ASSESSMENT OF LIPIDS AND ITS ARTHEROGENIC INDEX IN FEMALE INFERTILITY IN OWERRI, SOUTH EAST NIGERIA

Chikezie Onyebuchi Desmond[1*], Maduka Ignatius Chinedu[2], Ezugwu Uchenna Modestus[1], Awalu Chimezie Joseph[1], Ezuma Kenneth Emeka[3]

[1]Department of Medical Laboratory Science, Chemical Pathology Unit College of Health Sciences and Technology, Nnamdi Azikiwe University, Nnewi Campus, **NIGERIA**
[2]Human Biochemistry, Nnamdi Azikiwe University, Nnewi Campus, **NIGERIA**
[3]Department of Medical Laboratory Science, Madonna University Elele Campus, Rivers **NIGERIA**

ABSTRACT

Female infertility has been a global problem. Dyslipidemia have also seen associated with infertility. This study aimed to evaluate the serum lipid profile and its atherogenic index in female infertility. This is a cross-sectional study that involved the recruitment of 220 female subjects (age 20-45 years) attending Federal Medical Centre (FMC), Owerri Nigeria. They include 110 already diagnosed infertile women and 110 of apparently healthy fertile women served as the control subjects. Total cholesterol (TC), triglycerides (TG), high density lipoprotein cholesterol (HDL-c), low density lipoprotein cholesterol (LDL-c) and very low density lipoprotein cholesterol (VLDL-c) were determined. Atherogenic indices, TC/HDL-c, LDL-c/HDL-c and log (TG/HDL-c) (AIP) was calculated. Mean serum concentration of TG, VLDL-c, AIP were significantly lower in the infertile group when compared to the fertile group ($P < 0.05$), but there was no significant difference in TC/HDL-c and LDL-c/HDL-c between the two groups. Furthermore, the mean concentrations of TC, HDL-c, and LDL-c were all higher in the infertile group compared to fertile group and were all statistically significant ($P < 0.05$). Correlation analysis showed significant positive associations between TC and LDL-c, TC/HDL-c, LDL/HDL-c in infertile women while the fertile group also showed the same pattern of association of TC and LDL-c, VLDL-c, TC/HDL-c, LDL/HDL-c and AIP ($P < 0.05$). The TG also shows a significant positive association with VLDL-c, TC/HDL and AIP ($P < 0.05$) in infertile group and the same in fertile group including TC. There were negative significant correlations in both infertile and fertile group when HDL-c was correlated with TC/HDL-c, LDL-c/HDL-c, and AIP ($P < 0.05$). LDL-c shows a positive significant association with TC/HDL-c and LDL-c/HDL-c in infertile women while in fertile group it is positively associated with TC/HDL-c, LDL-c/HDL-c and AIP ($P < 0.05$). In both infertile and fertile group, the same pattern of association was seen in VLDL-c with TC/HDL-c and AIP; TC/HDL-c with LDL-c/HDL-c and AIP; and LDL-c/HDL-c with AIP ($P < 0.05$). In conclusion, infertile women have almost the same pattern of lipid distributions and artherogenic risk predicting factor with the fertile women. A triglyceride based index (AIP) can significantly add value when assessing the risk of developing atherosclerosis in Nigeria than other atherogenic indices.

Keywords: Lipid profile, atherogenic index of plasma (AIP), female infertility, dyslipidaemia, atherosclerosis.

INTRODUCTION

Infertility has been a global problem of public health importance both in Nigeria and many part of Africa, especially sub-Saharan region. This is not only because of the high prevalence but also due to the important social effect on affected couples and family. In Africa, children are the fabric of society, without which no meaningful social and economic progress is

considered worthwhile. Infertility is seen in both men and women, yet women, particularly in developing countries may bear the sole blame for barren marriages; and in many areas infertility is a socially acceptable basis for divorce by the husband[1]. Infertility in women is fundamentally the inability to conceive a baby after 12 months of contraceptive-free intercourse[2]. Infertility is defined as the failure to conceive after one year of regular intercourse in women < 35 years not using contraception and after six months in women > 35 years[3]. Demographers tend to define infertility as childlessness in a population of women of reproductive age, whereas the epidemiological definition refers to "trying for" or "time to" a pregnancy, generally in a population of women exposed to a probability of conception[4]. Existing definitions of infertility lack uniformity, rendering comparisons in prevalence between countries or over time problematic. A couple that tries unsuccessfully to have a child after a certain period of time (often a short period, but definitions vary) is sometimes said to be sub fertile, meaning less fertile than a typical couple but experience shows that many of these supposedly "infertile" couples will eventually conceive, even without treatment.

