Changes in serum proteins and urea during the oestrous cycle in Red Sokoto goats

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ABSTRACT

The study was conducted to determine the effect of oestrous cycle phase and ovarian steroids activity on serum proteins and urea profile during the oestrous cycle in Red Sokoto goats (RS). Eleven (n=11) clinically healthy RS goats aged between 1.5-3 years and weighing 15-25 kg were used for the study. The goats were synchronized with a single injection of 7.5 mg of PGF\textsubscript{2α}. The goats were bled by jugular venipuncture on d 0 (EE) or oestral, d 1-2 (LE) or late oestral, d 7-10 (M/ED) or metoestrus/early dioestral, d 11-15 (MD) or mid-dioestral and d 16-22 (LD/PE) or late diestrus/proestrus phases of oestrous cycle. Blood samples collected were analyzed for serum total protein, albumin, globulin and urea. Duplicate sera from the same blood samples were analyzed for progesterone and oestradiol-17\textbeta concentrations. Serum total protein was significantly lower at EE than MD phase of the cycle and globulin concentration was lower at EE than MD phase. Serum urea concentration was higher at EE than LE, M/ED, MD and LD/PE phases of the cycle (P < 0.05). Total protein and globulin concentrations were positively correlated during the oestrous cycle (r = 0.868; P < 0.05). While, globulin and progesterone concentrations were inversely correlated during the cycle (r = -0.868; P < 0.05). The result of present study has revealed that fluctuations in serum total protein, globulin and urea may be due to variation in ovarian steroids during the oestrous cycle. Therefore it is recommended that interpretation of plasma proteins and urea should take into consideration the oestrous cycle phase.

Keywords: Red Sokoto goat, oestrous cycle, serum proteins, urea.

INTRODUCTION

Several factors have been reported to affect plasma protein concentrations. These include; dehydration, external haemorrhage, inflammatory disorders, stress and stage of oestrous cycle [1, 2, 3]. Fluctuations of plasma proteins have also been reported during pregnancy [2, 4]. Haematological and biochemical parameters are indicators of the health and nutritional status of
an animal [5], which in turn affects its reproductive capability. Variations in progesterone and oestrogen levels in the blood regulate the oestrous cycle, which in turn affect cardiovascular and serum biochemical indices [6, 7]. Clinical interpretation of haematological and serum biochemical parameters cover a range of normal, high normal and low normal values. These physiological variations are attributed to exercise, season, sex and reproductive status [8, 5, 9]. There is paucity of information on the effect of oestrous cycle phase on serum proteins and urea profile in Red Sokoto goats.

Therefore, this study was carried out to determine serum proteins and urea profiles and the relationship with ovarian steroids concentrations during the oestrous cycle in RS goats.

MATERIALS AND METHODS

Location of the Experiment
The experiment was performed at Small Ruminant Research Programme of National Animal Production Research Institute (NAPRI), Ahmadu Bello University, Shika, Zaria, located on latitude 11° 12’ N, longitude 7° 33’ E, and altitude 610 m. The experiment was carried out in the rainy, hot-humid months of June – July, 2008.

Experimental Animals
Eleven cycling and clinically healthy RS does aged between 1.5 – 3 years and weighing about 15 – 25 kg were obtained from local livestock market in Chiromawa, Giwa Local Government area of Kaduna State, Nigeria. They were precondition at NAPRI for a period of two weeks, during which animals were prophylactically treated with albendazole and long – acting oxytetracycline at a dose rate of 7.5 mg/kg and 20 mg/kg, respectively. Blood and faecal samples were collected for haemoparasitic and helminthic screening and only clinically healthy animals were used. The goats were confined and fed with Digitaria smutsi hay as basal diet and supplemented with concentrate ration of ground maize, cotton seed cake and wheat offal at 300 g/ head/ day. The animals were provided with water and salt lick ad libitum.

Oestrus Synchronization
Experimental animals were synchronized with PGF\textsubscript{2}α (Lutalyze\textsuperscript{®} Pharmacia, South Africa) at a dose rate of 7.5 mg/animal intramuscularly. Each animal was weighed prior to commencement of oestrus synchronization treatment.

Oestrus Detection
An apronised teaser buck was used for detecting oestrus twice daily (at 08:00 and 16:00 hours). Heat detection began from the first day of 2PGF oestrus (day 0) to first day of the third post-treatment (3PGF Oestrus).

