



**EVALUATION OF LIPID PROFILE OF HUMAN IMMUNO DEFICIENCY VIRUS (HIV)
AND TUBERCULOSIS (TB) POSITIVE PREGNANT WOMEN**

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ABSTRACT

Pregnancy is the time during which one or more offspring develops and is characterized by significant changes in metabolism, fluid balance, organ function and blood circulation. This study was set to evaluate the Lipid Profile of Human Immuno Deficiency Virus and Tuberculosis positive pregnant Women. A total of four hundred (400) subjects were recruited for this study which comprised of 150 HIV positive pregnant women, 150 TB positive pregnant women and 100 apparently healthy women which served as the control. Fasting Blood samples (4mls) were collected using standard aseptic technique into plain tubes for lipid profile estimation. The results revealed that the mean cholesterol values of HIV (197.45 ± 38.56 mg/dl) and TB (185.65 ± 18.88 mg/dl) positive pregnant women were not statistically significant ($p > 0.05$) as compared to the controls (193.90 ± 53.68 mg/dl). The mean values of Triglyceride (mg/dl) of the HIV positive pregnant women (181.53 ± 26.31 mg/dl) was significantly higher ($p < 0.05$) when compared with the control (148.32 ± 49.98 mg/dl) and the TB positive pregnant women (128.55 ± 22.64 mg/dl). High Density Lipoprotein was significantly higher ($p < 0.05$) for HIV (47.75 ± 7.53 mg/dl), but it was significantly lower for TB (37.95 ± 3.60 mg/dl) when compared with the control (41.53 ± 6.42 mg/dl). The mean values for low Density Lipoprotein for HIV (129.85 ± 26.27 mg/dl) and TB (127.10 ± 18.16 mg/dl) positive pregnant subjects was not statistically significant ($p > 0.782$) when compared with the control (124.30 ± 47.59 mg/dl). There was no statistical significance ($p < 0.05$) in the BMI (kg/m^2) values of the HIV (25.35 ± 2.21 kg/m^2) and TB (24.30 ± 2.36 kg/m^2) positive pregnant women when compared with the control group, (25.35 ± 2.21 kg/m^2). However, this study revealed significant alterations in the lipid profile of HIV and TB positive pregnant women and hence could serve as a pointer in diagnosing and monitoring the progress of such patients.

KEYWORDS: Pregnancy, Lipids, Infections, Transmission.

INTRODUCTION

Pregnancy is the time during which one or more offspring develops inside a woman (Wylie, 2015). It can occur by sexual intercourse or assisted reproductive technology (Abman and Steven 2011). Childbirth typically occurs around 40 weeks from the last menstrual period (LMP) (Abman and Steven 2011). An embryo develops offspring during the first eight weeks following conception, after which, the term fetus is used until birth (Abman and Steven 2011). Symptoms of early pregnancy may include missed periods, tender breasts, nausea, vomiting hunger and frequent urination. Pregnancy is typically divided into three trimesters. The

first trimester is from week 1 through week 12 and includes conception (Abman and Steven 2011). Conception is when the sperm fertilizes the egg. The fertilized egg then travels down the fallopian tube and attaches to the inside of the uterus, where it begins to form the embryo and placenta. The first trimester carries the highest risk of miscarriage. The second trimester is from week 13 through week 28. Around the middle of the second trimester, movement of the fetus may be felt. At 28 weeks, more than 90% of babies can survive outside of the uterus if provided with high-quality medical care. The third trimester is from 29 weeks through 40 weeks (WHO 2015). Prenatal care improves

pregnancy outcomes and may include taking extra folic acid, avoiding drugs and alcohol, regular exercise, blood tests, and regular physical examinations. Complications of pregnancy may include high blood pressure of pregnancy, gestational diabetes, iron-deficiency anemia, severe nausea and vomiting among others. Term pregnancy is 37 to 41 weeks, with early term being 37 and 38 weeks, full term 39 and 40 weeks, and late term 41 weeks. After 41 weeks, it is known as post term. Babies born before 37 weeks are preterm and are at higher risk of health problems such as cerebral palsy. Delivery before 39 weeks by labor induction or caesarean section is not recommended unless required for other medical reasons (WHO 2015).