Lipid Profile or lipid panel, is a panel of blood tests that serves as an initial broad medical screening tool for abnormalities in lipids, such as cholesterol and triglycerides[5]. Lipid profile consists of a group of biochemical tests often used in predicting, diagnosing and treating lipid-related disorders including atherosclerosis[6]. The results of this test can identify certain genetic diseases and can determine approximate risks for cardiovascular disease, certain forms of pancreatitis, and other diseases[7]. The lipid profile typically includes low-density lipoprotein cholesterol (LDL-c), high-density lipoprotein cholesterol (HDL-c), triglycerides (TG) and total cholesterol[8]. Cholesterol is important for life; it is needed to build and repair all animal and human cell membranes and to make bile acids and steroid hormones, such as estrogens, progesterone, and testosterone.

LITERATURE REVIEW

Dyslipidemia is becoming increasingly prevalent all around the world. Many of its possible causes are not linked to hormonal disequilibrium. However, hyperlipidemia can be related to various hormonal diseases such as diabetes, polycystic ovary syndrome and metabolic syndrome. Besides the evident consequence of cardiovascular disease, dyslipidemia and the associated syndromes have also seen associated with infertility[9, 10]. Generally, the hyperlipidemias are of interest to the physician in the context of risk factors for ischaemic heart disease (IHD) and peripheral vascular diseases[11]. The first step in diagnosis of hyper- and hypolipoproteinaemias is to define the lipoprotein pattern by chemical analysis of the plasma lipids and lipoproteins[12]. Abundant evidence has accumulated, relating the concentrations of lipids (total cholesterol and triglycerides) and their associated blood transporting lipoproteins (HDL-c, LDL-c, VLDL-c) with the occurrence of atherosclerosis in general and coronary artery disease (CAD) in particular[13]. The strong association between the risk of coronary artery diseases (CAD), high levels of LDL-c and low levels of HDL-c has been well established[14, 15]. However the enormous contributions of triglycerides (TG) to cardiovascular risk have been underestimated especially in our environment[16]. Indeed high levels have been associated with an increased incidence of CAD[17] and an increased population of small dense LDL-c particles[18]. A lot of work has been done on the relationship between TG and HDL-C, and it has been shown that the ratio of TG to HDL-C was a strong predictor of myocardial infarction[19]. Universally, atherogenic index of plasma (AIP) calculated as log (TG/HDL-C) has been used by some practitioners as a significant predictor of atherosclerosis[20, 21].

In Nigeria, fertility rates have been put at approximately six children per woman[22]; notwithstanding a "high rate of pregnancy wastage" [23]. These data seems odd with growing concerns about infertility as such. As Hollos, [24] put it, "The problem of infertility in sub-Saharan Africa (including Nigeria) received comparatively little attention until recently". Hollos further emphasized that the problem of infertility "was obscured by the region's high fertility rates, which gave rise to a global climate of concern over population growth and high fertility that is not conducive to the perception of infertility as a real problem."[24]

Understandably, the main focus has been on how to tackle these problems and this invariably results to neglecting the effect of lipids on female fertility. In this study, we adopted atherogenic lipoprotein profile characterized by high ratio of LDL-c to HDL-c, TC to HDL-c and specifically triglyceride based index as an indicator and a significant adjunct for predicting atherosclerosis as well as cardiovascular diseases (CVD) in female infertility.

MATERIALS AND METHODS

Subjects and Methods

This is a cross-sectional study that involved the recruitment of 220 female subjects (age 20-45 years) attending Federal Medical Centre (FMC), Owerri Nigeria. They were selected by simple random sampling. They include one hundred and ten (110) already diagnosed infertile women who are not on any form of medication as the test subjects. One hundred and ten (110) of apparently healthy fertile women served as the control subjects. Informed consent was obtained from both groups and ethical clearance was obtained from the relevant authority of Federal Medical Centre (FMC), Owerri (Ethical approval number: 017871). Only primary infertility was considered in sample collection for already diagnosed infertile women. The control subjects consist of women who had been confirmed pregnant in the last one year. The following was excluded from the control group: pregnant women, breast feeding or lactating mothers, diabetic mothers, infertile women and women taking oral contraceptives. The following was excluded from the test group: women taking oral contraceptives or who have been on any steroid, ACTH, gonadotropin and estradiol medication.

