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**ANAEMIA PREVALENCE AND NUTRIENT INTAKE AMONG WOMEN IN
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ABSTRACT

Anaemia among women is a major public health concern globally. In developing countries, nutritional anaemia may be due to poor bioavailability of dietary iron, haemoglobinopathies, or intestinal parasites. The study objectives were to determine the prevalence of anaemia and current nutrient intakes essential to erythropoiesis among women of reproductive age (WRA) in peri-urban settlements in the Ga-East Municipality, Accra, Ghana. This cross-sectional study assessed the nutrient intake, sickling and haemoglobin levels of 134 women aged 15-49 years enrolled in a peanut-based field trial at baseline. A pretested semi-quantitative food frequency questionnaire was used to assess energy and nutrient intake using the Food Processor (ESHA) software. Whole blood was used to determine full blood count using the haematology automated analyser. Sickling was determined by blood staining. Anaemia was classified based on recommended cut-offs. Chi-square analysis was used as a test of independence between anaemia and age groups. Linear regression was used to determine predictors of haemoglobin concentration. The mean age of the women was 29 ± 8 years. The mean total caloric intake was 2315 ± 915 kcal, whilst protein and fat intakes were 67 ± 27 g and 68 ± 30 g, respectively. Almost three-quarters of the women met the recommended dietary allowance (RDA) for iron and vitamin C. However, only a third met the RDA for fat, and about two-thirds met their needs for energy and protein. None of the women met the RDA for folate which is integral for haem formation. Also 17% (23/134) of the women were sickle cell anaemia positive. Mean haemoglobin concentration was 12.1 ± 1.8 g/dL. Mild, moderate and severe anaemia due to iron deficiency was present in 35.8%, 6.7% and 1.5%, of the women respectively. All categories of anaemia was present in 44% of the women. Anaemia, prevalence increased during the early stages of the reproductive age (15-29 years) and declined towards the end of the reproductive age period (40-49 years). An increase in age of one year was significantly associated with a 0.056 g/dL rise in haemoglobin level ($p=0.014$). Anaemia due to iron deficiency exists as a public health problem among women in peri-urban settlements in the Ga-East Municipality, Accra, Ghana. Dietary diversity to include green leafy vegetables which are rich in folate and pro-vitamin A may reduce the current level of prevalence.

Key words: Anaemia, Nutrients, Peri-urban, Women, RDA, sickling test, Food Processor software



INTRODUCTION

Globally, anaemia is the most common nutritional deficiency and has received much attention for targeted improvements. Nonetheless, its prevalence remains unacceptably high. About one-third of the global population (over 2 billion) is anaemic [1]. Age-specific anaemia prevalence estimates of 47% in children under 5 years, 30% in non-pregnant non-lactating women and 42% in pregnant women has been reported by McLean *et al.* [2]. Anaemia prevalence among women of reproductive age (WRA: 15-49 years) in West and Central Africa was 48% as at 2011 [3]. Statistics in Ghana indicate that 4 out of every 10 WRA are anaemic [4].

Anaemia is a condition in which the body does not have enough healthy red blood cells or decreased normal quantity of haemoglobin (Hb) in the blood. Anaemia aetiology can be due to deficiency of one or more essential nutrients needed for erythrocyte formation as well as potential risk factors such as malaria, infectious diseases, parasitic infections and haemoglobin related disorders [5, 6]. For women, other risk factors include recurrent menstrual loss, pregnancy demands, short birth intervals, poor access to antenatal care and inadequate micronutrient supplementation [7]. Deficiencies in haematopoietic nutrients (iron, folic acid and vitamin A, vitamin B₁₂) have been described by the World Health Organization as nutritional anaemia. Increasing intake of foods rich in anti-nutritive factors (phytic acid, trypsin inhibitors, oxalates, tannins, polyphenols, haemagglutinin) as well as low fruit and vegetable intake has brought about a decline in bioavailable haematopoietic nutrients such as iron and its absorption enhancers including vitamin C [8]. Folic acid is required for the synthesis and maturation of the erythrocytes. Vitamin A has also been shown to increase haemoglobin concentration [9]. Nutritional anaemia contributes to high maternal mortality and morbidity [10]. Annually, anaemia is estimated to contribute to more than 115,000 maternal deaths [3].