Lipid are group of naturally occurring molecules that include fats, waxes, sterols, fat-soluble vitamins (such as vitamins A, D, E, and K), monoglycerides, diglycerides, triglycerides, phospholipids, and others. The main biological functions of lipids include storing energy, signaling, and acting as structural components of cell membranes (Fahyet *et al.*, 2009). Lipids have applications in the cosmetic and food industries as well as in nanotechnology (Subramaniam *et al.*, 2011). They may be hydrophobic or amphiphilic small molecules; the amphiphilic nature of some lipids allows them to form structures such as vesicles, multilamellar/unilamellar liposomes, or membranes in an aqueous environment. Biological lipids originate entirely or in part from two distinct types of biochemical subunits or "building-blocks": ketoacyl and isoprene groups. Lipids may be divided into eight categories: fatty acids, glycerolipids, glycerophospholipids, sphingolipids, saccharolipids, and polyketides (Michelle *et al.*, 1999; Fahyet *et al.*, 2009). Although humans and other mammals use various biosynthetic pathways both to break down and to synthesize lipids, some essential lipids cannot be made this way and must be obtained from the diet.

Human Immunodeficiency Virus (HIV) and tuberculosis infections have reached epidemic proportions in Nigeria, where increasing numbers of people now die from these diseases. TB is an important cause of morbidity and mortality worldwide (Corbett *et al.*, 2003). Approximately one-third of the world's population is infected with *Mycobacterium Tuberculosis*, the Bacillus that causes tuberculosis (TB). In 2014, it is estimated that 3.2 million women were infected with TB, 480 000 women died from TB in 2014, including 140 000 deaths among women who were HIV-positive (WHO 2014). TB is the cause of death during pregnancy for somewhere between one in six and one in sixteen women, and pregnant women living with HIV are at especially high risk of dying due to TB (Getahun *et al.*, 2012). TB in a pregnant woman also increases the risk of babies being born prematurely or with a low birth weight. TB during pregnancy is also associated with an increase in prenatal deaths. Among pregnant women with TB, when a pregnant woman is co-infected with HIV, TB doubles the risk of vertical transmission of HIV to the unborn child

(WHO 2009). TB and TB-HIV co infection are associated with poor pregnancy outcomes, including intrauterine growth retardation, prematurity and foetal death, and infant and maternal disease and death (Adhikari, 2009). Gupta *et al.*, (2010) studied the contribution of maternal TB as a risk factor for mother-to-child transmission (MTCT) of HIV infection. Limiting MTCT of HIV infection is a major public health priority, because every infant infected with HIV represents a preventable event. Early initiation of life-long antiretroviral therapy saves lives and improves the outcomes in HIV-infected infants (Violari *et al.*, 2008), but this comes at a huge cost to the health care system and a multitude of long-term adverse effects. Apart from the significant independent association demonstrated between maternal TB and infant HIV infection, the clustering of maternal TB diagnosis and infant HIV infection diagnosis also suggests a possible causal relationship. It demonstrates that prevention of TB among HIV-infected mothers should be considered as part of a well-functioning prevention of HIV MTCT program. The exceptionally high TB disease and transmission risk provides additional motivation to carefully monitor all HIV-infected women for TB during and after pregnancy (Marias *et al.*, 2009). More operational research is required to clearly define the feasibility, benefits, and risks of providing routine TB preventive therapy to HIV-infected pregnant mothers in areas where TB is endemic (Atun *et al.*, 2010). However, there is limited information on the assessment of lipid profiles of HIV and TB pregnant women in Esan Land. Hence, this study intends to evaluate the lipid profile of pregnant women to see how significant they predispose pregnant women in Esan Land, Edo State.

