Special Conference Edition, June, 2023

http://dx.doi.org/10.4314/bajopas.v14i1.70S

Bayero Journal of Pure and Applied Sciences, 13(1): 421 - 436
ISSN 2006 – 6996

STUDY OF SOME CARDIAC ENZYMES AND LIPID PROFILE N HEALTHY PREGNANT WOMEN IN JODHPUR, NORTH-WESTERN INDIA.

Shamsuddeen Haruna[1*], Fatima A. Chiroma[1], Yahaya Usman[1], Fatima Z. Abacha[2], and Jelani Ismail[3]

[1] Department of Medical Laboratory Science, Faculty of Allied Health Science, Ahmadu Bello University, Zaria Kaduna state Nigeria.

[2] Department of Medical Laboratory Science, Faculty of Allied Health Science, Collage of Medical Science, University of Maiduguri, Borno, State-Nigeria.

[3] School of Medical Laboratory Science, Usmanu Danfodiyo University, Sokoto, Sokoto, State-Nigeria

*Correspondence: shamsman19@gmail.com

ABSTRACT

Pregnancy is known to alter biochemical metabolic process involved in lipid and lipoprotein metabolism. The aim was to evaluate the levels of serum cardiac enzymes and lipid profile in healthy pregnant women, and compare with non-pregnant women. Serum cardiac enzymes and lipid profile were assayed using biochemical methods. Concentrations of total LDH were not significantly increased (P > 0.05) as compared with the non-pregnant control (345.50 ± 71.60 U/L, 334.76 ± 70.50 U/L and 281.52 ± 36.39 U/L). The levels of SGOT did not significantly differ (P > 0.05) in both second and third trimester of pregnant women when compared with that of control group (19.52 ± 2.64 U/L, 20.44 ± 3.13 U/L and 17.94 ± 2.44 U/L respectively). The pregnant women had a higher (p < 0.05) mean concentration of total cholesterol and HDL-C in both second and third trimester as compared with the non-pregnant control (261.06 ± 44.95 mg/dl, 258.94 ± 43.60 mg/dl, and 80.88 ± 10.87 mg/dl, 81.08 ± 11.23 mg/dl versus 145.84 ± 24.21mg/dl and 55.62 ± 14.98 mg/dl respectively). The mean concentration of serum triglycerides was significantly higher (P < 0.05) in third trimester of pregnant women than the control group (192.70 ± 28.01 mg/dl and 119.31 ± 19.89 mg/dl respectively). This study has found that hyperlipidaemia is a facet in pregnant women. Cardiac enzymes and lipid profile panel estimation and monitoring should be made part of routine investigation during antenatal period.

Key words: Pregnancy, hyperlipidaemia, Cardiac enzymes, Jodhpur, North-Western India

INTRODUCTION

Cardiovascular disease (CVD) affects approximately 0.2% to 4% of Pregnant Women (Regitz-Zagrosek *et al.,* 2011). Maternal mortality in pregnant women with CVD is about 1%, which is 100 times higher than women without CVD (Roos-Hesslink *et al.*, 2013). In Western countries, CVD is increasing (Roos-Hesslink *et al.*, 2013; Johns, 2013) and is a major cause of maternal mortality in pregnancy (Regitz-Zagrosek *et al,* 2011). Careful monitoring through pregnancy is required as there are altered physiological demands on the women's body including cardiovascular system, glucose, cholesterol, creatine kinase and coagulation homeostasis (Regitz-Zegrosek *et al,* 2011). India accounts for the maximum number of maternal deaths in the world 17 percent or nearly 50,000 of the 2.89 lac women who died as a result of complications due to pregnancy or childbearing in 2013. Nigeria has high maternal mortality rate with 1,047 deaths par 100,000 live birth in 2020 (World Bank, 2022).

The trend of maternal deaths in India is different in different states - Rajasthan, Uttar Pradesh, Madyha Pradesh, Orissa, Chathisgarh and Jharkhand still have high maternal mortality while Southern states have done reasonably well in reducing their levels (Meh *et al.,* 2022). According to SRS maternal mortality ratio of Rajasthan is 388 per 100,000 live births (Goli *et al.*, 2022).