In 2012, the 65th World Health Assembly approved an action plan with commitment to halve anaemia prevalence in WRA by 2025 from the 2011 levels [3]. This calls for the implementation of proven effective nutrition intervention strategies. For effective management and evaluation of prioritized country-specific nutrition interventions to curtail anaemia, information is needed about haemoglobin status in the population of concern. Also consumption and preparation of iron, folate and vitamin A rich foods varies between urban and rural dwellers. However, there is paucity of data on consumption and nutrient intake among peri-urban dwellers in Ghana. This study therefore, seeks to determine the prevalence of anaemia and the intake of nutrients essential for erythropoiesis among WRA in peri-urban settlements in the Ga-East Municipality in the Greater Accra region of Ghana.

PARTICIPANTS AND METHODS

Design and setting

This was a cross-sectional study conducted among women of reproductive age (WRA: 15-49 years) who reside in five peri-urban communities in the north-eastern horn of the Ga-East municipality in the Greater-Accra region of Ghana (Ayimensa, Kweiman, Danfa, Adoteiman and Otinibi). The municipality was created in December 2004 with a



land area of 166 square metres and Abokobi as the capital. The population density of the municipality is 1633.34 with an estimated population of 259,668 of which 51% are females [11]. The main occupation of the women in these communities is farming, gari production and petty trading. A tarred road links these communities. A borehole system provides all the communities with a source of drinking water. This is provided through Water and Sanitation Development Boards for piped schemes. A central health centre at Danfa called Danfa Health Centre provides all the communities with medical services. A signed informed consent was obtained from all recruited participants after the study protocol had been explained to them. The study protocol was approved by the institutional review board of Noguchi Memorial Institute for Medical Research, University of Ghana, Legon and permission also obtained from the Ga-East Municipal Chief Executive.

Data collection

Data on background characteristics, dietary and biochemical were collected following standard procedures. A simple population proportion formula assuming underweight prevalence of 10.1% [4], alpha level of 0.05 and 80% power was used to obtain a sample size of 121 and this was rounded up to 134 using a margin of error of 10% to improve the precision of the estimates [12]. Cluster sampling was used to select houses in each of the communities and any WRA living in the houses was recruited. Non-pregnant, non-lactating healthy WRA who reside in Ayimansa, Kweiman, Danfa, Adoteiman and Otinibi were recruited as study participants from January to February, 2012. For a woman (15-49 years) to be included, she should not have been on vitamin-mineral supplements at least 6 months prior to recruitment.

Information on participants' background characteristics was obtained using a pre-tested questionnaire. The initial questionnaire was pre-tested in another community in Ga-East municipality called Amarhyia, a community with similar characteristics as the study communities. Thirty questionnaires were used for the pre-testing after which questions that were difficult to understand by participants were reworded to ensure that all the questions in the questionnaire were easy to understand. Questions on age, level of education, marital status and occupation were obtained from the respondents through one-on-one interview. A semi-quantitative validated food frequency questionnaire was used to obtain information on participants' usual dietary intake in the previous one month [four weeks] prior to the study. For a particular food item, the frequency of intake in a week was recorded. Household food measures, food models and other eating wares were shown to the participants to improve estimation of portion sizes.

Blood sampling, Haemoglobin and Sickling test

A 2ml sterile syringe was used to draw venous blood from each participant by a trained phlebotomist on the third and fourth weekends in February 2012. The sampled blood was dispensed into a labelled ethylenediaminetetraacetic acid (EDTA) tube and transported on ice to the laboratory. The EDTA tubes containing 2 ml of blood were vortexed to ensure uniform mixing of the cellular components using Coultre mixer (UK). Haemoglobin (Hb) was measured using Olympus sysmex kx – 21N haematology fully automated analyser (France) [13]. Haemoglobin (Hb) was determined within two days after sample collection.



A thin film of blood from the EDTA containing tube was put on a slide and a drop of sickling fluid (sodium metabisulphite) was added to the blood. The blood-metabisulphite mixture was smeared on the slide and covered with a cover slip and allowed to dry. The prepared slide was observed under the electron microscope (Olympus –Germany) to identify at least a sickle shaped red blood cell in order to declare the participant as being sickling positive. All blood samples that tested positive for sickling were further examined through electrophoresis to determine those who had sickle cell disease or were carriers of the sickle cell trait. This was done with the help of a certified laboratory technologist.

