



SHORT COMMUNICATION

Blood gas analyses and other components involved in the acid–base metabolism of rats infected by *Trypanosoma evansi*



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ABSTRACT

The aim of this study was to investigate the effects of *Trypanosoma evansi* infections on arterial blood gases of experimentally infected rats. Two groups with eight animals each were used; group A (uninfected) and group B (infected). Infected animals were daily monitored through blood smears that showed high parasitemia with 30 trypanosomes per field (1000×) on average, 5 days post-infection (PI). Arterial blood was collected at 5 days PI for blood gas analysis using an automated method based on dry-chemistry. Hydrogen potential (pH), partial oxygen pressure (pO₂), oxygen saturation (sO₂), sodium (Na), ionic calcium (Ca ionic), chlorides (Cl), partial dioxide carbon pressure (pCO₂), base excess (BE), base excess in the extracellular fluid (BE_{ecf}), bicarbonate (cHCO₃), potassium (K), lactate, and blood total dioxide the carbon (tCO₂) were evaluated. The levels of pH, pCO₂, BE, BE_{ecf}, cHCO₃, and tCO₂ were significantly decreased ($P < 0.05$) in group B compared to group A. Additionally, the same group showed increases in Cl and lactate levels when compared to uninfected group. Therefore, it is possible

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to state that the infection caused by *T. evansi* led to alterations in the acid–base status, findings that are correlated to metabolic acidosis.

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Introduction

Trypanosoma evansi, the etiological agent of a disease known as “Surra” or “*Mal das Cadeiras*” in horses, is a hemoflagellate with wide geographic distribution in tropical and subtropical regions [1–3]. The parasite is transmitted primarily by blood sucking insects and possibly by vampire bats [4]. In horses, dogs, and camels the disease progresses to death, except in rare cases. The main clinical signs of the disease include: fever, anemia, swollen lymph nodes, jaundice, weight loss, and edema of hind limbs. Horses, cats, and rats show progressive weakness and motor disorders at chronic stages of the disease [5,6]. Rats infected by *T. evansi* without treatment usually die within 4–6 days post-infection (PI), and usually show seizures, few hours before death [7].

One of the main pathological findings in animals infected by *T. evansi* is anemia [3,6,7], which can lead to major changes in blood, as well as acid–base imbalance [8]. Acid–base disturbances are commonly observed in many infections and metabolic disorders, drawing the attention to the need of a precise description of these disorders in humans and animals [8]. The arterial blood gas determination plays an important role in diagnosing acid–base status disturbances, oxygenation, and ventilation [9]. Therefore, the aim of this study was to assess the levels of blood gases and other components involved in the metabolic acid–base status during an acute infection in rats experimentally infected by *T. evansi*.

Material and methods

T. evansi isolate

In this experiment, *T. evansi* isolate was obtained from a naturally infected dog [10] kept in liquid nitrogen. One rat (R_1) was inoculated with cryopreserved parasites in order to reactivate the *T. evansi* isolate.

Animal model

Sixteen female rats (Wistar) with mean age of 70 days weighing approximately 200 (± 10 g) were used. They were housed in cages on a light/dark cycle of 12 h in an experimental room with controlled temperature and humidity (25 °C; 70% respectively), fed with commercial feed and water *ad libitum*. All the animals were submitted to a period of 15 days for adaptation. The procedure was approved by the Animal Welfare Committee of The Federal University of Santa Maria, under protocol number 065/2012.

Experimental design

Rats were divided into two groups with eight animals each: group A was used as a negative control (uninfected animals), while group B was used as a test group (animals infected by

T. evansi). The infection was induced intraperitoneally with 0.1 mL of blood from rat (R_1) containing 2.7×10^6 trypanosomes (Day 0).

Parasitemia evolution and sampling

The rats were observed during 5 days with the evolution of parasitemia monitored daily through blood smears. For this procedure, each slide was prepared with fresh blood collected from the tail vein, stained by the panoptic method, and visualized at a magnification of 1000 \times according to the methodology described by Da Silva et al. [11]. On day 5 PI the animals were anesthetized in a chamber with isoflurane for blood sampling (an average of 7 mL per animal by intra-cardiac puncture of the left ventricle) using syringes of 0.7 \times 25 mm and 22 gauge needles (BD Preset Eclipse®). A part of the blood was stored in tubes with ethylenediamine tetraacetic acid (EDTA) for hematological analyses and other part was stored in sodium fluoride for lactate and gas analyses. All analyses were immediately performed using fresh samples. After collection, the animals were decapitated as recommended by the Ethics Committee.

Hematological analyzes

The hematocrit was determined by centrifugation of microhematocrit tubes in a microhematocrit centrifuge (Sigma Laborzentrifugen, Osterode am Harz, Germany) for 5 min at 19,720g. Erythrocytes count and hemoglobin concentration were determined using an electronic counter (CELM CC-550).

Blood gas analyses and other components involved in the acid–base status

The samples were stored in a cold water bath (0 °C) and they were analyzed within 45 min as recommended by Takada et al. [12]. Initially, the negative logarithm of hydrogen ions (pH) activity in a blood gas analyzer (OMNI C® Roche Diagnostics, Brazil) was performed. Subsequently, the other variables were determined using the Vitros 250 analyzer (Ortho-Clinical Diagnostics) by the method of dry chemistry. The pH (hydrogen potential), pCO₂ (partial dioxide the carbon pressure), pO₂ (partial oxygen pressure), BE (bases excess), BE_{ecf} (base excess in the extracellular fluid), cHCO₃ (bicarbonate), sO₂ (oxygen saturation), tCO₂ (blood total dioxide the carbon), Na (sodium), K (potassium), Ca ionic (calcium ionic), Cl (chlorides), and lactate were carried out in all blood samples.

Statistical analysis

Data of blood gas analyses and other components involved in the acid–base status were first analyzed descriptively; measures of central tendency and dispersion were computed. Further, all variables were submitted to Shapiro and Wilk's test. Since

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