Fasting blood samples (5.0mls) were collected by venopuncture from the antecubital vein, into sterile plain tubes, under aseptic conditions. The blood samples were allowed to clot and centrifuged at 4000 rpm for 5 minutes. Serum was stored frozen at −20° C and the analysis was carried out within one week of sample collection. The serum was used for the analysis of total cholesterol, triglycerides, and HDL-cholesterol levels. Total cholesterol was measured using established enzymatic methods of Allain *et al*[25] with the Randox cholesterol kit (Randox England). HDL-c was estimated by HDL-c precipitant method[26]. Triglyceride was assessed enzymatically[27]. LDL-C was calculated using the Friedewald formula[8]. VLDL-c was calculated based on the formula VLDL-c=TG/5.5[8]. Specific lipid rations, such as TC to HDL (TC/HDL), LDL to HDL (LDL/HDL), Log TG to HDL (Log TG/HDL) were also calculated. The principle of the method for measuring total cholesterol involves the use of three enzymes: Cholesterol Esterase (CE), Cholesterol Oxidase (CO) and Peroxidase (POD). In the presence of the former, the mixture of phenol and 4-aminoantipyrine (4-AA) are condensed by hydrogen peroxide to form a quinoneimine dye, the intensity of which is proportional to the concentration of cholesterol in the sample[25]. The HDL-c measuring technique uses a separation method based on the selective precipitation of low density lipoproteins (LDL-c and VLDL-c) and chylomicron fractions quantitatively by the addition of phosphotungstic acid in the presence of magnesium ions. After centrifugation, the cholesterol concentration in HDL fraction, which remains in the supernatant, is determined[26]. The principle of the

method for measurement of triglyceride is that sample triglycerides incubated with lipoprotein lipase (LPL), liberate glycerol and free fatty acids. Glycerol is converted to glycerol-3-phosphate (G3P) and adenosine-5-diphosphate (ADP) by glycerol kinase and ATP Glycerol-3-phosphate is then converted by glycerol phosphate dehydrogenase (GPO) to dihydroxyacetone phosphate (DAP) and hydrogen peroxide (H2O2). In the last reaction, hydrogen peroxide reacts with 4-aminophenazone (4-AP) and p-Chlorophenol in the presence of peroxidase (POD) to give red colored dye the intensity of which is proportional to the triglycerides concentration in the sample[27]. The LDL concentration was estimated using the equation of Friedewald *et al* [8] because women with TG concentrations greater than 399 mg/dL (4.5 mmol/L) were excluded from the analyses since this equation is not applicable in those with TG levels greater than 400 mg/dl.

Statistical Analysis

The data obtained were analyzed using SPSS version 21[28]. Student t-test was used to compare the differences in the parameters measured between the two groups, results were presented in mean and standard deviation (Mean ± SD). The Pearson's correlation coefficient was used to assess the level of association among the variables. Significant level was considered at P - value less than 0.05 ($P < 0.05$).

RESULTS

Table 1 shows comparison of mean level of TC, TG, HDL-c, LDL-c, VLDL-c, TC/HDL-c, LDL/HDL-c and AIP levels in the serum of infertile and apparently healthy fertile female group (mean ±SD). Mean serum concentration of TG, VLDL-c, AIP were significantly lower in the infertile group when compared to the fertile group ($P < 0.05$), but there was no significant difference in TC/HDL-c and LDL-c/HDL-c between the two groups. Furthermore, the mean concentrations of TC, HDL-c, and LDL-c were all higher in the infertile group compared to fertile group and were all statistically significant ($P < 0.05$). **Table 2** shows correlation values among TC, TG, HDL-c, LDL-c, VLDL-c, TC/HDL-c, LDL-c/HDL-c and AIP in serum of infertile and apparently healthy fertile female. Correlation analysis showed significant positive associations between TC and LDL-c, TC/HDL-c, LDL/HDL-c in infertile women while the fertile group also showed the same pattern of association of TC and LDL-c, VLDL-c, TC/HDL-c, LDL/HDL-c and AIP ($P < 0.05$). The TG also shows a significant positive association with VLDL-c, TC/HDL and AIP ($P < 0.05$) in infertile group and the same in fertile group including TC. There were negative significant correlations in both infertile and fertile group when HDL-c was correlated with TC/HDL-c, LDL-c/HDL-c, and AIP ($P < 0.05$). LDL-c shows a positive significant association with TC/HDL-c and LDL-c/HDL-c in infertile women while in fertile group it is positively associated with TC/HDL-c, LDL-c/HDL-c and AIP ($P < 0.05$). In both infertile and fertile the same pattern of association was seen in VLDL-c with TC/HDL-c and AIP; TC/HDL-c with LDL-c/HDL-c and AIP; and LDL-c/HDL-c with AIP ($P < 0.05$).

