



# Reductions in the water supply to crossbred Santa Inês ewes in the Brazilian semi-arid: Apparent nutrient digestibility, water and nitrogen balance, and performance

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## ABSTRACT

**Summary:** This study aimed to evaluate the intake and nutrient digestibility, water, and nitrogen balance of Santa Inês crossbred ewes under confinement receiving reductions in the water supply. Thirty-two Santa Inês crossbred ewes, at an average body weight of  $32.2 \pm 7.4$  kg and age of  $2.3 \pm 0.99$  years, were distributed in a completely randomized design, with four treatments, and eight animals per treatment, the treatments were 100%, 80%, 60%, and 40% of ad libitum water. Nutrient intake and digestibility, water and nitrogen balance, and performance were evaluated. The reductions in the water supply did not promote significant alterations, in the nutrient intake, or apparent digestibility of the animals ( $P > 0.05$ ). A linear decreasing effect was observed for free water intake ( $P < 0.001$ ), total water intake ( $P < 0.001$ ), total urine output ( $P = 0.008$ ), water excreted via urine ( $P = 0.006$ ), water excreted via feces ( $P = 0.006$ ), total water losses ( $P < 0.001$ ), water absorbed ( $P < 0.001$ ), and nitrogen excreted via feces ( $P = 0.002$ ). The water and nitrogen balance and performance were not influenced by water offers ( $P > 0.05$ ). It was concluded that water offers of up to 40% of ad libitum intake, do not promote negative effects on intake and apparent nutrient digestibility, mineral intake, nitrogen balance, and water balance in Santa Inês ewes.

## 1. Introduction

Arid and semiarid regions are characterized by high temperatures, high levels of evapotranspiration, low indices, and irregular rainfall. The same conditions are intrinsic to the Brazilian Semiárido, this added to the characteristics of the geological structure, which confer a crystalline shield that does not allow the accumulation of sufficient water in the subsoil, which is the largest source of water available to the region, resulting in less water availability, this being a limiting factor for livestock productivity, not only in the Brazilian Semiárido but also worldwide (Iniguez, 2011; Vosooghi-Postindoz et al., 2018; Albuquerque et al., 2020).

On the other hand, native or naturalized small ruminants in arid and semiarid regions are efficient water-using animals, due to their small size and better use of ingested and excreted water, giving them greater tolerance to water restriction. This fact is a result of adaptive mechanisms that increase the efficiency of water use and reabsorption along the digestive tract (Araújo et al., 2019). However, water deficit can affect the physiological homeostasis of animals, leading to reduced nutrient intake and digestibility, changes in electrolyte balance and plasma concentration of solutes, and consequent alteration of water and nitrogen balance to maintain body and tissue water levels without further impairment (Al-Ramamneh et al., 2012; Nejad et al., 2017; Akinmoladun et al., 2019).

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Given this scenario, research has been developed to seek solutions aimed at decreasing the effects of reduced water supply, and to determine the minimum water requirement required to maintain production rates without causing harm to animal welfare (D'Ambrosio et al., 2018; Kalyan et al., 2020; Araújo et al., 2021). However, most have studied the effect of intermittent water offerings, or water restriction at a fixed level, with supplementation of water requirement via succulent feed, or the use of antioxidant components, on intake, water and nitrogen balance, blood metabolites, and performance.

Therefore, improving the knowledge of the effects of reduced water supply at different levels for a period comprising an entire production cycle on animal metabolism may guide the possibility of using new strategies for water management under conditions of low water availability to maintain viable production. Therefore, this study aimed to evaluate the intake and digestibility of nutrients, the water and nitrogen balance, and performance in Santa Inês ewes subjected to reduced water supply in a confinement system.

## 2. Material and methods

### 2.1. Study site description

The study was conducted in the Laboratory of Animal Requirements and Metabolism, located in the Campus of Agricultural Sciences of the Federal University of São Francisco Valley (UNIVASF) in the city of Petrolina, Pernambuco, Brazil. This area is in the Caatinga biome, in the city of Petrolina - PE, Brazil. The average annual rainfall is 570 mm, during the experimental period, the maximum and minimum temperatures observed were 33.83 and 24.56 °C, respectively with relative humidity between 50.50% and 73.56%.

Before being carried out, the study was evaluated and approved by the Committee on Ethics and Deontology in Studies and Research of the Federal University of Vale do São Francisco (UNIVASF) (approval no. 0002/241017).

