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Title: Adolescent pregnancy and the risk of *Plasmodium falciparum* malaria and anaemia—a pilot study from Sekondi-Takoradi metropolis, Ghana

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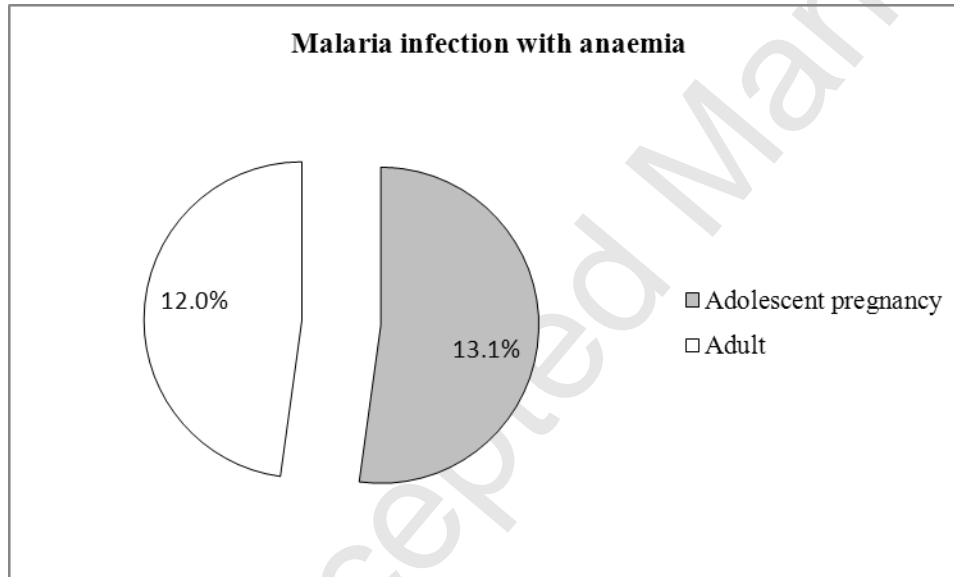
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Graphical abstract**Summary**

Taken together, these data suggest that adolescent pregnant girls were more likely to have malaria and anaemia compared to their adult pregnant counterpart



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4 Short communication
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9 **Adolescent pregnancy and the risk of *Plasmodium falciparum* malaria and anaemia – a**
10 **pilot study from Sekondi-Takoradi metropolis, Ghana**
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ABSTRACT

The problem of malaria in adolescence has been surpassed by the immense burden of malaria in children, most especially less than 5. A substantial amount of work done on malaria in pregnancy in endemic regions has not properly considered the adolescence. The present study therefore aimed at evaluating the prevalence of *Plasmodium falciparum* and anaemia infection in adolescent pregnant girls in the Sekondi-Takoradi metropolis, Ghana. The study was carried out at four hospitals in the Sekondi-Takoradi metropolis of the western region of Ghana from January 2010 to October 2010. Structured questionnaires were administered to the consenting pregnant women during their antenatal care visits. Information on education, age, gravidae, occupation and socio-demographic characteristics were recorded. Venous bloods were screened for malaria using RAPID response antibody kit and Geimsa staining while haemoglobin estimations were done by cyanmethemoglobin method. The results revealed that adolescent pregnant girls were more likely to have malaria infection than the adult pregnant women (34.6% versus 21.3%, adjusted OR 1.65, 95% CI, 1.03-2.65, $P = 0.039$). In addition, adolescent pregnant girls had higher odds of anaemia than their adult pregnant women equivalent (43.9% versus 33.2%; adjusted OR 1.63, 95% CI, 1.01-2.62, $P = 0.046$). Taken together, these data suggest that adolescent pregnant girls were more likely to have malaria and anaemia compared to their adult pregnant counterpart. Results from this study shows that proactive adolescent friendly policies and control programmes for malaria and anaemia are needed in this region in order to protect this vulnerable group of pregnant women.

Keywords: Adolescent pregnancy; Malaria; Anaemia; IPTp-SP; Ghana.