Table 1: Comparison of mean level of TC, TG, HDL-c, LDL-c, VLDL-c, TC/HDL-c, LDL/HDL-c and AIP levels in the serum of infertile and apparently healthy fertile female group (mean ±SD)

	GROUP			
	INFERTILE (n=110)	FERTILE (n=110)		
PARAMETER	(mg/dl)	(mg/dl)	t-value	p-value
TG	129.57 ±20.94	141.78 ±27.97	3.664	0.000*
TC	219.37 ±32.44	211.13 ±25.95	2.082	0.039*
HDL-c	46.67 ±7.19	44.50 ±3.52	2.846	0.005*
LDL-c	146.85 ±31.53	138.27 ±24.78	2.244	0.026*
VLDL-c	25.85 ±4.23	28.42 ±5.49	3.893	0.000*
TC/HDL-c	4.77 ±0.86	4.79 ±0.69	0.139	0.890
LDL-c/HDL-c	3.21 ±0.80	3.15 ±0.63	0.667	0.506
AIP	0.43 ±0.11	0.48 ±0.10	3.528	0.001*

*Significant at P < 0.05; TG means Triglyceride; TC means Total cholesterol; HDL-C means High density lipoprotein cholesterol; LDL-C means Low density lipoprotein cholesterol; VLDL-C means Very low density lipoprotein cholesterol and AIP means atherogenic index of plasma.

Table 2: Correlation values among TC, TG, HDL-c, LDL-c, VLDL-c, TC/HDL-c, LDL-c/HDL-c and AIP in serum of infertile and apparently healthy fertile female.

	GROUP			
	INFERTILE (n = 110)		FERTILE (n = 110)	
PARAMETER	ɤ - value	p – value	ɤ - value	p-value
TC vs TG	0.154	0.108	0.348	0.000*
TC vs HDL-c	0.169	0.078	0.019	0.844
TC vs LDL-c	0.971	0.000*	0.966	0.000*
TC vs VLDL-c	0.147	0.125	0.341	0.000*
TC vs TC/HDL-c	0.663	0.000*	0.818	0.000*
TC vs LDL-c/HDL-c	0.714	0.000*	0.753	0.000*
TC vs AIP	-0.065	0.502	0.376	0.000*
TG vs HDL-c	-0.142	0.138	0.064	0.505
TG vs LDL-c	0.057	0.553	0.135	0.159
TG vs VLDL-c	0.998	0.000*	0.999	0.000*
TG vs TC/HDL-c	0.239	0.012*	0.244	0.010*
TG vs LDL-c/HDL-c	0.156	0.105	0.057	0.553
TG vs AIP	0.753	0.000*	0.873	0.000*
HDL-c vs LDL-c	-0.036	0.712	-0.137	0.152
HDL-c vs VLDL-c	-0.142	0.140	0.077	0.427
HDL-c vs TC/HDL-c	-0.588	0.000*	-0.534	0.000*
HDL-c vs LDL-c/HDL-c	-0.534	0.000*	-0.534	0.000*
HDL-c vs AIP	-0.678	0.000*	-0.346	0.000*
LDL-c vs VLDL-c	0.049	0.608	0.125	0.192
LDL-c vs TC/HDL-c	0.786	0.000*	0.878	0.000*
LDL-c vs LDL-c/HDL-c	0.837	0.000*	0.851	0.000*
LDL-c vs AIP	-0.012	0.899	0.252	0.008*
VLDL-c vs TC/HDL-c	0.228	0.016*	0.230	0.015*
VLDL-c vs LDL-c/HDL-c	0.144	0.132	0.045	0.642
VLDL-c vs AIP	0.747	0.000*	0.861	0.000*
TC/HDL-c vs LDL-c/HDL-c	0.988	0.000*	0.936	0.000*
TC/HDL-c vs AIP	0.477	0.000*	0.507	0.000*
LDL-c/HDL-c vs AIP	0.382	0.000*	0.328	0.000*

TG means Triglyceride; TC means Total Cholesterol; HDL-c means High Density Lipoprotein Cholesterol; LDL-c means Low Density Lipoprotein Cholesterol; VLDL-c means Very Low Density Lipoprotein Cholesterol; VS means Verse and AIP means atherogenic index of plasma.