Blood Sampling
5 ml of blood sample was collected from each animal via jugular venipuncture into anticoagulant free sample bottles and allowed to clot. The sera were then centrifuged at 2000 x g for 10 minutes and 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 d16-22 or late dioestral/proestrus (LD/PE).
Determination of Serum Protein and urea

Total serum protein, albumin and urea were determined using the Bayer Express Clinical Chemistry Autoanalyzer (Bayer®, Germany), while globulin was calculated from the value of total serum protein and albumin.

Determination of Serum Oestradiol and Progesterone

The serum oestradiol and progesterone values were determined using commercial Enzyme-Linked Immunosorbent assay (ELISA) kits (CLINITECH®, Canada).

Table 1: Mean (±SEM) Serum Total Protein, Albumin, Globulin and Urea during the Oestrous Cycle in Red Sokoto goats (n = 11)

<table>
<thead>
<tr>
<th>Day/Cycle Phase</th>
<th>Total Proteins (g/L)</th>
<th>Albumin (g/L)</th>
<th>Globulin (g/L)</th>
<th>Urea (µmol/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Day 0 (EE)</td>
<td>65.0 ± 2.0 a</td>
<td>34.0 ± 1.0 a</td>
<td>32.0 ± 2.0 a</td>
<td>315.4 ± 11.9 a</td>
</tr>
<tr>
<td></td>
<td>(57.0 – 72.0)</td>
<td>(28.0 – 39.0)</td>
<td>(25.0 – 44.0)</td>
<td>(255.9 – 392.7)</td>
</tr>
<tr>
<td>Day 1 – 2 (LE)</td>
<td>69.0 ± 2.0 ab</td>
<td>37.0 ± 2.0 a</td>
<td>32.0 ± 2.0 a</td>
<td>220.2 ± 11.9 b</td>
</tr>
<tr>
<td></td>
<td>(58.0 – 80.0)</td>
<td>(27.0 – 49.0)</td>
<td>(22.0 – 38.0)</td>
<td>(166.6 – 267.8)</td>
</tr>
<tr>
<td>Day 7 – 10 (M/ED)</td>
<td>69.0 ± 1.0 ab</td>
<td>33.0 ± 1.0 a</td>
<td>36.0 ± 2.0 ab</td>
<td>226.1 ± 11.9 b</td>
</tr>
<tr>
<td></td>
<td>(62.0 – 76.0)</td>
<td>(30.0 – 36.0)</td>
<td>(31.0 – 43.0)</td>
<td>(166.6 – 279.7)</td>
</tr>
<tr>
<td>Day 11 – 15 (MD)</td>
<td>74 ± 1.0 b</td>
<td>34.0 ± 1.0 a</td>
<td>39.0 ± 2.0 b</td>
<td>232.1 ± 17.9 b</td>
</tr>
<tr>
<td></td>
<td>(68.0 – 81.0)</td>
<td>(29.0 – 39.0)</td>
<td>(30.0 – 52.0)</td>
<td>(125.0 – 327.5)</td>
</tr>
<tr>
<td>Day 16 – 22 (LD/PE)</td>
<td>68.0 ± 2.0 ab</td>
<td>35.0 ± 1.0 a</td>
<td>33.0 ± 1.0 a</td>
<td>232.1 ± 11.9 b</td>
</tr>
<tr>
<td></td>
<td>(60.0 – 72.0)</td>
<td>(30.0 – 41.1)</td>
<td>(28.0 – 41.0)</td>
<td>(172.6 – 291.6)</td>
</tr>
<tr>
<td>Mean (± S.E.M)</td>
<td>69.0 ± 1.0</td>
<td>34 ± 1.0</td>
<td>34 ± 1.0</td>
<td>244.0 ± 6.0</td>
</tr>
<tr>
<td></td>
<td>(57.0 – 81.0)</td>
<td>(27.0 – 49.0)</td>
<td>(22.0 – 52.0)</td>
<td>(125.0 – 392.7)</td>
</tr>
</tbody>
</table>

n = Number of animals sampled, ( ) = range

a,b = values with different superscripts within column are statistically significant (P < 0.05).

