SERUM ALPHA FETO PROTEINS OF PREGNANT WOMEN IN SELECTED PRIMARY HEALTH CENTERS IN BUKURU PLATEAU STATE

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Abstract

Background: Alpha Feto Protein (AFP) is a Tumour biomarker normally produced in the yolk sac, epithelial cells of the gastrointestinal tract and liver during embryonic development. In pregnancy, AFP enters the amniotic fluid through fetal blood, via the placenta into maternal blood. The serum concentration of maternal AFP predicts foetal and/or maternal disorders during gestation.

Aim: The study sought to evaluate serum levels of non-virally infected women in first, second and third trimester

Method: A total of 130 participants: 95(73.1%) pregnant women and 35(26.9%) non pregnant women as controls, all above 15 years, were enrolled in this study. All sera samples were analysed using Enzyme Linked Immunosorbent Assay (ELISA) technique. All data were subjected to statistical analyses using SPSS version 21.0 with statistical significance set at $P<0.05$ and these statistical instruments were used to analyse the data. Mean, Standard Deviation of Mean, ANOVA and Independent student's T-test was used to test for significance of parameters.

Results: Elevated serum AFP was recorded in 1(0.77%) among all the participants. There was a statistical significance between mean of AFP at first (:53.00 ± 31.37), second (29.58±4.80) and third trimesters (94.16±29.2), all in mg/ml. There was a significant difference between control and test participants ($p<0.05$) except between first and second trimesters. Mean±SD weight of the pregnant women based on their trimesters was 60.14±5.49, 64.79±10.92 and 72.42±15.13 (kg) respectively.

Conclusion: This work reveals increase in serum AFP with age of pregnancy with no variation with age of the subjects

Key words: Maternal serum alpha feto protein; Neural tube defect; Down syndrome: Pregnancy

Introduction

Alpha-feto Protein (AFP) is a glycoprotein produced in yolk sac, epithelial cells of the gastrointestinal tract and liver during embryonic development in humans. In pregnancy, AFP enters amniotic fluid through fetal blood, and passes the placenta, thus going into maternal blood. In healthy adults, AFP can be found in the blood in very little concentrations. Normal sera concentrations appear 9 months after birth. Elevated serum AFP levels (above 10 ng/ml) in adults can be found in the patients with acute viral hepatitis, liver cirrhosis, obstructive icterus, and in some malignant diseases, as pancreatic cancer, lung cancer or gastric cancer. *(*Murray *et al.,* 2011*)*AFP serum estimation is done to help prenatal diagnosis of some certain fetal conditions (germ cell tumour, neural tube defect and Downs syndrome). The level increases in neural tube defects (NTD), gastroschisis (open abdominal defects), and fatal death while it decreases in Down's syndrome, multiple pregnancies (triplet, twins).

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The length of gestation, mother's weight, smoking habit and other variables affects the level of AFP. Unexpected increase or decrease level of AFP can be confirmed using ultrasound, amniocentesis to confirm the actual disorder but AFP is a screening test not a diagnostic test (Hulya *et al.,* 2013). Measurement of maternal serum alpha-fetoprotein (AFP) during the first and second trimesters of pregnancy has been shown to be useful in screening for aneuploidies, neural tube defects and adverse pregnancy outcome including fetal death, pre-eclampsia (PE), fetal growth restriction and preterm birth (Nicolaides *et al.,* 2013, Bredake *et al.,* 2012, Huang *et al.,* 2010).

Methodology

Study population

This study was performed in women attending routine ante-natal care at primary Health Care Bukuru Express, Plateau State Hospital Management Board (PSHMB) and Mercy Seat Hospital Angle-Di Salama, Bukuru Express, over a period of 8 months during between 2015 and 2016. One hundred and thirty (130) women were enrolled for the study after a written consent was obtained. Antenatal women from first trimester to third trimester were included in the study. Dating was based on the last menstrual period.

Exclusion Criteria

Patients with HIV or viral diseases, such as hepatitis, female of non-child bearing age, , women not sure of their gestational period were excluded from the study.

Ethical clearance

The ethical clearance was obtained from the ethical clearance committee of Jos University Teaching Hospital (JUTH), Plateau State Specialist Hospital, and Jos South Local Government, Secretariat.

Sample collection

A semi-structured questionnaire was formulated, and data were collected at every antenatal visit. The data collected includes; Age of mother, Gestational age, weight, Ethnicity, family history of child defect, diabetics, use of harmful drugs and smoking. The blood samples were collected between 8- 11am after an overnight fast, from the ante-cubical vein of the fore arm using 2ml syringe which were put into a chemically clean plain redtop test tube**.** The samples were allowed to clot at room temperature and were centrifuged at 3000rpm for 5minutes, the clear unhaemolysed sera were separated from the clotted blood, the serum albumin were stored at -28°C, which were later used to measure serum AFP, the analysis was in batches. The serum AFP were determined using automated solid phase ELISA sandwich method.