DISCUSSION

Female infertility has been a global problem. It is a social menace in our society. It has economic, social and psychological impact on the individual, and our society at large. In Africa, female infertility produces social consequences for African women and these consequences are particularly profound for women as compared to men, regardless of the cause of infertility[1]. This study was carried out to evaluate lipids and its atherogenic implications in female infertility.

Findings of this study shows significantly lower TG, VLDL-c and AIP in the infertile group when compared to the fertile group ($P < 0.05$) but higher TC, HDL-c, and LDL-c in the infertile group compared to fertile group ($P < 0.05$). The results do not agree with the study of Mahmood[29], which found that infertile female have a significantly elevated TG compared to the control. This pattern of dyslipidemia is mostly found in different causes of infertility. Some studies reported that high TG levels are found in both obese and non obese women suffering from infertility[30, 31]. High serum levels of TC and LDL-c in infertile female is an indication that steroid genesis may not be taking place adequately and this could be the reason of infertility in that group. This agrees with a study which found that women with different causes of infertility had increase TC levels[32]. Although cholesterol is a precursor of steroid hormones, high level of it can cause infertility in female[33]. This means that increase in mean serum levels of total cholesterol in infertile female could be the reasons for their infertility. In general, high free cholesterol was associated with more difficulty getting pregnant and lower levels of fertility[33]. Abnormalities of LDL-c had not been found consistently in some cases of infertility. Even those with normal LDL-c level had shown increase VLDL-c relative to control group[34]. LDL- c which is high in infertile group may be as a result of dyslipidemia. Besides, the evident consequences of cardiovascular diseases, dyslipidemia and the associated syndromes have also been associated with infertility. The influence of abnormal lipoprotein metabolism on female infertility has not been thoroughly explored, despite observations suggesting a potential role for plasma lipoproteins, especially HDL-c[35]. Lipoproteins transport between tissues a number of lipids (e.g., cholesterol, steroid hormones, and vitamin E) that either directly, or indirectly through their metabolic products, play key roles in fertility. Several lipid ratios have been proposed as simple clinical indicators because of the integrative information of the multiple variables. These lipid ratios are strong indicators of cardiovascular risk by its expressions of imbalance between atherogenic and protective lipoproteins[36, 37]. According to Grover *et al*, the ration of LDL-c/HDL-c or TC/HDL-c is the best related predictor of future cardiovascular events[38]. Later TG/HDL-c was show to be a more accurate predictor of heart diseases[39]. In this study, there is no significant difference between the two groups in the atherogenic makers measured by TC/HDL-c and LDL-c/HDL-c but a significant low serum level of AIP was seen in infertile group when compared with fertile group. This implies that the fertile group has higher risks of developing cardiovascular problem than the infertile group using AIP as atherogenic marker while both groups have equal chances of developing the problem using TC/HDL-c and LDL-c/HDL-c as atherogenic marker.

Correlation analysis showed significant positive associations between TC and LDL-c, TC/HDL-c, LDL/HDL-c in infertile women while the fertile group also showed the same

pattern of association of TC and LDL-c, VLDL-c, TC/HDL-c, LDL/HDL-c and AIP ($P < 0.05$). This implies that increase in serum level of TC have a positive effect on the levels of atherogenic risk in both group. Based on this findings, both group of women in our environment are thus at a high risk of developing cardiovasculalar disease although the fertile group have more chances than the other having AIP which is considered a better predictor to atherogenic risk positive to TC in correlation. The TG also shows a significant positive association with VLDL-c, TC/HDL and AIP ($P < 0.05$) in infertile group and the same in fertile group including TC. The same pattern of relationship is also seen in both groups, showing that infertility as a medical condition does not have influence on the atherogenic risk that is difference from apparently healthy fertile women but dyslipidiamia due to LDL-c could be a factor to infertility. There were negative significant correlations in both infertile and fertile group when HDL-c was correlated with TC/HDL-c, LDL-c/HDL-c, and AIP ($P < 0.05$). The inverse pattern of this result is in line with other metabolic syndrome[40] which implies that decrease serum level of HDL-c increases atherogenic predicting factors. LDL-c shows a positive significant association with TC/HDL-c and LDL-c/HDL-c in infertile women while in fertile group it is positively associated with TC/HDL-c, LDL-c/HDL-c and logarithmatically transformed ratio of plasma TG to HDL-c (AIP) ($P < 0.05$). This could serve as an indicator of the atherogenic lipoprotein phenotype[20]. In both infertile and fertile women, the same pattern of association was seen in VLDL-c with TC/HDL-c and AIP; TC/HDL-c with LDL-c/HDL-c and AIP; and LDL-c/HDL-c with AIP ($P < 0.05$).

