Full Length Research Paper

The micro-minerals composition in serum of rabbits (Oryctolagus cuniculus) infected with Trypanosoma congolense

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Sixteen (16) rabbits aged between 6 to 12 months were infected with fresh stock of Trypanosoma congolense (Gboko strain) intravenously at the rate of 1.0 × 10^6/ml. Animals were classified into two groups; groups A were infected, while group B served as uninfected controls. Samples from the infected and the uninfected controls showed a significant increase in the levels of sodium (Na\(^+\)), calcium (Ca\(^{2+}\)), phosphate (PO\(_4^{3-}\)) and blood urea nitrogen (BUN) (P < 0.05) and a significant decline in the levels of potassium (K\(^+\)) and bicarbonate (HCO\(_3^-\)) (P < 0.05). Therefore, the alterations in the compositions of these micro-minerals in the serum of rabbits may suggest that, they could have a role in the pathogenesis of trypanosomosis due to T. congolense infection.

Key words: Rabbits, micro-minerals, Trypanosoma congolense, pathogenesis.

INTRODUCTION

Trypanosomosis (trypanosomiasis), a disease caused in domestic and wild animals as well as humans by tsetse fly-borne protozoan parasites belonging to the genus Trypanosoma, remains a serious setback to improved and profitable livestock and mixed-crop livestock farming in tropical Africa (Kristjanson et al., 1999; Swallow, 2002; Irunu et al., 2002; Shamaki et al., 2002). In animals, the characteristic clinical features of the disease, which is also referred to as African animal trypanosomosis or Nagana, include intermittent fever, anaemia, anorexia, poor hair coat, emaciation, lethargy, enlarged lymph nodes, abortion, infertility decreased milk yield, submandibular oedema, ascites and ocular discharge and mortality (Shamaki et al., 2002). The most pathogenic trypanosome species responsible for the disease in domestic livestock are Trypanosoma vivax (T. vivax), Trypanosoma congolense and Trypanosoma brucei in cattle, sheep and goats and Trypanosoma simiae in pigs (Nantulya, 1990). Losos and Ikede (1972) estimated that, of all disease due to trypanosomoses in Africa, sheep and goats accounted for 2%; the low incidence rate may be due to their resistance to trypanosomes. However, over time, the prevalence rates have changed to 8.6% for sheep and 8.1% for goats (Onyia,1997) in Nigeria and in Ogbomoso area of Oyo State, the prevalence rate in sheep, goats and cattle was 4.7, 3.5 and 3.9%, respectively (Ameen et al., 2008). Numerous physio-logical factors like diseases (such as trypanosomosis) can produce variation in micro-mineral concentrations in the blood of healthy sheep and cattle (Tartour, 1973; Moodie, 1975). Wellde et al. (1989) showed that, serum iron and serum-iron binding capacity for cattle decreases when infected with T. congolense. Cattle infected with T. congolense showed elevated levels of serum iron (SI), total iron binding capacity (TIBC), plasma iron turn over rates (PITR) and plasma iron clearance (PIC). In treated animals, SI and TIBC falls with the level of SI returned to preinfection level faster than TIBC. Dargie et al. (1979b) reported that, in trypanosomosis due to T. congolense in cattle, there was loss of 40 to 45% of iron in circulation. However, Sarror (1976) found that neither iron nor copper deficiency play any important role in the pathogenesis of anaemia in cattle infected with T. vivax and found no change in the levels of these microminerals in the serum of the cattle.

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Copper level fluctuates but within normal limits (Anosa, 1988a, b; Joshua et al. 1994). Kalu et al. (1989) reported that, the level of calcium in the serum of goat increased significantly during acute stage of trypanosomosis. After chemotherapy, there were constant decrease levels of calcium, which was similar with the increase in total protein level (Kalu et al., 1987). Anosa (1988a) reported a decrease in phosphate levels in cattle infected with *T. congolense*. The depression of calcium observed was thought to be due to thyroid gland damage. It is only imperative to carry out more detailed studies on micro nutrients of rabbits infected with *T. congolense* so that more definite and conclusive statements could be made on the possible effects of the trypanosome infection on the serum levels of some minerals and possibly, on the pathophysiology of some reported disorders in trypanosome infected animals. This study was carried out to determine the serum compositions of sodium, potassium, calcium, bicarbonate, phosphate and blood urea nitrogen of rabbits infected with *T. congolense*.