### 2.2. Animals, treatments, and experimental diet

Thirty-two Santa Inês crossbred ewes, with a mean body weight of  $32.2 \pm 7.4$  kg, and mean age of  $2.3 \pm 0.99$ , were distributed in an entirely randomized design with four treatments and eight repetitions per treatment. The performance trial lasted 58 days, and the metabolic trial lasted 15 days, with 10 days for adaptation and 5 days for data collection. The treatments were 100%, 80%, 60%, and 40% of ad libitum water intake.

The animals were previously identified by wearing ear tags, weighed, treated against endoparasites and ectoparasites, and then housed in metabolism cages equipped with feeders and drinkers made of black polyvinyl chloride (PVC). Samples of the water offered to the animals were collected every fifteen days for physical-chemical analysis (Table 1).

The diet was formulated according to the NRC (2007) to obtain daily

gains of  $157 \text{ g day}^{-1}$ , based on fresh Elephant grass cv. Cameron (*Pennisetum purpureum*, Schum) and concentrate consisting of corn bran, soybean meal, urea, and mineral salt (46:54 roughage: concentrate ratio - dry matter based) (Table 2). Feed was offered daily, at 0900 h and 1500 h, and water, at 0900 h. The amount of feed supplied was calculated based on the previous day's intake, not allowing leftovers of more than 15% of the amount offered (Table 3).

### 2.3. Sample collection

Weighing of the food and water supplied, as well as of the leftovers, feces, and urine was performed daily, and samples of these materials were collected for further analysis, to determine the intake and digestibility of the nutrients, as well as the water and nitrogen balances.

Samples of the feed and leftovers were collected during the five days of the collection period. Feces were collected using collection bags attached to the animals before the collection period, and the bags were weighed and emptied twice daily. Urine was collected once a day in plastic buckets containing 100 ml of 10% sulfuric acid to avoid the volatilization of nitrogen.

The samples were weighed, identified, and stored at  $-15^\circ\text{C}$ , and at the end of the collection period, they were homogenized to obtain one composite sample per repetition. The samples were then pre-dried in an oven with forced circulation at  $65^\circ\text{C}$  for 72 h. The food samples, leftovers, and feces were ground in a Willey knife mill with a 1.0 mm sieve for further chemical analysis.

### 2.4. Chemical analyses

The collected samples were submitted to chemical analyses according to the methods described by the Association of Official Analytical Chemists (AOAC, 2016) to determine the dry matter (DM; Method 967.03), mineral matter (MM; Method 942.05), crude protein (CP; Method 981.10). The ether extract (EE) content was obtained using a fat extractor (ANKOM TX-10, Macedon - NY, United States), according to the method of the American Oil Chemists' Society (AOCS, Champaign, 2017). Neutral detergent fiber corrected for ash and protein (using thermo-stable alpha-amylase, without sodium sulfite) (NDFap; Mertens,

**Table 2**  
Ingredients and chemical composition of the experimental diet.

| Ingredients  | g kg <sup>-1</sup> dry matter |          |              |       |
|--|-------------------------------|----------|--------------|-------|
| Elephant grass                                     | 460                           |          |              |       |
| Corn meal  | 381                           |          |              |       |
| Mineral Salt <sup>1</sup>                          | 20                            |          |              |       |
| Urea   | 7                             |          |              |       |
| Soybean meal                                       | 132                           |          |              |       |
| Nutrients (g kg <sup>-1</sup> )                    | Elephant grass                | Cornmeal | Soybean meal | Diet  |
| Dry matter <sup>2</sup>                            | 272.0                         | 889.3    | 886.1        | 617.2 |
| Organic matter <sup>3</sup>                        | 919.9                         | 987.1    | 935.2        | 919.1 |
| Mineral matter <sup>3</sup>                        | 80.1                          | 12.9     | 64.8         | 80.9  |
| Crude Protein <sup>3</sup>                         | 50.4                          | 89.9     | 487.4        | 131.7 |
| Ether extract <sup>3</sup>                         | 12.2                          | 45.1     | 19.0         | 24.6  |
| Neutral detergent fiber <sup>3</sup> <sub>ac</sub> | 714.1                         | 111.6    | 154.6        | 421.7 |
| Acid detergent fiber                               | 398.6                         | 33.7     | 88.5         | 206.7 |
| Total carbohydrates                                | 857.2                         | 859.9    | 42.8         | 762.8 |
| Non-fibrous carbohydrates                          | 143.2                         | 642.0    | 27.85        | 367.7 |
| Hemicellulose                                      | 315.5                         | 77.9     | 66.1         | 215.0 |
| Total digestible nutrients                         | 570.1                         | 850.0    | 800.48       | 821.4 |

