

# Exploring the potential of phenolic and antioxidant compounds identified and quantified of *Caesalpinia coriaria* fruits and their impacts on lambs' performance and health

Pedro Enrique Hernández Ruiz<sup>a</sup>, Mona Mohamed Mohamed Yasseen Elghandour<sup>a\*</sup>, José Luis Ponce-Covarrubias<sup>b</sup>, Blanca Celia Pineda Burgos<sup>b</sup>, Moyosore Joseph Adegbeye<sup>c</sup>, Miguel Mellado<sup>d</sup>, Mohamed Z.M. Salem<sup>e</sup>, Abdelfattah Z.M. Salem<sup>a\*</sup>

## Abstract

This study aimed to assess the impact of the identified phenolic, flavonoid, and fat-soluble vitamin compounds found in *Caesalpinia coriaria* fruits (EC) aqueous extract on growth performance, helminth parasitic load, feeding behavior, and physiological variables in lambs. The fruits were extracted using water, and three doses of the extract as no extract (EC0 mL/lamb/day), 30 mL of aqueous extract of *Caesalpinia coriaria* fruits (EC30 mL/lamb/day), or 60 mL of aqueous extract of *Caesalpinia coriaria* fruits (EC60 mL/lamb/day) were orally administered to 24 growing crossbred lambs weighing 18-20 kg for 60 days. The HPLC analysis revealed that ferulic acid, ellagic acid, syringic acid, quercetin, and hesperidin were the predominant compounds in *Caesalpinia coriaria* fruits. Additionally, water-soluble vitamin compounds such as thiamine, pantothenic acid, and niacin were detected in the *Caesalpinia coriaria* fruit extract. The dietary administration of EC improved ( $P < 0.05$ ) average daily gain and feed and water intake. EC30 lambs spent more time ( $P < 0.03$ ) eating and less time ruminating. The aqueous extract also showed a dose-dependent reduction ( $P = 0.048$ ) in fecal parasite egg count, with a steady decrease ( $P = 0.0001$ ) as the extract was applied. EC0 lambs exhibited the highest ( $P < 0.0001$ ) respiratory frequency and EC30 the lowest. A strong positive correlation was identified between rectal and abdomen temperature in the morning and afternoon measurements. The presence of phenolic and antioxidants in the aqueous extract of *C. coriaria*, up to 60 mL, demonstrated beneficial effects, including improved productivity, anti-parasitic activity, and mitigation of body temperature. Thus, farmers without access to synthetic drugs can use this plant extract to treat their animals during fattening to reduce gastrointestinal parasites, improve growth rate, and reduce drug residue in animal products. Further studies need to be carried out on the individual components of *Caesalpinia coriaria* fruits to ascertain their effect on body temperature/physiology and to know if the effects are individualistic or synergistic.

**Keywords:** Antioxidant; Phenolic; Flavonoid; *Caesalpinia coriaria*; Growth performance; Lambs

<sup>a</sup>Facultad de Medicina Veterinaria y Zootecnia, Universidad Autónoma del Estado de México, Estado de México, México

<sup>b</sup>Escuela Superior de Medicina Veterinaria y Zootecnia No. 3, Universidad Autónoma de Guerrero (UAGro), Tépam de Galeana, Guerrero, México

<sup>c</sup>Department of Animal Production and Health, University of Africa, Toru-Orua, Sagabama, Bayelsa, Nigeria

<sup>d</sup>Autonomous Agrarian University Antonio Narro, Department of Animal Nutrition, Saltillo, Mexico

<sup>e</sup>Forestry and Wood Technology Department, Faculty of Agriculture (EL-Shatby), Alexandria University, Alexandria, Egypt

### \* Corresponding Authors:

mmohamede@uaemex.mx (M.M.Y. Elghandour)  
salem@uaemex.mx (A.Z.M. Salem)

