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Research Article

Association of Total Antioxidant Status with Severity of Anaemia in Pregnancy in Ogun state, Nigeria

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Anaemia in pregnancy is a global public health challenge and contributes to increased generation of free radicals and decreased antioxidant status of pregnant women. This study evaluates the level of total antioxidant status (TAS) in uncomplicated pregnancy and its relationship with the levels of anemia severity in pregnant women.. Full blood count and total antioxidant status (TAS) were determined in 1200 pregnant women recruited from three health institutions in Shagamu, Ijebu Ode and Abeokuta in Ogun State, Nigeria. Total antioxidant status was analyzed using reagent supplied by Randox Laboratories, Crumlin, England while full blood count was assayed using Coulter counter cell dye auto-analyzed by Abott diagnostics (Wiesbaden, Germany). The demographic and clinical history was obtained using structured questionnaire. The levels of the measured variables were compared between anaemic and non-anaemic pregnant women while TAS was correlated with the severity of anaemia. Total antioxidant status was significantly lower ($p<0.001$) in anaemic than non-anaemic pregnant women. Among the anaemic pregnant women with normal levels of TAS, 92(46.9%) had mild anaemia, 100(51.0%) had moderate while 4(2.0 %) had severe anaemia. Out of those with abnormal TAS, 228(69.9%) had mild, 94(28.8%) had moderate, and 4(1.6%) had severe. A total of 196 anaemic pregnant women had normal TAS while abnormal low levels were observed among 326 anaemic pregnant women. The levels of TAS correlated significantly with anaemia severity from mild anaemia ($r=0.128;p=0.05$) to moderate anaemia ($r=0.237;p=0.02$) while insignificant correlation was observed among subjects with severe anaemia ($r=0.321;p=0.6$). . Pregnancy care should be organized in order to prevent anaemia and reduce free radical generation and maintain adequate antioxidant status of women to avoid complications.

Keywords: Anaemia, pregnancy, total antioxidant status.

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Introduction

Anaemia in pregnancy is a global public health problem which is associated with maternal morbidity and mortality [1]. Although pregnancy is a natural phenomenon, it is often associated with increased susceptibility to oxidative stress especially in developing countries where nutritional deficiency is common. During pregnancy, a number of physiological and metabolic changes occur to sustain and nurture the growth and development of the foetus [2]. It is a period that is associated with increased nutritional and oxygen demand leading to the generation of increased levels of free radicals and elevated levels of antioxidant defense system is also required to scavenge the free radicals [3]. Oxidative stress occurs when there is an imbalance between pro-oxidants (free radicals) generation and the antioxidants system resulting in the damage to lipids and biomolecules [4]. The placental is rich in mitochondria and poly unsaturated fatty acids (PUFFAs); therefore, it favors the development of oxidative stress because numerous metabolic and respiratory processes take place to provide nutrients and oxygen for the developing foetus. Previous studies have shown increased oxidative stress in uncomplicated pregnancy compared with levels in non-pregnant women [5-7], and anaemia in pregnancy [8]. The latter evaluated some oxidative stress markers in 50

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anaemic pregnant women and 25 non-pregnant women. The limitations of most of these studies were small sample size, and those were conducted at single and urban centres. There is however paucity of information on the association of total antioxidant status (TAS) with severity of anemia among pregnant women in Nigeria.

Anaemia is the most of the most common disorders in pregnancy, and it leads to about 56% maternal deaths in developing countries [9] and up to 50% of the cases due to iron deficiency [1,10]. Report from northern Nigeria suggested that about 60% of pregnant women were anemic [11]. Anaemia is considered as a major risk factor for an unfavourable pregnancy outcomes; such as premature labour, low birth weight and perinatal mortality [12,13]. Anaemia induced hypertension, placenta praevia, haemorrhage and cardiac failure [14,15]. According to the World Health Organization, the minimum non-acceptable haemoglobin (Hb) during pregnancy is 11g/dL [16]. Levels of severity of anaemia in developing countries were grouped as mild (10-10.9g/dL), moderate (7-9.9g/dL) and severe (<7.0g/dL). Anaemia in pregnancy may promote oxidative stress because of inadequate oxygen supply to the tissues which in turn could exacerbate the pathogenesis of iron deficiency anaemia. This study seeks to evaluate the level of total antioxidant status (TAS) and correlates with severity of anaemia in pregnant women.

