

Acid-base equilibrium of Brazilian locally adapted sheep under heat stress in a semi-arid region

Débora Andréa Evangelista Façanha¹, Fabrício Xavier Morais², José Moreira de Oliveira Filho², Chromácio Calafange Medeiros², Wallace Sóstene Tavares Silva³, Luis Alberto Bermejo Asensio⁴
José Ernandes Rufino de Sousa³

¹Docente do Centro de Ciências Agrárias - Grupo de Conservação da Genética Adaptada ao Semiárido da Ufersa, Campus de Mossoró, RN. e-mail: deboraf@ufersa.edu.br; ernandes@ufersa.edu.br

² Discentes de Medicina Veterinária – Ufersa, Campus de Mossoró.

³ Doutorandos do Programa de Pós-Graduação em Ciência Animal – Núcleo Pró-Criar: Pesquisa e Conservação da Genética Adaptada ao Semiárido, Campus de Mossoró, RN.

⁴ Agricultural Economy, Sociology and Policy Unit. Department of Engineering, Production and agricultural Economy - La Laguna University, Tenerife – Spain

Abstract: The Morada Nova is a Brazilian non-wool sheep, used for meat production that can present three coat colors: the black, the white and the red coat. However, black coated and white coated animals are in risk of extinction, once both are considered less adapted to warm conditions. The aim of this work was to evaluate thermoregulatory responses and their influence on acid-base equilibrium in red, black and white varieties of Morada Nova sheep, before, during and after the exposure to heat stress. Rectal temperature (RT, °C), respiratory rate (RR, breaths.minute⁻¹), skin temperature on the neck (STN, °C), skin temperature on the flank (STF, °C), cutaneous evaporation on the neck (NEv, W.m⁻²) and on the flank (FEv, W.m⁻²), were collected once a week, from 5 to 6 AM, from 12 to 1 PM and from 5 to 6 PM. The hemogasometric tests consisted in: blood pH (pH), carbon dioxide pressure (PCO₂), Oxygen pressure (PO₂), base excess (BECEF), concentration of bicarbonate ions (HCO₃⁻), Oxygen saturation (SO₂). Meteorological data were registered, and Black Globe Humidity Index and Radiant heat Load were estimated. The lower means of rectal temperature and respiratory rate were registered before the exposure to heat stress and important increases of RT and RR were registered during heat stress. The three varieties used intensively respiratory thermolysis during heat stress. The black animals showed most intensive thermoregulatory responses, followed by the red ones, however they did not keep the homeothermy. The white ones exhibited lower warm and kept normal temperature, what is advantageous for animals raised under high radiation. No differences in cutaneous evaporation were verified among black, red and white coated animals. During heat stress, the pH increased and the PCO₂ and HCO₃⁻ decreased in white and in red-coated ewes, characterizing a respiratory alkalosis, whereas the black animals presented also an increase of HCO₃⁻, which suggests respiratory and metabolic alkalosis. Four hours after heat stress peak the animals recovered homeothermic and normal blood gases patterns. We concluded that respiratory thermolysis is the most important mechanism used for sheep; however, the excessive respiratory effort during heat stress can cause acid-base disequilibrium.

Keywords: genetic resources, blood gases, adaptability

Os autores deste trabalho são os únicos responsáveis por seu conteúdo e são os detentores dos direitos autorais e de reprodução. Este trabalho não reflete necessariamente o posicionamento oficial da Sociedade Brasileira de Biometeorologia (SBBiomet).

The authors of this paper are solely responsible for its content and are the owners of its copyright. This paper does not necessarily reflect the official position of the Brazilian Society of Biometeorology (SBBiomet).

Introduction

According to FAO's Global Plan of Action, the sustainable management of animal genetic resources (AnGR) remains a vital and challenging task and one of the most important strategies for local breeds' conservation in hot environments emphasizes their capacity to cope with climatic changes and global warming. Producing meat in stressful tropical environments can be both complex and difficult, and the anticipated effects of climate change are expected to increase the environmental stress in all livestock systems. Strategies to improve productivity will involve genetics that deliver a product of high quality, from environmentally and socially sustainable production systems.

The Morada Nova is a Brazilian locally adapted breed of non-wool sheep, normally used for meat production in extensive systems, in the semi-arid region of Brazil. The animals have a small size, weight at about 30 – 40 kg and can present three coat colors: the black, the white and the red coat. However, the black coated animals are discarded from the genealogic book, and white coated animals are in risk of extinction, once both are considered less adapted to warm conditions than the red animals.