**MATERIALS AND METHODS**

**Experimental design**

Sixteen (16) rabbits of ages between 6 to 12 months were used for this study. They were housed in a fly proof hutch and fed with variety of fresh grass cut supplemented with spent grains daily. *T. congolense* was obtained from Nigerian Institute of Trypanosomiasis Research (NITR). The parasites were continually maintained in the mice until the donor animal was infected. Rabbits were inoculated with *T. congolense* as follows: group A: there were eight animals in this group and each animal was inoculated with 1.0 × 10^8/ml parasites intravenously. The animals were treated with berenil® at the end of the 5th week post infection after the first wave of parasitaemia. The drug was given intramuscularly; group B: this group of eight served as uninfected controls.

**Blood sampling**

Parasitological examinations were carried out with 2 ml of blood collected via the ear vein in a Bijou bottle with EDTA as the anticoagulant. The collection was done twice a week.

**Serum collection**

5 ml of blood was collected via the external jugular vein from each animal in the different groups. Samples were collected on the same day with that for parasitological examinations. Serum was harvested in a sterilized Bijou bottle and stored at -20°C till further needed.

**Determination of micro-minerals**

Serum sodium, calcium and potassium concentrations were determined by flame photometry using Gallenkamp Flame Analyser (FGA-330). Serum phosphate and bicarbonate were estimated according to Toro and Ackermann (1975). Blood urea nitrogen concentrations were measured by continuous flow analysis using an autoanalyser (Technicon, UK).

**RESULTS**

In this study, changes in serum micro-mineral compositions as a result of experimental *T. congolense* infection in rabbits were observed. The mean period to detection of trypanosomes in the peripheral blood of rabbits following experimental infection was week 1 with initial value of 10^3.9 trypanosomes ml^-1. Thereafter, the rabbits exhibited fluctuating parasitaemia with peaks of 10^3.9 trypanosomes ml^-1 at week 5 (Table 1). The mean serum sodium concentration of the infected animals increased by 4.45% (from pre-infective value of 146.00 ± 0.41 to 152.50 ± 0.29 mmol/l), while that of potassium decreased by 10.44% (from pre-infective value of 3.83 ± 0.01 to 3.4 ± 0.03 mmol/l). The mean serum calcium concentrations increased by 25.96% (from preinfective values of 5.70 ± 0.13 to 7.18 ± 0.03 mmol/l), the mean serum bicarbonate concentration was declined by 21.97% (from pre-infective value of 4.65 ± 0.03 to 3.91 ± 0.29 mmol/l) (Table 2).

**DISCUSSION**

The serum micro-minerals have been studied relatively by various workers in animals infected with *T. congolense*. Sheep infected with *T. congolense* was studied by Joshua et al. (1994) and Yankasa sheep infected with *T. congolense* was studied by Neils et al. (2006). In this study, there were statistically significant increase (P < 0.05) in serum sodium, calcium, blood urea nitrogen and phosphate concentration up to the 5th week post infection.
and it correlated with high peak of parasitaemia. This investigation showed that, rabbits were susceptible to freshly isolated *T. congolense* infection and that anaemia produced from trypanosome infection in rabbits may be related to the level of parasitaemia, however, parasitaemia fluctuated at intervals as described by Joshua (1990). The increase serum sodium concentration may therefore, be due to the failure of Zona glomerulosa of adrenal cortex to produce aldosterone hormone that therefore, may be due to the failure of Zona glomerulosa of (1990). The increase serum sodium concentration may therefore, be due to the failure of Zona glomerulosa of adrenal cortex to produce aldosterone hormone that regulates sodium in the extracellular fluid (Ogunsanmi et al., 1984; Zilva and Pannal, 1984). The concentration of calcium in the infected animals was found to increase and it correlated with the high peak of parasitaemia. This agrees with earlier observations made by Kalu et al. (1987) and Neils et al. (2006). It was obvious that, calcium increased in serum but the source remains unclear; it could be that, calcium ions from the extra cellular fluid that binds cell membranes together with ions in serum were partly mobile elevated concentration (Georgievskii et al., 1982). Phosphate (PO₄³⁻) level was also found to fluctuate in similar pattern to that of calcium, but the increase in the value was significant (P < 0.05) in the infected and the controls. Phosphate is found distributed in almost all organs of the body and plays a role in the production of ATP especially in muscles. During the course of infection, there was reduction in the production of ATP, thus, the probable increase of phosphate in serum. Anosa (1988a) suggested that, calcium in conjunction with phosphorus, depressed thyroid cells, but the actual roles of calcium and phosphorus during trypanosomosis are not yet known. In this study, there was an increase in blood urea nitrogen (BUN) in all the infected groups but there were significant differences with their corresponding controls. It is a by-product of protein catabolism. Increased BUN levels were consistent with the results from monkeys infected with *Trypanosoma rhodesiense* (Sadun et al., 1973) and human infected with *Trypanosoma gambiense* (Awobode, 2006). BUN is a product cleared from the body through the kidneys and as such, their measurements during disease are good indicators of renal function (Ramakrishnan et al., 1995). The causes of elevated BUN levels include kidney disease such as glomerulonephritis and excessive protein catabolism and febrile conditions. Fever and glomerulonephritis are common features of trypanosomosis and presumably act together to elevate BUN. Similar defects in renal function during trypanosomosis have been observed in man (Basson et al., 1977). Indeed, gross and histological changes affecting kidneys have been demonstrated in trypanosome-infected dogs (Murray et al., 1975) and humans (Anosa, 1988a, b), which could explain the observed changes in kidney function in this study. Progressive decrease in serum potassium levels (hypo-kalemia) was observed in infected animals and may suggest depletion of the body’s potassium stores or from a redistribution of potassium from the ECF into the ICF space (Brobst, 1986) and it may be due to excessive renal loss of potassium which results from the action of mineralocorticoid excess and as the result of altered renal tubular function in infected animals with renal tubular acidosis or post obstructive states. The mean serum bicarbonate levels in all the