Priyanka and chetan (2014) reported that a total of 54 maternal deaths occurred in 18 months period, in a ratio of 202 maternal deaths per 100,000 live births in S.N. Medical College Hospital, Jodhpur. This finding is in conformity with that of the Annual Health Survey (2011-12) (Office of the Registrar General & Census Commissioner, India, 2013)

Fact sheet for Jodhpur. Pregnancy is known to alter biochemical metabolic processes involved in lipid and lipoprotein metabolism among others (Bodnar *et al,* 2005). Cardiac disease complicates 1% to 4% of pregnancies in women without preexisting cardiac abnormalities Hague (2003) reported that maternal serum homocysteine (Regitz-Zagrosek *et al.,* 2014) concentrations fall Disturbances of maternal and fetal homocysteine metabolism have been associated with neural tube defects Vasculopathy, such as pre-eclampsia and abruption, and with recurrent pregnancy loss. It was recently postulated by Lavie *et al* (1993). This present study was set to evaluate the levels of maternal serum cardiac biomarkers and lipid profile with a view to assessing the contribution of certain abnormalities to maternal mortality and morbidity.

MATERIALS AND METHODS

Study Area

The location of the study was the Antenatal Clinic (ANC) of the Vasundhara Hospital and Fertility Research Centre, Jodhpur, India. Latitude and longitude coordinates are: 26.263863, 73.008957. Jodhpur is a very large city located northwest of the Luni River in the state of Rajasthan, northwestern India.

Sample Size Determination

The sample size for the study was determined using a standard formula (Oyejide, 1992)

$$N = \frac{(z\text{-}a)^2 (p) (1\text{-}P)}{(d)^2}$$

Where; $n = (d)^2$ minimum sample size, z-a = the value of standard normal deviation which at 95% confidence level has been found to be 1.96, P=the best estimate of the population prevalence obtained from the above literature review d= the difference between the true population rate and sample that can be tolerated, that is the absolute precision required (in percentage point) on either side of the population. In this case is taken to be = 0.05.

(World Health Organisation, 2017) reported a prevalence of 1.5-5.5% maternal mortality in developing world.

Therefore, using 5% as the prevalence (P) the minimum sample size n was calculated as follows

$$n = \frac{(1.96)2\ (0.05)\ (1\text{-}0.05)}{(0.05)2}$$

$$n = \frac{3.8 \times 0.05 \times 0.95}{0.0025} = 72$$

The calculated minimum sample size was 72 Subjects. Therefore, 72 healthy pregnant women were recruited for the study.

Due to lack of client's compliance among the both test and control groups 50 healthy pregnant women in their second and third trimester each, and 30 non pregnant (Controls) were recruited for the study.

Study Population

The study was conducted at Vasundhara Hospital and Fertility Research Centre, Jodhpur, India. A total 130 female subjects were recruited for this study. This consisted of 100 healthy pregnant women (50 in the second trimester and 50 in their Third trimester) and 30 apparently healthy non-pregnant women of reproductive age as control. The target population were adult females who were pregnant (both prinigravidae and multigravidae) in their reproductive age of 19-45 years attending Antenatal clinic of Vasundhara Hospital and Research Center apparently non-pregnant subjects (control group) from the population of clients who visited General outpatient Department (GOPD) of Vasundhara Hospital and Fertility Research Centre, Jodhpur.

Blood Collection and Processing

About 10 ml of blood specimen was collected from a peripheral vein (antecubital venepunture). The serum was separated from the cells and transferred to (sample) vial and then stored frozen until the time for analysis.

Estimation of Serum Creatine Kinase Myocardial-Band CKMB N-acetyl-cystein- (NAC ACT)

Immunoinhibition/Modified IFCC (Horder,1990)

CK-M Fractions of the CK-MM and the CK-MB in the sample are completely inhibited by an anti CK-M antibody present in the reagent. Then the activity of the CK-B fraction is measured by the CK (NAC act) method. The CK-MB activity is obtained by two multiplying the CK-B activity by two.