Subjects and Methods

Study Area and study design

This is a multicenter cohort study of pregnant women recruited from Sagamu, Abeokuta and Ijebu-Ode in Ogun State, South-West, Nigeria. Pregnant women attending the antenatal clinics of the Olabisi Onabanjo Teaching Hospital (OOTH), General Hospital Ijebu-ode (GHI) and General Hospital Abeokuta (GHA) were randomly recruited in the study. A structured questionnaire was administered to obtain demographic and clinical information of the participants. A total of 1200 pregnant women (400 from each of the health institutions) were randomly enrolled into the study, but 78 of the participants were later excluded due to the medical disorders which may affect their haemoglobin levels. The medical disorders included asthma, fibroid, gestational diabetes, hypertension, leg ulcer, peptic ulcer, sickle cell anaemia, typhoid fever, and bleeding. All pregnant women attending the antenatal clinics for the first time irrespective of the gestational age were enrolled into the study. Whole blood specimen was used for complete blood count determination while serum was used for the assay of total antioxidant status. On the average, biochemical analyses were carried out every 3 weeks to allow for collection of a sample size good enough to run a pack of kit. The subjects were grouped into mild, moderate and severe anaemia based on their haemoglobin level. A balanced number of subjects from the strata Hb 7-8.9 g/dL, Hb 9.0-10.9 g/dL, Hb 11.0-14.9 g/dL, and all with Hb15.0 g/dL and above were included [17].

Inclusion and exclusion criteria

Pregnant women with Systemic diseases or malignancies or those who do not give consent were excluded. All pregnant women on vitamin C were also excluded from the study. The pregnant women included in the study were those diagnosed clinically and biochemically who gave informed consent, without other medical disorders or pregnancy related complications.

Sample preparation

Five milliliters (5mls) of venous blood sample was aseptically collected with 2mL dispensed into Ethylene diamine tetra acetic acid (EDTA) anti-coagulated sample bottle and 3ml emptied into plain bottle. The concentration of potassium EDTA was 1.5- 2.0mg/mL of blood. The tubes were inverted several times (8-10) to ensure thorough mixing and proper anticoagulation. The anti-coagulated blood was used to analyze the full blood count while the blood in the plain bottle was left to clot for at least 30 minutes. The clotted specimen was centrifuged at 3000rpm for 10minutes and the serum was separated into another plain specimen container. The serum was stored at -20ºC until analyzed. The complete blood count was analyzed using Coulter counter cell dye auto-analyzed by Abott diagnostics (Wiesbaden, Germany) while TAS was assayed using reagents supplied by Randox Laboratories, Crumlin, England.

Ethical consideration

The study protocol was reviewed and approved by the ethics committees of Olabisi Onabanjo Teaching Hospital (NHREC/08/10/2012; dated 22nd September, 2015), General Hospital Ijedu-Ode (PER/A/Vol1/130; dated 27th May 2015 and General Hospital Abeokuta (2265/03; dated

23rd April, 2015). The participants gave informed consent before they were enrolled in the study.

Table 1. Comparison of TAS and measured haematological parameters in all anaemic and non-anaemic pregnant women.

Measured variables	Non- Anaemic pregnant women n =600	Anaemic pregnant women n =522	*p*-value
Age (Years)	22.5±0.2	22.2±0.3	p=0.15
Hb value (g/dl)	11.2 ± 0.05	9.3 ±0.11	p <0.001
PCV (%)	34.4 ± 0.14	29.9±0.18	p <0.001
Platelet (x10^9/L)	235.5± 4.23	243.7±3.91	p =0.05
TWBC (x10^6/L)	7.1±0.22	9.1±0.10	p =0.05
Absolute neutrophil count (x 10^6 /L)	6.52±0.61	8.22±0.60	p =0.005
Absolute lymphocyte count (x 10^6/L)	2.96±0.57	3.28±0.59	p =0.6
Absolute eosinophil count (x 10^6/L)	0. 29±0.11	0.34±0.14	p =0.7
Absolute monocyte count (x 10^6/L)	0.19±0.13	0.22±0.15	p =0.9
Basophil (x 10^6/L)	0.01 ± 0.00	0.014±0.51	p =0.9
TAS (mmol/L)	1.1± 0.001	0.9±0.001	p <0.001

Hb=haemoglobin; PCV= packed cell volume; TWBC- total white blood cell; TAS= total antioxidant status.