Several lipoprotein ratios or "atherogenic indices" have been defined in an attempt to optimize the predictive capacity of the lipid profile. The total/high-density lipoprotein (HDL) cholesterol ratio, known as the atherogenic or Castelli index and the LDL/HDL cholesterol ratio are two important components and indicators of vascular risk, the predictive value of which is greater than the isolated parameters. In this respect, an increase in total cholesterol concentration, and specifically LDL cholesterol, is an atherogenic lipid marker, whereas reduced HDL cholesterol concentration is correlated with numerous risk factors, including the components of the metabolic syndrome, and probably involves independent risk[41]. As total cholesterol/HDL ratio is considered a more sensitive and specific index of cardiovascular risk than total cholesterol, the Canadian working group has chosen this lipid ratio as a secondary goal of theraphy[42]. The LDL/HDL cholesterol ratio appears to be as useful as the total/HDL cholesterol ratio. Their similarity can be explained by the fact that approximately two thirds of plasma cholesterol is found in LDL-c and, consequently, total and LDL cholesterol are closely related. Like the total/HDL cholesterol ratio, LDL/HDL cholesterol may have more predictive power if triglyceridemia is taken into account[43]. Although, the increase in these ratios predicted a greater cardiovascular risk in a wide range of cholesterol or triglyceride concentrations, the risk is significantly higher when hypertriglyceridemia is present, as shown by the Helsinki Heart Study[44]. Individuals with a high total/HDL cholesterol or LDL/HDL cholesterol ratio have greater cardiovascular risk owing to the imbalance between the cholesterol carried by atherogenic and protective lipoproteins. This may be due to an increase in the atherogenic component contained in the numerator, a decrease in the anti-atherosclerotic trait of the denominator, or both[45]. Some studies have calculated inverse ratios, i.e., HDL/total cholesterol and HDL/LDL cholesterol. Although their predictive value is identical but in the opposite order, few authors suggest they more clearly express the proportion of HDL cholesterol (protective) compared with the rest[43]. It is concluded that infertile women have almost the same pattern of lipid distributions and artherogenic risk predicting factor with the fertile women. . A triglyceride based index (AIP) can significantly add value when assessing the risk of developing atherosclerosis in Nigeria than other artherogenic indices.

REFERENCES

[1]. Inhorn, M.C. and Frank, V. (2002) Infertility around the globe: New thinking on childlessness, gender, and reproductive technologies. Berkeley: University of California Press. Pp. 159-175.

[2]. Cooper, T.G., Noonan, E. and von Eckardstein, S. (2010) World Health Organization Reference values for human semen characteristics. *Human Reproduction Update*, *16* (3): 231–245.

[3]. Practice Committee of the American Society for Reproductive Medicine (2008) Definitions of infertility and recurrent pregnancy loss. *Fertility and Sterility*, *90* (5): S60.

[4]. Gurunath, S., Pandian, Z., Anderson, R.A and Bhattacharya, S. (2011) Defining infertility: A systematic review of prevalence studies. *Human Reproduction Update, 17* (5): 575–588.

[5]. National Cholesterol Education Program (NCEP) Expert Panel (2002) Third report of the National Cholesterol Education Programme (NCEP) Expert Panel on detection, evaluation, and treatment of high blood cholesterol in adults (Adult Treatment Panel III) final report. *Circulation, 106* (25): 3143–3421.

[6]. Brites, F. D., Bonavita, C.D., Cloes, M, Yael, M. J., Fruchart, J.C., Castro, G.R and Wikinski, R.W. (1998) VLDL Compositional changes and plasma levels of triglycerides and high density lipoproteins. *Clinica Chimica Acta, 269*(2):107–124.

[7]. Sidhu, D and Naugler, C (2012). Fasting time and lipid levels in a community-based population. *Archives of Internal Medicine;* **1**: 1–4.

[8]. Friedewald, W.T., Levy, R.I and Fredrickson, D.S. (1990) Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. *Clinical Chemistry, 18*: 499 -502

[9]. Ben-Shlomo, I., Grinbaum, E and Levinger, U (2008). Obesity-associated infertility-the earliest known description. *Reproductive. Biomedicine. Online*, *17*(1):5-6.