Biochemical analysis

The serum AFP were determined using automated solid phase ELISA sandwich method. The test kits were commercially available and were purchased from Hotgen Biotech (Beijing, People's Republic of China)

Statistical analyses

Statistical analyses were performed using SPSS software version 21 for windows 10.0. The differences of pregnancy outcomes (first, second and third trimester) and control were carried out with ANOVA, Student's *t*-test. P value <0.05 was considered statistically significant.

Results

This study carried out to determine the serum concentration of AFP among pregnant women attending primary heath facilities in Bukuru, Plateau state had a total of one hundred and thirty (130) pregnant women enrolled. Out of the 130 participants, 95 were pregnant women while thirty five were non pregnant women who served as control. The demographic characteristics of the one hundred and thirty participants investigated shows that age group 15-24 had a total of 24(25.3%) women, age group 25-34 and greater than 35age group were 34(35.8%) and 37(38.9%) women respectively. In addition, based on the different trimesters,1st trimester had 7(7.4%) participants,2nd and 3rd trimesters were 38(40%) and 50(52%) respectively. On incidence of Diabetes in family,12(12.6%) had history of diabetes mellitus in their family while 83(87.4%) had no history of diabetes mellitus in their family history.

This work also shows that all the participants had not been involved with harmful drugs like marijuana and cigarette smoking however, 2(2.1%) had a history of congenital anomalies while 93(97.9) had no history of congenital anomalies as shown in Table 1.

Table 1. Demographic characteristics of participant

Participants	Frequency (%)	n=130(100%)
Total number of participants		
Age Distribution(years)		
15-24	24 (25.3)	
25-34	34 (35.8)	
>35	37 (38.9)	
Trimesters		
1st	7 (7.4)	
2nd	38 (40.0)	
3rd	50 (52.6)	
Diabetics		
Yes	12 (12.6)	
No	83 (87.4)	
Involvement with Drugs(marijuana)		
Yes	0 (0.00)	
No	95 (100.0)	
Family History of Congenital anomalies		
Yes	2 (2.1)	
No	93 (97.9)	
Smoking Habit		
Yes	0 (0.0)	
No	95 (100.0)	

n –number of subject (total)

The mean ±SD of serum AFP based on the different trimesters and age group is shown in Table 2.The 1st,2nd and 3rd trimesters had a mean±SD of 53±31.37,29.58±4.80 and 94.16±29.2 respectively. On the other hand, serum AFP based on age group indicates that age group 15-24years had mean±SD of 2.08±0.85,age group 25-34years had mean±SD of 2.06±0.85 while the age >35years had a serum mean±SD AFP of 2.08±0.64.

Table 2. Mean ± SD of Serum AFP concentration of pregnant women based on their trimesters and age group

Participants	Number of subjects (%)	Mean ± SD of AFP (ng/ml)
1st trimester	7 (7.4)	53.00±31.37
2nd trimester	38 (40.0)	29.58±4.80
3rd trimester	50 (52.6)	94.16±29.2
Age Group(years)		
15-24	24 (25.3)	2.08±0.72
25-34	34 (35.8)	2.06±0.85
>35	37 (38.9)	2.08±0.64

On comparing the weights of the participants, Table 3 shows that the mean±SD of their weights based on different trimesters had 1st trimester with 60.14±5.49kg,2nd trimester 64.79±10.92kg and 3rd trimester with 72.42±15.13kg.The different age groups had 60.13±6.82kg, 69.76±13.09kg and 72.65±13.68kg for age group 15-25years, 25-35years and >35years respectively.

Table 3. Mean± SD of weight of pregnant women based on their trimesters and age grouping

Participant	Number of Subjects (%)	Weight (kg)
1st Trimester	7 (7.4)	60.14±5.49
2nd Trimester	38 (40.0)	64.79±10.92
3rd Trimester	50 (52.6)	72.42± 15.13
Age Group (years)		
15-24	24 (25.3)	60.13±6.82
25-34	34 (35.8)	69.76±13.09
>35	37 (38.9)	72.65± 13.68

On comparison between Control vs1st trimester, their mean±SD for AFP was 18.11±44.45,53.00±31.37(P=0.05),Controlvs2ndtrimester had mean AFP of 18.11±44.45,29.58±4.80(p= 0.01).The Control vs 3rd trimester had mean±SD AFP 18.11±44.45,94.16±29.2(p= 0.01).In addition, comparison between 1st vs 2nd trimester mean±SD gav53.00 ±31.37,29.58±4.80(p= 0.21),while 1st vs 3rd trimester comparison was 53.00±31.37,94.16±29.20(p= 0.001) as shown in table 4.