Analytical method

Randox TAS kit was used for the quantitative in-vitro determination of total antioxidant status in serum (Randox Laboratories, Crumlin, England). The results are expressed as mmol Eq Trolox/L of plasma. Principle of assay is ABTS (2, 2'-Azino-di-[3-ethylbenzthiazoline sulphonate]) is incubated with a peroxidase (met myoglobin) and H_2O_2 to produce the radical cation ABTS. This has a relatively stable blue-green color, which is measured at 600nm. Antioxidants in the added sample cause suppression of this color production to a degree which is proportional to their concentration. The results are expressed as mmol Eq Trolox/L of plasma.

Statistical Analysis

The statistical analysis was conducted using SPSS 20 version and Unpaired Students t-test was used to compare the mean levels of measured haematological parameters and TAS between anaemic and non-anaemic pregnant women. Pearson correlation coefficient and Chi square were used to correlate the levels of TAS with severity of anaemia. Values of $p<0.05$ was considered as statistically significant. Data were expressed as mean ± standard error of mean.

Results

Table 1 shows the comparison of measured variables between anaemic and non-anaemic pregnant women from the three study centers. The mean levels of Hb and PCV

were significantly lower (p<0.001) in anaemic than non-anaemic pregnant women while total white blood cell, neutrophil and lymphocyte counts were significantly higher in anaemic than non-anaemic pregnant women. Total antioxidant status (p<0.001) and platelet count (p=0.05) were significantly lower in anaemic pregnant women than non-anaemic pregnant women.

Table 2 shows the medical conditions of the pregnant women. There was significant association between medical disorders and communities (p<0.001). Majority of the pregnant women (93.5%) had no adverse medical disorders while 6.5% had various medical disorders ranging from fibroid (0.8%), gestational diabetes 0.2%, hypertension 0.5%, hepatitis 0.2% peptic ulcer 0.5% and HIV infection 2.5%. The pregnant women with medical disorders were more in Shagamu (9.2%) than Abeokuta (5.5%) and Ijebu-Ode (4.7%).

Table 2. Medical disorders distribution among pregnant women overall and in the individual communities.

Medical disorders	Overall		Abeokuta		Ijebu-Ode		Sagamu	
	Freq	(%)	Freq	(%)	Freq	(%)	Freq	(%)
NAMD	1122	93.5	378	94.5	381	95.3	363	90.8
Asthmatic	6	0.5			2	0.5	4	1.0
Bleeding	2	0.2	2	0.5				
Fibroid	10	0.8	2	0.5	1	0.25	7	1.75
Gestational diabetes	2	0.2	2	0.5				
Hepatitis	2	0.2	2	0.5				
HIV	30	2.5	8	0.7	10	2.5	12	3.0
Hypertension	6	0.5	4	1.0	2	0.5		
Infection	6	0.5					6	1.5
Leg pain	2	0.2					2	0.5
Peptic ulcer	6	0.5	2	0.5	2	0.5	2	0.5
Sickle cell disease	4	0.3			2	0.5	2	0.5
Thyphoid fever	2	0.2					2	0.5
Total	1200	100	400	100	400	100	400	100

(X^2 = 607.00, df = 52, p < 0.001); NAMD- No adverse medical disorder.