<sup>1</sup> Guarantee levels per kilogram of the product according to the manufacturer: Calcium (min.) 190 g; Phosphorus (min.) 75 g; Magnesium (min.) 10 g; Chlorine (min.) 218 g; Sulfur (min.) 70 g; Sodium (min.) 143 g; Copper (min.) 300 mg; Cobalt (min.) 405 mg; Iron (min.) 500 mg; Iodine (min.) 80 mg; Manganese (min.) 1100 mg; Selenium (min.) 30 mg; Zinc (min.) 4600 mg; Fluorine (max.) 0.87 g; Phosphorus (P) solubility in 2% citric acid (min): 95%. <sup>2</sup> In g/kg natural matter. <sup>3</sup> Neutral detergent fiber corrected for ash and crude protein.

**Table 1**  
Physical and chemical characteristics of the water offered.

| Parameters   |      |
|--|------|
| Calcium (mmol L <sup>-1</sup> )                        | 0.63 |
| Magnesium (mmol L <sup>-1</sup> )                      | 0.74 |
| Sodium (mmol L <sup>-1</sup> )                         | 0.27 |
| Potassium (mmol L <sup>-1</sup> )                      | 0.18 |
| Carbonates (mmol L <sup>-1</sup> )                     | 0.0  |
| Bicarbonates (mmol L <sup>-1</sup> )                   | 0.32 |
| Sulfates (mmol L <sup>-1</sup> )                       | 0.51 |
| Chlorides (mmol L <sup>-1</sup> )                      | 0.66 |
| pH   | 6.98 |
| Electrical conductivity (ds m <sup>-1</sup> )          | 0.08 |
| Water hardness CaCO <sub>3</sub> (mg L <sup>-1</sup> ) | 3.44 |

**Table 3**  
Mineral composition of ingredients and experimental diet.

| Minerals                         | Roughage | Concentrate | Diet   |
|----------------------------------|----------|-------------|--------|
| Phosphorus (g kg <sup>-1</sup> ) | 4.51     | 11.67       | 8.37   |
| Potassium (g kg <sup>-1</sup> )  | 20.35    | 16.80       | 18.43  |
| Calcium (g kg <sup>-1</sup> )    | 6.77     | 15.83       | 11.66  |
| Magnesium (g kg <sup>-1</sup> )  | 3.21     | 1.95        | 2.52   |
| Sodium (g kg <sup>-1</sup> )     | 0.26     | 4.8         | 2.71   |
| Sulfur (g kg <sup>-1</sup> )     | 1.51     | 3.06        | 2.34   |
| Boron (mg kg <sup>-1</sup> )     | 9.63     | 7.46        | 8.45   |
| Copper (mg kg <sup>-1</sup> )    | 10.79    | 131.13      | 75.77  |
| Iron (mg kg <sup>-1</sup> )      | 97.16    | 269.85      | 190.41 |
| Manganese (mg.kg <sup>-1</sup> ) | 47.96    | 79.51       | 64.99  |
| Zinc (mg kg <sup>-1</sup> )      | 42.94    | 136.50      | 93.46  |

2002; Licitra et al., 1996), and acid detergent fiber (ADF) were determined as described by Van Soest et al. (1991). Hemicellulose (HEM) was calculated by the following equation: HEM = NDF – ADF.

Total carbohydrates (TC) were estimated according to Sniffen et al. (1992), and non-fibrous carbohydrates (NFC) were estimated using the equations recommended by Hall (2003), with NDF corrected for ash (a) and protein (p) (NDFap). Total digestible nutrients (TDN) were estimated based on apparent digestibility data and calculated according to Sniffen et al. (1992). The TDN of the diets was converted into digestible and metabolizable energy using the equations described by the National Research Council (NRC, 2001).

Regarding the quantification of the mineral content of the samples, nitrogen (N) followed the methodology described by AOAC (2016; Method No. 981.10). Sodium (Na) and calcium (Ca) concentrations were performed using a flame spectrophotometer (AOCS, 1984). Potassium (K) and magnesium (Mg) contents were determined according to Harris (1991) methodology. Phosphorus (P) was determined in a spectrophotometer following a dilution of the ash extract (1:20) and after a reaction with ammonium molybdate (Arabi et al., 2014). While the concentrations of iron (Fe) and zinc (Zn) were determined according to the methodology of Khan et al. (2017) with the aid of a mass spectrometer. Copper (Cu) and sulfur (S) were determined by atomic absorption spectrometry (Irschik et al., 2013; Zambrzycka and Godlewska-Żyłkiewicz, 2014).