[10]. Chughtai, B., Lee, R.K., Te, A.E and Kaplan, S.A. (2011) Metabolic syndrome and sexual dysfunction. *Current Opinion in Urology, 21* (6): 514-518

[11]. Goldberg, D.M. (1981) The Plasma Lipoprotein system. *Clinical Biochemistry Reviews*, *2:* 341–353.

[12]. Burtis, C.A and Ashwood, E.R. (1996) Lipids, Apolipoproteins and Lipoproteins. In: Tietz Fundamentals of Clinical Chemistry. 4th ed. Philadelphia, Pa: WB Sunders, pp. 375–386.

[13]. Cummings, K.C. (2003) Lipid and Cardiac Risk profiles. *Clinical Chemistry*, *47*:407–409.

[14]. Castelli, W.P. (1988) Cholesterol and lipids in the risk of coronary artery disease-the Framingham Heart study. *Canadian Journal of Cardiology, 4*:5–10.

[15]. Igweh, J.C., Nwagha, I.U and Okaro, J.M. (2005) The Effects of Menopause on the Serum lipid profile of Normal Females of South East Nigeria. *Nigerian Journal of Physiological Sciences*, *20*(1–2):48–53.

[16]. Nwagha, U.I and Igweh, J. C. (2005) Atherogenic Index of Plasma: A significant indicator for the onset of Atherosclerosis during menopause in hypertensive females of South East Nigeria. *Journal of College of Medicine*, *10*(2):67–71.

[17]. Hokanson, J.E and Austin, M.A. (1996) Plasma triglyceride level is a risk factor to cardio vascular disease independent of high density lipoprotein cholesterol level: a meta analysis of population based prospective studies. *Journal of Cardiovascular Risk*, *3* (2):213–219.

[18]. Guerin, M., Legoff ,W., Lassel, T.S., VanTol, A., Steiner, G and Chapman, M.J. (2001) Proatherogenic role of elevated CE transfer from HDL to VLDL and dense LDL in type 2 diabetics. *Arteriosclerosis, Thrombosis and Vascular Biology, 21* (2):282–287.

[19]. Gaziano, J.M., Henne kens, C.H., O'Donnell, C.J., Breslow, J.L and Buring, J.E. (1997) Fasting triglycerides, high density lipoprotein, and risk of myocardial infarction. *Circulation, 96*:2520–2525.

[20]. Dobiasova, M and Frohlich, J. (2001) The plasma parameter log (TG/HDL-C) as an atherogenic index: correlation with lipoprotein particle size and esterification rate in apo B- lipoprotein-depleted plasma (FERHDL). *Clinical Biochemistry, 34* (7):583–588.

[21]. Tan, M.H., Johns, D and Glazer, N.B. (2004) Pioglitazone reduces atherogenic index of plasma in patients with type 2 diabetes. *Clinical Chemistry, 50* (7):1184–1188.

[22]. Larsen, U. (1995) Trends in infertility in Cameroon and Nigeria. *International Family Planning Perspective, 21*(4):138–142.

[23]. Adekunle, L. (2002) Infertility: A sociological analysis of problems of infertility among women in a rural community in Nigeria. *African Journal of Medical Sciences*, 31(3):263–266.

[24]. Hollos, M. (2003) Profiles of Infertility in Southern Nigeria: Women's voices from Amakiri. *African Journal of Reproductive Health***,** *7* (2): 46-56.

[25]. Allain, C.C., Poon, L.S., Chan, C.S.G., Richmond, W and Fu, P.C. (1974) Enzymatic Determination of total serum cholestrol. *Clinical Chemistry, 20* (4):470–475.

[26]. Burstein, M., Scholnick, H.R and Morfin, R. (1980) Rapid method for the isolation of lipoproteins from human serum by precipitation with polyanions. *Scandinavian Journal of Clinical and Laboratory Investigation, 40*: 583-595.

[27]. Buccolo, G and David, H. (1973) Quantitative determination of serum Triglycerides by use of enzymes. *Clinical Chemistry, 19* (5):476–482.

[28]. Levesque, R. (2007) A guide for SPPSS and SAS users, SPSS programming and data management, (4th Edition), 233 south wacker drive, 11th floor, Chicago, United State of America. Pp. 2-540.

[29]. Mahmood, A R. (2009) Association between Lipid Profile, BMI, and Some Pituitary Hormones Abnormalities in Sera of Iraqi Infertile Females. *Ibn Al- Haitham Journal for Pure and Applied Science, 22* (4): 135-145

[30]. Marther, K. J., Kwan, F and Corenblum, B. (2000) Hyperinsulinemia in polycystic ovary syndrome correlates with increased cardiovascular risk independent of obesity. *Fertile Sterile, 73* (1): 150-156.

