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Full Length Research Paper

Effect of oestrous cycle on serum electrolytes and liver enzymes in Red Sokoto goats

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This study was carried out to determine changes in serum electrolytes and liver enzymes during the oestrous cycle in Red Sokoto goats. Eleven (11) apparently healthy Red Sokoto goats were synchronized with a single injection of 7.5 mg of $PGF_{2\alpha}$. The goats were bled via jugular venipuncture in oestrus or late oestrus, metoestrus/early dioestrus, mid-dioestrus and late dioestrus/proestrus. Mean serum sodium, potassium and chloride levels non-significantly fluctuated during the oestrous cycle. Calcium levels in oestrus or late oestrus were higher ($P < 0.05$) than in late diestrus/pro-oestrus. There was a positive correlation between oestradiol and calcium concentrations ($r = 0.771$; $P < 0.05$). Aspartate aminotransferase (AST) and alanine aminotransferase (ALT) concentrations non-significantly fluctuated during the oestrous cycle. Progesterone and AST concentrations were correlated ($r = 0.925$; $P < 0.05$) during the oestrous cycle. In conclusion, some serum electrolytes and liver enzymes fluctuated and correlated with ovarian steroids in different phases of the oestrous cycle. The information generated may be useful as physiologic reference values for reproductive herd health management.

Key words: Red Sokoto goats, oestrous cycle, electrolytes, liver enzymes.

INTRODUCTION

Blood biochemical values is an important tool for assessment of the health status of animals and this has been shown to vary even in healthy animals due to differences such as: sex, season and oestrous cycle phase (Tamukai et al., 2011; Yaqub et al., 2011). Reproductive status and sex variations in blood electrolytes and liver enzymes have been reported in several studies (Tamukai et al., 2011; Stojevic et al., 2005). These electrolytes are very important in homeo-stasis, nerve impulse transmission, muscle contraction, ovarian steroidogenesis and the process of ovulation (Peracchia 1978; Carnegie and Tsang, 1984).

The involvement of oestradiol in the regulation of fluid and electrolytes balance has long been recognized (Khan, 1993). Oestradiol influences salt retention in systemic circulation and alter ion permeability in various epithelial cells (Zeitlin et al., 1989; Sweezey et al., 1996). The oestrous cycle in domestic goats is characterized by cyclic fluctuations in major ovarian steroids. During the follicular phase of oestrous cycle, oestradiol concentration increases and then declines to nadir during the luteal phase (Gaafar et al., 2005). There is paucity of information on changes in serum biochemical parameters during the oestrous cycle in Red Sokoto goats.

Therefore, this study was designed to evaluate changes in serum electrolytes and liver enzyme concentrations during the oestrous cycle in Red Sokoto goats. The data generated from this study may serve as a veritable input for reproductive herd health programme in goats.

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MATERIALS AND METHODS

Location of the experiment

The experiment was performed at Small Ruminant Research Programme of the National Animal Production Research Institute (NAPRI), Ahmadu Bello University, Shika, Zaria, Nigeria located on latitude 110 12' N, longitude 70 33' E, and altitude 610 m. The experiment was carried out in the rainy, hot-humid months of June - August, 2009.

Experimental animals

Eleven (11) cycling and apparently healthy Red Sokoto does, aged between 1.5 to 3 years and with body weight ranging between 15 and 25 kg were obtained from local livestock market located at a distance of approximately 45 km from the location of experiment. They were housed in semi-opened concrete floored pens and were preconditioned for two weeks. During preconditioning, the animals were prophylactically treated with albendazole and long-acting oxytetracycline at dose rates of 7.5 and 20 mg/kg, respectively. Blood and faecal samples were collected for haemoparasitic and helminthic screening, and only clinically healthy animals were used. The animals were confined and fed with *Digitaria smutsi* hay as basal diet and supplemented with concentrate ration of ground maize (12%), cotton seed cake (24%), wheat offal (62%), bone meal (1.5%) and salt (0.5%) at 300 g/head/ day. The animals were provided with water and salt lick *ad libtum*.

Oestrus synchronisation

Experimental animals were synchronised with $PGF_2\alpha$ (Lutalyse® Pharmacia, South Africa) at a dose rate of 7.5 mg/animal intra-muscularly. Each animal was weighed prior to commencement of treatment for oestrus synchronisation.

Oestrus detection

An apronised teaser buck was used for detecting oestrus twice daily at 08:00 and 16:00 h. Heat detection was carried out daily between the first day of the second oestrus (d 0) and the first day of the third oestrus following treatment with $PGF_{2\alpha}$.