In fact, some studies conducted at Mossoró, RN (Maia et al., 2014) showed that when solar radiation levels increased from 300 to 1000 W.m⁻² the heat losses through latent flow increased in both, respiratory and cutaneous surface of goats, independent of the coat color. Costa et al, (2015) verified an increase of respiratory rate (RR) in Red Morada Nova ewes, when compared to the white ones. Morais-Leite et al., (2017) found that Morada Nova ewes were able to maintain RR in low stressful levels (40 to 60 breaths/minute) over different environmental conditions. On the other hand, Silva et al., (2017) verified the increase of respiratory thermolysis as the most important mechanism triggered for Morada Nova ewes under intense heat stress conditions. Therefore, these findings must be confirmed and better investigated, once maintained RR in high levels can cause some physiological disturbance, such as acid-base disequilibrium and respiratory alkalosis.

In the present study, we hypothesized that even dark colored animals can compensate the additional heat gain and the thermoregulatory effort and then recover acid-base equilibrium in few hours after return to a thermal comfortable situation. Therefore, we aimed to evaluate thermoregulatory responses and their influence on acid-base equilibrium in red, black and white varieties of Morada Nova sheep, before, during and after the exposure to natural heat stress in an open field, in semi-arid environment.

Material and Methods

This research was conducted at the Small Ruminant Research Centre of Semi-Arid Federal University (UFERSA), in Mossoró, Brazil, with the geographic coordinates 5°12' south, 37°19' west, into Brazilian semi-arid region. The climate in the region is tropical semi-árid, with the rainy season from January to May and the dry season from June to December. The annual average of temperature and humidity are 27.2 °C and 70% respectively, with a mean annual rainfall of 414.7 mm (Silva et al., 2017). The experiment was carried out during two months in the dry season, and two months in the rainy season. The animals were kept in an open field since 24 hours before measurements and received *Cynodon dactylon* hay, concentrate mix composed by corn grain, soybean meal, mineral and vitamin complex and water ad libitum.

Data were collected once a week, before heat stress, from 5 to 6 AM (T1), during the peak of heat stress, from 12 to 1 PM (T2) and after heat stress, from 5 to 6 PM (T3), according to the findings of Silva et al., (2017). The thermoregulatory traits evaluated included: rectal temperature (RT, °C), measured with a digital clinical thermometer; respiratory rate (RR, breaths/minute), with a stethoscope; skin temperature on the neck (STN, °C), skin temperature on the flank (STF, °C), both with an infrared thermometer. Cutaneous evaporation on the neck (NEv, W.m⁻²) and cutaneous evaporation on the flank (FEv, W.m⁻²), were also measured, with a ventilated capsule. In the same times of thermoregulatory evaluations was taken a venous blood sample of each animal, in heparinized tubes for, aiming to evaluate the acid-base equilibrium. The hemogasometric tests consisted in: blood pH (pH), carbon dioxide pressure (PCO₂), Oxygen pressure (PO₂), base excess (BE), concentration of bicarbonate ions (HCO₃⁻), Oxygen saturation (SO₂), total concentration of carbon dioxide (TCO₂), hematocrit (Hct), hemoglobin (Hb), sodium (Na⁺), calcium (Ca⁺). At the same times of animals samplings, meteorological data were registered, and Black Globe Humidity Index (BGHI) and the Radiant heat Load (RHL) were estimated, using the following equations: BGHI = T_{bg} + 0.36 + 41.5 t_{pd} (T_{bg} = Black globe temperature, °C; T_{pd} = Dew point temperature, °C); RHL = σ (MRT)⁴ (Radiant Heat Load, w.m⁻²; σ = Stefan-Boltzmann constant, 5.67 .10⁻⁸ w. 2.k m⁻¹; MRT = mean radiant temperature, ° K).

Data were analyzed by mixed models, using the Statistical Analysis System (SAS Institute, 1995), according to the following statistical model: $Y_{ijk} = \mu + TD_i + V_j + V(TD)_{ij} + \varepsilon_k$; where TD: effect of time of the day; V: effect of variety; V(TD): effect of variety within time of the day and ε: experimental error. The

means were compared using Tukey test, except for BE, which was submitted to Kruskal Wallis test, once showed a non-parametric behavior. A Principal Component Analysis was carried out, including thermoregulatory and hemogasometric traits.

Results and Discussion

According to Table 1 we verified that the lower means of rectal temperature (RT) and respiratory rate (RR) were registered during the first sampling (T1), before the exposure to heat stress. We detected significant and important increases of RT and RR during the (T2), when the animals were exposed to the most stressful thermal conditions. The three varieties used intensively respiratory thermolysis during heat stress. The black animals showed most intensive thermoregulatory responses, followed by the red ones; however both varieties didn't keep the homeothermy. On the other hand, the white ones exhibited lower warm and kept body temperature inside of homeothermic range and this can be considered advantageous for animals raised under high radiation exposition, once they have pigmented skin.