1. Introduction

Adolescence is an age of opportunity for children with about 1.2 billion individuals aged 10-19 years (UNICEF, 2011). Adolescent pregnancy (usually within the ages of 13-19) is a worldwide concern particularly in Latin America, the Caribbean, Asia, sub-Saharan Africa and high-income countries like the United States of America (WHO, 2011). In low- and middle-income countries about 10% of girls become mothers by age 16 years, with the highest rates in sub-Saharan Africa, south-central and south-eastern Asia (Bongaarts and Cohen, 1998; WHO, 2011). About 16 million women between the ages of 15 and 19 years old give birth each year, comprising 11% of all births worldwide (WHO, 2011). A large proportion of adolescents in the developing countries enter pregnancy with a poor nutritional status with the likelihood of sub-optimal dietary intake during pregnancy (King, 2003). Furthermore, the growing foetus competes with the growing adolescent mothers for limited dietary nutrients (King, 2003; Scholl et al., 1994).

Pregnancy during adolescence has been found to increase the risk of adverse birth outcomes, poor foetal growth, infant and maternal health, and mortality (Kurth et al., 2010; Malamitsi-Puchner and Boutsikou, 2006; Stewart et al., 2007). In addition, many health problems have been associated with the negative consequences of adolescence pregnancy including anaemia, malaria, human immunodeficiency virus (HIV) and other sexually transmitted infections, postpartum haemorrhage and mental disorders, such as depression (WHO, 2011). In many malarious countries, 50% of first pregnancies occur during adolescence (Lalloo et al., 2006) often associated with maternal and foetal consequences (Steketee et al., 2001) who are prone to being primigravidae (Desai et al., 2007; Fleming, 1989). In 2000, World Health Organization (WHO) ranked malaria as the second most common cause of death in adolescence,

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4 accounting for 7.4% of all adolescent deaths globally (WHO, 2000). Adolescents seem to have
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6 higher risk of severe disease compared with other age groups. In KwaZulu-Natal, the peak age of
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8 admission with malaria was in adolescents with case fatality over the age of 12 years (Soni and
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10 Gouws, 1996). Severe morbidities and mortalities have been reported from different malaria
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12 transmission settings among adolescent (Ejov et al., 1999; Luxemburger et al., 1997; Singh et al.,
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14 1992).

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19 In Ghana, some works have been done on malaria and anaemia in pregnancy but not
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21 much were specifically targeted to adolescent pregnancy. Previous data showed that the
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23 prevalence of anaemia ranged from 34% in a district in Accra to 75% in Kassena-Nankana
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25 district in the northern region of Ghana (Clerk et al., 2009; Engmann et al., 2008). Similarly,
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27 much work have been done on the prevalence of malaria in pregnant women, with 19.7%
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29 reported in Dagme-west district of Accra and 47% in Kassena-Nankana district (Clerk et al.,
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31 2009; Ofori et al., 2009). These observations led us to this pilot study with view of determining
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33 the association between adolescent pregnancy, anaemia and malaria in Sekondi-Takoradi
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35 metropolis of the western region of Ghana.
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2. Materials and methods

This study was carried out in the Sekondi-Takoradi metropolis, Ghana. Sekondi-Takoradi, comprising the twin cities of Sekondi and Takoradi, is the administrative capital of the western region of Ghana with a land area of 385 square kilometres. It is Ghana's fourth largest city and an industrial and commercial centre with a population of about 335,000. The metropolis is an urban centre surrounded by towns and villages. Temperatures are high with an average of 22°C. It has a mean annual rainfall of 2.350 millimetres, which is experienced heavily in May and June with the minor rains occurring between September and October. Like most areas in Ghana, the metropolis is endemic for malaria.

Pregnant women attending their antenatal care (ANC) visits were strategically sampled from four hospitals in the metropolis with the intention of recruiting pregnant women from sub-urban and rural communities of the city. These hospitals included Effia-Nkwanta Regional Hospital, Essikado Hospital, Takoradi Hospital and Jemima Crentil Hospital. This cross-sectional study was carried out from the month of January to the month of October, 2010. Each facility was visited once every week on their routine antenatal days. Pregnant women, cross-checked with ultra-sound or with clinical evidence of pregnancy, were included in the study while pregnant women with significant bleeding were excluded from the study. Each consenting pregnant woman was asked of her demographic characteristics, past and present obstetrics history. History of fever and any other illness during the pregnancy were asked. HIV screening for the pregnant women were performed and status of the women were obtained from the preventing mother to child transmission (PMTCT) clinic in the hospital. Sero-statuses were determined by applying the national diagnostic algorithm of two rapid antibody tests and western blot confirmation while indeterminate cases were confirmed in the reference laboratory of the

