Biochemical Changes Associated with Protein Energy Malnutrition among Pregnant Women in Enugu Metropolis of Nigeria

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Abstract

Serum total protein, albumin, urea, total cholesterol, creatinine and calcium were evaluated in three groups of female subjects as part of an investigation on the biochemical changes associated with protein energy malnutrition (PEM) in pregnant women. The first group were 52 pregnant women with low total protein (<52g/l), the second group were 50 pregnant women with normal total protein (>52g/l) while the third group were 50 nonpregnant, non-lactating, apparently healthy women with normal total protein (>63kg). All the subjects were resident in Enugu metropolis and aged between 20 to 40 years. The pregnant subjects were in different gestational stages of pregnancy, having different parity and attending the antenatal clinic of Parklane Specialist Hospital, Enugu. The results show that there was no significant difference between the mean serum total protein of the different age groups (p>0.05). Parity (ie the number of children had by mother) correlated negatively (p<0.05) with serum total protein, urea, total cholesterol, creatinine and calcium. Gestational stage of pregnancy in trimesters correlated negatively and significantly with serum total protein and serum calcium (p < 0.05). The results also revealed that serum total cholesterol did not correlate significantly with serum total protein, urea, total cholesterol, creatinine and calcium (p>0.05 in each case). Urea levels correlated significantly and positively with serum total protein (r = + 0.246, p<0.05), and creatinine (r = +0.275, p<0.05). creatinine correlated positively with serum total protein (r = +0.497, p < 0.05), urea (r = +0.275, p < 0.05) and calcium (r = +0.356, p < 0.05). Calcium negatively and significantly correlated with gestational stage of pregnancy in trimesters (r = -0.288, p<0.05) and correlated positively and significantly with serum total protein (r = +0.681, p<0.05) and creatinine (r = +0.0356, p<0.05).

Keywords: Protein energy malnutrition (PEM), Pregnancy, Biochemical, Gestation.

1.1 Introduction

Protein Energy Malnutrition (PEM) results when the body's need for protein, energy or both cannot be satisfied by the diet. Worldwide, an estimated 852 million people are undernourished with most (815 million), living in developing countries (WHO, 2002; FAO, 2004) Poverty is the main underlying cause of malnutrition and its determinants (Sachs and McArthur, 2005). The degree and distribution of Protein Energy Malnutrition (PEM) in a given population depends on many factors – the political and economic situation, level of education and sanitation, the season and climate conditions, food production, cultural and religious food customs, breastfeeding habits, prevalence of infectious diseases, the existence and effectiveness of nutrition programmes and the availability and quality of health services (FAO, 2004; Salama *et al.*, 2004).

Malnutrition continues to be a major health burden in developing countries. It is globally the most important risk factor for illness and death with hundreds of millions of pregnant women and young children particularly affected (Muller and Krawinkel, 2005). Poor nutrition in pregnancy in combination with infections is a common cause of maternal and infant mortality and morbidity, low birth weight and intrauterine Growth Retardation (IUGR) (Pena and Bacalao, 2002). In Nigeria, maternal death per 100,000 births is put at 800 while percentage low birth weight stands at twenty (Enwonwu *et al.*, 2004).

The nutritional status of a person depends on food consumption and not solely the production and availability of food. Dietary energy supply measurement assume that available food is distributed and consumed in relation to requirement which is often not the case (WHO, 2001).

Low birth weight babies have increased risk of mortality, morbidity and development of malnutrition. Children who suffer from malnutrition are more likely to have slowed growth, delayed development, difficulty in school and high rates of illness and they may remain malnourished to adulthood (Scrimshaw, 1998; Abidoye and Eze, 2000). IUGR is associated with poor cognitive and neurological development for the infant and in adulthood, susceptibility to cardiovascular disease, diabetes and renal disease (De Onis *et al.*, 1998).

Malnutrition remains one of the world's highest priority health issues not only because its effects are so widespread and long lasting but also because it can be eradicated. Eradication is best carried out at the preventive stage. Hence the need to identify groups of pregnant women at greater risk of developing PEM. Such high-risk groups can be targeted in any planned intervention programme.

Pregnancy is a normal physiological process associated with major alterations affecting every maternal organ, system and metabolic pathway (McGanity *et al.*, 1994). This physiological process results in increased plasma volume and red blood cells, decreased concentration of circulating nutrient-binding proteins and other micronutrients (Ladipo, 2000).

The aim of this study is to investigate some of the biochemical changes associated with protein energy malnutrition (PEM) in pregnant women in Enugu metropolis of Nigeria.