Statistical analysis

The responses obtained on participants' dietary intake from the semi-quantitative food frequency questionnaire were analysed using ESHA FPRO (Athens, Georgia, USA) version 10.0.1. The total weekly dietary intake was calculated and the average of the weekly intake used as the usual daily nutrient intake of the participant. Each participant's nutrient intake was classified. The classification was done as follows: a participant was classified as having a low intake when the recommended dietary allowance (RDA) for a particular nutrient was not met, and at least meeting the RDA when the woman's dietary intake was within or above the RDA. The RDA values were obtained from Whitney and Rolfe [14].

Data were coded, entered into SPSS version 21 (Athens, Georgia, USA) and cleaned before analysis. Haemoglobin (Hb) concentration was classified as severe anaemia (<7.0 g/dL), moderate anaemia (7.0-9.9 g/dL), mild anaemia (10.0-11.9 g/dL), all forms of anaemia (<12.0 g/dL) and non-anaemic (≥ 12.0 g/dL) [4]. Descriptive statistics was performed for the socio-demographic characteristics of the women. Independent-sample T-Test was used to test for differences in mean nutrient intakes and haemoglobin levels (anaemia and non-anaemia). Pearson's chi-square was used to examine associations between categorical variables. Pearson's correlation was used to determine relationship between haemoglobin and the different continuous variables. Linear regression was used to determine predictors of haemoglobin concentration. For the regression analysis, dummies were created for all categorical variables and were put in the model together with the continuous variables. The result of the most stable linear regression model has been presented. Minimum 95% confidence intervals and p-value of less than 0.05 were considered significant.

RESULTS

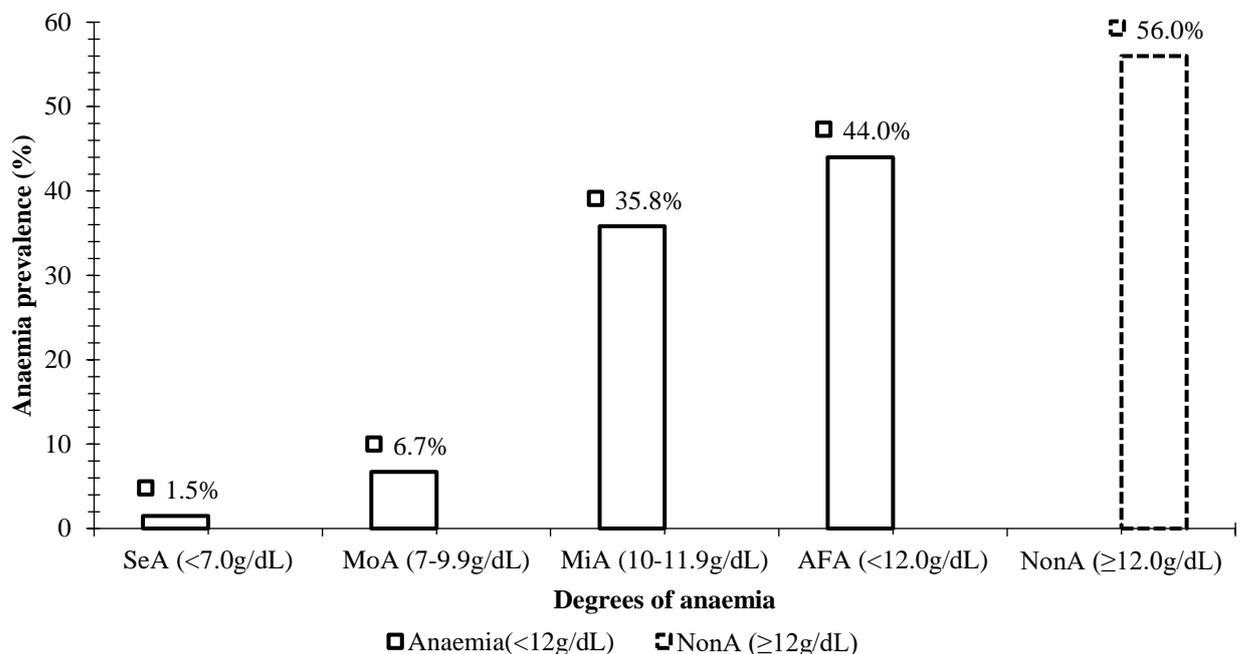
A total of 134 WRA were enrolled in the study with mean age of 29 ± 8 years. About 50% of the participants were in the 20-29-year age category. Majority had basic education (67.2%), most of them were employed (61.2%) and married (53.7%). Sickle cell trait was identified in 17.2% (23/134) of the participants; 16.4% were sickle cell carriers and one participant had sickle cell Hb C disease (Table 1).