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4 Regional Hospital. Five mls of venous blood were collected from the pregnant women by a
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6 trained laboratory technician from the median cubital vein. Blood samples were collected into an
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8 EDTA bottle and temporarily stored in an ice chest and were transported to a designated
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10 reference laboratory for same day analysis and storage.
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14 Laboratory diagnosis of malaria was performed using fast RAPID response antibody kit
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16 (Premier Medical Corporation Ltd) and Geimsa staining. The brand of the RAPID response kit
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18 was specific for the detection of *Plasmodium falciparum* antigens. The presence of two lines in
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20 the text kit well indicated positive for *P. falciparum* malaria. The RAPID response kit contained
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22 a membrane strip pre-coated with monoclonal antibody specific for histidine rich protein 2
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24 antigen of *P. falciparum*. For proper confirmation of malaria parasites, thick and thin smear with
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26 Geimsa staining were performed and examined microscopically using 100 power fields under oil
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28 immersion. Malaria parasites were counted against 200 leukocytes, read independently by two
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30 competent microscopists and where they had discordances, a third microscopist reassessed the
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32 slide. Malaria diagnosis was defined on the identification of any asexual blood stages of *P.*
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34 *falciparum* species in the thick and thin smears while a slide was pronounced negative when 100
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36 high power fields have been examined using x100 oil immersion objective lens. Haemoglobin
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38 estimation was performed using cyanmethemoglobin method (Bhaskaram et al., 2003). Anaemia
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40 was defined based on WHO criteria haemoglobin levels of <11 g/dL (WHO, 1989). All
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42 laboratory personnel were blinded to the status of patients. Written informed consents were
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44 received from the recruited pregnant women and ethical clearance for the study was received
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46 from the Ghana Health Service Ethical Committee.
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55 Characteristics of the pregnant women were analysed by Pearson's Chi-square (χ^2) tests
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57 and ANOVA for the comparisons of proportions and mean, respectively. The analyses were
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4 focused on adolescent pregnant girls who are at increased risk of adverse birth outcomes
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6 (Malamitsi-Puchner and Boutsikou, 2006). To examine the independent effect of anaemia and
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8 malaria among the pregnant women, adolescent pregnant girls were compared with their adult
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10 pregnant women counterpart and univariable and multivariable logistic regression analyses were
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12 performed. The final parsimonious multivariable model was selected after an assessment of
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14 regression assumptions and confounding, based on biological plausibility and the results of
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16 univariable analysis. Associations were quantified using odds ratios (OR) with 95% confidence
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18 intervals (CI) to measure the strength of association. All tests were two-tailed and statistical
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20 significance was defined as $P < 0.05$. Data and statistical analyses, including binary logistic
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22 regression, were performed using IBM SPSS Statistics version 17.0 (SPSS Inc., IL, USA).
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3. Results

In total, 866 pregnant women were screened for this epidemiological survey. Adolescent pregnant girls represented 12.4% (107/866) of pregnant women in this study population while 87.6% (759/866) were adult pregnant women. Table 1 describes the characteristics of the pregnant women stratified by adolescence and adult. There were significant differences in mean age of the pregnant women as the mean age of the adult was higher [27.35 years (95% CI, 26.99-27.70) versus 17.79 years (95% CI, 17.56-18.01)]. The median age of the adolescent pregnant girls was 18 years (range, 15-19) while that of the adult pregnant women was 27 years (range, 20-46). There were no statistical differences with mean haemoglobin, trimester, HIV status and the use of intermittent preventive treatment of malaria during pregnancy (IPTp) with sulphadoxine-pyrimethamine (SP) between the adolescent pregnant girls and adult pregnant women.

However, there were statistical differences with education, gravidae, malaria infection and maternal anaemia in the proportion of adolescent pregnant girls compared to the adult pregnant women. All the adolescent pregnant girls were primigravidae 100% (107) while 53.4% of the adult pregnant women were primigravidae, 36.4% secundigravidae and 10.2% were multigravidae. The proportion of adolescent pregnant girls with malaria infection was 34.6% while 65.4% were malaria negative. Similarly, the proportion of adult pregnant women with malaria infection was 21.3% while those without malaria infection were 78.7%. Anaemia was detected in 43.9% of the adolescent pregnant girls while 56.1% had haemoglobin >11 g/dL. In the adult pregnant women, 33.2% had anaemia and 66.8% were non-anaemic. Though no statistical significance differences were observed with IPTp-SP use and SP doses between the adolescent pregnant girls and their adult pregnant counterpart, 24.5% (13/107) of adolescent

