

JoSVAS 2023 September Vol 5 Issue 1: 26-33 ©2023 College of Veterinary Medicine, Michael Okpara University of Agriculture, Umudike, Nigeria

Original Research

Effect of pregnancy and lactation on the serum lipid profiles: cholesterol and triacylglycerol ratios of apparently healthy large white sows

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ABSTRACT

This study evaluated the effects of pregnancy and lactation on the serum lipid profiles of large white (LW) sows. Twenty (20) sexually matured LW sows and three (3) LW boars, of mean weight 45.38 ± 1.29 kg, aged between 8 and 10 months were used for this study. The sows were randomly assigned to five groups (A to E) of four sows per group in a completely randomized design (CRD). Group A: Non pregnant (control), Group B: Early gestation (38 days), Group C: Mid gestation (78 days), Group D: Late gestation (112 days) and Group E: Post gestation (14 days postpartum). Blood samples were collected during five stages of gestation. The mean total cholesterol (TC) and high density lipoprotein cholesterol (HDL-C) concentrations of group A (non-mated or control group) were significantly (p < 0.05) higher than groups B and C. The mean total cholesterol (TC/HDL-C) and low density lipoprotein cholesterol:high density lipoprotein cholesterol (TC/HDL-C) and low density lipoprotein cholesterol (LDL-C/HDL-C) ratios of the postpartum sows were significantly (p < 0.05) lower than group C. There were however no significant (p > 0.05) variations in the mean triacylglycerol (TAG), low density lipoprotein cholesterol (LDL-C), triacylglycerol:high density lipoprotein cholesterol (TAG/HDL-C) ratio and very low density lipoprotein cholesterol (VLDL-C) concentrations among all the groups. This present study has presented preliminary information on the changes of serum lipid profiles, cholesterol and triacylglycerol ratios during gestation in LW sows that can be sought by the scientific community.

Keywords: Gestation, large white pigs, lipid profiles, serum

INTRODUCTION

Blood serum biochemical profile testing is a pre-symptomatic diagnostic tool to assess the swine herd's nutritional status and other productive and reproductive disorders (Pathan *et al.*, 2011). The profile may vary according to factors such as origin, climate, management practices, geographical distribution, season and reproductive stage of animals (Cozzi *et al.*, 2011; Mahima *et al.*, 2013; Pal & Acharya, 2013). So, it is important to determine the blood biochemical profile for the clinical interpretation of laboratory findings especially in the post parturient stages at which the sows are more likely to suffer from metabolic disorders.

Physiological changes in pregnancy are primarily due to the changes in the hormonal dynamics (Chandra *et al.*, 2012). Pacheco *et al.* (2016) and Souza *et al.* (2018) emphasized the physiological changes related to nutrient expenditure with foetal growth, placental functioning, increased foetal covering and fluid, uterine wall and mammary glands,

highlighting the increase in the metabolic rate that occurs during gestation due to the higher requirement of energy and proteins.

Vyas *et al.* (2015) and Sanchis-Gomar *et al.* (2016) opined that the quantity of cholesterol in the serum could cause hypercholesterolaemia, a predisposing factor for cardiovascular diseases like atherosclerosis and myocardial infarctions.

Cholesterol is an important sterol in mammals. It participates in many physiological processes including the formation of cell membranes and regulates signal transduction in the cells. Cholesterol is required for normal cell functions and it is essential for the structural integrity of the cell membranes. Increasing concentration of cholesterol decreases the membrane fluidity and thereby controls or regulates the permeability of the cell membranes. Cholesterol is also required for differentiation of cells, their proliferation and interaction, development of the embryo and foetus. It is essential for the synthesis of progesterone and estradiol hormones that actively participate to sustain pregnancy and parturition respectively. It is known that the level of total cholesterol (TC) may have important long-term regulatory effect on steroidogenesis in the ovaries (Rabiee *et al.*, 1999).