MATERIALS AND METHODS

Study Area

This study was carried in Esan land, Edo Central senatorial district of Edo state in south Southern Nigeria. This area is located between latitude $6^{\circ} 10'$ and $6^{\circ} 45'$ north of the equator and between longitudes $6^{\circ} 10'$ and $6^{\circ} 30'$ east of the Greenwich Meridian (Akinbode, 1983). The 2006 national census put the population of the study area at 591,534 people Projected to 2014 at 2.8 percent national growth rate, the 2014 population of the study area is 734,583 (World Gazetteer, 2007).

Study Design

The research was designed as prospective cross-sectional study to assess the lipid profile of HIV and TB positive pregnant women attending ANC in Irrua Specialist Hospital, Central Hospital Uromi and ST. Camillus Hospital Uromi, Edo State, Nigeria. After obtaining verbal informed consent to be enrolled into the research, the height and weight of the subjects were measured using approved standard techniques to calculate the BMI. Fasting blood samples (4mls) were collected into plain vacutainer tubes for lipid profile which includes; Total Cholesterol, Triglyceride, High Density Lipoprotein, Low Density Lipoprotein.

Data Collection and Anthropometric Measurement

The data collected were age, number of pregnancies (gravidity) and the age (trimester) of the pregnancy. Weight and height were measured by pre-defined procedure to evaluate the Body Mass Index (BMI) which indicates the usual health and nutritional status of individual (WHO, 1995).

Ethical Approval

Ethical approval was sought from the Ethics and Research committees of Ambrose Alli University, Ekpoma, Edo State.

Study Population

A total of One hundred subjects were recruited for this study which comprised of forty (40) pregnant women without HIV and TB (controls), Forty (40) HIV positive pregnant women and twenty (20) TB positive pregnant mothers. The subjects were pregnant women attending Antenatal Clinics (ANC) in Irrua Specialist Hospitals Irrua, Central Hospital Uromi and ST. Camillus Hospital Uromi, Edo State.

Informed Consent

A verbal informed consent was sought for from each pregnant woman and control subjects, assuring them of the confidentiality of the information provided and results beyond the scope and purpose of the study.

Inclusion Criterion

HIV and TB positive pregnant women and HIV and TB negative pregnant women attending Irrua Specialist Hospital Irrua, Central Hospital Uromi and ST. Camillus Hospital Uromi who gave consent were purposefully included in the study.

Exclusion Criterion

All women who refused to give an informed consent and those pregnant women with serious complications attending ANC and ART Clinic in Irrua Specialist Hospital, Central Hospital Uromi and St Camillus Hospital Uromi were excluded from this study.

Sample Collection and Processing

Fasting Blood samples (4mls) were collected using standard aseptic technique into plain tubes for lipid profile estimation. The samples were allowed to clot and centrifuged at 3000 rpm for 5 minutes to obtain the serum.

Sample Analysis

Total Cholesterol, Triglyceride, HDL, LDL were assayed using RANDOX – kit (Abell Kendell method) by automation method as described by RANDOX Laboratories, (2010a). Body Mass Index was calculated as Weight (kg)/ h (m²). Underweight = <18.5kg/m², Normal/healthy weight = 18.5 – 24.5kg/m², Overweight 25 – 29.9kg/m² and Obesity = 30kg/m² and above (WHO, 1995).

RESULTS

A total of one hundred (100) pregnant women at different trimesters were recruited in this study, among these were 40 HIV positive pregnant women, 20 TB positive pregnant women and 40 HIV and TB negative pregnant women that served as the control group. The age group 20 – 29 years had the highest number with 65% (26), this was followed by 30 – 40 years with 35% (14). According to the number or occurrence of pregnancy (Parity/Gravidity) in the study groups, for No or one (± 1) pregnancy; control amounted to 22.5% (9), HIV amounted to 22.5% (9) and TB 77.5% (5), while for 2 – 4 pregnancies; control amounted to 77% (31), HIV amounted to 77.5% (31) and TB amounted to 75% (15). According to their BMI categorization, none had BMI less than 18.5kg/m² for all subjects, those within 18.5 – 24.9kg/m² were 47.5% (19), 32.5% (13) and 60% (12) for control, HIV and TB groups respectively, while those with BMI between 25.0 – 29.9kg/m² were 45% (18), 60% (24) and 40% (8) for control, HIV and TB groups respectively. 7.5% (3) of both the control group and HIV group had BMIs of 30.0kg/m² and above (Table 1).