DOI: 10.2478/ebtj-2024-000

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## Introduction

Trees, such as those bearing leaves and fruits, are crucial sources of animal feedstuff. However, the global campaign against using medically necessary antibiotics in livestock nutrition necessitates researchers and farmers to find alternatives for managing livestock for productive performance and health. This situation has led livestock researchers to explore nutraceuticals, such as herbs or plants, for non-nutritional purposes such as immunity, anthelmintic activity, antibiotics, stress alleviation, and rumen manipulation. It has been proposed that local plant resources will play a significant role in agricultural productivity and resource-use efficiency. Jack et al. [1] and Jimoh et al. [2] have attested to the positive impact of local herbal supplements on growth promotion, reduction in fecal egg count, optimization of ruminal fermentation, and heat stress in small ruminants. However, little research has been done on the impact of *Caesalpinia coriaria* fruits, a leguminous species of the tropics in Mexico.

This plant contains bioactive ingredients such as tannins, gallic acid, and flavonoids. It has been used in mitigating methane *in vitro* [3], as an anthelmintic *in vitro* [4], as well

as an alternative feed ingredient in goat nutrition [5]. The positive effects of these herbs, either as fruits, have been attributed to their plant secondary metabolites, such as tannins, or their nutrient content. However, no study has been conducted on the impact of this leguminous plant on gastrointestinal helminths, performance, feeding behavior, and its influence on body temperature. Investigating the influence of this plant on endoparasites can help increase the benefits that animals can derive from nutrient intake and reduce farmers' dependence on synthetics [6]. Furthermore, investigating the impact of plants on body site temperature can provide a physiological view of their impact on animal health. Body temperature status can indicate heat stress or disease, which can affect growth rate, feed intake, digestion, and metabolism [7,8].

However, evaluating the biological and physiological impact of compounds from *Caesalpinia coriaria* fruit extract on the growth performance, feeding behavior, and body characteristics of growing lambs is pertinent.

## Materials and Methods

### 2.1. Preparation and characterization of *Caesalpinia coriaria* fruits

#### 2.1.1. Preparation of extract from fruits

The fruits of the Cascalote (*Caesalpinia coriaria* (Jacq) Willd) were collected manually in the municipality of Tlapehuala, belonging to the Tierra Caliente region of Guerrero, Mexico. Fruits of *Caesalpinia coriaria* were dried at room temperature and subsequently ground in a hammer mill with a particle size of 1 mm.

To prepare the aqueous extract, dried fruits of the *Caesalpinia coriaria* were collected and ground in the hammer mill to 1 mm. 1 kg of *Caesalpinia coriaria* ground fruits were soaked in 2.5 L of distilled water. The mixture was left to stand for 72 h [34], and after that, the contents were filtered to obtain the aqueous extract. Each week of the experiment, the extract was prepared to be offered orally to the lambs, and the extract was refrigerated. The calculated extract percentage was calculated as follows: Extract percent = [extract amount (g)/dry fruit sample amount (g)] × 100. The extract percentage was 17%.

#### 2.1.2. HPLC for chemical characterization of the exported extract

##### Phenolic and flavonoid compounds

The phenolic and flavonoid compounds from the aqueous extracts of *Caesalpinia* were identified by Agilent ChemStation (HPLC- (Agilent, Santa Clara, CA, USA), which is composed of a quaternary pump, UV/V detector, and C18 column (125 mm × 4.60 mm, 5 µm particle size). Chromatograms were obtained and analyzed using HPLC. Phenolic compounds were separated using a mobile gradient phase of Solvent A: water/acetonitrile/glacial acetic acid (980/20/5 µl, v/v/v, pH 2.68) and solvent B: acetonitrile/glacial acetic acid (1000/5 µl, v/v). The apparatus was operated at 30°C with a 1 mL/min flow rate and detected at 325 nm [9,10]. The gradient profile was 4% B to 33% B linearly

in 90 min, a linear increase to 100% B at 95 min, followed by 5 min isocratic, a return to 4% B at 105 min, and 5 min isocratic to re-equilibrate [11]. All chemical standards (high-performance liquid chromatography (HPLC grade) were from Sigma-Aldrich (St. Louis, MO, USA). The standard phenolic compounds catechol, caffeic acid, ferulic acid, gallic acid, syringic acid, cinnamic acid, and ellagic acid, as well as the standard flavonoid compounds, chrysoeriol, rutin, naringin, quercetin, kaempferol, luteolin, hesperidin, and catechin, were used.