Table 4Mean±SD of serum AFP of different trimesters in relation to control

Group	Mean ±SD	p-Value
Control vs 1st trimester	18.11±44.45,53.00±31.37	0.05
Control vs 2nd trimester	18.11±44.45,29.58±4.80	0.01
Control vs 3rd trimester	18.11±44.45,94.16±29.2	0.01
1stvs 2nd trimester	53.00 ±31.37,29.58±4.80	0.21
1stvs 3rd trimester	53.00±31.37,94.16±29.20	0.001
2ndvs 3rd trimester	29.58±4.80, 94.16±29. 20	0.01

P<0.05=significant

When the multiple of median was analysed based on the different trimesters, the Ist trimester had the least multiple of the median of 0 for the range of median 1.5-1.9ng/ml while the highest was 3(42.8%) for the range 2-2.5ng/ml.For the 2nd trimester, the range 0.9-1.4ng/ml was the highest with multiples of 18(47.4%) and 2(5.3%) multiples was the case for 2.0-2.5ng/ml range. For the 3rd trimester,34 (68%) had a range of between 0.9-1.4ng/ml while 1(2%) had range of between 2-2.5ng/ml this is shown in table 5.

Table 5. Distribution of Multiple of the Median (MoM) based on the different trimesters

Trimesters	Multiple of the Median for AFP (ng/ml)				
	0.1-0.4MoM	0.5-0.8MoM	0.9-1.4MoM	1.5-1.9MoM	2.0-2.5MoM
1st	1(14.3%)	1 (14.3%)	2(28.6%)	0 (0.0%)	3 (42.8%)
2nd	6(15.8%)	8(21.0%)	18(47.4%)	4(10.5%)	2(5.3%)
3rd	2(4%)	13(26%)	34(68%)	0(0.0%)	1(2%)

Discussion

This study determined the AFP level of pregnant women in various trimesters. Table showed that there was a statistical significance between the Test participants and the Control participants (P<0.05). These imply that the parameter assessed is increased in pregnant women than the control.

From table 2, Maternal serum AFP concentration in this study increased with increase gestational age which correlates with the study of Foteini *et al* (2011) which revealed a decreases with maternal weight, smoking status, regional difference, racial origin and diabetes (Foteini *et al.*, 2011).This study also correlates with the study done by Deborah *et al* (2014). Most cases with advanced pregnancy had higher AFP levels.

Similarly, the sera AFP concentration of first (3-100ng/ml), second (19-130ng/ml) and third (40-150ng/ml) trimester, falls within the work done by Kratz *et al., 2004* (first 18-119ng/ml, second 96-302ng/ml, and third 160-550ng/ml trimester). The difference might be due to geographical area or race.

Table 3, this study shows the mean maternal weight for second trimester has 64.74±10.92kg. But a study reported by Hsu *et al., (1998),* that mean maternal weight was 54.95 ±7,36kg in Taiwanese pregnant women at the second trimester while Esin et al.,(2014) observed the mean maternal weight at second trimester as 65, 13±11,10kg in Gaziantep population. This correlate with the work of Esin *et al. (*2014) with a slight difference. The differences observed in various studies may be attributable to the gestational ages of the enrolled populations, maternal age differences and regional differences in the subpopulation of women (Hsieh et al.(2007), and Esin *et al*.(2014).

In table 4, there was a significance difference (p <0.05) between the non-pregnant (control) and the trimesters, it could be due to the normal formed foetus in the pregnant women which is not so in non-pregnant women. Within the trimesters, there is a gradual increase from the first to the third trimester it might also be due to the normal growth of the foetus in the womb or due to the presence of disorder with the foetus of the mother. The mean and standard deviation of serum alpha feto protein of first, second, and third trimesters were found to be 60.14 ± 5.49ng/ml (n=7), 64.79± 10.92ng/ml (n=38), and 72.42 ±15.13 ng/ml (n=50) respectively. Maternal serum AFP rises from weeks 10 to 32 and then declines.

Table 5 showed the calculated Multiple of Median (MoM) values in normal pregnancies for AFP at second trimester (13-24weeks) correlate with the result of Sheth *et al 2008* with a MoM value of 1.0 for AFP but at 14-20 weeks. The little increase in the MoM in these study might be due to the gestational week or the number of pregnant women in each study differs.

This study shows there is no high significant association between elevated maternal serum AFP levels (in this study the normal cut off defined as 2.0-2.5 MoM). Only about 14.3% of pregnant women at their first trimester were having MoM values of 0.1- 0.4 while 15.8% and 4% second and third trimester respectively have MoM values of 0.1-0.4. Women with Maternal Serum AFP (MSAPF) greater than 2.5 or less than 2.0 could be offered increased antenatal surveillance in the form of ultrasound scans to confirm it.

Conclusion

Our work has brought to light the fact that the serum concentration of AFP increases with gestational age although this is not affected by the age of pregnant women. There is therefore need for further work to be done on post delivery serum AFP concentrations.

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