Estimation of Serum LDH

Modified IFCC method (Schumann, 2002)

Lactate dehydrogenase catalyzes the reduction of pyruvate with NADH to form NAD. The rate of oxidation of NADH to NAD is measured as a decrease in absorbance which is proportional to the LDH activity in the sample

$$\text{Pyruvate} + \text{NADH} + H^+ \xrightarrow{\text{LDH}} \text{Lactate} + \text{NAD}+$$

Estimation of Serum SGOT

Modified IFCC method (Hafkenscheid 1979)

$$\text{L-Aspartate} + \text{2-Oxoglutarate} \xrightarrow{\text{AST}} \text{Oxaloacetate} + \text{L-Glutamate}$$

$$\text{Oxaloacetate} + \text{NADH} \xrightarrow{\text{MDH}} \text{Malate} + \text{NAD}$$

$$\text{Sample Pyruvate} + \text{NADH} \longrightarrow \text{L-Lactate} + \text{NAD}$$

Estimation of Serum Total Cholesterol

Modified Roeschlau's method (1974)The estimation of total cholesterol involves the following enzymes.

1. $\text{Cholesterol ester} \xrightarrow{\text{Cholesteryl esterase}} \text{Cholesterol} + \text{Fatty Acid}$
2. $\text{Cholesterol} + O_2 \xrightarrow{\text{Cholesterol oxidase}} \text{Cholest 4-en-3-one} + H_2O_2$

3. $2H_2O_2$ + 4-aminoantipyrine + Phenol $\xrightarrow{\text{Peroxidase}}$ Quinonemine + $4H_2O$

Absorbance of Quinoneimine so formed is directly proportional to cholesterol concentration in the sample.

Estimation of serum Triglycerides

Colorimetric enzymatic method with glycero-phosphate oxidase. This reagent is based on the method of Wako and the modifications by Mc Gowan et al (1983)

Triglyceride+ H_2O $\xrightarrow{\text{LPL}}$ Glycerol + Free Faty Acids

Glycerol + ATP $\xrightarrow{\text{GK}}$ Glycerol-3-Phosphate + ADP

Glycerol-3-Phosphte + O_2 $\xrightarrow{\text{GPO}}$ DAP + H_2O_2 (Peroxidase

H_2O_2 + 4AAP + TOOS $\longrightarrow$ Quinoneimine dye + $2H_2O$

The intensity of chromogen (Quinoneimine) formed is proportional to the triglyceride cancentration.

Estimation of HDL: Cholesterol

HDL Direct Method without the preliminary separation of the HDL Containing and fraction

The assay is based on a modified polyvinyl sulfonic acid (PVS) and Polyethytene-glycol-methylether (PEGME) Coupled classic precipitation method with the improvements in using optimized quantities of PVS/PEGME and selected detergents. LDL, VLDL and chylomicron (CM) react with PVS and PEGME and the reaction results in inaccessibility of LDL, VLDL and CM by cholesterol oxidase and cholesterol esterase. The enzymes selectively react with HDL to produce H_2O_2 which is detected through a Trinder reaction.

HDL+LDL+VLDL+CM $\xrightarrow{\text{PVS, PEGME}}$ HDL+(LDL+VLDL+CM) + PVS/PEGME

HDL $\xrightarrow{\text{CHOD, CHER}}$ Fatty Acid + H_2O

$2H_2O_2$ + 4-AA + TODB $\xrightarrow{\text{Peroxidase}}$ Quinone + $5H_2O$

Estimation of Serum LDL-Cholesterol

Freidwald formular;

LDL-Cholesterol was calculated using the freidwald equation. Freidwald equation was first developed in 1972.

LDL = Total Cholesterol - (HDL Cholesterol + 1/5 TG) mg/dl

Data Analysis

The data obtained were analyzed using SPSS statistical software version 19 The mean values of CK-MB, LDH, SGOT, Total Cholesterol, Triglycerides, HDL and LDL of each group and their standard deviations were determined. Chi-squared test, student t-test, and analysis of variance (ANOVA) were used where applicable in comparing values between groups. P-value <0.05 was considered to be statistically significant.

RESULTS AND DISCUSSION

A total of one hundred and thirty (130) female subjects were recruited for the study aged 19 to 45 years. These were made up of one hundred (100) healthy pregnant women 50 in their second trimester, and 50 in There Third Trimester and thirty (30) non-pregnant women as controls.

Table1. Means of Cardiac Enzymes and Lipid fractions in two of the trimester of pregnancy compared to the control.