The mean caloric, protein and fat intake were 2315 kcal, 67 g and 68 g, respectively (Table 2). These mean intakes were higher than their corresponding RDA for calories



but protein was within the RDA range whilst fat was below the RDA range. The mean iron intake of 24 mg was above the RDA of 15-18 mg whilst folate (119 µg) was far below the RDA (400 µg). From Table 2, it was also evident that the mean vitamin A intake was below the RDA whilst that of vitamin C was above the RDA. A significant number of women (41.8%) did not meet their RDA for calories. The number of women who did not meet their fat intake was twice (65.7%) the number that did not meet their RDA for proteins. Caloric and vitamin A intakes were much higher for non-anaemic women as compared to anaemic women (Table 2). There was no significant difference in iron intake for anaemic and non-anaemic women. In Table 3, all the women did not meet their RDA for folate and a greater majority (74.6%) also ate less vitamin A rich foods. Majority (73.9%) and 75.4% met their iron and vitamin C daily requirements, respectively. Nutrient intake generally increased from 15 to 29 years and then declines from 30 to 49 years. Nonetheless, there was no significant difference between nutrient intake and the age of the respondents as shown in Table 3.

The mean haemoglobin (Hb) concentration was 12.1 ± 1.8 g/dL. Hb levels increased from 11.7 g/dL to 12.7 g/dL from the 20-29 year group to the 40-49 year group. However, this increase was not different among the various age groups (Table 4). Anaemia prevalence was 44.0% in this study population. Of this percentage, 35.8% were mildly anaemic, 6.7% were moderately anaemic and 1.5% were severely anaemic (Figure 1).



Key: SeA=Severe anaemia; MoA=Moderate anaemia; MiA=Mild anaemia; AFA=All forms of Anaemia; NonA=Non-anaemic

Figure 1: Anaemia prevalence among study participants

Anaemia prevalence increased during the early stages of the reproductive age (15-29 years). This was followed by a decline in anaemia rates towards the end of the reproductive age period (40-49 years). There was a significant difference in anaemia and non-anaemic rates within an age group (Figure 2).