MATERIALS AND METHODS

2.1 Study Subjects

Three groups of female volunteers were involved in this study. The first group were 52 pregnant women with low total protein (<52g/l), the second group were 50 pregnant women with normal total protein (>52g/l), while the third group were 50 non pregnant, non lactating, apparently healthy women (>52g/l). All the subjects were between 20-40 years of age. All the pregnant subjects were attending antenatal clinic of Parklane Specialist Hospital, were in different gestational stages of pregnancy with different parity. Subjects with complications such as hypertension, diabetes, HIV/AIDS on admission were excluded.

2.2 Collection of Blood Samples and Preparation of Serum

Blood (2.5mls) was collected from each volunteer by venepuncture and delivered into clean and duely labelled specimen containers. The blood was allowed to clot and then centrifuged at 5000 rpm for 10 minutes. Using a Pasteur pipette serum was separated from the cells and delivered into a clean and dry bottle. It was stored frozen at -20° C until it was used.

2.3 Preparation of Reagents

The reagents used were high performance enzymatic colorimetric commercial analytical kits (Biosystems Reagents and Instruments, Barcelona, Spain). These commercial kits were purchased and used according to the manufacturer's direction for all the parameters assayed.

2.4 Determination of serum total protein

Principle: The serum total protein was determined using the Biuret method. The protein in the sample reacted with copper II ion in alkaline medium forming a coloured complex that was measured spectrophotometrically

Determination of serum protein was carried out according to the method of Gornall *et al.* (1949) and Berg *et al.* (1984).

2.5 Determination of Serum Albumin Concentration

The measurement of serum albumin was by the quantitative method using (B.C.G) Bromocresol green. This method is based on the quantitative binding of albumin in the sample to the indicator 3, 3', 5, 5' – tetra bromo-m cresol sulphonephthalein (BCG) (Bromocresol Green). This will form a complex known as the Albumin – BCG complex. This complex absorbs maximally at 578nm, the absorbance (A) being directly proportional to the concentration of Albumin in the sample.

The determination of serum albumin concentration was done according to the method of Doumas et al. (1971).

2.6 Determination of Serum Total Cholesterol

The total cholesterol concentration of the test individuals was determined using cholesterol enzymatic endpoint method. Cholesterol is determined after enzymatic hydrolysis and oxidation. The indicator quinoneimine is formed from hydrogen peroxide and 4-aminoantipyrine in the presence of phenol and peroxide. The free and esterified cholesterol in the sample originates by means of the coupled reactions described below, a coloured complex is formed that can be measured spectrophotometrically.

The concentration of serum total cholesterol was determined according to the methods of Allain *et al.* (1974) and Meiattini *et al.* (1978).

2.7 Determination of serum urea concentration

The urea concentration of the test individuals were determined by the urease – salicylate enzymatic method. This uses the enzyme urease to hydrolyze urea. The ammonia produced reacts with alkaline hypochlorite and phenol in the presence of a catalyst to form indophenol. The coloured complex is measured spectrophotometrically. The determination of serum albumin concentration was done according to the method of Cheestbrough (1998).

2.8 Determination of serum creatinine concentration

Creatinine in the sample reacted with picrate in alkaline medium to form a coloured complex. The coloured complex is measured spectrophotometrically. It is measured within a short period to avoid interference from non creatinine substances.

The serum concentration of serum albumin concentration was done according to the methods of Bartels and Bohmer (1971) and Fabiny and Ertingshausen (1971).

2.9 Determination of serum calcium concentration

Calcium in the sample reacted with methylthymol blue in alkaline medium to form a coloured complex. The coloured complex was measured spectrophotometrically. Hydroxyquinoline was included in the reagent to avoid magnesium interference.

The determination of serum albumin concentration was done according to the methods of Gindler and King (1972) and Barnett *et al.* (1973).

Statistical Analysis

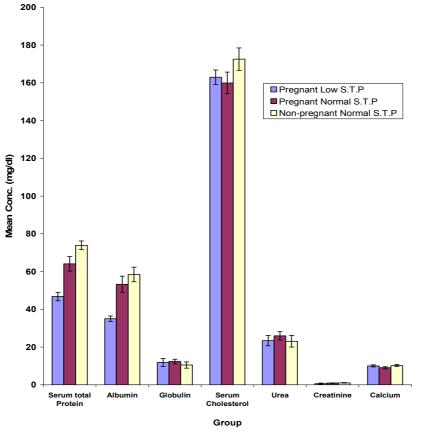
The data were analysed using the SPSS package of windows version 11.00 (SPSS Corporation, IL). Differences between the means were separated and analysed for statistical difference using the one way ANOVA while correlations between parameters were calculated using the Pearsons correlation coefficient. Difference in means with p values ≤ 0.05 were accepted as significant. Data were presented as means \pm standard deviations.