## 2.5. Intake and apparent digestibility of nutrients

The nutrient intake was calculated by the difference between the amount of nutrient present in the feed supplied and the amount of nutrient present in the leftovers, all based on the percentage of dry matter. The apparent nutrient digestibility coefficient was calculated as described by Silva and Leão (1979).

Total digestible nutrient (TDN) intake was calculated according to Sniffen et al. (1992):  $TDNi = (CPI - CPF) + 2.25 * (EEi - EEf) + (TCi - TCf)$ , so that CPI, EEi, and TCi mean crude protein, ether extract, and total carbohydrate intake, respectively, while CPF, EEf, and TCf refer to the excretion of CP, EE, and TC in feces. To calculate ME (Mcal of ME/kg of DM), initially, the digestible energy (DE) was calculated as the product between the TDN content and the factor 4.409/100 considering the concentration of ME 82% of ED.

## 2.6. Temperature and humidity index

Air temperature and relative humidity were recorded and stored every minute using a HOBO® U12-013 Temp/RH 2EXT data logger (temperature measurement range between –20 °C and 70 °C, and an accuracy of +/- 0.35 °C, and relative humidity between 5% and 95%, with an accuracy of +/- 2.5%) installed at a height of 1.5 m from the ground. The hourly averages of these parameters were obtained, and the Temperature and Humidity Index (THI) was calculated according to the equation proposed by Thom (1959):

$$THI = aT + 0.36 dpT + 41.5$$

Where: THI = temperature and humidity index; aT = air temperature (°C); dpT = dew point temperature (°C).

## 2.7. Water and nitrogen balance

The water was weighed before being fed and again 24 h later. The evaporated water was estimated using buckets randomly arranged around the experimental shed, with the same amount of water available for each treatment, and was determined by the difference in weight over 24 h. The water lost by evaporation was also considered in the calculation of water intake.

The water balance was evaluated using the following equations: Total water intake (kg day<sup>-1</sup>) = water consumed (corrected for evaporation) + water in feed; Total water production = water excreted by urine + water excreted by feces; Water balance = total water intake - total water production (Church, 1976).

Apparent nitrogen balance (NB) was calculated using the following equations described by Silva and Leão (1979), NB = N ingested - (N feces + N urine); N absorbed = N ingested - N feces; N ingested = N offered - N refuses, the data were expressed in g day<sup>-1</sup>.

## 2.8. Productive performance

The animals were weighed at the beginning and end of the experimental period, after a 12 h period of deprivation of solid food (with access to water) to obtain the initial body weight and final body weight, and then it was possible to calculate the total weight gain, average daily gain, average daily gain per metabolic weight unit.

## 2.9. Statistical analysis

Regression analysis was performed by PROC REG using the Statistical Analysis System version 9.0 (SAS University), considering probabilities less than 5% as significant.

The following statistical model was adopted:

$$Y = \mu + T_j + e_{ij}$$

Where: Y = observed value of the variable;  $\mu$  = overall mean;  $T_j$  = effect of water supply level j;  $e_{ij}$  = residual error.

## 3. Results

The water supply did not influence nutrients, digestible energy, and metabolizable energy intake, as well as digestibility coefficients of nutrients ( $P > 0.05$ ) (Table 4). The THI index remained steadily under 80 between 1:00 am and 7:00 am. Subsequently, between 8:00 a.m. and 10:00 p.m., a remarkable range of THI values between 80 and 89 was observed. It is important to emphasize the period between 3:00 p.m. and 6:00 p.m., during which THI values exceeded the 90 thresholds. In particular, the highest value (91.3) was recorded at 4 pm (Fig. 1).

A linear decreasing effect was observed on free water intake ( $P < 0.001$ ), total water intake ( $P < 0.001$ ), total urine output ( $P = 0.008$ ), water excreted via urine ( $P = 0.006$ ), water excreted via feces ( $P = 0.006$ ), total water losses ( $P < 0.001$ ) and water absorbed ( $P < 0.001$ ) (Table 5). A decrease in the nitrogen excreted via feces was observed ( $P < 0.001$ ). The other variables related to nitrogen balance were not influenced by water supply ( $P > 0.005$ ) (Table 5). Performance was not influenced by water supply ( $P > 0.05$ ) (Table 6).