The TAS distribution is presented in table 3. Among the anaemic pregnant women with normal values of TAS, 92(46.9%) had mild anaemia, 100 (51.0 %) had moderate while 4(2.0 %) had severe anaemia. Out of those with abnormal TAS (values below the lower limit of the reference range), 228(69.9%) had mild, 94(28.8%) had moderate, and 4(1.2%) had severe anaemia. A total of 196 anaemic pregnant women had normal TAS while abnormal low levels were observed among 326 anaemic pregnant women. The levels of TAS was significantly associated with severity of anaemia in pregnancy (r=0.094;p <0.05). A greater proportion of anaemic pregnant women (326) had abnormal (lower) TAS levels than those (196) with TAS levels wihin the normal reference range. The mean levels of TAS among pregnant women with abnormal mild anaemia, moderate anaemia and severe anaemia were 0.9±0.001; 0.7±0.001 and 0.6±0.006 respectively.

Table 3. Test of association between severity of anaemia classes and TAS.

Anti-oxidant status	Mild	Moderate	Severe	Total	Chi square	*p* -value
Normal	92 (46.9%)	100 (51.0%)	4(2.04%)	196(100.0%)		
Abnormal	228 (69.9%)	94(28.8%)	4 (1.2%)	326 (100.0%)		
					33.936	0.001
TAS(mmol/L)	0.9±0.001	0.7±0.001	0.6±0.006			
Total	320 (61.3%)	194.7 (37.2%	8 (1.5%)	522(100.0%)		

TAS=total anti-oxidant status; Reference Range (1.2-1.77mmol/L). Abnormal refers to the pregnant women with TAS levels below the lower limit of the reference range.

Table 4 shows the correlation between severity of anaemia and TAS in anaemia pregnant women. The levels of TAS decrease with increasing severity of anaemia (decreasing concentrations of haemoglobin). The levels of TAS correlated significantly with anaemia severity from mild anaemia (r=0.128;p=0.05) to moderate anaemia (r=0.237;p=0.02) while insignificant correlation was observed among subjects with severe anaemia (r=0.321;p=0.6).

Table 4. Correlation of total antioxidant status with anaemia severity in anaemic pregnant women.

Parameters	R	*p*-value
Mild anaemia and TAS	0.128	0.05
Mild anaemia and TAS	0.237	0.02
Severe anaemia and TAS	0.321	0.6

Discussion

Anaemia in pregnancy is the commonest medical disorder that affects pregnant women especially in developing countries in the world. It confers a number of deleterious effects on both mothers and foetus including maternal morbidity and mortality, abnormal foetal development, low birth weight, premature labour and pre-term delivery [18]. About 50% of all anaemic cases in pregnant women have been attributed to iron deficiency, because iron requirements are not readily met by dietary intake alone especially if iron bioavailability is poor in the pregnant women [19]. In addition, decreased red blood cell survival which is secondary to an increased susceptibility to oxidant damage has been observed in anaemia [20]. In this study we observed a proportion of 46.5% (522/1122) pregnant women to be anaemic. The TAS levels were significantly lower in anaemic than non-anaemic pregnant women which was also correlated with severity of anaemia in the participants.

The mean TAS level among anaemic pregnant women

was lower ($p<0.001$) than in non-anaemic pregnant women. This suggested that in the non-anaemic pregnant women, the generations of free radicals may be lower than the anaemic group. Free radicals are generated in the body at physiological levels, but when their production rates overwhelm the synergistic actions of available antioxidants, several deleterious or harmful conditions may ensue [21,22]. However, the mean TAS levels observed in this study was higher than previously reported in Benin and Iran [7,21]. The differences in the concentrations of TAS in this study and the previous may be attributed to small sample of previous studies. In addition, Idogun et al.[7] in their study enrolled women in third trimesters only while Asemi et al. [21] enrolled primigravid subjects only. Anaemia was reported to worsen the levels of TAS, thus increasing the oxidative stress of pregnant women. This observation is consistent with previous studies elsewhere [8, 23,24].Hassan et al.[8] reported a positive association between anaemia in pregnancy and total antioxidant capacity by measuring malondialdehyde, total peroxide and oxidative stress index. Increased requirement of oxygen, reduced activity of antioxidant enzymes requiring iron for their activity may result in increased free radical generation and damage to biomolecules [25]. The observed significantly lower TAS levels in anaemic pregnant women may be attributed to the increased production of free radicals and/or decreased antioxidant levels which are associated with severity of anaemia in pregnancy. Increased levels of free radicals especially hydrogen peroxide, apart from depleting the levels of other antioxidants, inhibits the activity of superoxide dismutase [26], an important antioxidant enzyme responsible for the dismutation of superoxide radicals. In the same vein, superoxide dismutase and catalase carry out their antioxidant function by detoxifying the peroxides (-OOH) and superoxide radicals. Catalase has been reported to provide vital pathway for the degradation of hydrogen peroxide in to water and molecular oxygen. Even though we did not analyze the individual antioxidant parameters in this study, the assay of TAS is a better indicator of antioxidant status since the determination of individual antioxidant levels may not provide the true antioxidants status of the organism [27]. Anaemia is an important contributing factor to lowering TAS levels in pregnancy. The placenta is also a major generator of free radicals because it is rich in PUFFAs which could make the pregnant women susceptible to increased oxidative stress [28]. Placenta oxidative stress has been associated with the pathogenesis of pre-eclampsia [29] and fetal growth retardation [5]. The need for the evaluation of oxidative stress in pregnant women to better understand the relationship between oxidative stress and pregnancy outcome cannot be overemphasized.