The results on lipid profile revealed that the mean cholesterol values of HIV (197.45 \pm 38.56 mg/dl) and TB (185.65 \pm 18.88 mg/dl) positive pregnant women were not statistically significant ($p > 0.05$) as compared to the controls (193.90 \pm 53.68 mg/dl). The mean values of Triglyceride (mg/dl) of the HIV positive pregnant women (181.53 \pm 26.31 mg/dl) was significantly higher ($p < 0.05$) when compared with the control (148.32 \pm 49.98 mg/dl) and the TB positive pregnant women (128.55 \pm 22.64 mg/dl). High Density Lipoprotein was significantly higher ($p < 0.05$) for HIV (47.75 \pm 7.53 mg/dl), but it was significantly lower for TB (37.95 \pm 3.60 mg/dl) when compared with the control (41.53 \pm 6.42 mg/dl). The mean values for low Density Lipoprotein for HIV (129.85 \pm 26.27 mg/dl) and TB (127.10 \pm 18.16 mg/dl) positive pregnant subjects was not statistically significant ($p > 0.782$) when compared with the control (124.30 \pm 47.59mg/dl). There was no statistical significance ($p < 0.05$) in the BMI (mg/m²) values of the HIV (25.35 \pm 2.21 kg/m²) and TB (24.30 \pm 2.36 kg/m²) positive pregnant women when compared with the control group, (25.35 \pm 2.21 kg/m²) (Table 2).

The total cholesterol levels were higher in the third trimester (213.50 \pm 46.06 mg/dl) as compared to the first (197.88 \pm 46.17 mg/dl) and second (190.00 \pm 7.64 mg/dl) trimesters and this observation was not statistically significant ($p > 0.05$). The mean values of Triglyceride of the different trimesters increased progressively in the first (162.38 \pm 11.98 mg/dl), second (179.64 \pm 22.19) and third (201.00 \pm 31.34) trimesters and the increase was statistically significant ($p < 0.05$). The mean values of High Density Lipoprotein increased from the 1st to the 2nd through the 3rd trimesters (43.13 \pm 2.36 mg/dl, 46.46 \pm 6.28mg/dl and 54.30 \pm 8.87 mg/dl) respectively, which was statistically significant ($p = 0.002$). The mean values of Low Density Lipoprotein were

129.63±21.77mg/dl in the 1st trimester, 128.36±23.77mg/dl in the 2nd trimester and 133.30±35.87mg/dl in the 3rd trimester. The difference was not statistically significant ($p>0.05$) (Table 3).

The mean total cholesterol values of the TB infected pregnant women were 187.43±17.15 mg/dl, 188.09±19.88mg/dl and 166.00±14.14mg/dl in the 1st, 2nd and 3rd trimesters respectively and this observation was not statistically significant ($p<0.05$). There was no significant difference ($p>0.05$) when the mean triglyceride levels of the TB infected pregnant women were compared in the first (133.86±26.36mg/dl), second

(124.00±22.05mg/dl) and third trimesters (135.00±14.14mg/dl). There was no significant difference ($p<0.05$) when the mean HDL values were compared across the first (39.14±2.67mg/dl), second (37.64±4.18mg/dl) and third (35.50±2.12mg/dl) trimesters. There was no significant difference when the Low Density Lipoprotein was compared across the first (131.34±11.47mg/dl), second (123.36±18.08mg/dl) and third (132.50±43.13mg/dl) trimesters. The mean BMI levels were not statistically significance across the first (24.71±2.62 kg/m²), second (24.36±2.37 kg/m²) and third (24.30±2.36 kg/m²) trimesters (Table 4).