Physiological and metabolic adaptations in animals occur during reproductive cycle. Internal redistribution of substrates under hormonal control is the main characteristic of metabolic adaptations to pregnancy and lactation. The concentration of cholesterol in the blood can be regulated by many processes that include - its absorption from food, endogenous synthesis in the liver, excretion of neutral steroids and bile acids from the body and/or its absorption and release from tissues. The dynamics of cholesterol in mammals of different species depends on the stage of the reproductive cycle (Kessler & Rawlins, 1983; Knopp et al., 1983; Quig & Zilversmit, 1983; Smith et al., 1998; Wright-Rodgers et al., 2005). It is considered that the achievement of the necessary levels of total cholesterol (TC) is essential to maintain pregnancy and optimal foetal development (Chiang et al., 1995; Qureshi et al., 1999; Ordovas et al., 2005; Saarelainen et al., 2006). The results of studies of various mammalian species, including humans, during pregnancy showed an increase in TC in the blood serum of some mammals in late pregnancy compared to the early stages (Piechota & Staszewski, 1992; Chiang et al., 1995; Dejager & Turpin, 1996; Qureshi et al., 1999; Wright-Rodgers et al., 2005; Ordovas et al., 2005; Saarelainen et al., 2006).

The concentrations of TC in rabbits, guinea pigs, cows, nonhuman primates decreases gradually toward the end of pregnancy (Kessler & Rawlins, 1983; Koritnik et al., 1984; Viard-Drouet et al., 1984; Spicer et al., 1993; Burdge & Postle, 1994; Wells et al., 1999; Francisco et al., 2003) and rises during postpartum (lactation) period (Francisco et al., 2003). All of these differences may be related to the level of metabolism in the body of different animals and with necessary changes of the metabolic demands of the foetus. Changes in the level of cholesterol in females in the postpartum period may be due to its direct participation in the reproductive processes and intensive use in the synthesis of milk (Francisco et al., 2003). Mechanisms by which some factors increase total cholesterol (TC) in the blood of different animals during pregnancy or lactation period are not well understood.

Some studies have been developed to characterize the dynamics of biomarkers in Santa Inês (Araujo *et al.*, 2014), Dorper sheep (Soares *et al.*, 2014), Morada Nova sheep (Santos *et al.*, 2014) and Saanen and Alpine Brown dairy goats (Oliveira *et al.*, 2019) during the transition period. These studies demonstrated that changes in metabolic response of several biochemical parameters which are influenced by the varying gestational period also served as

tools for the diagnosis of metabolic disorders seen in the transition period. Currently, there are no data available on the serum lipid changes that occur during the transition period in LW sows; hence the aim of this study was to determine the effect of pregnancy and lactation on the serum lipid profiles, cholesterol and triacylglycerol ratios in LW sows kept in this eco-zone.

MATERIALS AND METHODS

EXPERIMENTAL ANIMALS

This experiment was carried out on LW sows and LW boars obtained and kept in Captain commercial breeding farm located in Amorji Nike, Enugu East LGA, Enugu State, Nigeria. Twenty (20) sexually matured cycling apparently healthy LW sows and three (3) adult LW boars (23 pigs), of mean weight 45.38±1.29 kg, aged between 8 and 10 months were used for this study. The sows and boars were acclimatized for three weeks during which they were dewormed using ivermectin at a dose of 300 µg/kg subcutaneous (SC) ones, for 'ecto' and 'endo' parasites and repeated 10-14 days for second batch of maturing parasites. The LW pigs were kept in well ventilated pens at room temperature (25-27^oC) and 12 hours light/darkness cycle maintained. Ethical approval for this study was obtained from the Research Ethical Committee of Michael Okpara University of Agriculture Teaching Hospital with ethical approval number; MOUAU/CVM/REC/202114. The animals were managed as prescribed in the Guide for the Care and Use of Large Animals of National Research Council.