[31]. Julie, L. S. (2003) Polycystic ovary syndrome and the metabolic syndrome. *Clinical Diabetes, 21* (4): 154-161.

[32]. Fiances, S.G and David, G.G. (2001) Basic and Clinical Endocrinology: Disorder of lipoprotein catabolism in female infertility. (6th ed). New York: McGraw-Hill, 20: 7230 - 7235.

[33]. Centers for Disease Control. (2014) Prospective parents with high cholesterol levels could be in for a long wait to become pregnant. *Journal of Clinical Endocrinology and Metabolism State, 1*:1-2.

[34]. Pirwany, I.R., Fleming, R., Greer, I.A., Packard, C.J and Sattar, N.(2001) Lipids and lipoprotein subfractions in women with PCOS: Relationship to metabolic and endocrine parameters. *Clinical Endocrinology, 54*: 447-453.

[35]. Helena, E. M., Helen, R and Monty, K. (2001) Abnormal lipoprotein metabolism and reversible female infertility in HDL receptor (SR-BI)–deficient mice. *Journal of Clinical investigation. 108*: 1717–1722.

[36]. Lemieux, I., Lamarche, B., Couillard, C., Pascot, A., Cantin, B., Bergeron, J., Dagenais, G.R and Després, J.P. (2001) Total cholesterol/HDL cholesterol ratio vs LDL cholesterol/HDL cholesterol Ratio as indices of ischemic heart disease risk in men: The Quebec Cardiovascular Study. *Archive of Internal Medicine*, *161* (22): 2685–2692.

[37]. Milla´n, J., Pinto,´ X., Mun˜oz, A., Zúñiga, M., Rubiés-Prat, J., Pallardo, L.F., Masana, L., Mangas, A., Hernández-Mijares, A., González-Santos, P., Ascaso, J.F and Pedro-Botet, J. (2009) Lipoprotein ratios: Physiological significance and clinical usefulness in cardiovascular prevention. *Vascular health and Risk Management*, *5*:757–765.

[38]. Grover, S.A., Levington, C and Panquest, S. (1999) Identifying adults at low risk for significant hyperlipidaemia: A validated clinical index. *Journal of clinical epidemiology*, *52*:49-55.

[39]. Gotto, A.M. (1998) Triglycerid, the forgotten risk factor. *Circulation*, *97*: 1027-1028.

[40]. Kalidhas, P., Kanniyappan, D., Gandhi, K and Aruna, R. (2011) Coronary artery disease risk factors in type 2diabetes with metabolic syndrome in the urban south Indian population. *Journal of Clinical Diagnostic Research*, *5*(3): 511-518.

[41].Dobiasova, M and Frohlich, J. (2001) The plasma parameter log (TG/HDL-c) as an atherogenic index. *Clinical Biochemistry*, *38*:583-588.

[42]. Ascaso, J., González, S. P., Hernández, M. A., Mangas, R. A., Masana, L., Millan, J., Pallardo, L.F., Pedro-Botet, J., Perez Jimenez, F., Pintó, X., Plaza, I., Rubiés, J and Zúñiga, M. (2007) Management of dyslipidemia in the metabolic syndrome. Recommendations of the Spanish HDL Forum. *American Journal of Cardiovascuar Drugs*, *7* (1): 39–58.

[43]. Genest, J., Frohlich, J., Fodor, G and McPherson, R. (2003) The Working Group on Hypercholesterolemia and Other Dyslipidemias: Recommendations for the management of dyslipidemia and the prevention of cardiovascular disease: 2003 update. Canadian Medical Association Journal, 169 (9):921–924.

[44]. Pintó, X and Ros, E. (2000) Lípidos séricos y predicción del riesgo cardiovascular: importancia de los cocientes colesterol total/colesterol HDL y colesterol LDL/colesterol HDL. *Clínica e Investigación en Arteriosclerosis*, *12*: 267–284.

[45]. Manninen, V., Tenkanen L, Koskinen P, Huttunen, J.K., Mänttäri, M., Heinonen, O.P and Frick, M.H. (1992) Joint effects of serum triglyceride and LDL cholesterol and HDL cholesterol concentrations on coronary heart disease risk in the Helsinki Heart Study. Implications for treatment. *Circulation, 85* (1):37–45.

[46]. Criqui, M.H and Golom, B.A. (1998) Epidemiologic aspects of lipid abnormalities. *American Journal of Mededicine*, *105* (1):48–57.