Parameter	Control	Cases	
		2nd Trimester	3rd Trimester
CKMB (NAC) (U/L)	19.12±1.96	21.20±2.04	21.16±6.34
LDH (U/L)	281.52±36.39	345.50±71.60	334.76±70.51
SGOT (U/L)	17.94±2.44	19.52±2.64	20.44±3.13
TotalCholesterol (mg/dl)	145.84±24.21	261.06±44.95*	258.94±43.60*
Triglycerol (mg/dl)	119.31±19.89	146.46±32.55	192.70±28.01*
HDL-C (mg/dl)	55.62±14.98	80.88±10.87*	81.08≠11.23*
LDL-C (mg/dl)	124.04±11.44	129.48±15.91	128.44±12.31

* Statistically significant difference.

It had been observed in the present study that in the 100 healthy women studied, the mean serum CK-MB (NAC), LDH and SGOT were not significant, higher in both second and third trimester of pregnancy when compared with the controls (P>0.05). Acute coronary events and myocardial infarction (MI) are rare in pregnancy, resulting from the low prevalence of coronary artery diseases (CAD) in women of child bearing age. In women aged 15 to 44 years, the incidence of MI unrelated to pregnancy is 5 per 100,000 womens years and the rate of cerebrovascular events is 10.07 per 100,000 women years (Walsh *et al.* 2010). However, coronary dissection and thromboembolic events are unusual causes of MI disproportionately represented in pregnancy. A literature review of 125 cases of MI in pregnancy found the highest incidence in Multigravida women over age 33 years (Marlo *et al.*, 2022). MI tended to occur in the third trimester, often during labor and delivery (Walsh *et al.*, 2010). Another study

identified 859 MIs related to pregnancy, which was a case event rate of 6.2 per 100,000 (Marlo *et al.*, 2022). The case fatality rate was 5.1%, demonstrating that although rare MI related to pregnancy carries a significant risk of mortality. Additionally, the risk was 30 times higher for women older than 40 years of age compared with women younger than 20 years of age (Walsh *et. al.,* 2010). As women delay child birth, the incidence of MI in pregnancy may rise (Kealey *et al.* 2010). The usual criteria for MI apply in pregnancy and the puerperium with the exception of myoglobin and creative kinase myocardial band (CK-MB), which may increase two fold in normal women within 30 minutes of delivery (Khan *et al.,* 2017) observed that knowing the pattern of these enzymes in the serum during labor and puerperium may prevent erroneous cardiac enzymes diagnosis of Myocardial ischemia or infarction changes in the pregnant patient may not be diagnostic for MI. Numerous authors have shown that an elevation of the creative kinase MB isoenzyme (CK-MB) fraction can result from sources other than the heart (Khan *et al.,* 2017). In normal pregnancy, the serum MB fraction may be elevated almost 100% from that of the non-pregnant state (2.1 versus, 1.1 units/l). Although the exact source of the elevated MB fraction remains unknown, most of the CK elevation seen after delivery is from the MM fraction secondary to skeletal muscle damage during labor: Some patients are also positive for the BB fraction, because the uterus contains only CK-BB (Kurapati & Soos, 2023). It is postulated that the elevated CK-MB may come from cord blood or from some small amount of myocardial damage associated with normal pregnancy (Cabaniss, 1990).

Table 2: Comparison of cardiac enzymes and Lipid Profile in the 2nd and 3rd Trimesters.

Parameter	2nd Trimester	3rd Trimester	P-Value
CKMB (NAC) (U/L)	21.20±2.04	21.16±6.34	>0.05
LDH (U/L)	345.50±71.60	334.76±70.51	>0.05
SGOT (U/L)	19.52±2.64	20.44±3.13	>0.05
Total Cholestrol (mg/dl)	261.06±44.95*	258.94±43.60*	>0.05
Triglycerol (mg/dl)	146.46±32.55	192.70±28.01*	>0.05
HDL-C (mg/dl)	80.88±10.87*	81.08≠11.23*	>0.05
LDL-C (mg/dl)	129.48±15.91	128.44±12.31	>0.05

This study also indicated that the serum levels of LDH and SGOT were not significantly increased in the second and third trimesters of pregnancy. He *et al* (1995) reported that in a study of 26 normal pregnant women, no significant changes were seen in LDH level. This also corroborated with work of Dev & Hemalatha, 2019 that found out that the concentration of LDH in third trimester of pregnancy remained normal.

This study indicated that there was increase in all lipid fractions in both second and third trimesters of pregnancy compared to non-pregnant state, except LDL-C, which is not statistically significant. Much of the lipid fractions increase were on total cholesterol (261.06 ± 44.95 and 258.94 ± 43.60 Vs 145.84 ± 24.21), and HDL-C (80.88 ± 10.87 and 81.08 ± 11.23 Vs 55.62 ± 14.98).