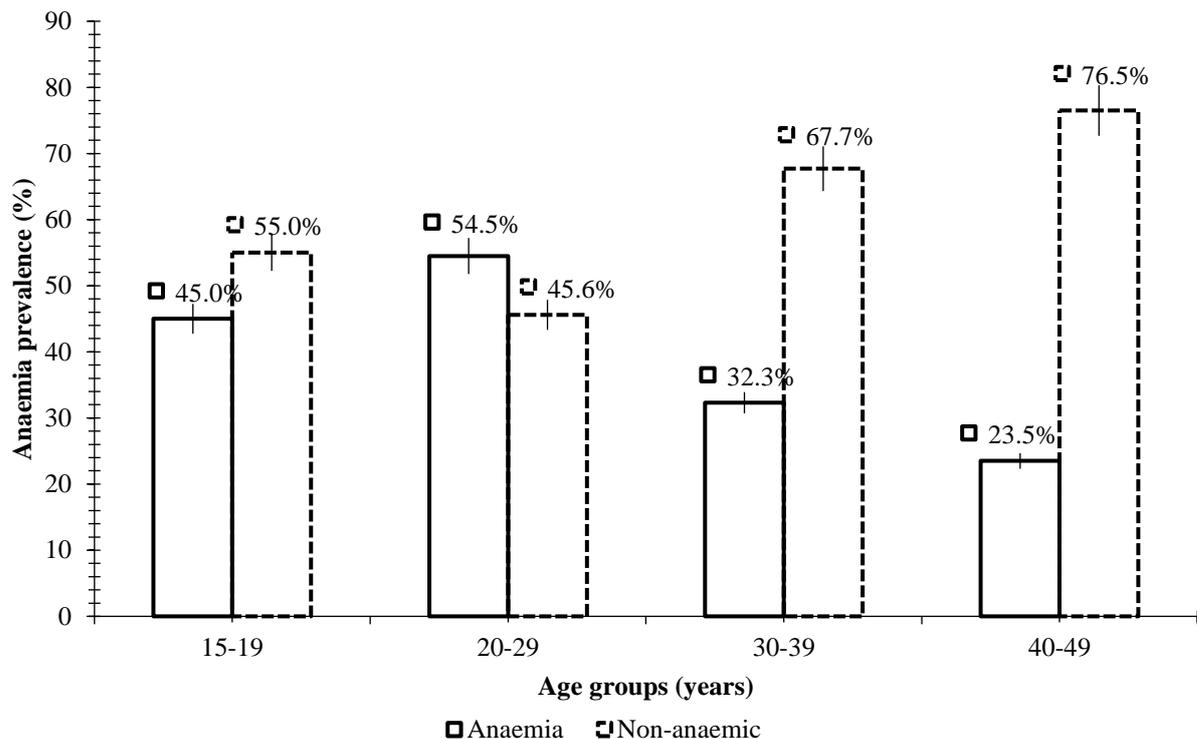


Figure 2: Anaemia prevalence by age groups

There was a weak but positive relationship between Hb and age ($r=0.175$, $p<0.05$) (Table 5). With the exception of vitamin A, there was no significant association between Hb and the other haematopoietic nutrients. A weak inverse relationship was observed between Vitamin A intake and Hb. An increase in age of one year was significantly associated with a 0.056 g/dL rise in Hb level (Table 6). Also a unit increase in calorie intake was significantly associated with a 0.002 g/dL rise in Hb levels. Nonetheless, a unit rise in fat intake was associated with a 0.020 g/dL fold decrease in Hb level. Women who did not have sickle cell as compared to those with sickle cell were more likely to have an increase in their haemoglobin level by 0.580 g/dL fold.

DISCUSSION

Prevalence of iron deficiency anaemia (IDA) defined as low haemoglobin (Hb) concentration exceeded the threshold of 30.2% stated for non-pregnant and non-lactating women globally [8]. Anaemia prevalence in this study was 44.0%. The global anaemia prevalence of 48% estimated for non-pregnant non-lactating women of reproductive age in Central and West Africa [3] compares well with the 44% anaemia prevalence reported in this study. Nonetheless, the 2014 Ghana Demographic and Health Survey (GDHS) reported that 42% of women aged 15-49 years have some form of anaemia [4]. Anaemia prevalence reported in this study is similar to that reported by the GDHS in 2014 [4]. Anaemia is higher in the urban areas than the rural areas in Ghana [4]. The observed prevalence suggests that in the peri-urban areas anaemia prevalence is high just as in the urban areas. Most of the women are mildly anaemic and this may not contribute to mortality and morbidity but may negatively affect reproduction and productivity. This

prevalence may be chronic considering that over 17% suffer from sickle cell disease [15]. In sickle cell, the circulating red blood cells (RBC) are fewer and have a lifespan of 10-20 days as compared to a normal RBC with a life span of 120 days. The presence of the sickle cell trait in some participants (17.2%) and a form of sickle cell disease (sickle cell haemoglobin C disease) in one participant could also contribute to anaemia rates in the study population. Both haemoglobinopathies can lead to haemolytic anaemia and because it is benign, its aetiology is different from nutrition related anaemia.

The cause of anaemia is multifactorial, the commonest being iron, folate, B12 and Vitamin A and C deficiencies, worm infestation and malaria as well as other infections and inflammations [6]. The high prevalence of anaemia in this study population could be attributed to the fact that Africa and for that matter Ghana is prone to malaria. Malaria contributes to anaemia by causing intravascular haemolysis with subsequent blood loss [6]. Also, malaria causes an immune response that suppresses erythropoietin and erythropoiesis [16].