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4 pregnant girls who used IPTp-SP had malaria infection as compared to 17.8% (68/759) of the
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6 adult pregnant women. However, the proportion of malaria infection among those who did not
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8 use IPTp-SP was 52.3% in adolescent pregnant girls and 33.8% among the adult pregnant
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10 women [Table 1].
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14 In order to assess if there are any notable association between malaria infection, anaemia
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16 and adolescent pregnancy, univariable and multivariable analyses were performed. Univariable
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18 analysis showed that the adolescent pregnant girls were more likely to have malaria infection
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20 than the adult pregnant women (OR 1.95, 95% CI, 1.26-3.01, $P = 0.003$) [Table 2]. After
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22 adjusting for maternal age, education, gravidae and IPTp-SP as potential confounders in the
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24 multivariable binary logistic regression analysis, the statistical significant effect remained
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26 (adjusted OR 1.65, 95% CI, 1.03-2.65, $P = 0.039$). Logistic regression analysis was used to
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28 assess the effect of educational level attained on the study population. Although adolescent
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30 pregnant girls were more likely to have primary and secondary education in univariable analysis,
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32 the statistical inference disappeared in multivariable logistic regression analysis signifying
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34 confounding. Interestingly, when pregnant women that did not receive IPTp-SP were selected for
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36 further analysis, adolescent pregnant girls were twice likely to have malaria infection than the
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38 adult pregnant women (adjusted OR 2.33, 95% CI, 1.14-4.76, $P = 0.020$) [Table 2]. Furthermore,
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40 multivariable regression analysis of the probability to have maternal anaemia was performed in
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42 the study population using the same logistic regression model. The data revealed that adolescent
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44 pregnant girls had higher odds of anaemia than the adult pregnant women (adjusted OR 1.63,
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46 95% CI, 1.01-2.62, $P = 0.046$) [Table 2]. When only the malaria infected pregnant women with
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48 anaemia were selected for further analysis, 13.1% (14/107) were adolescent pregnant girls while
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4 12% (91/759) were adult pregnant women and no statistical significant differences were
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6 observed (Figure 1).
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4. Discussion

Our results revealed that the overall proportion of malaria infection and anaemia were statistically higher among the adolescent pregnant girls suggesting a significant public health peril. Although the aetiology of anaemia is multifaceted; due to malnutrition, iron deficiency, HIV, malaria and other parasitic diseases (hookworm, ascaris and schistosomiasis), vitamins B₁₂ and A deficiency, foliate deficiency, thalassemia and sickle cell anaemia (WHO, 2008), our data indicated that adolescent pregnant girls were more likely to suffer from malaria infection and anaemia than the adult pregnant women. Anaemia is a common finding in pregnant women worldwide especially in sub-Saharan Africa (WHO, 1992) and it is a very common complaint seen in nearly all pregnant women worldwide. During pregnancy, many women lack the sufficient amount of iron needed for the second and third trimesters as the body needs more iron than it has available. Evidence showed that child-bearing during adolescence hinder growth and nutritional status of the adolescent girls (Rah et al., 2008). Nutrition in pregnancy demands extra care due to the growing foetus, placing a high demand on the mother's energy, protein and micronutrients (Butte and King, 2005). Good nutrition is the best way to prevent anaemia during pregnancy but young still-growing women are highly disadvantaged due to competition for nutrients between the mother and the foetus (Scholl et al., 1994).

The interaction between malaria infection, anaemia and nutritional deficiencies is complex but remains important to an adolescent pregnant girl. The higher risk seen in adolescent pregnancy could be compounded by their limited access to antenatal care services, neglect in addressing sexual health issues, immaturity, and lack of nutritional and educational initiatives. The extent of the contribution of malaria and anaemia in adolescent pregnancy remains uncertain and additional research would be needed particularly among adolescent boys to further