Blood sampling

Blood sample (5 ml) was collected from each animal by jugular venipuncture into anticoagulant free sample bottles and allowed to clot. The serum samples were then centrifuged at 2000 x *g* for 10 minand serum harvested. Sampling was done on day 0 or early oestral phase (EE), day 1-2 or late oestral phase (LE), day 7-10 or metoestrus/early dioestral phase (M/ED), day 11-15 or mid-dioestral phase (MD) and day 16-22 or late dioestral/proestral phase (LD/PE).

Determination of serum oestradiol and progesterone

The serum level of oestradiol-17β and progesterone values were determined using commercial Enzyme-Linked Immunosorbent assay (ELISA) kits (CLINITECH®, Canada). The sensitivity of the assay for oestradiol and progesterone were 10 ρ/ml and 0.05 ng/ml, respectively.

Determination of serum electrolytes

The serum electrolytes, $Na^{+,}$ Cl^{-}, K^{+} and Ca^{2+} were determined using flame photometry method (Dacie and Lewis, 1991).

Determination of serum liver enzymes

Serum liver enzymes were assayed using Bayer Express Clinical Chemistry Autoanalyzer (Bayer®, Germany). The serum enzymes assayed were alanine aminotransferase (ALT) and aspartate aminotransferase (AST). The assay was carried out as described in Bayer Express Plus Clinical Chemistry Autoanalyzer Manual (2000).

Data analysis

Values obtained were expressed as mean (± SEM) and subjected to one-way analysis of variance (ANOVA), followed by Tukey's post-hoc test and Pearson's correlation analysis. The statistical package used was GraphPad Prism version 4.0 for windows (2003) from GraphPad software, San Diego, California, USA (WWW. Graphpad.com). Values of $P < 0.05$ were considered significant.

RESULTS

Changes in serum electrolyte levels during the oestrous cycle

Mean sodium, potassium, calcium and chloride levels during the oestrous cycle were 139.4 ± 0.5 mmol/l; 4.2 ± 0.1; 2.5 ± 0.2 mmol/l and 97.4 ± 0.4 mmol/l, respectively (Table 1). Sodium, potassium and chloride levels were highest at days 11-15 (MD) of the oestrous cycle (140.2 ± 1.1, 4.5 ± 0.1 and 98.2 ± 0.6 mmol/l, respectively). The decreased sodium and potassium levels were observed at days 1-2 (LE) (138.7 ± 1.1 l; 40.0 ± 0.2 mmol/l, respectively), while the serum chloride level was decreased at day 7-10 (LD/PE) (97.0 ± 0.8 mmol/l). No changes were found in serum calcium level at day 0 (EE) and day 1-2 (LE), while significantly decreased calcium levels were observed at days 7-10 (M/ED), days 11-15 and days 16-22 (LD/PE) of the oestrous cycle.

There was a significant positive correlation between sodium and potassium concentrations during the oestrous cycle ($r = 0.736$; $P < 0.05$). Similarly, serum calcium and oestradiol concentrations were significantly and positively correlated during the cycle ($r = 0.771$; $P < 0.05$) (Table 2).

Changes in serum liver enzyme levels during the oestrous cycle

Mean serum AST concentration during the oestrous cycle was 23.6 ± 1.4 IU/l. Serum AST concentration was highest at day 0 (EE) and days 16-22 (LE/PE) and, lowest at days 11-15 (MD) phases of the oestrous cycle (24.4 ± 1.4 IU/l vs 21.8 ± 1.3 IU/L; $P > 0.05$) (Table 3). There was a

Table 1. Mean (± SEM) serum electrolytes during the oestrous cycle of Red Sokoto goats (n = 11).

Day/cycle phase	Sodium (mmol/l)	Chloride (mmol/l)	Potassium (mmol/l)	Calcium (mmol/l)
Day 0 (EE)	139.8 ± 0.7 [a]	98.1 ± 0.6 [a]	4.1 ± 0.2 [a]	2.6 ± 0.1 [b]
	(136 - 144)	(94 - 100)	(3.0 - 4.7)	(2.3 - 2.9)
Day 1 - 2 (LE)	139.1 ± 1.1 [a]	97.5 ± 1.1 [a]	4.0 ± 0.2 [a]	2.6 ± 0.1 [c]
	(133 - 146)	(90 - 100)	(3.6 - 5.3)	(2.3 - 2.7)
Day 7 - 10 (M/ED)	138.7 ± 1.1 [a]	96.2 ± 1.3 [a]	4.0 ± 0.2 [a]	2.5 ± 0.04 [a]
	(132 - 144)	(90 - 102)	(2.8 - 4.6)	(2.3 - 2.7)
Day 11 - 15 (MD)	140.2 ± 1.1 [a]	98.2 ± 0.6 [a]	4.5 ± 0.1 [a]	2.5 ± 0.1 [d]
	(136 - 146)	(96 - 100)	(3.5 - 5.0)	(2.2 - 2.6)
Day 16-22 (LD/P/E)	139.2 ± 1.6 [a]	97.0 ± 0.8 [a]	4.3 ± 0.2 [a]	2.4 ± 0.02 [bc]
	(133 - 151)	(90 - 102)	(3.0 - 5.5)	(2.3 - 3.5)
Mean (± S E M)	139.4 ± 0.5	97.4 ± 0.4	4.2 ± 0.1	2.5 ± 0.02
	(132 - 151)	(90 - 102)	(2.8 - 5.5)	(2.2 - 2.9)