Other authors highlighted the need for pre-pregnancy and early pregnancy assessment of antioxidant status in women. They also suggested that the outcome of an uncomplicated pregnancy may be dependent on the amount of free radicals produced during parturition on the antioxidant status of the pregnant women. Early assessment of antioxidant status of pregnant women was therefore advocated [30]. Increased oxidative stress predisposes pregnant women to various pregnancy associated disorders such as malaria parasitaemia, infections and infestations [30]. Oxidative stress can also down-regulates T-mediated or adaptive immune response even though low immunity is needed to tolerate genetically different foetal tissues [6], which increased the susceptibility of pregnant women to infection.

The study indicated significantly lower TAS levels in anaemia pregnant women than non-anaemic counterparts which correlated with severity of anaemia. Pregnancy care should be organized in order to prevent anaemia, reduce free radical generation and maintain adequate antioxidant status of the pregnant women.

Conflicts of interest

None

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References

1. Allen LH. Anaemia and Iron deficiency: Effects on pregnancy outcome. Am J Clin Nutri 2000; 71(5):1280S-1284S

2. Qanungo S, Mukherjee M, Ontogenic profile of some antioxidants and lipid peroxidation in human placental and foetal tissues. Mol.

Cell Biochem., 2000; 215(1):11-9.

3. Knapena MFCM, Zuster Zeel PLM, Peters WHM, Steegers EAP. Glutathione and glutathione related enzymes in reproduction: A review. Eur J. obstet-Gynaecol. Reprod biol,1999; 82(2):171-84.

4. Agrawal A, Gupta S, Sharma RK. Role of oxidative stress in female reproduction. Reprod. Biol. Endocrinol, 2005; 3(1). 28-47.

5. Idonije OB, Festus O, Okhai O, Ukpamu U. A comparative study of the status of oxidative stress in pregnant Nigerian women. Res J Obstet Gynecol 2011; 4:28-36.

6. Ogbodo SO, Okaka ANC, Nwagha UI, Ejezie FE. Free radicals and antioxidants status in pregnancy: Need for pre- and early pregnancy assessment. Am J Med Med Scis 2014; 4(6):230-35..

7. Idogun ES, Odiegwu ME, Momoh SM, Okonofua FE. Effect of pregnancy on total antioxidant capacity in Nigerian women. Pak J Med Sci 2008;24(2):292-5.

8. Hassan S, Shaheen S. Association between oxidative stress and anaemia in pregnancy. Int Arch Biomed Clin Res 2017;3(2):547.

9. DeMayer EM, Tegman A. Prevalence of anaemia in the World.World Health Organ Qlty 1998; 38(3): 302-16.

10. Racknagel RD, Ghosal AK. Quantitative estimation of peroxidative degeneration of rat liver microsomal and mitochondrial lipids alter carbon tetrachloride poisoning. Exp Mol Pathol., 1966; 5(suppl 3):413–26.

11. Nwizu EN, lliyasu Z, Ibrahim SA, Galadanci HS. Socio Demographic and maternal factors in Anaemia in pregnancy at booking in Kano, Northern Nigeria. Afr J Reprod Health 2011; 15(4):33-41.