The augmentation in the maternal lipid fractions is in reaction to the maternal change from carbohydrate to fat metabolism; which is an alternative conduit for energy production due to high energy demand (Folkert *et al,.* 2016). The maternal hormonal changes in pregnancy (rise in 17-B estradiol, progesterone, insulin and Human Placental Lactogen) exaggerated the lipid fraction levels. Other maternal factors that may also have significant effects on lipid metabolism and plasma levels included, pre-pregnancy lipid levels, BMI (body mass index), maternal weight gain in pregnancy, maternal nutrition and various medical complications of pregnancy (David *et al*, 2010).

A persistent increase in HDL-C was observed as the pregnancy progresses, but with no significant difference between 2nd and 3rd trimesters. This is to postulate that the increase started in the first trimester but essentially plateau in the last part of 2nd and in the 3rd trimester. However, a study by Deepak and Digisha (2011) reported serum HDL-C that showed a biphasic pattern, an initial rise and then decline in later third trimester of pregnancy. This third trimester drop was also found in another study from Israel (David *et al*, 2010). But this decline was not noticeable in this present study. But Poveda et al. (2018) observed that the possible initial increase level of HDL-C is estrogen dependent, while fall in HDL-C in the last half of pregnancy, correlate with increasing levels of human placental lactogen, insulin and insulin resistance (Poveda *et al,* 2018), hence, the variation in the levels of these hormones, among pregnancies, may explain the differences in the various studies (Saliu *et al*, 2022). The higher level of HDL-C in the pregnant subjects could provide defense against the risk of coronary heart disease since pregnancy is associated with hemodynamic changes (Saliu *et al,* 2022). Total cholesterol, triglyceride and HDL-C levels of the test subjects in the third trimester were higher than those of the control subjects. This is conformity with results of another study (Som-Pilley *et al*, 2016), in which it was suggested that the high levels may be implicated in development of fetal organ in the third trimester.

CONCLUSION

Estimation of cardiac enzymes and lipid profile panel should be part of routine investigation during antenatal period of pregnant women in Jodhpur in order to assess the aetiology of cardiovascular abnormalities; these results should always be assessed in conjunction with the patient's medical history, clinical examinations and other findings.

REFERENCES

Bodnar LM, Ness RB, Markovic N, Roberts JM (2005). The risk of preeclampsia rises with increasing pregnancy body mass index. Ann Epidemiol 15:475-482.

Cabaniss, C. D. (1990, January 1). *Creatine Kinase - Clinical Methods - NCBI Bookshelf*. Creatine Kinase - Clinical Methods - NCBI Bookshelf. https://www.ncbi.nlm.nih.gov/books/NBK352/

Chemmitz G, Nevermann : Schmidt E, Schmidt FW Lobers J. ClinBiochem 1979 Dec, 12 (6) : 277-81.

Deepak P. Digsha P stetus of Lipid in pregnancy. National Journal of Medical Research, 2011 : 1, 10-12.

Dev, S. V., & Hemalatha, C. R. (2019, December 15). *Evaluation of maternal and perinatal outcomes in preeclampsia and eclampsia in correlation with LDH - Indian J Obstet Gynecol Res*. Evaluation of Maternal and Perinatal Outcomes in Preeclampsia and Eclampsia in Correlation With LDH - Indian J Obstet Gynecol Res. https://doi.org/10.18231/j.ijogr.2019.108

Folkert Kuipers, Henkjan J. Verkade, A. W., Werner, A., Kuipers, F., & Verkade, H. J. (2013, January 1). *Fat Absorption and Lipid Metabolism in Cholestasis - Madame Curie Bioscience Database - NCBI Bookshelf*. Fat Absorption and Lipid Metabolism in Cholestasis - Madame Curie Bioscience Database - NCBI Bookshelf. https://www.ncbi.nlm.nih.gov/books/NBK6420/

Goli S, Puri P, Salve PS, Pallikadavath S, James KS (2022) Estimates and correlates of district-level maternal mortality ratio in India. PLOS Glob Public Health 2(7): e0000441. https://doi.org/10.1371/journal.pgph.0000441

Hafkenscheid J.C.M., Dijt C.C.M. Determination of serum aminotransferases: activation by pyridoxal-5′-phosphate in relation to substrate concentration. *Clin. Chem.* 1979;**25**:55–59.

