The study was conducted at MRRH in antenatal clinic. This hospital serves as a teaching hospital for Mbarara University of Science and Technology and is also a regional referral hospital. It is located in Mbarara municipality in Mbarara district about 250 km south west of the capital city Kampala. The catchment area of the hospital includes the neighboring countries of Rwanda, Northern Tanzania and Democratic Republic of Congo but for ANC, the majority of the mothers come from the nearby districts of Mbarara, Isingiro, Ntungamo, Kiruhura, Ibanda, Kamwengye, Lyantonde, Buhweju, Rubirizi, Sheema and Bushenyi all together with an

approximate total population of two million fifty two thousand, six hundred forty seven (Geopolitical Entities and Codes 2011).

The Antenatal Care Clinic is an out patient clinic however the very sick patients identified in the clinic are usually referred to the maternity ward for admission. According to the ANC clinic records the clinic attendance averages fifty patients per day from Monday to Friday. The hospital offers specialized services including a hematology section, and transfusion services are also available. This makes it suitable for handling even complicated cases.

Study Design: This was a cross sectional study where eligible women attending ANC were interviewed, blood, urine and stool samples taken off and a proportion of mothers with anemia and the morphological types of anemia were determined.

Study Population: The study population consisted of pregnant women attending ANC at MRRH and fitting the criteria for recruitment.

Inclusion Criteria: The study participants were confirmed pregnant women attending ANC at MRRH and consented to participate in the study.

Exclusion Criteria: Pregnant women who were too sick to participate in the study and those who could not consent.

Sample Size Determination: The sample size was determined by Kish and Leslie formula, (1965) and the total number of women recruited was 354.

Sampling procedure and Collection of laboratory samples: To ensure a good representation of the population study, systematic sampling was done. Routinely, patients were registered on arrival and then collected in a common room for triage and health education talk. I utilized the time of health education to emphasize the need to screen for anemia in pregnancy. Sampling was done and participants sent to the examination room where they found the research assistant. Every fifth patient on leaving the health education talk was taken up for recruitment if they met the inclusion criteria.

The study participants would be further informed and counseled about the study and informed consent got in writing. An Interviewer administered questionnaire would there after be filled through asking questions about the participant's sociodemographics, obstetric and medical factors followed by physical examination with major interest in pallor of mucus membranes and symphysiofundal length. 4mls of blood were collected for hemoglobin estimation, blood smears for malaria parasites and possible thin film incase the participant was found to be anemic. A stool sample was collected from all the participants to analyze for worm infestation and occult blood. Urine sample was also taken off from all participants and urinalysis done for features of urinary tract infection. We used midstream urine technique to collect the urine with the help of wide neck leak proof urine specimen containers. Severely anemic patients were referred immediately to admission ward for prompt management. The results of all the tests were availed to the in charge of ANC clinic so that the patients with derangements could be managed appropriately.

Data collection: Data was collected by the principal investigator and the research assistants by use of a pre-tested interviewer administered questionnaire. The questionnaire helped collect data on some of the study variables that included participant's social demographics, obstetric and medical factors, and the rest of the information/variables were got from the laboratory results that were obtained after examination.

The primary outcome variable is anemia: One was considered to have anemia if they had a low hemoglobin of less than 11.0g/dl during the first and third trimester and hemoglobin less than 10.5g/dl during the second trimester. In this study hemoglobin was estimated using Drabkins method;

The independent variables were age, place of residence, employment, ethnicity, level of education, gravidity, gestation age in completed WOA, presence of features of infection in respondent's urine, history of use of IPT by respondent, history of use of mosquito nets (ITNs) by respondent, history of blood transfusion of the respondent, presence of fecal occult blood in the respondent's stool, presence of Hookworms in the respondent's stool, presence of malaria parasites in the respondents blood, history of renal disease, history of liver disease, Respondent's HIV serostatus and use of AZT (Azidothymidine), presence of splenomegaly, BMI of the respondent.

Quality control: Research assistants were registered midwives selected from the ANC clinic and trained by the principal investigator to ensure uniform data collection. Qualified laboratory personnel were involved in laboratory sample collection and examination. All the laboratory tests were done at medical ward MRRH side laboratory and every twentieth sample would be taken to an independent laboratory (MRRH main laboratory) for comparison of results. The questionnaire was pre tested before the study began.

Data entry and analysis: The filled questionnaire would be checked for completeness before they were put in a lockable cupboard. Data was entered into Microsoft Access Computer Programme by the PI and then exported to STATA 11 for analysis with the help of a biostastician. The number of participants that were anemic under each independent variable was determined and presented inform of frequencies and percentages. Bivariate and Multivariate analysis was done to identify the factors that were associated with anemia. The association was measured using Odds ratio, and a P-value of 0.05 was considered significant.