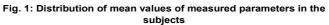
3.0 **RESULTS**

Three groups of female subjects were involved in this study. The first group represented 52 pregnant women with low total protein, the second group represented 50 pregnant women with normal total protein while the third group represented 50 non pregnant, non lactating apparently healthy women. All the subjects were aged between 20 and 40 years. All the pregnant subjects were in various gestational stages of pregnancy, and different parity. The mean age was 28.90 ± 5.31 for all the subjects studied.

3.1 Distribution of Mean Values of Measured Parameters in the Subjects

Fig. 1 shows the means values of serum total protein, albumin, globulin, cholesterol, urea, creatinine and calcium of all the subjects studied.

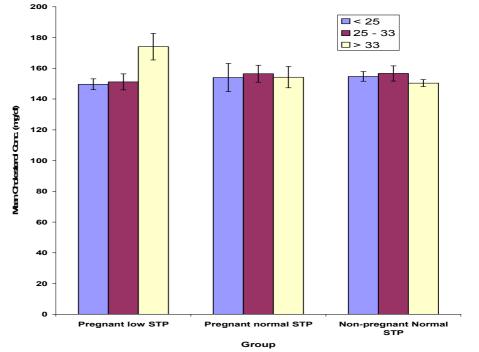


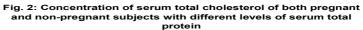


3.2 Effect of Age on the Different Parameters Measured

Figs. 2 to 5 show the results of all parameters measured for mothers in different age groups divided according to

their level of serum total protein. There was no significant difference in the means of the serum total protein of mothers of different age ranges and other parameters measured (p>0.05). Therefore age may not affect the level of serum total protein of a mother and also other parameters measured. A test of correlation showed that age of mother did not correlate significantly with serum total protein. However age of mother correlated positively and significantly with parity only (r = +0.545) (p<0.05) and no other parameter measured (p>0.05 in each case).





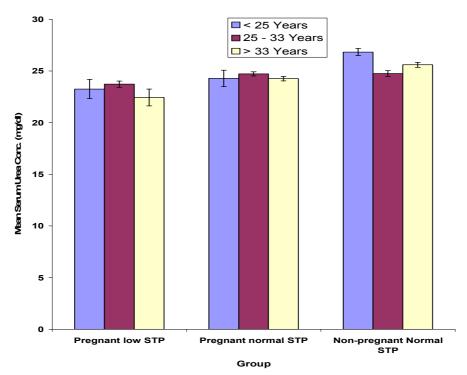


Fig. 3: Concentration of serum urea of both pregnant and nonpregnant subjects with different levels of serum total protein

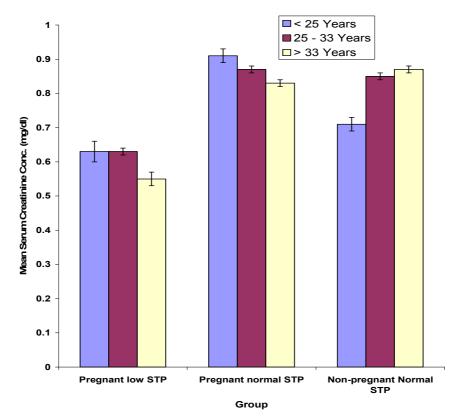


Fig. 4: Concentration of serum creatinine of both pregnant and nonpregnant subjects with different levels of serum total protein

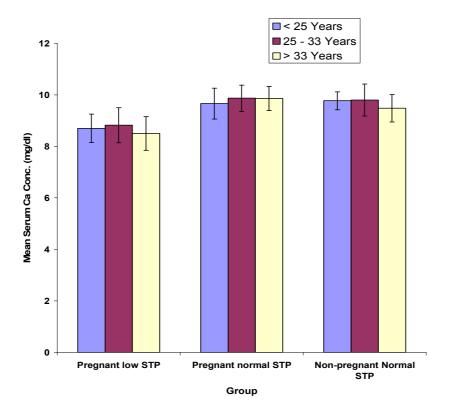


Fig. 5: Concentration of serum calcium of both pregnant and nonpregnant subjects with STP

DISCUSSION

Pregnancy is a physiological condition severely aggravated by protein energy malnutrition. This makes protein energy malnutrition (PEM) the most widespread and disabling public health problem among women especially in developing countries like Nigeria.