## 4. Discussion

The biological mechanisms highlighted as reducing productivity in the presence of a stressor agent, like the proposed treatments, are mainly

**Table 4**

Daily intake of nutritional components and apparent nutrient digestibility in crossbred Santa Inês ewes submitted to a reduction in the water supply.

| Variables  | Water offering levels (%) |      |      |      | SEM   | P-value<br>L |
|--|---------------------------|------|------|------|-------|--------------|
|  | 100                       | 80   | 60   | 40   |       |              |
| <i>Intake</i>  |                           |      |      |      |       |              |
| Dry matter (kg day <sup>-1</sup> )                       | 1.15                      | 1.10 | 1.10 | 1.08 | 0.044 | 0.574        |
| Organic matter (kg day <sup>-1</sup> )                   | 0.95                      | 0.88 | 0.88 | 0.86 | 0.034 | 0.373        |
| Crude protein (kg day <sup>-1</sup> )                    | 0.16                      | 0.15 | 0.15 | 0.15 | 0.006 | 0.478        |
| Neutral detergent fiber (kg day <sup>-1</sup> )          | 0.44                      | 0.42 | 0.43 | 0.41 | 0.018 | 0.715        |
| Acid detergent fiber (kg day <sup>-1</sup> )             | 0.20                      | 0.20 | 0.20 | 0.19 | 0.009 | 0.734        |
| Ether extract (kg day <sup>-1</sup> )                    | 0.03                      | 0.03 | 0.03 | 0.03 | 0.001 | 0.489        |
| Non-fibrous carbohydrates (kg day <sup>-1</sup> )        | 0.44                      | 0.42 | 0.42 | 0.41 | 0.016 | 0.559        |
| Total carbohydrates (kg day <sup>-1</sup> )              | 0.87                      | 0.83 | 0.83 | 0.81 | 0.034 | 0.598        |
| Total digestible nutrient intake (kg day <sup>-1</sup> ) | 0.83                      | 0.84 | 0.81 | 0.81 | 0.029 | 0.748        |
| Digestible energy intake (Mcal kg <sup>-1</sup> )        | 3.64                      | 3.68 | 3.55 | 3.56 | 0.129 | 0.755        |
| Metabolizable energy (Mcal kg <sup>-1</sup> )            | 2.98                      | 3.02 | 2.91 | 2.92 | 0.105 | 0.749        |
| <i>Digestibility coefficient (g kg<sup>-1</sup> DM)</i>  |                           |      |      |      |       |              |
| Dry matter   | 0.72                      | 0.74 | 0.74 | 0.75 | 0.007 | 0.363        |
| Organic matter   | 0.71                      | 0.73 | 0.72 | 0.72 | 0.008 | 0.780        |
| Crude protein  | 0.83                      | 0.89 | 0.89 | 0.89 | 0.010 | 0.148        |
| Neutral detergent fiber                                  | 0.63                      | 0.63 | 0.64 | 0.64 | 0.012 | 0.675        |
| Acid detergent fiber                                     | 0.56                      | 0.58 | 0.60 | 0.58 | 0.015 | 0.628        |
| Ether extract  | 0.70                      | 0.73 | 0.71 | 0.69 | 0.010 | 0.559        |
| Non-fibrous carbohydrates                                | 0.84                      | 0.88 | 0.87 | 0.87 | 0.008 | 0.167        |
| Total carbohydrates                                      | 0.74                      | 0.76 | 0.76 | 0.76 | 0.007 | 0.355        |

SEM = standard error of means; L = significance for linear effect.

reduced DM intake, rumination, nutrient absorption, and water loss by evaporative mechanisms, mediated by changes in the endocrine profile, aimed at conserving body water and maintaining homeostasis (Al-Ramamneh et al., 2012; Santos et al., 2019).

Nutrient intake is one of the main factors influencing animal production efficiency (Araújo et al., 2019). It is known that small ruminants are animals adapted to arid and semiarid regions, in the present study used Santa Inês crossbred sheep, known for their productivity and resilience to challenges such as water and food scarcity, high temperatures, and high levels of radiation (Azambuja, González-garcía 2016), which certainly was a preponderant factor for the nutrient intake unchanged.

However, in studies with animals considered adapted to semiarid regions, there was a reduction in nutrient intake and digestibility due to water restriction conditions. When submitted to a water restriction of 50%, it was observed that Baluchi ewes had reduced DM intake by

39.75% and OM digestibility reduction by 2.72% (Vosooghi-Postindoz et al., 2018).

The lack of nutrient digestibility changes with water treatments, especially the NDF, denotes a positive aspect since this fraction comprises the majority of dietary DM in the present experiment, being considered the main energy source for ruminants, which suggests that the animals activated adaptive mechanisms that allowed maintaining the intake and efficiency of the digestive process.