EE = Early Oestral Phase; LE = Late Oestral Phase; M/ED = Metoestrus/Early Dioestral Phase; MD = Mid-dioestral Phase and LD/PE = Late Dioestrus/Pro-oestrus.

Table 2: Relationships between serum total protein, albumin, globulin, urea, Progesterone and oestradiol concentration during the oestrous cycle

<table>
<thead>
<tr>
<th>Correlated Variables</th>
<th>Pearson’s correlation, r</th>
<th>P – value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Oestradiol and Total Protein</td>
<td>0.1982</td>
<td>P &gt; 0.05</td>
</tr>
<tr>
<td>Oestradiol and Albumin</td>
<td>-0.2251</td>
<td>P &gt; 0.05</td>
</tr>
<tr>
<td>Oestradiol and Globulin</td>
<td>0.2985</td>
<td>P &gt; 0.05</td>
</tr>
<tr>
<td>Oestradiol and Urea</td>
<td>0.2182</td>
<td>P &gt; 0.05</td>
</tr>
<tr>
<td>Progesterone and Total Protein</td>
<td>-0.6569</td>
<td>P &gt; 0.05</td>
</tr>
<tr>
<td>Progesterone and Albumin</td>
<td>0.6797</td>
<td>P &gt; 0.05</td>
</tr>
<tr>
<td>Progesterone and Globulin</td>
<td>-0.8682</td>
<td>P &lt; 0.05</td>
</tr>
<tr>
<td>Progesterone and Urea</td>
<td>0.3739</td>
<td>P &gt; 0.05</td>
</tr>
<tr>
<td>Total Protein and Albumin</td>
<td>-0.06552</td>
<td>P &gt; 0.05</td>
</tr>
<tr>
<td>Total Protein and Globulin</td>
<td>0.8682</td>
<td>P &lt; 0.05</td>
</tr>
<tr>
<td>Albumin and Globulin</td>
<td>-0.5514</td>
<td>P &gt; 0.05</td>
</tr>
</tbody>
</table>
Values obtained were expressed as Mean (± SEM) and were 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

Serum total protein profile during the oestrous cycle.
Mean serum total protein concentration rose gradually from 65.0 ± 2.0 g/L on d 0 (EE) to a peak value of 74.0 ± 1.0 g/L on d111-15 (MD) (Table 1). Thereafter, total protein concentration declined to 68.0 ± 2.0 g/L. Mean serum total protein concentration was significantly higher at MD than EE (P < 0.05) (Table 1). The lowest serum total protein value was recorded on day EE.
and the highest on day MD. Mean serum total protein concentration during the oestrous cycle was 69.0 ± 1.0 g/L.

Serum albumin concentration during the oestrous cycle.
Mean serum albumin concentration remained fairly constant during the cycle. The highest albumin level occurred at d1-2 (LE) (37.0 ± 2.0 g/L), and the lowest level was obtained d 7-10 (33.0 ± 1.0). Mean serum albumin concentration during the oestrous cycle was 34.0 ± 1.0 g/L.

Serum globulin profile during the oestrous cycle.
Mean serum globulin concentration remained fairly constant during the cycle with the highest level occurring at d11-15 (MD) (39.0 ± 2.0 g/L) and the lowest levels at d0 (EE) (32.0 ± 2.0 g/L) and d1-2 (LE) (32.0 ± 2.0 g/L), respectively (Table 1). Mean serum globulin concentration during MD phase was significantly higher than at EE, LE and LD/PE phases of the cycle (39.0 ± 2.0 g/L vs 32.0 ± 2.0 g/L, 36.0 ± 2.0 g/L and 33.0 ± 1.0 g/L, respectively, P < 0.05) (Table 1.0). Mean globulin concentration was positively correlated with total protein (r = 0.868; P < 0.05) and inversely correlated with progesterone (r = - 0.868; P < 0.05) during the oestrous cycle (Table 2).