The demand for both energy and nutrient is increased during pregnancy and for well nourished women only a small amount of additional energy is required (WHO, 1999). Pregnancy is also associated with major alterations in every maternal organ, system and metabolic pathway. Values of biochemical parameters may change as the pregnancy advances from first to third trimester and to parturition and then return towards normal during post partum period. The two major physiological forces driving these changes are:

- (1) The increase in plasma volume, increase in red blood cells and decreased concentrations of circulating nutrient-binding proteins and micronutrients.
- (2) The ever increasing levels of estrogen and progesterone as well as other placental related hormones, which have particular impact on maternal lipids (cholesterol) (McGanity *et al.*, 1994).

These two physiological modifications result in two dominant effects: the first reduces levels of biochemical substances such as albumin and haemoglobin which return to normal 8–10 weeks post partum. The second causes lipids to rise during pregnancy and return to normal at post partum. The major consequences of protein energy malnutrition (PEM) are mainly poor weight gain in pregnancy, anaemia leading to high risk delivery and low birth-weight babies that fail to thrive.

The result of this investigation showed that all the biochemical parameters measured viz serum total protein, Albumin, urea, total cholesterol, creatinine and calcium were significantly reduced in pregnant PEM individual compared with pregnant and non-pregnant controls.

This agrees with the studies of Onyeneke *et al.* (2003) which suggests that abnormalities in serum levels of biochemical parameters occur in any form of PEM and are related to the severity of the condition (Fig. 1).

The result of this investigation also showed there was no significant difference in the mean serum total protein in the different age groups (see Figs. 2–5). Therefore age of mother may not be a factor and may not affect the level of serum total protein and other parameters measured. This agrees with the study of Okwu *et al.* (2007), which showed that the lower age groups (below 20 years and 20–24 years) presented higher prevalence of PEM than other age groups, with the effect more prominent in rural areas than in urban areas.

In addition, Lapido (2000) suggested that many pregnancies in developing countries are unplanned, coupled with inadequate dietary intake due to dietary taboos associated with pregnancy, gender and other cultural beliefs.

The results from assessment of biochemical parameters showed that serum total cholesterol did not significantly correlate with serum total protein or any other parameters measured (p > 0.05 in each case). Serum total cholesterol therefore may not be a factor in changes associated with low serum total protein and protein energy malnutrition. These findings are in consonance with the view of Toigo *et al.* (2000) that low serum cholesterol in PEM may reflect energy imbalance. Total serum cholesterol therefore is a useful marker for energy intake but not for protein intake.

The serum urea levels were found to correlate significantly and positively with serum total protein (r = +0.246, p < 0.05) and serum creatinine level (r = +0.275, p < 0.05) only. Its correlation with other parameters were not significant (p > 0.05 in each case). This suggests that as the serum total protein increases, serum urea and creatinine levels increase with it and vice versa.

A test of correlation further revealed that serum creatinine correlated positively and significantly with serum total protein level (r = +0.497, p < 0.05), serum urea level (r = +0.275, p < 0.05) and serum calcium levels (r = +0.356, p < 0.05). This suggests that as serum total protein increases, serum creatinine, serum urea and serum calcium levels increase and vice versa.

However, serum calcium was found to positively and significantly correlate with serum total protein level (r = + 0.681, p<0.05) and serum creatinine level only (r = + 0.0356, p < 0.05). This suggests that as serum total protein increases, serum calcium levels and serum creatinine levels increase with it and vice versa.

These factors are pointers to Protein Energy Malnutrition (PEM). Toigo *et al.* (2000) reported that in individuals with PEM, there is low serum concentration of creatinine, suggesting a decreased skeletal muscle mass or a low dietary protein intake.

Summary/Conclusion

In summary, the serum total protein which is known to correlate positively and significantly with protein energy malnutrition was found to also correlate positively and significantly with serum levels of urea, creatinine and calcium. Serum total cholesterol did not correlate significantly with serum total protein or any other parameter measured (p>0.05 in each case).

The serum levels of urea, creatinine, calcium and cholesterol have been implicated in this study as Biochemical indices or Biochemical Markers of PEM in pregnant women. Serum total cholesterol may however be a useful marker for energy intake and not for protein intake. This agrees with the work of Toigo *et al.* (2000) which listed

urea, creatinine, calcium and cholesterol as serum Biochemical indices or serum biochemical markers of PEM.

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