Serum biochemical values of *Trypanosoma vivax*-infected cattle and the effects of lactose in saline infusion

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ABSTRACT

Experiments were carried out on twelve young zebu cattle (*Bos indicus*) aged from 12 to 18 months, divided into 3 groups of 4 animals. Following experimental infection with Samaru strain of *Trypanasoma vivax*, parasitemia and serum, biochemical values - bilirubin, glucose, and blood urea nitrogen (BUN) and aspartate serum transaminase (AST) - were monitored in all groups. The strain of *T. vivax* used caused an acute infection, with parasites appearing in circulation on day 2 with infection and peaking on day 5 post-infection (p.i.). However, parasites disappeared from circulation on day 7 p.i. only to reappear, with a second peak occurring on day 13 p.i. In the group infused with lactose, parasitemia persisted without apparent remission until day 13 p.i. when the experiment was terminated. The effects of *T. vivax* on biochemical values of infected animals (group B) indicated significant increases (P<0.05) on serum glucose and bilirubin levels following first peak of parasitemia, remaining low thereafter until the end of the experiment. This situation was ascribed to possible cellular or organ damage following the peak parasitemia earlier observed. Lactose in saline infusion at peak parasitemia in group C animals caused normal values of serum BUN and AST. This is indicative of low-level tissue and cellular damage, or probably haemodilution arising from the infusion of lactose in saline.

Key words: lactose, Trypanosoma vivax, zebu cattle, Bos indicus, biochemistry

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Introduction

Trypanosome infections have been reported to elicit several changes in the host metabolic system (SEED et al., 1985). The changes have been associated with the level of parasites in the blood, especially during the early phase of the disease (ANOSA, 1988), and with tissue or organ damage (FIENNES, 1946; MURRAY, 1974; NOK et al., 1992; OGUNSANMI et al., 1994).

Recently, blood cellular damage (UMAR et al., 1998) and histopathological changes in liver, kidney, spleen and other related organs were found in cattle infected with T. *vivax*. Improvements in these organs were however observed following infusion of lactose in saline solution into infected cattle (IBRAHIM et al., personal communication). These observations made it necessary to investigate the various serum biochemical parameters associated with cellular and organ damage in cattle infected with T. *vivax* and infused with lactose in saline.

Materials and methods

Experimental animals. Twelve male zebu (*Bos indicus*) calves aged 12-18 months with masses of 80-90 kg were purchased from Talata Mafara, an area known to be tsetse-free in North Western Nigeria. The animals were housed at the Faculty of Veterinary Medicine, Ahmadu Bello University, Zaria. They were screened for ecto and haemo parasites; vaccinated against prevailing diseases, and fed on grass hay, wheat offal and cotton seed cake. Water and salt lick was provided *ad libitum*. Animals were allowed to acclimatize for one month before the experiment commenced.

Parasites/Infection. A stock of *Trypanosome* was isolated from a cow with a pure natural infection in Samaru village, Zaria and was identified as Samaru strain of *T. vivax.* Two millilitres of the infected cow's blood was used to infect a clean Sahel goat, which served as donor animal for the experimental calves.

The 12 calves were divided into 3 groups of four. Group A served as control animals. Group B served as infected controls, and Group C served as infected lactose in saline infused group. Groups B and C were infected with 5 ml of goat blood containing Samaru strain of *T. vivax* 1.0 10^6 /ml, of parasites.

Lactose in saline infusion. In Group C, lactose in saline was infused after parasitemia had been established at a dosage of 0.5g/kg body mass daily at the rate of 18 ml/min for 1 hr. The lactose in saline was given as a

function of animal mass, their blood volume being about 6-7% their body mass, as outlined by UMAR (1997).

Sample collection. For parasite estimation, 5 ml of blood were collected daily from cattle in all 3 groups into clean vacutainers with ethylene diamino tetracetate (EDTA) as anticoagulant. Another set of 5 ml was collected into clean vacutainers without anticoagulant; serum was harvested from clotted blood within 1 hr by centrifugation (200g/15 min), and immediately stored at -20 °C prior to analysis.