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4 understand their interaction. Adolescents in malaria stable region, like in Ghana, are still
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6 developing their immunity to malaria, and are still susceptible to malaria infection compared to
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8 older adults that have fully acquired partial immunity to malaria. They enter pregnancy with a
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10 double risk of malaria and anaemia. The higher risk of malaria and anaemia in adolescent
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12 pregnancy observed in this study is in agreement with the earlier findings in northern district of
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14 Ghana (Clerk et al., 2009). In a similar study, malaria and anaemia were common among
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16 pregnant women especially primigravidae (Enato et al., 2009). In another study, 44.4% of
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18 adolescent pregnancies had one form of pregnancy-associated complication including anaemia in
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20 a case-controlled study (Adeyinka et al., 2010). Furthermore, heavy burden of malaria and
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22 anaemia were found to be significantly higher in communities with intermittent access to health-
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24 care (Grenfell et al., 2008). The findings of this study and others call for an urgent contemplation
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26 of the problem of adolescent pregnancy in the country with view of instigating programmes that
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28 would benefit adolescence age.
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36 Concerted effort cutting across malaria, anaemia, HIV, reproductive information,
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38 consequences and intervention should be accentuated as most control programmes do not
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40 specifically involve adolescents (Grietens et al., 2010; Lalloo et al., 2006). Targeted education
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42 to women of child-bearing age need more emphasis as evidences have shown their influential
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44 impacts on malaria control (Hwang et al., 2010; Iriemenam et al., 2011). In addition, proper
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46 monitoring of this target group is essential as timing of malaria infection seem to be harmful at
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48 the beginning and at the end of pregnancy (Huynh et al., 2011). Data showed that in areas with
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50 high proportion of first pregnancies to young girls, ANC for pregnant adolescents need to be
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52 improved to avoid adverse birth outcome and morbidity (Brabin et al., 1998). Recently, a study
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54 from Ghana indicated that IPTp-SP regimen is useful in preventing malaria and anaemia among
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4 pregnant women (Wilson et al., 2011). This intervention needs to be scaled-up as our data and
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6 others (Gies et al., 2009; Grietens et al., 2010) revealed the need in adolescent pregnancy. Major
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8 limitations of this study include the lack of CD4 count as potential confounders and none
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10 measurement of other likelihood causes of anaemia. In addition, low birth weight was not
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12 measured as we did not follow the pregnant women till delivery. The small sample size of
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14 adolescent pregnant girls may have limited our analysis. In the view of these, our results should
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16 be interpreted with reference to the observed.
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21 In conclusion, these data suggest that adolescent pregnant girls were at increased risk of
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23 malaria and anaemia in the study population. It is imperative to prioritize and implement
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25 adolescent friendly policies and control programmes for malaria and anaemia in this region.
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27 Additional measures such as the use of insecticides treated mosquito bed nets (ITN), appropriate
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29 treatment of *Plasmodium* infections, iron supplementation, community health education targeted
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31 at adolescence should be deployed and incorporated within the national programmes in order to
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33 facilitate surveillance and monitoring. These approaches would ensure widespread coverage of
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35 this target group.
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48 **Conflict of interest**

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50 The authors declare that they have no conflict of interest.
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Figure legend

Fig. 1. Proportion of malaria infected pregnant women with anaemia.

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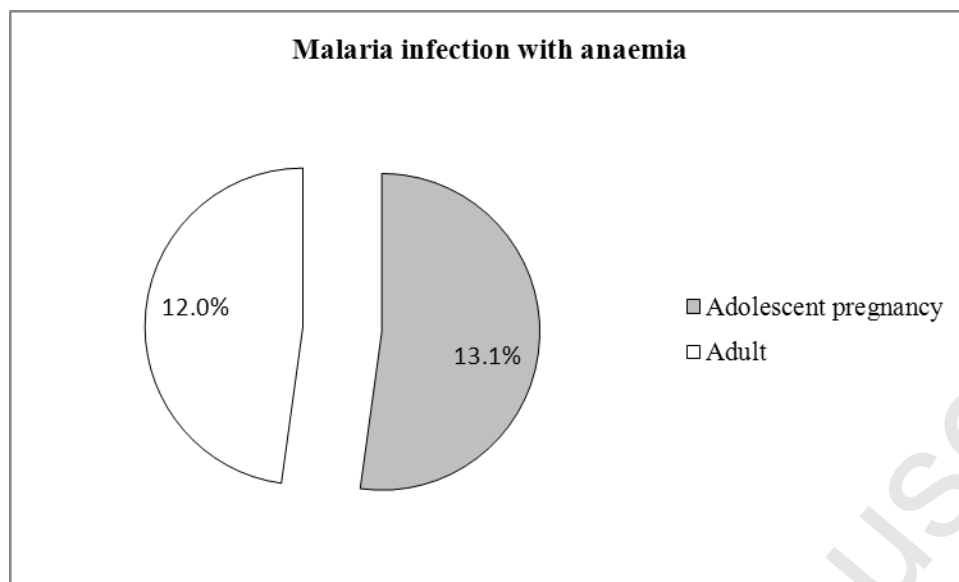


Fig. 1.

Table 1

Characteristics of the pregnant women stratified by adolescent and adult pregnancy.