Lower water intake may be associated with reduced rumen motility and decreased passage rate with a consequent increase in the digestibility of structural carbohydrates (Silanikove, 1992). However, the fact that the most severe water restriction treatment did not affect the intake and digestibility of fibrous carbohydrates may denote that, possibly the retention time did not change in the rumen, once the intake of DM and fibrous fractions are similar and there was no change in the digestibility of these.

The temperature and humidity index is of significant importance in identifying the degree of environmental comfort and thermal stress to which the animal is subjected. The findings of the present study highlight the temporal dynamics and intensity fluctuations of the THI index throughout the designated time intervals. The behavior of peak THI values was as expected, indicating that between 3 pm and 6 pm, the animals were exposed to severe stress, while at other times, the animals experienced moderate stress (Fuquay, 1981).

Although the ewes were under water restriction, and heat stress conditions, evidenced by the THI values, there were no losses in nutrient intake and digestibility, as well as in water and nitrogen balance and performance (explained in more detail below), these results are considered positive since these reflect the adaptability of Santa Inês crossbred ewes.

The decreasing linear effect observed on free water and total water intake was expected, as these result from the supply of 80%, 60%, and 40% of free water about control, reducing approximately 19.39; 39.94%, and 59.44% in free water intake.

Bearing the total intake of water results from the amount between free water intake and water intake by food, the reduction of water supply is directly reflected in the total water intake, where a decrease of 13.11 was observed; 22.72% and 34.42% in total water intake, values close to the proposed reduction in treatments. The decrease in total urinary production, water excreted via urine, excretion of water via feces, total water losses, and absorbed water are closely linked.

The lower the level of water excretion via urine, the higher the concentration of blood electrolytes, and with this, there are increased water requirements. Water in the blood plasma conducts toxic and excess substances or metabolic residues in the body to be excreted (Wakchaure et al., 2015). Given that the intake of nutrients, including N,

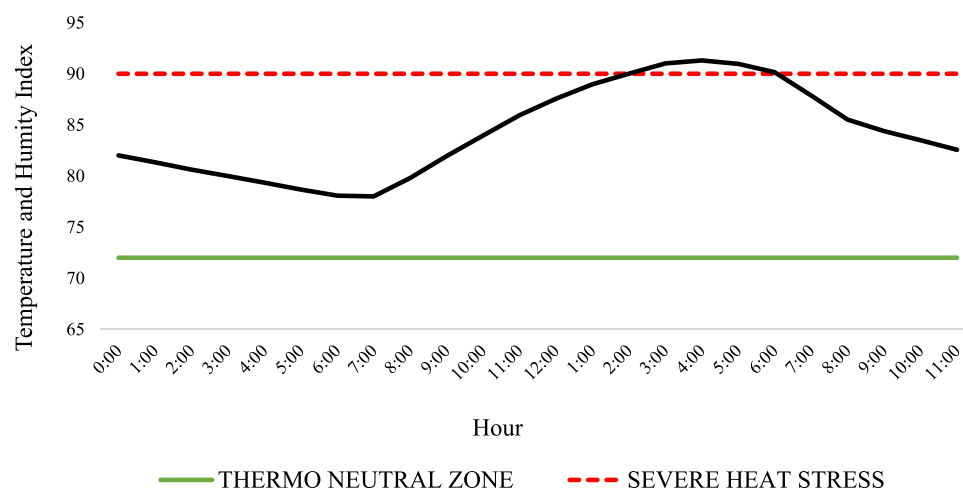


Fig. 1. Temperature and Humidity Index at the different times of the day.



**Table 5**

Water and nitrogen balance in crossbred Santa Inês ewes submitted to a reduction in the water supply.