Sample analysis. Trypanosomes were detected in blood samples by the microhematocrit technique and parasite score estimated as described by WOO (1969). Serum glucose was estimated by oxidation method, serum urea nitrogen (BUN) by diacetyl monoxime method. Serum bilirubin values were estimated by modified Jendressik method, while serum aspartate amino transferase (AST) was estimated using the Reitman & Frankel method as described by CHEESEBROUGH (1991).

Statistical analysis was done using student t-test and values of P< 0.05 were considered significant.

Results

Parasitemia. Parasites were detected in the blood of infected cattle on day 2 post-infection (2 p.i.) with peak parasitemia on day 5 p.i., followed by a decline at day 6 p.i. Parasites disappeared from circulation on day 7 p.i. Subsequently, reappearance of parasites was noticed on day 10 p.i. and increased to a second peak on day 13 p.i., when the experiment was terminated (Fig. 1). In the infected infused animals (Group C), parasites also appeared on day 2 p.i., peaking on day 5 p.i. but decreasing on day 6 p.i., remaining in circulation at very low levels on days 7 to 9 p.i., the days lactose in saline was infused. Subsequently, parasites increased to a second peak on day 13 p.i. (Fig. 1).

Serum biochemical changes. Serum glucose levels were significantly low (P<0.05) on days 3, 4 and 5 p.i., corresponding with the first parasitemia build up, followed by a significant increase (P<0.05) on days 7 and 8 p.i. This also coincided with disappearance of parasites from blood. Subsequently, a gradual decrease from day 10 p.i. to below normal values was observed on days 11 to 13 p.i. In group C animals, during lactose infusion on days 6 to 10 p.i., serum glucose levels were within the normal values of the control group A animals (Fig. 1).

Serum bilirubin values for infected group B animals were within normal values on days 3 to 6 p.i., followed by intermittent significant increases (P<0.05) on days 7, 9 and 13 p.i. In group C, during lactose in

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Fig. 1. Mean serum levels of glucose, bilirubin, blood urea nitrogen (BUN), and aspartate amino trensferase (AST) in control zebu calves (group A), *T. vivax* infected calves (group B), and *T. vivax* infected lactose-in-saline infused calves (group C), in relation to the parasitemia

saline infusion on days 6 to 10 p.i., serum bilirubin values remain within normal values of the control group A animals, although it subsequently increased on days 11 and 13 p.i. (Fig. 1).

Serum urea nitrogen (BUN) values for group B animals was within normal levels on days 4, 5, and 6 p.i., followed by significantly (P<0.05) low values from days 7 to 13 p.i. as disease progressed. The pattern of change was similar in the lactose in saline infused group C animals (Fig. 1).

Serum aspartate amino transferase (AST) rose with increased parasitemia on days 4 to 6 p.i., but decreased significantly (P<0.05) on days 7 to 10 p.i. In group C animals the pattern of change for AST was similar to that of the other group during lactose in saline infusion, in that the serum levels of AST remained significantly low (Fig. 1).

Discussion

In this study, infection with the Samaru strain of *T. vivax* caused an acute disease with a very short prepatent period. This was probably due to the high dose of parasites given (MURRAY and DEXTER, 1988). Similarly, the high parasitemia observed coincided with significantly (P<0.05) low levels of glucose. This situation could be explained by the parasites' need for glucose for their cellular metabolism through their glycolytic pathway (OPPERDOES et al., 1986). Subsequent significance increases in glucose when parasites have disappeared from blood seem to further agree with the above suggestions of a parasitic/glucose relationship. However, during lactose in saline infusion, low levels of parasites were seen in blood and the glucose levels were within normal values of the control group, agreeing with the postulation that lactose may have been hydrolyse to galactose and glucose by host enzymes and its glucose section therefore provides a ready source of energy for the circulating parasites (UMAR et al., 1998), thereby no depletion of host reserve.

The insignificant changes of bilirubin values on days 3 to 6 p.i. as first parasitemia build up, suggest that even though red blood cell destruction may be taking place due to the parasitemia (IGBOKWE, 1994), there may be enhancement of liver reserves to conjugate the excess bilirubin produced during this period (COLES, 1986). However, the significant intermittent decreases as the disease progresses may be indicative of a gradual inability of the liver to effectively conjugate the bilirubin in circulation. However, lactose in saline infusion appears to bring the high levels to within normal values of the control group, probably by reducing blood cellular and organic damage, or that volume infused may have caused haemodilution of the high levels in circulation.