Ethical considerations: The proposal was presented to the department of obstetrics and gynecology at MUST and ethical approval was sought from Mbarara University Institutional Review Committee.

Study limitations: It was not possible to analyze diet because the participants were mainly on the same diet.

RESULTS OF THE STUDY

A total of 355 pregnant women were recruited in the study of which 223 participants were found to be anemic.

Prevalence of anemia

Hb	***≤14WOA (07.04%)***	***15-28WOA (37.75%)***	***≥29WOA (55.21%)***	***Total***
Normal	10(40%)	46(34.33%)	76(38.78%)	132(37.18%)
Anemic	15(60%)	88(65.67%)	120(61.22%)	**223(62.82%)**
Total	**25(100%)**	**134(100%)**	**196(100%)**	**355(100%)**

Participants who were between 15-28 WOA were considered anemic if their Hb was below 10.5gm/dl while the rest were considered anemic if their Hb was below 11gm/dl, and the prevalence anemia was found to be 62.82%.

Factors associated with Anemia in pregnancy (on bivariate analysis).

Factor	***OR***	***95% CI***	***P-value***
Having UTI	1.95	1.158-3.309	0.0105
Positive B/S	7.45	2.940-12.919	0.0252
Use of IPT	0.72	0.453-1.153	0.1713
Low Income	2.69	1.292-5.602	0.0059
Use of Hematinics	0.54	0.153-2.791	0.0283
Taking AZT	1.30	0.691-16.091	0.1506
Having Hookworms	0.76	0.358-1.681	0.4640
History of APH	0.91	0.411-2.004	0.8110
Contraception use	0.64	0.414-0.991	0.0439
ITN use	0.79	0.485-1.730	0.7850
Occult blood (stool)	3.77	1.078-13.212	0.0254
BMI	1.11	0.829-1.400	0.4895

Factors significantly associated with anemia in pregnancy before adjusting for confounders were found to be UTI (p=0.0105), Malaria parasitaemia (positive B/S) with p=0.0252, low income (<30 dollars per month) with p=0.0059 and having Occult blood in stool (p=0.0254). Factors that were found protective against developing anemia included use of hematinics during pregnancy (OR=0.54) and history of prior use of family planning contraception (OR=0.64).

Factors independently associated with Anemia in pregnancy (Multivariate analysis)

Factors associated	**Coefficient**	**95% CI**	**P-value**
Positive B/S	3.9951	2.82302-8.27258	0.007
Having UTI	1.5703	0.58004-3.18941	0.038
Occult blood (stool)	2.9199	1.85660-6.02558	0.002
Low Income	0.23192	0.07361-13.90231	0.094
Contraception use	-0.11764	-0.27261-15.37320	0.136
Hematinics use	2.54881	0.58729-8.77532	0.031

Following adjusting for confounding factors, the factors significantly associated with anemia in pregnancy in our study population were; malaria parasitaemia (p=0.007), UTI (p=0.035), Occult blood in stool (p=0.002), and use of hematinics was protective against developing anemia (p=0.031).

The morphological types of anemia

Red cell morphology was determined by doing a thin film for all the blood samples of the participants that were found to have anemia, and the results are as shown.

RED CELL MORPHOLOGY: Macrocytic hypochromic anemia constituted 8.7%%, microcytic hypochromic anemia-76.7%, normocytic normochromic anemia-13.5% and normocytic hypochromic anemia-1.2%.

Majority of the participants had microcytic-hypochromic anemia (probably due to Iron deficiency anemia) and a number of thin films had a dimorphism picture especially the microcytic ones; some had poikilocytosis and anisocytosis. This was reported as spherocytes, ovalocytes, pencil cells and stomatocytes amidst the general appearance as microcytic, macrocytic, normocytic, hypochromic, hyperchromic and normochromic.

DISCUSSION

In this study we found that the prevalence of anemia in pregnancy was 62.82%. Urinary tract infections during pregnancy, having malaria parasitaemia, passing of Occult blood in stool, and failure to use hematinics were significantly associated with anemia. The patient social-demographics were not found to be associated with anemia.

Prevalence of anemia

We found that the prevalence of anemia among pregnant women attending antenatal care at Mbarara Regional Referral Hospital was 62.82% and this is comparable to 64% of the UDHS report of 2006. The same was found in a cross-sectional study done in Mulago Uganda National Referral Hospital that the prevalence of anemia was 64.6% (Mukaya et al., 2009). In our study anemia was common among pregnant women who had UTI (OR 1.95), malaria parasitaemia (OR 7.45), Occult blood in stool (OR 3.77) and those who did not use hematinics during antenatal care.