Characteristics	Adolescent pregnancy (<19 years) n = 107 (%)	Adult (>20 years) n = 759 (%)	P value
Mother's age (years)			
Mean \pm SD	17.79 \pm 1.19	27.35 \pm 4.97	<0.001
Haemoglobin (g/dL)			
Mean \pm SD	11.33 \pm 1.03	11.50 \pm 0.96	0.09
Education			
None	11 (10.4)	156 (20.9)	0.005
Primary	18 (17.0)	106 (14.2)	
Secondary	77 (72.6)	451 (60.4)	
Tertiary	0 (0)	34 (4.6)	
Gravidae			
Primigravidae	107 (100)	403 (53.4)	<0.001
Secundigravidae	0 (0)	275 (36.4)	
Multigravidae	0 (0)	77 (10.2)	
Trimester			
1st	15 (14.4)	168 (23.0)	0.14
2nd	73 (70.2)	466 (63.8)	
3rd	16 (15.4)	96 (13.2)	
Malaria infection			
Positive	37 (34.6)	162 (21.3)	0.002
Negative	70 (65.4)	597 (78.7)	
Maternal anaemia			
<11 g/dL	47 (43.9)	248 (33.2)	0.03
>11 g/dL	60 (56.1)	498 (66.8)	
HIV status			
HIV-	99 (92.5)	681 (89.7)	0.37
HIV +	8 (7.5)	78 (10.3)	
IPTp-SP use			
Malaria positive	13 (24.5)	68 (17.8)	0.24
Malaria negative	40 (75.5)	314 (82.2)	
No IPTp-SP use			

Malaria positive	23 (52.3)	92 (33.8)	0.018
Malaria negative	21 (47.7)	180 (66.2)	
IPTp-SP doses			
0	44 (45.4)	272 (41.6)	0.41
1	42 (43.3)	262 (40.1)	
2	10 (10.3)	108 (16.5)	
3	1 (1.0)	12 (1.8)	

Note. SD = standard deviation. IPTp-SP = Intermittent preventive treatment of malaria during pregnancy with sulphadoxine-pyrimethamine. P values derived from Pearson Chi-Square test for categorical variables and ANOVA for the mean of continuous variable.

For education; adolescent pregnancy (n = 106), adult (n = 747).

For gravidae; adult (n = 755).

For trimester; adolescent pregnancy (n = 104), adult (n = 730).

For maternal anaemia; adult (n = 746).

For IPTp-SP use; adolescent pregnancy (n = 53), adult (n = 382).

For No IPTp-SP use; adolescent pregnancy (n = 44), adult (n = 272).

For IPTp-SP doses; adolescent pregnancy (n = 97), adult (n = 654).

Table 2

Multivariable logistic regression analysis of the risk of malaria and anaemia among the pregnant women.

Variable	Adolescent/Adult pregnancy (%)	Unadjusted OR (95 % CI)	P value	Adjusted OR (95% CI)	P value
Malaria					
Negative	65.4/78.7	1		1	
Positive	34.6/21.3	1.95 (1.26-3.01)	0.003	1.65 (1.03-2.65)	0.039
Education					
None	10.4/20.9	1		1	
Primary	17.0/14.2	2.41 (1.09-5.30)	0.029	1.71 (0.67-4.36)	0.27
Secondary	72.6/60.4	2.42 (1.26-4.67)	0.008	1.02 (0.48-2.16)	0.97
Tertiary	0/4.6	-	-	-	-
IPTp-SP					
No	45.4/41.6	1		1	
Yes	54.6/58.4	0.86 (0.56-1.32)	0.48	0.81 (0.59-1.10)	0.17
IPTp-SP doses					
0 doses	45.4/41.6	1		1	
<2 doses	43.3/40.1	0.99 (0.63-1.56)	0.97	1.16 (0.71-1.89)	0.57
≥2 doses	11.3/18.3	0.57 (0.28-1.14)	0.11	0.66 (0.32-1.38)	0.27
No IPTp-SP use					
Malaria negative	47.7/66.2	1		1	
Malaria positive	52.3/33.8	2.14 (1.13-4.08)	0.020	2.33 (1.14-4.76)	0.020
Maternal anaemia					
>11 g/dL	56.1/66.8	1		1	
<11 g/dL	43.9/33.2	1.57 (1.04-2.37)	0.031	1.63 (1.01-2.62)	0.046

OR = odds ratio, CI = confidence interval. IPTp-SP = Intermittent preventive treatment of malaria during pregnancy with sulphadoxine-pyrimethamine.