### Table 2. Mean ± SD of micro-mineral compositions in serum of rabbits infected with *T. congolense*.

<table>
<thead>
<tr>
<th>Micro-mineral</th>
<th>Status</th>
<th>Pre infection/week</th>
<th>Post-Infection/week</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>Na (mmol/l)</td>
<td>Infected</td>
<td>146.00 ± 0.4</td>
<td>148.00 ± 0.4</td>
</tr>
<tr>
<td></td>
<td>Control</td>
<td>146.08 ± 0.4</td>
<td>146.00 ± 0.4</td>
</tr>
<tr>
<td>Ca (mmol/l)</td>
<td>Infected</td>
<td>5.70 ± 0.1</td>
<td>6.15 ± 0.1</td>
</tr>
<tr>
<td></td>
<td>Control</td>
<td>5.75 ± 0.1</td>
<td>5.63 ± 0.1</td>
</tr>
<tr>
<td>K (mmol/l)</td>
<td>Infected</td>
<td>3.83 ± 0.01</td>
<td>3.78 ± 0.01</td>
</tr>
<tr>
<td></td>
<td>Control</td>
<td>3.84 ± 0.01</td>
<td>3.85 ± 0.03</td>
</tr>
<tr>
<td>PO₄ (mmol/l)</td>
<td>Infected</td>
<td>4.65 ± 0.03</td>
<td>4.93 ± 0.08</td>
</tr>
<tr>
<td></td>
<td>Control</td>
<td>4.56 ± 0.03</td>
<td>4.63 ± 0.03</td>
</tr>
<tr>
<td>HCO₃ (mmol/l)</td>
<td>Infected</td>
<td>23.50 ± 0.2</td>
<td>21.50 ± 0.3</td>
</tr>
<tr>
<td></td>
<td>Control</td>
<td>23.88 ± 0.24</td>
<td>24.13 ± 0.3 b</td>
</tr>
<tr>
<td>BUN (mg/dl)</td>
<td>Infected</td>
<td>15.38 ± 0.1</td>
<td>16.75 ± 0.1 a</td>
</tr>
<tr>
<td></td>
<td>Control</td>
<td>15.43 ± 0.1</td>
<td>15.50 ± 0.04 b</td>
</tr>
</tbody>
</table>

a, b, means a significant increase (P < 0.05) between the infected and control groups in serum compositions of Na⁺, Ca²⁺, PO₄³⁻ and BUN of infected rabbits and means a significant decline (P < 0.05) between the infected and control groups in serum compositions of K⁺ and HCO₃⁻ of the infected rabbits.
infected rabbits were observed to have a sharp drop during post infection. This observation agreed with the report of Goodwin and Guy (1973) who reported a decrease in serum bicarbonate in T. brucei infected rabbits and disagrees with Ogunsanmi et al. (1984) who observed elevated bicarbonate and a sharp drop on day 56 in T. brucei infection in sheep. The sharp drop in the serum bicarbonate levels might be due to acidosis associated with anaemia, renal malfunction and the release of toxic metabolites such as free acids by trypanosome (Anosa, 1988a, b).

In conclusion, T. congolense was found to be pathogenic to rabbits. The increase or decrease in micro-minerals concentrations fluctuated with peaks of parasitaemia. The actual roles played by these micro-minerals in the pathogenesis of trypanosomosis due to T. congolense infection are not clear and therefore, may need further investigation.

REFERENCES