EXPERIMENTAL DESIGN

Twenty (20) LW sows were randomly assigned to five groups (A to E) of four sows per group in a completely randomized design (CRD). Group A: Non pregnant (control), Group B: Early gestation (38 days), Group C: Mid gestation (78 days), Group D: Late gestation (112 days) and Group E: Post gestation (14 days post-partum). The sows were synchronized using $PGF_{2\alpha}$ given twice, 11 days apart (Akusu & Egbunike, 1984) at a dose of 10 mg/kg intramuscular (IM). One boar each was introduced to naturally serve the sows in groups B, C and D (pregnant groups). Following successful mating, pregnancy was confirmed by ultrasonography (B-Ultrasound scanner, Korea) (Ali & Fahmy, 2008) between days 22-23. The pregnant sows were identified with tag letters thus, B₁₋₄, C₁₋₄, D₁₋₄, and kept in separate pens until farrowing and are maintained in these separate pens throughout lactation. The lactating sows were also kept in separate pens and identified thus; E₁₋₄ while the control sows were identified thus A₁₋₄ and kept together in a pen. Sows were fed twice daily while the piglets received udder milk from the lactating sows until the end of lactation. Sows and piglets were provided with clean fresh water ad libitum throughout the period of the study. Four milliliters (4ml) of blood was collected from each sow in a group from the femoral vein and dispensed into plane test tubes and allowed to clot in a slanting position and centrifuged at 2,500 RPM for 5minutes. The resulting sera were aspirated, stored at -20^oC and used to determine serum total cholesterol concentration (Artiss & Zak, 1997), triacylglycerol (TAG) concentration (Rifai et al., 1999) and high density lipoprotein (HDL) cholesterol (Rifai & Warnick, 1994). The serum total cholesterol:high density lipoprotein cholesterol (TC/HDL-C), triacylglycerol:high density lipoprotein cholesterol (TAG/HDL-C) and low density lipoprotein cholesterol:high density lipoprotein cholesterol (LDL-C/HDL-C) ratios were obtained by calculation.

Serum low density lipoprotein cholesterol (LDL-C) was calculated using Friedewald's equation (Friedewald *et al.*, 1972).

 $LDL-C = [TC - {HDL-C+ (TAG/5)}]$

Serum very low density lipoprotein cholesterol (VLDL-C) was calculated by dividing TAG by five (Wilson *et al.*, 1981; Kenneth, 2001).

VLDL = 0.2 x TAG (where TAG is triacylglycerol).

DATA ANALYSIS

The data collected for each of the lipid parameters were subjected to One Way Analysis of Variance (ANOVA) using

statistical package for social sciences (SPSS) version 20.0. Variations in means were separated using Duncan's New Multiple Range Test (Steel and Torrie, 1980). Probability values < 0.05 were considered significant.

RESULTS

The results of serum lipid parameters of LW sows in this study are shown in figures 1-5 while the results of serum total cholesterol:high density lipoprotein cholesterol, triacylglycerol:high density lipoprotein cholesterol and low density lipoprotein cholesterol:high density lipoprotein cholesterol and low density lipoprotein cholesterol:high density lipoprotein cholesterol.

The mean total cholesterol (TC) (Figure 1) and high density lipoprotein cholesterol (HDL-C) concentrations (Figure 3) of group A (non-mated or control group) were significantly (p < p0.05) higher than groups B and C. The mean total cholesterol:high density lipoprotein cholesterol (TC/HDL-C) and low density lipoprotein cholesterol:high density lipoprotein cholesterol (LDL-C/HDL-C) ratios of the postpartum sows were significantly (p < 0.05) lower than group C (table I). There were however no significant (p > 1)0.05) variations in the mean triacylglycerol (TAG) (Figure II), low density lipoprotein cholesterol (LDL-C) (Figure III), triacylglycerol:high density lipoprotein cholesterol (TAG/HDL-C) ratio (Table 1) and very low density lipoprotein cholesterol (VLDL-C) concentrations (figure V) among all the groups.