Serum urea profile during the oestrous cycle.
Mean (±SEM) serum urea concentration dropped from the highest level on day 0 (EE) (315.4 ± 11.9 µmol/L) (Table 1) to the lowest concentration on day 1-2 (LE) (220.2 ± 11.9 µmol/L). Thereafter, serum urea concentration rose slightly through d 7-10 (M/ED) (226.1 ± 11.9 µmmol/L) to a marginally higher level at the end of the cycle at d16-22 (LD/PE), 232.1 ± 11.9 µmmol/L. Mean serum urea concentration was significantly higher at EE than LE, M/Ed, MD and LD/PE phases of the cycle (315.4 ± 11.9 µmmol/L Vs 220.2 ± 11.9 µmmol/L, 226.1 ± 11.9 µmmol/L, 232.1 ± 17.9 µmmol/L and 232.1 ± 11.9 µmmol/L, respectively, P < 0.05).

Figure 3: Relationship between urea and progesterone profiles during the oestrous cycle of Red Sokoto goats (n = 11).
DISCUSSION

The mean serum total protein obtained during the oestrous cycle in this study fell within the range earlier reported for the Savannah Brown goats [4], Nubian goats [5], and West African Dwarf goats [10, 11].

The significantly higher total protein concentration observed at MD than EE phase of this study agrees with earlier reports of higher luteal than follicular phase total protein in cows [3] and human [12]. Lower serum total protein has also been reported during the follicular phase in Baboon [13]). The decrease in serum total protein may be due to increase in blood flow to the uterus along with serum protein during the oestral phase [14]). Thus causing decreased serum total protein but increased protein concentrations in the uterus during the oestral phase.

The mean globulin concentration recorded in this study also agrees with values earlier reported in goats [2]. The significantly lower serum globulin concentration observed in this study at EE and LE phases of the oestrous cycle agrees with an earlier report in the cow [3]. The lowering of serum globulin during the oestral phase is considered to be due to relatively lower serum total

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protein at that phase. Lower fibrinogen concentration had also been reported during the follicular phase of menstrual cycle [15]. The positive and significant correlation between serum total protein and globulin observed in this study indicates that changes in serum total protein levels invariably affects serum globulin concentration during the oestrous cycle. The lower serum globulin observed in this study during the EE and LE may be due to movement of globulin from general circulation to the uterine fluid due to effect of oestrogen. The negative correlation between serum progesterone and globulin concentrations suggests inhibition of globulin metabolism during luteal phase of oestrous cycle in Red Sokoto goats. Globulin is an acute phase protein and the uterus has a more active defense mechanism during the follicular phase than the luteal phase of the oestrous cycle [16]. Therefore, significantly lower serum globulin during EE and LE phases as compared to MD phase may be due to increased mobilization of serum globulin from the systemic circulation to the uterine lumen during the oestral than luteal phase of the oestrous cycle in RS goats. From the clinical perspective, lower EE than MD serum globulin is suggestive of lower systemic immune capability during oestral than dioestral phase of the oestrous cycle.

The mean serum albumin concentration in this study fell within the range earlier reported in goats [1,10, 5]. Serum albumin concentration was relatively constant during the different phases of oestrous cycle. There was a very low correlation between serum total protein and albumin concentrations during the oestrous cycle in this study. This is indication that changes in serum total protein concentration did not affect changes in albumin concentration during the oestrous cycle in RS goats. The mean serum urea concentration recorded in this study was similar to values earlier reported in other animal species [17, 10, 18]. However the values were lower than those reported in pregnant ewes [3] and cows [19]. Mean serum urea concentration was highest at EE, lowest at LE, and it showed a marginal increase during MD and LD/PE phases of this study. This finding is at variance with the observation of lower serum urea concentration obtained during follicular than luteal phase of menstrual cycle in lower primate [13]. The difference may be due to peculiarities of protein metabolism between domestic animals and primates. The high level of serum urea observed during EE phase agrees with observation in lactating dairy cows [19]. This may be due to more efficient digestion of dietary protein at the oestral than luteal phase of the oestrous cycle. High concentration of plasma urea nitrogen causes elevated ammonia and urea concentration in the preovulatory follicles and uterine fluid [19]. This situation compromises fertility by impairing embryo viability in lactating cows [20, 21]. However, serum urea concentration was lowest at LE phase, which is the phase of ovulation and embryo development.

Based on the findings of the study, it is concluded that fluctuations in serum total protein, globulin and urea may be due to variation in ovarian steroids concentrations during the different phases of oestrous cycle. It is, therefore, recommended that variation in oestrous cycle phases should be taken into consideration when interpreting data on serum proteins and urea during the oestrous cycle in RS goats.

REFERENCES