Hague W. M. (2003). Homocysteine and pregnancy. *Best practice & research. Clinical obstetrics & gynaecology*, *17*(3), 459–469. https://doi.org/10.1016/s1521-6934(03)00009-9oyej

He S, Bremme K, Kalliner A, Blomback M. GynaecolObseftInvest : 1995, 39(4) : 234-8.

Horder, Mogens & Elser, R & Gerhardt, W & Mathieu, M & Sampson, E. (1990). International Federation of Clinical Chemistry (IFCC): Scientific Division, Committee on Enzymes. IFCC methods for the measurement of catalytic concentration of enzymes. Part 7. IFCC method for creatine kinase (ATP: creatine (N-phosphotransferase, EC 2.7.3.2). IFCC Recommendation. The Journal of automatic chemistry. 12. 22-40. 10.1155/S1463924690000049.

https://data.worldbank.org/indicator/SH.STA.MMRT?contextual=max&locations=NG

Khan, A., Rathore, B., Singh, K., Singh, M., & Misra, A. (2017, March 28). *Association of creatine kinase and its isoenzymes (CK-MB, CK-BB) activity with high risk pregnancy | International Journal of Research in Medical Sciences*. Association of Creatine Kinase and Its Isoenzymes (CK-MB, CK-BB) Activity With High Risk Pregnancy | International Journal of Research in Medical Sciences. https://doi.org/10.18203/2320-6012.ijrms20171247

Lavie, O, Beller, l, Neuman, M. et al. (1993) Maternal serum creatine kinase.a possible predictor tubal pregnancy. AM.J. Obstet. Gynaecol, 169-1149-1150.

Leisesomitz GS, Evans AT, Samuels SJ, Omand K, Kost GJ.J Report Med. 1992 Nov., 37 (11) 910-6.

Lupton M oteng-Nitm E, Ayida G, steer P. Current option in obsetrics and Gynaecology, 2002, 14:137-43.

Lupton M, Oteng-Nitim E, Ayida G, Steer P. Cardiac disease in pregnancy. Current opinion in obstetris and gynaecology.2002, 14:137-43.

Mcfaul, P. Dornan J, Lanki, H et al. A Review of complicated by Maternal heart disease. A review of 519 women.Br.J. obsfetGynaecol.95, 861-67.

McGowan, M.W., Artiss, J.D., Strandbergh, D.R., & Zak, B. (1983). A peroxidase-coupled method for the colorimetric determination of serum triglycerides. *Clinical chemistry, 29 3*, 538-

Merlo, A. C., Rosa, G. M., & Porto, I. (2022). Pregnancy-related acute myocardial infarction: a review of the recent literature. *Clinical research in cardiology : official journal of the German Cardiac Society*, *111*(7), 723–731. https://doi.org/10.1007/s00392-021-01937-5

Meh, C., Sharma, A., Ram, U., Fadel, S., Correa, N., Snelgrove, J. W., Shah, P., Begum, R., Shah, M., Hana, T., Fu, S. H., Raveendran, L., Mishra, B., & Jha, P. (2022). Trends in maternal mortality in India over two decades in nationally representative surveys. *BJOG : an international journal of obstetrics and gynaecology*, *129*(4), 550–561. https://doi.org/10.1111/1471-0528.16888.

Mez et al., C-reactive proteinincreases with gestational age during pregnancy among chinese women. AM J. Hum Biol., 2016.

Office of the Registrar General & Census Commissioner, India. (2013). *ANNUAL HEALTHE SURVEY*. Retrieved August 24, 2023, from https://www.im4change.org/docs/679Rajasthan_Bulletin-2011-12.pdf

Oyejide, O. (1992). *Health Research Methods for Developing country scientists.* (1st ed., Vols. 59–63). codat publication. http://www.sciepub.com/reference/290248

Park K. preventive Medicine in obsetricpaediatics and genatrics, park' Text book of preventive and social medicine 20th Edition, Jabalpur.M/S Banarasi Das Bhanot 2009. P. 479-483.

Priyanka Kalra, Chetan Prakash Kachhwaha (2014) Indian Journal of public Heal Volume 58, issue 3 page, 199-201.