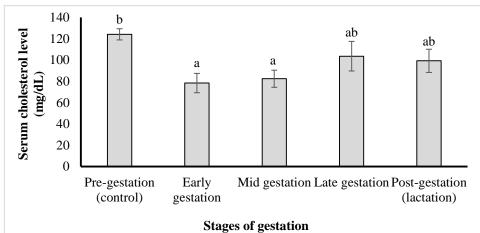
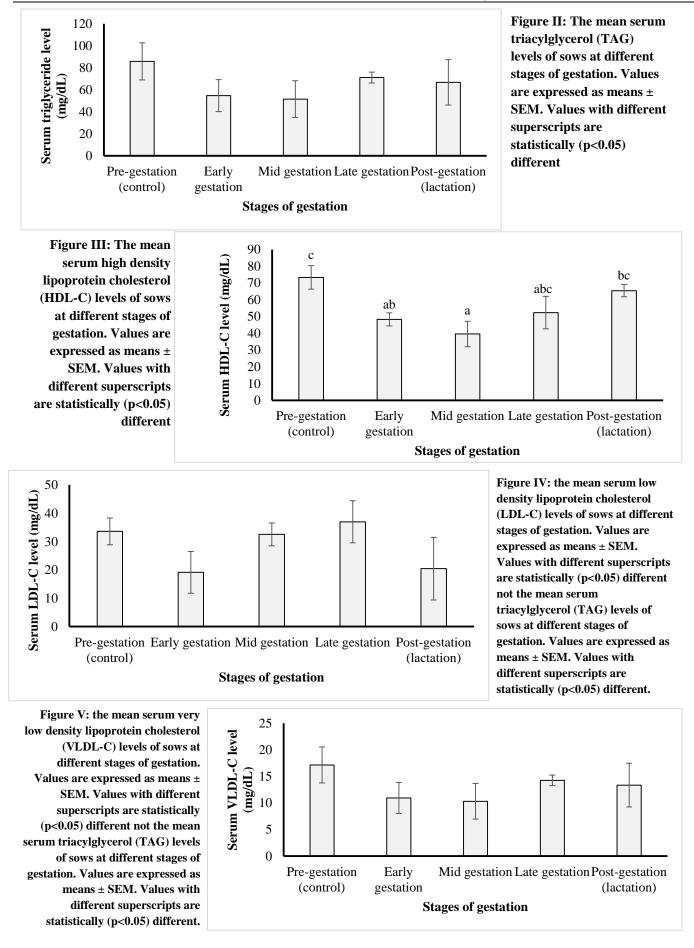


Figure 1: The mean serum total cholesterol levels of sows at different stages of gestation. Values are expressed as means \pm SEM. Values with different superscripts are statistically (p<0.05) different

Table 1: The mean total cholesterol: high density lipoprotein cholesterol, triacylglycerol: high density lipoprotein
cholesterol and low density lipoprotein: high density lipoprotein cholesterol ratios of LW sows at different stages of
gestation. Values are expressed as means \pm SEM in the table

Parameters	Pre-gestation (Non-pregnant control)	Early gestation	Mid gestation	Late gestation	Post-gestation (lactation)
TC/HDL-C	$1.74{\pm}0.17^{ab}$	1.61 ± 0.06^{a}	2.23 ± 0.27^{b}	2.08 ± 0.23^{ab}	$1.52{\pm}0.17^{a}$
TAG/HDL-C	1.28 ± 0.40	1.20±0.39	1.44 ± 0.51	1.59 ± 0.44	0.98 ± 0.24
LDL-C/HDL-C	0.48 ± 0.11^{ab}	$0.37{\pm}0.14^{a}$	0.95 ± 0.22^{b}	$0.76{\pm}0.15^{ab}$	$0.32{\pm}0.18^{a}$

^{ab}Mean values in the same row with different superscripts are significantly different (P<0.05)



DISCUSSION

Variations in blood cholesterol content have been observed during pregnancy, as precursor of the steroid hormones (Iriadam, 2007). Lipid profiles have been used to predict peripartum diseases; circulating blood triacylglycerols contribute significantly to milk fat synthesis (Nazifi *et al.*, 2002). Cholesterol is needed for normal functioning of the body (Shukla *et al.*, 2002), and it plays an essential role in cell membrane formation, hormone production, and the production of fat-soluble vitamins (Okonkwo *et al.*, 2010).

The total cholesterol concentrations of the treated groups (early and mid-gestations) was significantly (p < 0.05) lower when compared to the control group and this result disagrees with Schlumbohm et al. (1997), Liberati et al. (2004), Wright-Rodgers et al. (2005), Piccione et al. (2009), Sandabe et al. (2011), Mohammadi et al. (2016) and AL-Hassan (2018) who reported increased cholesterol concentrations during pregnancy in sahel goats, rat, dog, makouei sheep and Aardi goats respectively. Insignificantly increased cholesterol levels in the blood of different animal species during gestation period especially at late stage had been reported by Waziri et al. (2010) and Antunovic et al. (2011) and these reports equally disagree with our findings. These changes were the result of normal endocrinological and physiological changes in pregnant animals (Watson et al., 1993). Differences in the serum cholesterol concentrations from previous studies can be attributed to differences in breed and nutritional status (Khan et al., 2013). Other researchers agree with our findings (Ozpinar & Firat, 2003; Khatun et al., 2011; Santos et al., 2014 and Soares et al., 2014).