Despite of black ewes had higher skin temperature; we did not verify statistical differences in cutaneous evaporation among black, red and white coated animals, especially during T2. It is important to emphasize that no differences were detected in cutaneous evaporation among the times of sampling, so that this mechanism was used with the same intensity before, during and after heat stress exposure. On the other hand, as thermal environment becomes more stressful, all varieties triggered respiratory thermolysis, specially the black coated ones. These results alert to the risk of some disturbance on acid-base balance, due to hyperventilation.

We verified that before heat stress (T1) the white and the red ewes tended to metabolic acidosis, once the pH and the HCO_3^- dropped. The black coated animals presented normal blood gases values. During heat stress, we verified an increase of pH and a decrease of PCO_2 and HCO_3^- in white and in red-coated ewes, characterizing a respiratory alkalosis, whereas the black animals presented also an increase of HCO_3^- , which suggests respiratory and metabolic alkalosis associated and emphasizes the necessity of protection from the excessive radiation exposure. An important finding was that four hours after heat stress peak all the animals recovered homeothermic and normal blood gases patterns, despite of the coat color.

We did not verify any change in hematocrit, among varieties and time of sampling, even when the animals used intensively the evaporative mechanisms and lost an important amount of water. The values of hemoglobin Ca^+ and Na^+ concentrations followed the same pattern and were always inside of the normal range.

Table 1. Means for thermoregulatory and hemogasometric traits in Morada Nova ewes (white, red and black coats) on each time of sampling

	Time of Day								
	05:00h - 06:00h (T1)			12:00h - 13:00h (T2)			17:00h - 18:00h (T3)		
	White	Red	Black	White	Red	Black	White	Red	Black
Thermoregulatory traits									
RT (°C)	38,19 ^B	37,9 ^B	38,65 ^A	39,46 ^B	39,78 ^A	40,03 ^A	38,69 ^A	38,69 ^A	38,93 ^A
RR (breaths/min)	26,96 ^A	29,26 ^A	34,46 ^A	134,73 ^B	144,46 ^B	161,01 ^A	28,53 ^A	29,6 ^A	31,65 ^A
STN (°C)	30,14 ^A	30,44 ^A	30,67 ^A	35,8 ^B	37,25 ^{AB}	38,93 ^A	31,78 ^A	31,81 ^A	31,55 ^A
STF (°C)	30,3 ^A	30,58 ^A	30,4 ^A	36,92 ^C	40,59 ^B	44,63 ^A	31,57 ^A	31,7 ^A	30,9 ^A
NEv (W.m ⁻²)	307,53 ^A	300,26 ^A	296,9 ^A	256,13 ^A	249,01 ^A	253,4 ^A	323,8 ^A	297,71 ^A	312,66 ^A
FEv (W.m ⁻²)	313,52 ^A	283,89 ^A	290,07 ^A	225,28 ^A	248,52 ^A	245,63 ^A	299,61 ^A	305,94 ^A	304,05 ^A
Hemogasometric traits									
pH	7,38 ^A	7,31 ^A	7,40 ^A	7,42 ^A	7,46 ^A	7,48 ^A	7,39 ^A	7,41 ^A	7,41 ^A
PCO_2 (mm.Hg ⁻¹)	38,24 ^B	39,58 ^{AB}	41,17 ^A	35,14 ^A	33,53 ^A	33,97 ^A	36,22 ^A	36,83 ^A	38,72 ^A
PO_2 (mm.Hg ⁻¹)	45,63 ^A	37,1 ^A	41,92 ^A	51,96 ^A	49,96 ^A	54,33 ^A	46,13 ^A	41,23 ^A	45,31 ^A
BE (mmol.L ⁻¹)	-2,51 ^B	-1,50 ^B	2,50 ^A	-1,00 ^B	-1,00 ^B	2,50 ^A	-2,55 ^B	-1,22 ^{AB}	0,06 ^A
HCO_3^- (mmol.L ⁻¹)	22,62 ^B	23,88 ^B	25,7 ^A	23,28 ^A	23,57 ^A	24,78 ^A	21,86 ^B	23,01 ^{AB}	24,41 ^A
SO_2 (%)	71,38 ^A	62,56 ^B	65,63 ^{AB}	79,93 ^A	76,71 ^A	79,85 ^A	73,28 ^A	66,23 ^B	70,45 ^{AB}
TCO_2 (mM)	23,96 ^B	24,96 ^B	26,87 ^A	24,31 ^A	24,15 ^A	25,97 ^A	22,85 ^B	24,15 ^{AB}	25,49 ^A