Poveda, N. E., Garcés, M. F., Darghan, A. E., Jaimes, S. A. B., Sánchez, E. P., Díaz-Cruz, L. A., Garzón-Olivares, C. D., Parra-Pineda, M. O., Bautista-Charry, A. A., Müller, E. Á., Alzate, H. F. S., Acosta, L. M. M., Sanchez, E., Ruíz-Parra, A. I., & Caminos, J. E. (2018). Triglycerides/Glucose and Triglyceride/High-Density Lipoprotein Cholesterol Indices in Normal and Preeclamptic Pregnancies: A Longitudinal Study. *International journal of endocrinology*, *2018*, 8956404. https://doi.org/10.1155/2018/8956404

Rao SP, miller S, Rosenbaum R, Lakier JB (August 1999)."Cardiac troponin 1 and Cardiac enzymes after electrophysiological studies, ablations and defibrillator implantations".AM J. Cardoil 84 (4): 470, Aq.

Regitz -Zagrosek V, BlomstromLundgvist C, Borghi C, Cifkova R, Ferreira R, Foidart J-M, et al. ESC Guidelines on the management of cardiovascular disease during pregnancy. The Task Force, on the management of cardiovascular diseases during pregnancy of the European Society of cardiology (ESC).European Heart Journal.2011, 32 (24) 3147-97.

Regitz-Zagrosek, Gohlke-Bärwolf, lung, & Pieper, G. (2014). *Management of cardiovascular diseases during pregnancy*. https://read.qxmd.com/read/24794710/management-of-cardiovascular-diseases-during-pregnancy?redirected=slug. Retrieved August 24, 2023, from https://read.qxmd.com/read/24794710/management-of-cardiovascular-diseases-during-pregnancy?redirected=slug

Roeschlau, P., Bernt, E., & Gruber, W. (1974). Enzymatic determination of total cholesterol in serum. *Zeitschrift fur klinische Chemie und klinische Biochemie*, *12*(5), 226.

Roos-Hesselink JW, Ruys TP, Stein JI, Thiten U, Webb GD, Niwa K et al. Outcome of pregnancy in patients with structural or ischaemic heart disease. Results of a registry of the European Society of Cardiology. European Heart Journal, 2013, 34:657-65.

Saliu, M. A., Salihu, A., Mada, S. B., & Owolabi, O. A. (2021). Dyslipidaemia-related cardiovascular risk among pregnant women attending Aminu Kano Teaching Hospital Kano: A longitudinal study. *Journal of Taibah University Medical Sciences*, *16*(6), 870–877. https://doi.org/10.1016/j.jtumed.2021.07.004

Schumann, G., Bonora, R., Ceriotti, F., Clerc-Renaud, P., Ferrero, C., Férard, G., Franck, P., Gella, F., Hoelzel, W., Jørgensen, P., Kanno, T., Kessner, A., Klauke, R., Kristiansen, N., Lessinger, J., Linsinger, T., Misaki, H. & Panteghini, M. (2002). IFCC Primary Reference Procedures for the Measurement of Catalytic Activity Concentrations of Enzymes at 37°C. Part 3. Reference Procedure for the Measurement of Catalytic Concentration of Lactate Dehydrogenase. , *40*(6), 643-648. https://doi.org/10.1515/CCLM.2002.111

Soma-Pillay, P., Nelson-Piercy, C., Tolppanen, H., & Mebazaa, A. (2016). Physiological changes in pregnancy. *Cardiovascular journal of Africa*, *27*(2), 89–94. https://doi.org/10.5830/CVJA-2016-021

Walsh, J., Murphy, C., Murray, A., O'Laoide, R., & McAuliffe, F. M. (2010). Maternal cerebrovascular accidents in pregnancy: incidence and outcomes. *Obstetric medicine*, *3*(4), 152–155. https://doi.org/10.1258/om.2010.10004342.

world bank. (2022). *Nigeria Maternal Mortality Rate 2000-2023*. Retrieved August 24, 2023, from https://www.macrotrends.net/countries/NGA/nigeria/maternal-mortality-rate#:~:text=Nigeria%20maternal%20mortality%20rate%20for,a%200.71%25%20increase%20from%202017.

world health organisation. (2017). *maternal and reproductive health*. Retrieved August 25, 2023, from https://www.who.int/data/gho/data/themes/maternal-and-reproductive-health