Non-significant (p > 0.05) reductions in triacylglycerol and serum cholesterol levels were observed in lactating sows. A reduction in serum cholesterol was observed in lactating sows compared to non-pregnant control in this current study and this agrees with the work of Amer et al. (1999) who reported lower postpartum cholesterol levels in does. Similar findings were observed in dairy cow due to increased energy demand (Marcos et al., 1990). Reductions in serum lipid profiles at lactation observed in this study could be attributable to the stimulation of lipogenesis by insulin which ultimately manifests as low serum cholesterol and triacylglycerol levels as described by Piccione et al. (2009). During lactation phase, the mammary gland uptake 80% of body metabolites to form milk, the increase in adrenalin cause a decrease in lipid (Quanes et al., 2012). The negative balance of energy during lactation causes a decrease in triacylglycerol and cholesterol (Antunovic et al., 2011). Cholesterol presented the lowest values at early gestation and the highest value in the control. Cholesterol is influenced by the state of pregnancy, the time and number of lactations according to studies by Pysera & Opalka (2000) and Nath et al. (2005). Rowlands et al. (1980) analyzed cholesterol levels which

were lower in the period immediately after calving and increase with lactation, reaching the highest values at the end of lactation, this trend was equally seen in this study. Ruginosu et al. (2011) showed that elevated cholesterol values indicate liver and metabolic malfunctions. Physiological status was examined on serum bio-chemistry in cows (Marcos et al., 1990), cats (Watson et al., 1995) and mares (Watson et al., 1993) where a decreased triglyceride, cholesterol and VLDL levels in pregnant ewe were documented (Yokus et al., 2006; Nazifi et al., 2003) and similar trend was seen in this study. Physiological status is also associated with a strong reduction in lipogenesis during the pregnancy and lactation periods (Yokus et al., 2006; Nazifi et al., 2003). All through pregnancy and lactation periods the number of total insulin receptors (TIR) decreases and insulin stimulation of lipogenesis becomes in-efficient (Burtis & Ashwood, 1999; Guesnet et al., 1999). A decreased triglyceride levels in pregnant sows could be related to the increase in insulin resistance.

The elevated TC/HDL-C ratio at mid gestation in this study probably shows more substantial alterations in metabolic indices predictive of ischemic heart disease risk (Kannel & Wilson, 1996; Boden, 2000; Lemieux et al., 2001; Teixeira et al., 2001). Although the HDL-C level in this study was higher at post gestation than at mid gestation, the TC/ HDL-C ratio is lower (p < 0.05) at post gestation than at mid gestation. Thus the risk of coronary heart diseases may be higher at mid gestation than at postpartum. Moreover, the prevalence of atherosclerosis is higher in mid pregnant sows than in other groups (Liu et al., 1986). Triacylglycerol values of 55.00±8.10 mg/dl are considered normal, 200 to 400 mg/dl mild elevation, 400 to 1000 mg/dl moderate elevation and greater than 1000mg/dl severe elevation (Johnson, 1989). In this study, the serum concentrations of triacylglycerol are normal in all the groups (figure 2). The triacylglycerol concentrations reported in this study (65.97±14.74 mg/dl) was lower than the values of 76.33±27.12 mg/dl reported in dog (Sezgin et al., 2002). The TC/HDL-C ratio reported in this study (1.84±0.18) was higher than the values of 1.48±0.12 reported for dog (Sezgin et al., 2002).

CONCLUSION

The serum lipid panels, total cholesterol:high density lipoprotein cholesterol and low density lipoprotein:high density lipoprotein cholesterol ratios of LW sows in this study were significantly influenced by gestation. The lipid profiles of sows used in this study are within the physiological range required for the maintenance of homeostasis and health essential for efficient reproduction in LW sows. Therefore the sows used in this study during gestation suggest having good lipid compositions capable of supporting optimal reproductive performance. This study has established the changes in the lipid profiles of LW sows during pregnancy and lactation that are consistent with normal health in these species.

ACKNOWLEDGEMENTS

The authors wish to acknowledge the efforts of numerous researchers, who have helped in improving our current understanding of the topic under study in one way or the other. The authors acknowledge the efforts of Mr Orazulume Nelson (Captain Farms) for providing us with his farms and other logistics used for this study. We would also like to express our thanks to Prof. Chike Oguejiofor for providing laboratory space and assistance during analysis of the samples.

CONFLICTS OF INTEREST

This manuscript was read and approved by all the authors. There is no conflict of interest

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