The mean iron intake was 24 ± 12 mg, a value higher than the daily requirements, but iron bioavailability greatly depends on the dietary composition. Besides, in the absence of severe malnutrition or co-morbidity, the diet in itself infrequently causes IDA [6]. This is because increased intake of dietary factors such as polyphenols (examples: tea, coffee and so on), phytates (whole grains and cereals) and calcium (dairy products) reduce the bioavailability of non-heme iron [17, 18]. Probably heme iron intake of the women was low as reported in Nepal [19]. This is probably due to their low level of education and financial difficulties since most are engaged in low income generating activities (personal observation). In times of financial difficulty, carbohydrate intake may be maintained through alternative food sources that may be cheaper whilst affordability of micronutrient food sources may have been reduced [20]. More importantly, the absorption of nutrients that promote haemopoiesis can be affected by physiological and pathological factors. For example, *helicobacter pylori* is associated with reduced iron stores. It has also been documented that infections and inflammations inhibit iron absorption even if it is readily available in the diet [21].

The other nutritional causes of anaemia in this population are probably due to the low intake of folate and vitamin A rich foods. All the women did not meet their RDA for folate whilst only 25% met their RDA for vitamin A. Hodges *et al.* [22] demonstrated that, adult subjects maintained on vitamin A deficient diets developed anaemia despite adequate iron intakes. This observation confirms the results of this study since majority (74%) met their daily iron requirement as compared to the 25% who met their daily requirement for vitamin A. Folate deficiency will account for macrocytic anaemia whilst vitamin A deficiency will decrease Hb concentration. Vitamin A mechanism in anaemia is by the enhancement of growth and differentiation of erythrocyte progenitor cells and mobilization of iron stores from tissues [23]. Thus, the low protein status among some participants (35.8%) could also contribute to anaemia.

A weak correlation was observed between iron intake and Hb. This suggests that the aetiology of the anaemia is not due to iron alone but other causes such as folate and vitamin A. A similar weak positive correlation between Hb and iron intake was reported in another study [24]. In children, dietary iron intake from 9-11 months was associated

with Hb concentration at 12 months but iron intake from 12-18 months was not associated with Hb concentration at 18 months [25].

In this study, there was a positive correlation between Hb and age ($r=0.175$, $p=0.04$; Table 5). It was realized that a year's increase in age was associated with a 0.056 g/dL increase in Hb concentration. This may be attributed to increased bone marrow haematopoiesis during the reproductive period. According to Kushang, Hb increases with age in WRA but a modest decline occurs after the reproductive age [26]. For the women in this study, Hb generally increased across their reproductive age group, which is in agreement with a study in rural India [27].

The negative correlation between vitamin A intake and Hb is at variance with what Zimmermann and his colleagues reported in 2006 [28]. This difference may be due to the amount of vitamin A consumed by the respondents in each of the studies. Whilst Zimmermann and his colleagues gave their study participants vitamin A supplements, the vitamin A intake reported in this study was from food, which had a lower concentration than the supplement.

Also a possible under-estimation could have resulted in the low vitamin A intake whilst over-estimation could have resulted in the high intake of carbohydrate and iron among the participants. This finding is in consonance with a similar study conducted by Schaefer *et al.* [29] using a food frequency questionnaire as is the case in this present study. Another study commented on the huge disparity between results of dietary assessment using food frequency questionnaire and those using the gold standards in a research they conducted [30]. Nonetheless, the challenge of underestimation or overestimation of food intake, which is associated with dietary assessment methods, is not different from the food frequency questionnaire used in this study. Underestimation and overestimation was minimised by using food models to help participants to estimate portion sizes. Another limitation of this study was that it was not possible to screen for malaria parasites and worm infections. Also, haemolytic anaemia and anaemia due to hormones and thalassemia were beyond the scope of this study. However, screening for sickle cell is one of the strengths of this study as most studies assessing Hb levels do not screen for it despite its strong effect on Hb values.

CONCLUSION

Anaemia due to iron deficiency exists as a public health problem among women in peri-urban settlements in the Ga-East Municipality, Accra, Ghana. Dietary diversity is important and needs to include green leafy vegetables which are rich in folate and vitamin A.