Table 1: Demographic Characteristics of the Study Population.

Variables	Pregnant Women Control (n = 40)	HIV Pregnant Women (n = 40)	TB pregnant women (n = 20)
Age groups (years)			
20 – 29	26 (65%)	14 (35%)	13 (65%)
30 – 39	13 (32.5%)	25 (62.5%)	7 (35%)
40+	1 (2.5%)	1 (2.5%)	0 (0%)
Gravidity			
±1	9 (22.5%)	9 (22.5%)	5 (25%)
2 – 4	31 (77.5%)	31 (77.5)	15 (75%)
BMI (kg/m²)			
<18.5	0 (0%)	0 (0%)	0 (0%)
18.5 – 24.9	19 (47.5%)	13 (32.5%)	12 (60%)
25.0 – 29.9	18 (45%)	24 (60%)	8 (40%)
30+	3 (7.5%)	3 (7.5%)	0 (0%)

KEY: BMI = Body Mass Index (kg/m²), n= Sample size, Gravidity = Number of pregnancies.

Table 2: Lipid Profile and BMI of HIV and TB Positive Pregnant Women in comparison with the control.

Parameters	Control (n=40)	HIV (n=40)	TB (n=20)	F-value	P-value	Sig
TC (mg/dl)	193.90±53.68 ^a	197.45±38.56 ^a	185.65±18.88 ^a	0.509	0.603	NS
TG (mg/dl)	148.32±49.98 ^a	181.53±26.31 ^b	128.55±22.64 ^c	17.585	0.000*	S
HDL (mg/dl)	41.53±6.42 ^a	47.75±7.53 ^b	37.97±3.60 ^c	17.787	0.000*	S
LDL (mg/dl)	124.30±47.59 ^a	129.85±26.27 ^a	127.10±18.16 ^a	0.246	0.782	NS
BMI (kg/m ²)	25.42±3.09 ^a	25.35±2.21 ^a	24.30±2.36 ^a	1.376	0.258	S

Key: Sig = Significance. NS= Not significant. S= Significant. *= Statistically significant at $p\leq 0.05$, BMI=Body Mass Index. HIV= HIV positive pregnant women. TB= TB positive pregnant women. Mg/dl= Milligrams per Decilitre n= Number of subjects; TC= Total Cholesterol, TG=Triglyceride, HDL=High Density Lipoprotein, LDL= Low Density Lipoprotein

Table 3: Lipid Profile and BMI of HIV-Infected Pregnant Women Based on Trimesters.

Parameters	First Trimester (n=8)	Second Trimester (n=22)	Third Trimester (n=10)	F-value	P-value	Sig
TC(mg/dl)	197.88±46.17	190.00±7.64	213.50±46.06	1.297	0.286	NS
TG(mg/dl)	162.38±11.98	179.64±22.19	201.00±31.34	6.237	0.005*	S
HDL(mg/dl)	43.13±2.36	46.46±6.28	54.30±8.87	7.476	0.002*	S
LDL(mg/dl)	129.63±21.77	128.36±23.77	133.30±35.86	0.116	0.891	NS
BMI(kg/m ²)	24.62±2.61	25.50±2.01	25.60±2.41	0.530	0.593	NS

Key: Sig = Significance, NS= Not significant, S= Significant. *= Statistically significant at $p\leq 0.05$, HIV= HIV positive pregnant women. TB= TB positive pregnant women. Mg/dl= Milligrams per Decilitre. n= Number of subjects. TC=Total Cholesterol. TG= Triglyceride. HDL= High Density Lipoprotein. LDL=Low Density Lipoprotein. BMI=Body Mass Index.

Table 4: Lipid Profile and BMI of TB-Infected Pregnant Women Based on Trimesters.