[a,b,c,d] = values with different superscripts within column are statistically significant (b = P < 0.01; c = P <.0.05); n = number of animals sample; values in parenthesis are the range; EE = Early Oestral Phase; LE = Late Oestral Phase; M/ED = Meteostrus/Early Dioestral Phase; MD = Mid-dioestral Phase and LD/PE = Late Dioestrus/Pro-oestrus.

Table 2. Relationships between progesterone, oestradiol and serum electrolytes during the oestrous cycle in Red Sokoto goats (n = 11).

Correlated variable	Pearson's correlation (r)	P - Value
Progesterone and chloride	0.2930	P > 0.05
Progesterone and calcium	0.2174	P > 0.05
Progesterone and potassium	-0.2237	P > 0.05
Progesterone and sodium	0.000	P > 0.05
Oestradiol and chloride	0.2693	P > 0.05
Oestradiol and calcium	0.7707	P < 0.05
Oestradiol and potassium	0.2756	P > 0.05
Oestradiol and sodium	0.2149	P > 0.05
Chloride and sodium	0.9178	P < 0.05
Sodium and potassium	0.7355	P < 0.05

positive correlation between serum AST and progesterone levels during the cycle (r = 0.924; P < 0.05) (Table 4).

Mean serum ALT concentration was 24.7 ± 0.6 mmol/l. Serum ALT was concentration was highest at d 1-2 (LE) and lowest at d 7-10 (M/ED) phases (27.3 ± 2.0 vs 22.6 ± 0.8; P > 0.05) .

DISCUSSION

Serum calcium level obtained in the present study lies within the range reported in Saanen goats (Temizel, et al., 2009), West African Dwarf goats (Daramola et al., 2005) and in the equine (Ali et al*.,* 2004). Peak serum calcium concentration observed at day 0 (EE) and day 1-2 (LE) may be due in part to high serum concentration of oestradiol recorded during these phases. This finding agrees with the observations of Ali et al. (2004), who reported high blood calcium concentration in mares during the oestral phase of the oestrous cycle. High oestradiol levels during oestral phase causes increased intestinal absorption of calcium (Brommage et al., 1993). In addition, the ability of oestradiol to retain salt and to alter ion transport in various other epithelial cells could be partly responsible for high serum calcium observed during the oestral phase. Furthermore, increased muscular activities during the oestral phase as a result of psychic manifestation of heat may be responsible for the increase in serum calcium level in extracellular fluid during the follicular phase of the oestrous cycle. This increase in serum calcium level in oestral phase may be necessary to support the increased neuromuscular activity, and ovarian hormone synthesis and release associated with this phase of the

Table 3. Mean (± S.E.M) serum aspartate aminotransferase and alamine aminotransferase during the oestrous cycle in Red Sokoto goats (n = 11).

Day/cycle phase	Aspartate aminotransferase (IU/l)	Alanine aminotransferase (IU/l)
Day 0 (EE)	24.4 ± 1.4 (18.0 - 31.0)	24.5 ± 1.4 (17.0 - 31.0)
Day 1 - 2 (LE)	24.3 ± 1.0 (18.0 - 28.0)	27.3 ± 2.0 (18.0 - 39.0)
Day 7 - 10 (M/ED)	22.8 ± 1.1 (18.0 - 29.0)	22.6 ± 0.8 (18.0 - 26.0)
Day 11 - 15 (MD)	21.8 ± 1.3 (16.0 - 30.0)	26.4 ± 1.1 (22.0 - 33.0)
Day 16 - 22 (LE/PE)	24.4 ± 1.4 (17.0 - 33.0)	22.9 ± 0.8 (18.0 - 26.0)
Overall Mean (± S.E.M)	23.6 ± 0.6 (16.0 - 33.0)	24.7 ± 0.6 (17.0 - 39.0)

n= Number of animals sampled values in parenthesis are the range; EE = Early Oestral Phase; LE = Late Oestral Phase; M/ED = Meteostrus/Early Dioestral Phase; MD = Mid-dioestral Phase and LD/PE = Late Dioestrus/Pro-oestrus.