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Table 1: Background characteristics of study participants (N=134)

Characteristics	n (%)
Age (years)	
15-19	20 (14.9)
20-29	66 (49.3)
30-39	31 (23.1)
40-49	17 (12.7)
Marital status	
Single	62 (46.3)
Married	72 (53.7)
Level of education	
Senior High School	21 (15.6)
Junior High School/basic education	90 (67.2)
None	23 (17.2)
Occupation	
Employed	82 (61.2)
Unemployed	52 (38.8)
Sickling status	
Positive	23 (17.2)
Carrier	22 (16.4)
Sickle	1 (0.7)
Negative	111 (82.8)

Table 2: Nutrient intake of study participants in comparison to RDA (N=134)

Nutrient intake	RDA	Mean \pm SD	Haemoglobin classification		
			Anaemic	Non-Anaemic	p-value
Calories (kcal)	2000-2200	2315 \pm 915	2257 \pm 860	2361 \pm 960	0.52
Protein (g)	50-82.5	67 \pm 27	65 \pm 24	68 \pm 29	0.46
Fats (g)	75-165	68 \pm 30	70 \pm 29	67 \pm 30	0.53
Iron (mg)	15-18	24 \pm 12	23 \pm 12	24 \pm 12	0.65
Folate (μg)	400	119 \pm 85	122 \pm 71	116 \pm 95	0.71
Vit. A (IU)	500	218 \pm 37	160 \pm 36	292 \pm 65	0.06
Vit. C (mg)	40-45	92 \pm 71	91 \pm 61	92 \pm 79	0.98

RDA- recommended dietary allowance (Whitney and Rolfes [14])

Table 3: Proportion of women who did not meet their RDA (N=134)

Nutrients	Age groups (years)				Total n (%)	p-value
	15-19	20-29	30-39	40-49		
	(n=20) n (%)	(n=66) n (%)	(n=31) n (%)	(n=17) n (%)		
Calories (kcal)	9 (6.7)	26 (19.4)	12 (9.0)	9 (6.7)	56 (41.8)	0.43
Proteins (g)	6 (4.5)	23 (17.2)	7 (5.2)	9 (6.7)	45 (33.6)	0.10
Fats (g)	12 (9.0)	35 (26.1)	26 (19.4)	15 (11.2)	88 (65.7)	0.98
Iron (mg)	7 (5.2)	18 (13.4)	5 (3.7)	5 (3.7)	35 (26.1)	0.10
Folate (μg)	19 (14.2)	54 (40.3)	38 (28.4)	22 (16.4)	133 (99.3)	0.31
Vit. A (IU)	15 (11.2)	37 (27.6)	30 (22.4)	18 (13.4)	100 (74.6)	0.62
Vit. C (mg)	3 (2.2)	11 (8.2)	13 (9.7)	6 (4.6)	33 (24.6)	0.37

Table 4: Mean haemoglobin concentration of the women by age groups

Age groups (years)	Haemoglobin (g/dL)
15-19	12.1 ± 1.1
20-29	11.7 ± 2.0
30-39	12.3 ± 1.9
40-49	12.7 ± 1.2
Total	12.1 ± 1.8
p-value	0.13

Table 5: Correlation between haemoglobin, age and nutrient intake

Variables	Haemoglobin (g/dL) r	p-value
Age (years)	0.18	0.043*
Calories (kcal)	0.09	0.33
Proteins (g)	0.08	0.34
Fats (g)	-0.04	0.62
Iron (mg)	0.09	0.28
Folate (µg)	-0.02	0.79
Vit. A (IU)	-0.24	0.006*
Vit. C (mg)	-0.02	0.80

*Significant at $p < 0.05$

Table 6: Linear regression showing the association between haemoglobin and nutrient intake and age of respondents

Variable	Regression Coefficient (β)	95% Confidence Interval		p-value
		Lower	Upper	
Age (years)	0.06	0.01	0.08	0.014*
Caloric intake (kcal)	0.00	0.00	0.00	0.012*
Protein intake (g)	-0.01	-0.04	0.02	0.52
Fat intake (g)	-0.02	-0.04	0.00	0.045*
Iron intake (mg)	-0.05	-0.12	0.01	0.08
Folate intake (μg)	-0.00	-0.01	0.00	0.49
Vit. A intake (IU)	-0.00	-0.00	0.00	0.002*
Vit. C intake (mg)	0.00	-0.01	0.00	0.75
Sickle cell				
Positive	1.00	Reference		
Negative	0.58	-0.22	1.37	0.15

Adjusted $R^2=0.11$; *Significant at $p<0.05$

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