Parameters	FIRST TRIMESTER (n=7)	SECOND TRIMESTRE (n=11)	THIRD TRIMESTER (n=2)	F-VALUE	P-VALUE	SIG
TC (mg/dl)	187.43±17.15	188.09±19.88	166.00±14.14	1.236	0.315	NS
TG(mg/dl)	133.86±26.36	124.00±22.05	135.00±14.14	0.468	0.634	NS
HDL(mg/dl)	39.14±2.67	37.64±4.18	35.50±2.12	0.883	0.432	NS
LDL(mg/dl)	131.43±11.47	123.36±18.08	132.50±43.13	0.492	0.620	NS
BMI(kg/m ²)	24.71±2.62	24.36±2.37	24.30±2.36	0.667	0.526	NS

Key: Sig = Significance, NS= Not significant, S= Significant. *= Statistically significant at $p \leq 0.05$, Mg/dl= Milligrams per Decilitre, n= Number of subjects. TC=Total Cholesterol, TG= Triglyceride, HDL= High Density Lipoprotein, LDL=Low Density Lipoprotein, BMI= Body Mass Index.

DISCUSSION

Pregnancy causes significant changes in metabolism, fluid balance, organ function and blood circulation which are driven by estrogen and the presence of feto-placental unit (Mahmoud *et al.*, 2013). These dramatic changes coupled with other illness like HIV and TB influence a wide variety of parameters. However, these changes are essential when interpreting the result of investigations to diagnose or monitor illness in pregnant women (Mahmoud *et al.*, 2013).

The use of ART during pregnancy is associated with several concerns, which include potential risk for the exposed and uninfected newborn, possible reduced efficacy of antiretroviral regimens in this particular condition, and safety considerations for the mother, including potentially increased risk of specific adverse events (Florida *et al.*, 2006). This study was carried out to evaluate the nutritional indices among HIV and TB positive pregnant women in Esan Land, Edo State.

In this study, none of the pregnant mothers were below 20 years with majority of them being within the age of 20 – 29 years and between 30-39 years of age. This finding reveals the age range that reflects the reproductive group. Also, none among the pregnant women studied had BMI less than 18.5kgm².

The study revealed that the mean total cholesterol was high in HIV positive pregnant women when compared with the control patients and the TB positive pregnant women. This however connotes significant alterations in the lipid profile among the pregnant women studied. This increase was not statistically significant ($p > 0.05$). The triglyceride and high density lipoprotein (HDLc) showed a statistically significant increase among pregnant women with HIV ($p < 0.05$) when compared with control and pregnant women with TB. This is in agreement with the findings of Khiangte *et al.*, (2007) but at variance with the study of Manafa *et al.*, (2012) who reported a decrease in lipid profile in HIV pregnant women in Nnewi Nigeria. Furthermore, Obirikorang *et al.*, (2014) reported a significant increase in serum triglyceride among HIV patients compared to controls, but their finding on a significant decrease in LDL-cholesterol by the same study is not in agreement with the results of this study.

The present study also showed lower values in the mean level of serum total cholesterol, triglyceride and HDL in TB positive pregnant women when compared to the controls and HIV positive pregnant women. Triglyceride and LDL-cholesterol are major constituents of cell membrane while HDL-cholesterol protects arterial walls of the blood circulatory system (Liesuy and Tomato, 1994). The low concentrations of triglyceride and total cholesterol observed among tuberculosis patients in this study correlates with the findings of previous studies (Kwiatkowska *et al.*, 1999, Perez-Guzman *et al.*, 2002, Mohamed and Heshman, 2012). Reddy *et al.*, (2009), have reported increased lipid peroxidation in all categories of TB patients. This might have caused the reduction in the concentration of serum lipid as noted in this study. The Reduced concentrations of these lipid fractions could have resulted from tissue and cells damage and consequently lead to wasting and weight loss which are often observed in TB patients. The lower levels of the lipid fractions observed in TB pregnant women when compared with HIV and control subjects could also be as a result of impaired rate of lipid production and enhanced rate of lipid catabolic rate associated with pulmonary TB infection.

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