| Variables (g day <sup>-1</sup> )     | Water offering levels (%) |         |         |         | SEM   | p-value<br>L         |
|--------------------------------------|---------------------------|---------|---------|---------|-------|----------------------|
|                                      | 100                       | 80      | 60      | 40      |       |                      |
| Free water intake                    | 1538.08                   | 1239.85 | 923.77  | 623.90  | 68.58 | < 0.001 <sup>1</sup> |
| Water intake via food                | 1723.53                   | 1594.28 | 1596.81 | 1514.94 | 56.80 | 0.226                |
| Total water intake                   | 3261.61                   | 2834.13 | 2520.56 | 2138.84 | 104.9 | < 0.001 <sup>2</sup> |
| Total urine output                   | 1102.32                   | 711.07  | 580.18  | 415.39  | 91.42 | 0.008 <sup>3</sup>   |
| Water excreted via urine             | 993.34                    | 623.07  | 512.38  | 345.99  | 82.83 | 0.006 <sup>4</sup>   |
| Production of feces (natural matter) | 940.71                    | 761.61  | 756.43  | 611.46  | 43.92 | 0.091                |
| Water excretion via feces            | 602.86                    | 477.80  | 468.57  | 350.68  | 31.05 | 0.006 <sup>5</sup>   |
| Total water losses                   | 1596.19                   | 1100.87 | 980.96  | 696.68  | 94.41 | < 0.001 <sup>6</sup> |
| Absorbed water                       | 2658.75                   | 2356.33 | 2051.99 | 1788.16 | 85.05 | < 0.001 <sup>7</sup> |
| Water balance                        | 1665.42                   | 1733.26 | 1539.61 | 1442.16 | 68.22 | 0.160                |
| Nitrogen intake                      | 23.89                     | 22.44   | 21.30   | 21.04   | 0.784 | 0.171                |
| Nitrogen in feces                    | 3.46                      | 2.57    | 2.23    | 1.96    | 0.193 | 0.002 <sup>8</sup>   |
| Nitrogen in urine                    | 2.32                      | 2.14    | 1.95    | 2.16    | 0.181 | 0.693                |
| Absorbed nitrogen                    | 20.43                     | 19.88   | 19.07   | 19.09   | 0.813 | 0.514                |
| Retained nitrogen                    | 18.11                     | 17.73   | 17.12   | 16.92   | 0.802 | 0.568                |
| Retained N/N absorbed                | 0.88                      | 0.88    | 0.89    | 0.87    | 0.010 | 0.989                |
| Retained N/N ingested                | 0.74                      | 0.78    | 0.80    | 0.80    | 0.014 | 0.122                |

SEM = standard error of means; L = significance for linear effect; (1) $\hat{y} = 15.293x + 10.882$ ,  $R^2 = 0.99$ ; (2)  $\hat{y} = 18.409x + 1400.13$ ,  $R^2 = 0.99$ ; (3) $\hat{y} = 10.958x - 64.848$ ,  $R^2 = 0.93$ ; (4) $\hat{y} = 10.263x - 99.749$ ,  $R^2 = 0.93$ ; (5) $\hat{y} = 3.829x + 206.959$ ,  $R^2 = 0.92$ ; (6)  $\hat{y} = 14.092x + 107.218$ ,  $R^2 = 0.94$ ; (7) $\hat{y} = 14.581x + 1193.16$ ,  $R^2 = 0.99$ ; (8) $\hat{y} = 0.024x + 0.855$ ,  $R^2 = 0.92$ .

**Table 6**

Performance of crossbred Santa Inês ewes submitted to the reduction in the water supply.

| Variables  | Water supply levels (%) |        |        |        | SEM    | P-value<br>L |
|--|-------------------------|--------|--------|--------|--------|--------------|
|  | 100                     | 80     | 60     | 40     |        |              |
| Initial weight (kg)                              | 29.19                   | 31.82  | 29.10  | 30.69  | 1.060  | 0.848        |
| Final weight (kg)                                | 35.73                   | 38.62  | 34.23  | 37.37  | 1.113  | 0.871        |
| Total weight gain (kg)                           | 6.54                    | 6.80   | 5.13   | 6.69   | 0.412  | 0.959        |
| Average daily weight gain (g day <sup>-1</sup> ) | 116.82                  | 121.43 | 91.67  | 119.39 | 7.352  | 0.960        |
| Weight gain (g kg <sup>-0.75</sup> )             | 543.12                  | 536.75 | 423.07 | 519.09 | 37.872 | 0.785        |

SEM: Standard error of the means; L = significance for a linear effect.

was not affected by the water supply, an increase in water intake would be necessary for the excretion of the metabolites generated from renal water resorption (Correia, 2015).

The results obtained agree with Vosooghi-postindoz et al. (2018), who observed a reduction of 84.97% in urine production when they submitted Baluchi lambs to 50% water restriction. However, a reduction of 52.84% in feces production (DM) was recorded.

The authors related what happened with lower dry matter intake and higher water absorption, resulting in feces with lower moisture content, which differs from the results of the present study for feces production. In the present study, feces production did not decrease and was expressed in natural matter. Nejad et al. (2017) also observed a reduction in urine production (volume) in 16.48% in 2 h of water restriction after feeding and 20.24% in 3 h of restriction in Corriedale ewes.

Conditions of low water intake can lead to hemoconcentration, with the reduction of blood flow in the kidneys, the resorption of water from the nephrons is activated at a higher level (Casamassima et al., 2008). This is directly linked to the presence of minerals such as sodium and potassium. In the Henle Loop, there is resorption of water and Na, by osmosis and co-transporter dependent on Na, K, and Cl. While in the distal tubule, there is resorption of water and Na and excretion of K (Reece, 2008).