Hct (%)	20,63 ^A	20,88 ^A	17,96 ^B	19,5 ^A	19,6 ^A	16,71 ^B	19,43 ^A	19,45 ^A	16,48 ^B
Hb (g.dL ⁻¹)	6,86 ^A	7 ^A	6,11 ^B	6,6 ^A	6,61 ^A	5,76 ^B	6,53 ^A	6,39 ^A	5,61 ^B
Na ⁺ (mmol.L ⁻¹)	146,31 ^A	145,83 ^A	144,67 ^B	148,13 ^A	147,23 ^{AB}	146,64 ^B	147,86 ^A	146,25 ^A	146,81 ^A
Ca ⁺ (mmol.L ⁻¹)	0,84 ^A	0,85 ^A	0,85 ^A	0,92 ^A	0,86 ^A	0,82 ^A	0,84 ^A	0,98 ^A	0,81 ^A

RT - Rectal temperature; RR - Respiratory rate; STN - Surface temperature of the neck; STF – Surface temperature of the flank; NEv - Cutaneous evaporation of the neck; FEv - Cutaneous evaporation of the flank. pH- hydrogenation potential; PCO₂ - venous oxygen pressure; PO₂ - venous pressure of carbon dioxide; BE - base excess; HCO₃⁻ - bicarbonate; SO₂ - oxygen saturation; TCO₂ - total concentration of carbon dioxide; Hct - hematocrit; Hb - hemoglobin; Na⁺-sodium; Ca⁺ - calcium.

The principal component analysis showed that the first component (Axis 1) was related to thermoregulatory traits, whereas the second component (Axis 2) was related to hemogasometric traits. (Fig 1. A). In Axis 1, the most important variables were body temperature (RT, STN, STF) and RR and an inverse relationship between these traits and FEv suggests that when the environment becomes stressful and the animals increase body temperature, they trigger preferentially respiratory thermolysis, instead of cutaneous thermolysis. These finding confirms that even no woolled sheep, witch theoretically can find less difficulties to loose heat from skin surface, used more intensively respiratory thermolysis. That is why the risk of an acid-base disequilibrium and its negative effects must be considered on the animals' homeostasis. The most important hemogasometric traits were TCO₂, HCO₃⁻ and BE and this can be due the drop of PCO₂ that occurred during respiratory alkalosis verified in all varieties of ewes, in T2.

The Fig 1 (B) showed that all parameters values changed from T1 to T2, as a response to heat stress. The animals triggered thermoregulatory mechanisms and increased respiratory rate hemogasometric parameters indicated acid-base disequilibrium when exposed to the most stressful conditions. After noon (T3) thermoregulatory mechanisms means decreased and the animals from all varieties recovered homeothermic patterns as well as acid-base equilibrium. According to Fig 1 (B) we observed that the white and the red animals had similar behavior when compared to black coated ones. These findings confirmed that dark colored animals, even from adapted animals must be protected from excessive solar radiation, once they can absorb excessive warm and, despite of their ability for thermoregulation, it can have negative effects on homeostasis and performance.

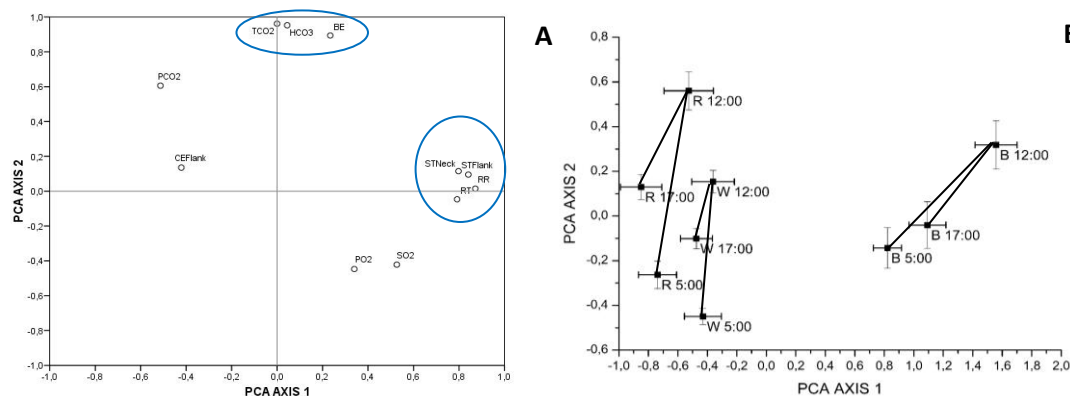


Figure 1 Principal Component Analysis (PCA) plot based on thermoregulatory and Hemogasometric traits of Morada Nova ewes under thermal comfort and thermal stress. T1 (5:00h); T2 (12:00h); T3 (17:00h). W; white variety; R: red variety; B: black variety. RT: respiratory temperature; STN: skin temperature on the neck; STF: skin temperature on the flank; RR: respiratory rate; BE: base excess.