##### Vitamins

Water-soluble vitamins were identified in *Caesalpinia coriaria* fruits extract as follows: powdered samples (0.5 g) were extracted by ultrasound (for 30 min at 25°C) using methanol/water (80%, v/v) and then filtered.

Analysis of vitamins was performed by HPLC-(Agilent 1100), which composed of two LC- pumps, and a UV/Vis detector. Chromatograms were obtained and analyzed using the Agilent ChemStation.

For water soluble vitamins; Chromatographic separation was achieved on column (Agilent ZORBAX C18; 250 × 4.6 mm i.d., 5 µm) through the isocratic delivery mobile phase (A/B 33/67; A: MeOH, B: 0.023 M H<sub>3</sub>PO<sub>4</sub>, pH = 3.54) at a flow rate of 1.0 mL/min. At room temperature, ultraviolet (UV) absorbance was recorded at 270 nm [35].

### 2.2. Biological and physiological evaluation of the exported extract

Twenty-four growing crossbred male lambs (18 to 20 kg fasted live weight) were individually allocated to wood pens for 60 days during the experiment. Lambs were divided into three experimental treatments (8 lambs per group) the control group was fed with the free access balanced diet (EC0), group 2 was fed the balanced diet and offered 30 mL of aqueous extract of *Caesalpinia coriaria* fruits (EC30), and group 3 fed the balanced diet and offered 60 mL of aqueous extract of *Caesalpinia coriaria* fruits (EC60). Lambs were adapted to the basal balanced diet for 15 days before the experimental period. Ingredients of the lambs' diet were (% of the diet): 50, 10, 20, 19, and 1% of mixed vitamins and minerals, in which the chemical composition (% on DM basis) was 98.12 organic matter, 16 crude protein, 2.6 ether extract, 38.0 neutral detergent fiber, and 15.9 acid detergent fiber. The extract was offered orally before the morning feeding at 7:00 a.m., and water was provided in the morning and afternoon (3 liters per shift). Lambs were weighed with a CRANE SCALE® digital hanging weight scale on days 1, 15, 30, 45, and 60 of the experiment using a harness that was adapted to each lamb to avoid stress.

Lambs were fed the balanced diet at 7:00 am and 4:00 pm: according to feed consumption, 100 grams more were added to what was consumed per day, and the surplus feed was measured in each shift. Regarding water consumption, 3 liters of water were offered in the morning and 3 liters in the afternoon. The control group was not provided any extract, group 2 was offered a total of 30 milliliters of aqueous extract per lamb from the group, a total amount of 240 mL/group, and group 3 was offered a total of 60 milliliters of aqueous extract per lamb from the

group, a total amount of 480 mL/group before the morning feed during the 60 days of the experiment.

Feeding behavior parameters (time spent eating, ruminating, resting, and other activities such as water intake and erratic movements) were measured on two consecutive days at days 1 and 60 of the experimental period for each lamb using Hi Look® brand video cameras. These were installed strategically to observe the lambs' behavior during the first 48 hours after the experiment began (day 1) and during the last 48 hours (day 60) of the experiment. The behavior of the 8 lambs in each group was recorded; the videos were saved on a hard disk for later review.