The increase in AST values in the disease on days 3 to 5 p.i. could not have been due to any tissue damage, but rather due to parasites secreting it as part of their metabolites into blood circulation, since alanine amino transferase ALT and AST have been observed in homogenates and suspensions of trypanosome (STEPHEN and GRAY, 1960; GRAY, 1961). However, the decreases following the second parasitemic build-up suggest a possible progressive liver fibrosis occurring as the disease progresses. Similarly, the increase in values of BUN early in the disease could be due to body catabolic breakdown of proteins as a result of the fever of the parasitemia (COLES, 1986), as has been reported in acute trypanosoma infections (ISOUN and ANOSA, 1979), while the significantly low levels observed as the disease progressed suggest possible kidney damage and accelerated kidney excretion of urea.

It seems, therefore, that the infection of cattle with *T. vivax* caused cellular and tissue damage, as indicated by the changes in the biochemical parameters observed, the changes being marked as the second parasitemia build up. However, lactose in saline infusion and the volume given appear not to have caused any tissue reaction in the animal, but probably caused reduced cellular/tissue damage and/or increased blood volume and thus, blood protein dilution.

Conclusion

Infection of zebu cattle (*Bos indicus*) with the Samaru strain of *T. vivax* in this study caused an acute infection and a variety of changes in the host biochemical parameters. These changes seem to vary with the level of parasites in blood early in the disease, but as the disease progressed the changes displayed no particular pattern, thus suggesting possible liver/kidney damage and the ineffective regulation of these biochemical parameters by the affected organs.

Lactose in saline solution infusion appears to have ameliorated the serum biochemical changes caused by the infection, probably by reducing the level of tissue damage, hence the normal values observed; also, the volume given may have caused a blood volume increase and hence the dilution of blood proteins, especially BUN and AST.

Therefore, one would surmise that lactose in saline infusion in T. *vivax* infected cattle, and the volume given, appeared to be useful to the animal, judging from the changes in the serum biochemical parameters.

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Pokusi su izvedeni na 12 mladih zebu goveda u dobi od 12 do 18 mjeseci, a podijeljenih u 3 skupine po 4 životinje. Nakon pokusne invazije sa Samaru sojem nametnika *Trypanasoma vivax*, u svih skupina su praćeni parazitemija i biokemijske vrijednosti bilirubina, glukoze, nebjelančevinskog dušika (BUN) i aspartat transaminaze (AST) u krvnom serumu. Korišteni soj nametnika T. vivax izazvao je akutnu invaziju u kojoj su se nametnici pojavili u krvnom optoku drugog dana, a vrhunac dosegli petog dana poslije invazije. Nametnici su se, međutim, izgubili iz optoka sedmog dana poslije invazije, da bi se ponovo pojavili s drugim vrhuncem trinaestog dana poslije invazije. U skupini koja je dobivala infuzije laktoze parazitemija je trajala bez remisije do 13. dana kada je pokus završen. Učinak nametnika T. vivax na biokemijske vrijednosti invadiranih životinja (skupina B) pokazao je značajni porast (P<0,05) razina serumske glukoze i bilirubina nakon prvog vrhunca parazitemije, dok su serumski BUN i AST pokazivali značajno (P<0,05) niske razine nakon prvog vrhunca i ostale su niske do kraja pokusa. To je pripisano mogućim oštećenjima stanica ili organa nakon vrhunca parazitemije. Infuzija mliječnog šećera u fiziološkoj otopini pri vrhuncu parazitemije u skupini C izazvala je normalne vrijednosti serumske glukoze i bilirubina, ali i značajno (P<0,05) niske razine BUN-a i AST-a. To ukazuje na lagano oštećenje tkiva i stanica, a vjerojatno i razređenje krvi zbog infuzije fiziološke otopine s mliječnim šećerom.

Ključne riječi: laktoza, mliječni šećer, fiziološka otopina, Trypanosoma vivax, zebu govedo, biokemija