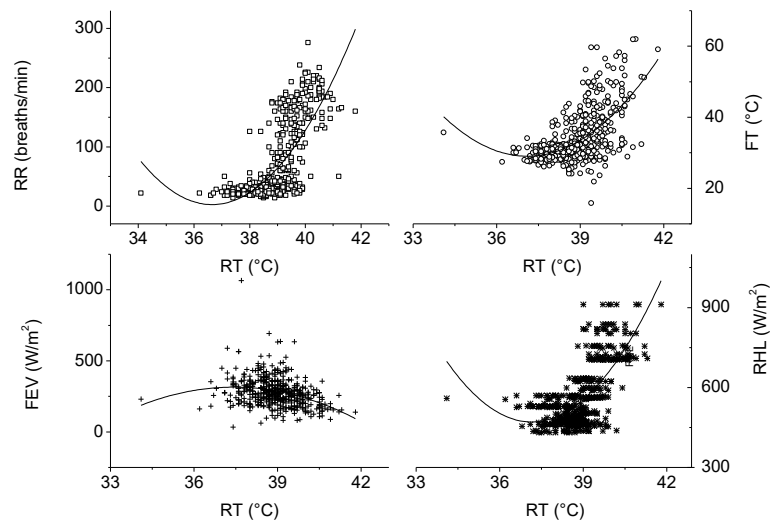


Figure 2. Variation of rectal temperature (RT °C), Flank evaporation (FEV, W.m⁻²), respiratory rate (RR), flank surface temperature (FT °C) and radiant heat load (RHL, W.m⁻²) produced; in Morada Nova ewes.

The rectal temperature (RT) showed a strong and positive relationship with radiant heat load and flank temperature was also strongly related to RT, maybe due to the absorption of solar radiation. These results confirm the necessity of protecting the animals from excessive radiation exposure (Fig. 2). The strong and positive relationship found between RT and Respiratory rate (RR) showed the importance of this mechanism in total thermolysis. On the other hand, RT was negatively related with flank evaporation (FEV). These results when RT overcame the normal range and exceeded 40°C, heat stressed animals triggered intensive respiratory thermolysis instead of cutaneous evaporation.

Conclusions

We concluded that respiratory thermolysis is the most important evaporative mechanism used for sheep, even in non-wooled breeds. However, the excessive respiratory effort can cause acid-base disequilibrium and disturb homeostasis.

Acknowledgements

We acknowledge the Embrapa – Goat and Sheep and The Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (grant number 88881030352/2013-1) for the financial support.

References

- SILVA, W. E.; LEITE, J. H. G. M.; SOUSA, J. E. R.; SILVA, W. S. T.; GUILHERMINO, M. M.; ASENSIO, L. A. B.; FAÇANHA, D. A. E. **Daily rhythmicity of the thermoregulatory responses of locally adapted Brazilian sheep in a semiarid environment.** *International Journal of Biometeorology* (Print), p. 1-11, 2017.
- LEITE, J. H. G. M.; FAÇANHA, D. A. E.; COSTA, W. P.; CHAVES, D. F.; GUILHERMINO, M. M.; SILVA, W. S. T.; BERMEJO, L. A. **Thermoregulatory responses related to coat traits of Brazilian native ewes: an adaptive approach.** *Journal of Applied Animal Research*, v. 1, p. 1-7, 2017.
- MAIA, A. S. C.; SILVA, R. G.; NASCIMENTO, S. T.; NASCIMENTO, C. C. N.; PEDROZA, H. P.; DOMINGOS, H. G. T. **Thermoregulatory responses of goats in hot environments.** *International Journal of Biometeorology*, v. 59, p. 1025-1033, 2014.
- COSTA, W. P.; FAÇANHA, D.A.E.; MORAIS, J. H. G.; PEIXOTO JUNIOR, G. N. A.; SOUZA, C. H.; CHAVES, D. F.; VASCONCELOS, A. M.; SOTO-BLANCO, B.; VALE, A. M.; PIMENTA-FILHO, E. C. **Thermoregulatory responses and blood parameters of locally adapted ewes under natural weather conditions of Brazilian Semiarid region.** *Semina. Ciências Agrárias* (Online), v. 36, p. 4589-4600, 2015.