The reduction of water excretion via feces is a direct effect of the reduction of water intake, with this the rate of passage through the digestive tract can be reduced, leading to an increase in the DM content of the feces – explained further below (Silanikove, 1996).

The observed results can be explained by the arguments described above by adding the total water losses that have been reduced, as well as the absorbed water. These adaptations were probably the way to keep the electrolyte balance in the body stable, this premise can be confirmed because the water balance has not been affected by the water supply.

Nintake responses are probably due to CP and N intake not being affected by water offerings. The reduction of nitrogen excretion via feces can be explained by the decrease in water excretion via feces.

The reduction of water excretion via feces resulted in feces with a higher DM content of 35.91; 37.26; 38.06%, and 42.65% for offers of 100, 80 60%, and 40% respectively. This resource may have been used to compensate for the reduction in water supply, since with reductions in water intake, the passage rate may decrease as previously mentioned, which leads to better utilization of nutrients, greater absorption, and efficiency in the use of these, which explains the reduction in the amount of N present in feces.

The results are supported by those found by Nejad et al. (2014), in which Corriedale ewes exposed to 2 and 3 h of water restriction, observed a reduction in fecal nitrogen excretion. According to the authors, the lower excretion of fecal nitrogen reflects the increase in the digestibility of CP (a fact not observed in the present study), resulting in greater nitrogen retention, which confers a positive nitrogen balance.

In the present experiment, there was no increase in the digestibility of CP, but rather the maintenance of intake and similar digestibility between treatments, this fact points out that the two theories together explain the reduction of N excreted in feces.

The fact that the nitrogen balance was not affected by the water offers suggests that there was synchrony between the availability of energy and protein for the rumen microorganisms (Araújo et al., 2019) and that possibly the animals did not need to mobilize the body protein reserves to meet nutritional needs. Meeting the nutritional needs would explain the body weight gain.

Moderate water restriction primarily affects body water loss, rather than mass loss with no deleterious effects on production or growth (Qinisa et al., 2011). In contrast, Jaber et al. (2011), observed that Awassi ewes subjected to water restriction and supplemented with vitamin C experienced weight loss between 22% and 26.2%.

In the present study, the average daily weight gain was similar among the four groups, which had satisfactory average body weight gain between 91 and 122 g day<sup>-1</sup>. Maintaining the performance of the ewes was a direct benefit of unchanged nutrient intake, a possible improvement in nutrient use efficiency, and an indirect benefit of reduced

passage rate.

Still, about this variable, it is important to highlight the characterization of the animals, mentioned previously, and the fact these are not typical meat animals, which corroborated the results observed, that despite the evident challenge imposed by thermal stress, and by water restriction reached efficient production rates, once again demonstrating the adaptability of crossbred Santa Inês ewes.

## 5. Conclusion

The water supply of up to 40% of ad libitum intake did not promote negative effects on the intake and apparent digestibility of nutrients, nitrogen balance, and water balance, as well as the performance of crossbred Santa Inês sheep under feedlot conditions. In addition, the study of other levels of hydric supply in animals at different physiological stages, age, and sex, as well as the measurement of more variables are necessary since this information can contribute to the clarification of the physiology and adaptation to hydric restriction in the complete production cycle in beef sheep breeding and will guide the real requirements in each phase for the Santa Inês breed.

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## CRediT authorship contribution statement

**Patrícia Rodrigues de Lima:** Formal analysis, Methodology, Investigation, Writing - original draft. **Cleyton de Almeida Araújo:** Formal analysis, Methodology, Investigation. **Vanúzia Gonçalves de Menezes:** Formal analysis, Methodology, Investigation. **Fleming Sena Campos:** Conceptualization, Supervision, Resources. **Neila Lidiany Ribeiro:** Implementation of the computer code and supporting algorithms. **Gherman Garcia Leal de Araújo:** Conceptualization, Methodology, Writing - original draft, Writing - review & editing, Project administration, Supervision, Resources, Visualization. **Daniel Ribeiro Menezes:** Conceptualization, Supervision, Writing - review & editing. **Maria Helena Tavares de Matos:** Conceptualization, Supervision, Resources. **Mário Adriano Ávila Queiroz:** Conceptualization, Supervision, Resources. **Edson Mauro Santos:** Conceptualization, Supervision.

## Declaration of Competing Interest

We wish to confirm that there are no known conflicts of interest associated with this publication, and there has been no significant financial support for this work that could have influenced its outcome.

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