Factors associated with anemia in pregnancy

These were found to be mainly infections (malaria and urinary tract infections), occult gastrointestinal bleeding and lack of supplementation/treatment with hematinics during pregnancy.

Urinary Tract Infections: There was a significant relationship found between urinary tract infections and anemia in pregnancy with p value of 0.038 and CI of 0.58004-3.18941. This is in agreement with a prospective case control study done in Ghana (Geelhoed et al., 2006) The same was found in a cross sectional study done in northern Tanzania (Hinderaker et al., 2002). Urinary tract infections are common during pregnancy, it is estimated that 30% of women have UTI (Duarte et al., 2008). This is comparable to the finding in this study that the proportion of women who had UTI was 26.67%.

The mechanism by which UTIs cause anemia is not clearly understood but there is hemolysis secondary to lipopolysaccharide induced red blood cell membrane damage (Areej et al., 2006) Also substances, known collectively as “leukocyte endogenous mediator (LEM)” or “leukocyte pyrogen”, are released by neutrophils and macrophages following infections and chronic inflammation and these substances interfere with erythropoesis (Anemia of chronic disease available at www.innvista.com/ailments/anemias/anemia-of-chronic-dis).

Malaria parasitaemia (positive blood smear for malaria parasites): The proportion of mothers with positive B/S who had anemia was high (92.31%) and on further analysis there was a significant association between anemia and malaria parasitaemia with a p value of 0.007 and CI of 2.82302-8.27258. This was the same finding in a prospective study which was conducted in Kisumu(western Kenya) (Ouma et al., 2007) and (Agan et al., 2010). In the same study, use of insecticide-treated mosquito net (ITN) was found to be protective against anemia and this is in agreement with the finding in our study that the big proportion (64.58%) of the participants who never used ITN had anemia.

Occult blood in stool: Twenty one study participants were found to have Occult blood in stool, though this prevalence seems low a big proportion (85.71%) of these participants had anemia. On bivariate analysis a pregnant woman with Occult blood in stool has 3.77 times the risk of developing anemia than their counterpart without Occult blood in stool. This agrees with Nakama et al., 2000, and also relates to hookworm infestation (Mukaya et al., 2009). In our study the prevalence of worms was 9.86% and 57.14% had anemia

Association between anemia and use of Hematinics: Use of Hematinics is protective against developing anemia in pregnancy as displayed by our finding with p value of 0.031 and CI of 0.58729-8.77532. This agrees with the finding in a study done to assess the effect of routine iron supplementation on anemia in pregnancy where it was found out that daily iron supplementation reduced the risk of developing anemia at term by 73%, (Yakoob et al., 2011). It is important to diagnose anemia in pregnancy so that a higher than the usual supplementation dose of iron can be given (ACOG, 2008).

The morphological type of anemia: The study revealed that the commonest morphological type of anemia among the participants was microcytic hypochromic anemia (76.68%), and matches the picture of iron deficiency anemia. This agrees with the study done in Mulago Uganda where they found that the common type of anemia was microcytic hypochromic anemia (Mukaya et al., 2009), and Jalilolghadr et al., 2004 in Iran with a prevalence of 77%. This prevalence can be compared to our finding that the proportion of mothers who had microcytic anemia was 76.68%.

CONCLUSIONS: The prevalence of anemia among pregnant women attending ANC at MRRH was 62.82% and this is high. The commonest morphological type of anemia was microcytic-hypochromic anemia though some had a dimorphism picture. The factors significantly associated with anemia in pregnancy were; (i) having malaria parasitaemia, (ii) passing of Occult blood in stool and, (iii) having a urinary tract infection. Taking of hematinics during pregnancy period was found to be protective against developing anemia.

RECOMMENDATIONS: There is need to investigate why routine screening for anemia by hemoglobin estimation for all pregnant women during ANC is not done. Examining for pallor alone is not effective. For all the patients that are found to be anemic in this hospital, it would be good that the following laboratory investigations be done routinely; (i) Blood smear for malaria parasites, Urinalysis for features of UTI, Stool analysis including testing for Occult blood.

Since the prevalence of anemia is high it would be useful to study its effect on maternal and perinatal outcome at MRRH.

There is need for more Health Education for the public to be more aware of the importance of hematinics and the pregnant women to be further encouraged to take hematinics.

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