12. Scholl TO, Stein TP. Oxidant damage to DNA and pregnancy outcome. Matern Fetal Med 2001;10:182-5.

13. Agarwal A, Gupta S, Sharma RK, 2005. Role of oxidative stress in female reproduction. Reprod Biol Endocrinol 2005;3: 28-47.

14. Poggi SBH. Postpartum hemorrhage and abnormal puerperium. In: AH DeCherney, L Nathan, TM Goodwin, N Laufer (Eds);Current Diagnosis and Treatment, Obstetrics and Gynecology, 10th edn. New York: NY: McGraw-Hill Medical, 2007; 477-84.

15. Allen LH, de Benoist B, Dary O, Hurrell R. Guidelines on food fortification with micronutrients. Geneva: World Health Organisation,2006; 3-15.

16. World Health Organization. Iron deficiency anaemia: Assessment, prevention and control. A Guide for programme Manager. 2001;5-10. Available from: http://www.who.int./ nutrition/publications/micronutrients/anaemia_iron_ deficiency/WHO .Accessed on 10/11/2017.

17. Hinderaker SG, Olsen BE, Bergsjø PB, Lie RT, Gasheka P and Kvåle G. Anemia in pregnancy in the highlands of Tanzania. Acta Obstet Gynecol Scand 2001; 80: 18-26

18. Bothwell TH. Iron requirements in pregnancy and strategies to meet them. Am J Clin Nutr. 2000;72:257S–64S.

19. Galan P, Cherouvrier F, Zohoun T, Chauliac M, Hercbeeg S. Iron absorption from typical West African meals containing contaminating Fe. Br J Nutr. 1990;64:5416.

20. Tiwari AKM, Mahdi AA, Zahra F, Chandyan S, Srivastava SVK, Negi MPS. Evaluation of oxidative stress and antioxidant status in pregnant anemic women. Indian J Clin Biochem 2010; 25: 411-8.

21. Asemi Z, Taghizadeh M, Sarahroodi S, Jazayeri S, Tabasi Z, Seyyedi F. Assessment of the relationship of vitamin D with serum antioxidant vitamins E and A and their deficiencies in Iranian pregnant women. Saudi Med J 2010; 31: 1119-23.

22. Szeto HH (2006). Mitochondria-targeted peptide antioxidants: novel neuroprotective agents. AAPS J 2006; 8:E521–E531.

23. Aslan M, Horoz M, Celik H. Evaluation of oxidative status in iron deficiency anemia through total antioxidant capacity measured using an automated method. Turk J Hematol., 2011;28(1): 42-6.

24. Kumar A, Mahmood T. A Study of Oxidative Stress in Pregnant Anemic Women in Rural Rajasthan. International journal of scientific research, 2015; 4(6):803-5.

25. Halliwell B, Gutteridge JM. Role of free radicals and catalytic metal ions in human disease: an overview. Method in Enzymology, 1990; 186(1): 1-85.

26. Isler M, Delibas N, Guclu M, Gultekin F, Sutcu R, Bahceci M et al.. Superoxide dismutase and glutathione peroxidase in erythrocytes of patients with iron deficiency anemia: effects of different treatment modalities. J Croat Med. 2002;43:16–9.

27. Erel O. A novel automated method to measure total antioxidant response against potent free radical reactions. Clin Biochem.,2004; 37(2):112-9.

28. Gitto G, Reiter RJ, Karbownik M, Tan DX, Gitto P, Barberi S et al.. Causes of oxidative stress in the pre and perinatal period. Biol. Neonate 2002; 81: 146-57.

29. Onah HE, Okaro JM, Umeh U, Chigbu CO. Maternal morality in health institutions with emergency Obstetric care facilities in Enugu state, Nigeria. J Obstet Gynaecol 2005;25(6):569-74.

30. Kamath U, Rao G, Kamath SU, Rai L. Maternal and fetal indicators of oxidative stress during pregnancy-induced hypertension (PIH). Intern J Appl Biol Pharmaceut Technol 2011;2(1):405-10.