Table 4. Relationships between serum oestradiol, progesterone, aspartate aminotransferase and alamine aminotransferase during the oestrous cycle in Red Sokoto goats.

Correlated variable	Pearson's correlation Coefficient (r)	P - Value
Progesterone and aspartate aminotransferase	0.9249	P < 0.05
Progesterone and alanine aminotransferase	0.1602	P > 0.05
Oestradiol and aspartate aminotransferase	-0.4427	P > 0.05
Oestradiol and Alanine aminotransferase	0.4446	P > 0.05
Aspartate aminotransferase and Alanine aminotransferase	0.1162	P > 0.05

oestrous cycle. Significant positive correlation between oestradiol and serum calcium concentrations suggests an enhancing effect of oestradiol on serum calcium homeostasis during the oestrous cycle. Similarly, increased serum calcium concentrations had been reported by Ayo et al. (2009) in Red Sokoto goats as a result of high muscular activities following road transportation stress.

The gradual but consistent decline in serum calcium recorded from M/ED phase to the end of the cycle (LD/PE) may be due to decline in peripheral oestradiol concentration and, thus, decreased intestinal calcium absorption during this period. Also, the decline in serum calcium may be due to a decrease in neuromuscular excitability during the dioestral phase of the oestrous cycle. Brommage et al. (1993) reported a decrease in intestinal calcium absorption during the diestral phase of the oestrous cycle in rats.

Serum sodium, potassium and chloride concentrations were within the range reported in goats (Ayo et al., 2009). The non-significant fluctuations in potassium, sodium and chloride concentrations during the oestrous cycle and the non-significant correlation with oestradiol and progesterone concentrations suggest that neither the cycle phase nor fluctuations in ovarian steroid levels had any significant effect on these serum electrolyte levels during the oestrous cycle in goats.

Aminotransferases act as catalyst during amino acid and carbohydrate metabolism. Changes in their activity in the blood can be a consequence of their increased activity in the cells or cell structure damage. The mean serum value of AST recorded in this study is similar to that obtained in the WAD goat (Daramola et al., 2005), but lower than the value reported for Sahel goats (Waziri et al., 2010), West African Dwarf sheep (Oduye and Adadevoh, 1976) and camels (Ayoub et al., 2003). This difference may be attributed to breed or species differences, geographical location and method used in analyzing the serum samples (Tibbo et al., 2004). The highest AST concentration in this study was observed at EE, LE and LD/PE phases of the oestrous cycle. This finding agrees with the result obtained by Marai et al. (2006), who recorded higher AST activity in oestrus than in pro-oestrus phase in

ewes. The highest AST activity recorded during the day 0 (EE) and day 1-2 (LE) phases may be due to the effect of ovarian steroids in increasing permeability of hepatocellular membrane to this enzyme. Furthermore, since AST is also a muscle enzyme, the relatively high AST activity recorded during the oestral phase in this study may have been caused by the heightened neuromuscular excitability, associated with this phase of the oestrous cycle. On the other hand, the positive correlation between serum AST and progesterone concentration recorded in this study indicates a stimulatory effect of progesterone on liver synthesis and secretion of AST; especially during the day 0 (EE), day 1-2 (LE) and days 16 to 22 (LD/PE) phases of the oestrous cycle, when progesterone concentrations were highest. This may also imply that progesterone activity affects serum AST activity during the oestrous cycle in goats.

The serum activity of ALT recorded in this study was slightly higher than the range reported for West African Dwarf goats (Daramola et al., 2005), Arsi-Bale goats, Long-eared Somalia goats, central Highland goats (Tibbo et al., 2008) and sheep (Oduye and Adadevoh, 1976), but lower than the value reported in rabbits (Mizoguchi et al., 2010). Changes in serum ALT activity observed in this study during the oestrous cycle did not follow a consistent pattern. This may imply that oestrous cycle phase and changes in ovarian steroid concentrations may not have a significant influence on serum ALT activities during the oestrous cycle in goats.

Conclusion

Based on the findings of this study, it is concluded that some biochemical parameters fluctuated and correlated with ovarian steroids during the different phases of oestrous cycle. The results obtained may serve as physiologic reference values for reproductive herd health programme.

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