Five lambs were collected at random from each group to obtain fecal samples by anal stimulation. Subsequently, the eggs per gram of feces (EPG) was evaluated in the laboratory of the faculty of veterinary medicine at period interval of day 0, day 10, day 30, and day 60 after the beginning of the study. The McMaster method was used to quantify EPG. Adapting the methodology of Thienpont et al. [15] and Madeira de Carvalho [16], two grams of feces were added to 28 mL of 25% saturated sodium chloride solution and homogenized with a glass rod. Subsequently, the solution was filtered through a tea strainer over a beaker. Then, the two compartments of the McMaster chamber were filled with a Pasteur pipette, and the eggs were counted inside the limits of the two grids. Considering the chamber volume (0.30 mL), the number of eggs counted was multiplied by the correction factor of 50.

### 2.3. Climatic variables

The local climate data was obtained through the recording of ambient temperature (AT; °C) and relative humidity (RH; %), which were measured 24 hours a day, these were obtained through electronic sensors connected to the station of the national meteorological system [36], Tecpan number 12233, latitude 17°15'00" N, longitude 100°34'07" W, altitude 262.0 MSNM in the municipality of Tecpan de Galeana, state of Guerrero, Mexico.

The heat stress evaluation of the experiment was carried out through the following indices:

TA and RH index (THI) temperature and humidity index: it relates climatic factors; it was calculated with the Kibler equation [37]:

$$\text{THI} = 1.8 \text{ AT} - (1 - \text{RH}) (\text{AT} - 14.3) + 32$$

where AT is the average temperature (°C), and RH is the relative humidity. The THI was calculated according to the temperature and RH records, obtaining daily averages, considering that values between 71 and 79 units indicate an alert condition, following what was indicated by the World Meteorological Organization [38] (Figure 8).

### 2.4. Physiological variables and skin temperatures

The physiological variables evaluated were: respiratory frequency (RF) rectal temperature in Celsius degrees (RT). RF was

measured by counting the number of movements of the right paralumbar fossa; HR was measured with a single bellstethoscope (3M™ Littmann®, Penlight; Shanghai, China), counting the number of repetitions per minute; RT was measured with a digital thermometer of veterinary use (Delta Trak®, Pleasanton, CA, USA). In addition, the following skin temperatures were measured (using a Fluke 59E®).

The temperatures of different anatomical parts of the lambs were taken, including the head, neck, scapula, jar (end of the last rib), abdomen, rump, leg, dorsum, and anus, for the skin temperatures (ST), total body photos of each ewe were taken from a distance of 30 cm. The measurements were taken every third day of the 60 days of the experiment four times a day before the morning feeding at 6:00 am, after the feeding at 11:00 am, at 2:00 pm before the afternoon feeding, and at 5:00 pm after the afternoon feeding.

### 2.5. Statistical analysis

Data from the biological and physiological evaluation of the exported extract of *Caesalpinia coriaria* fruits (feed and water intake, growth performance, gastrointestinal egg counts, and physiological constants) were analyzed using the PROC MIXED procedure of SAS [39]. A completely randomized design with repeated measures of two experimental periods was used. Two experimental periods (day 1 and day 60) per treatment were considered for each parameter during the 60 days of experiments. For each treatment (i.e., 0, 30, and 60 mL of extract per lamb per day), 8 growing lambs were used, and each animal was used as an experimental unit.

The following statistical equation was used:

$$Y_{ij} = \mu + T_i + P_j + E_{ijk}$$

where  $Y_{ij}$  is every observation of the  $j$ th lamb assigned to the  $i$ th treatment,  $T_i$  is the treatment effect,  $P_j$  is the period effect and  $E_{ijk}$  is the residual error. Comparisons of results were performed using Tukey's test at  $P < 0.05$ ). The Tukey test was used for the multiple comparisons of means, and polynomial (linear and quadratic) contrasts were used to examine the extract dose responses to increasing levels of *Caesalpinia coriaria* fruit extract in the diet. Significance was declared at  $P < 0.05$ , and a trend at  $P \leq 0.10$ . The Pearson correlation was conducted among all the physiological constants.

## Results

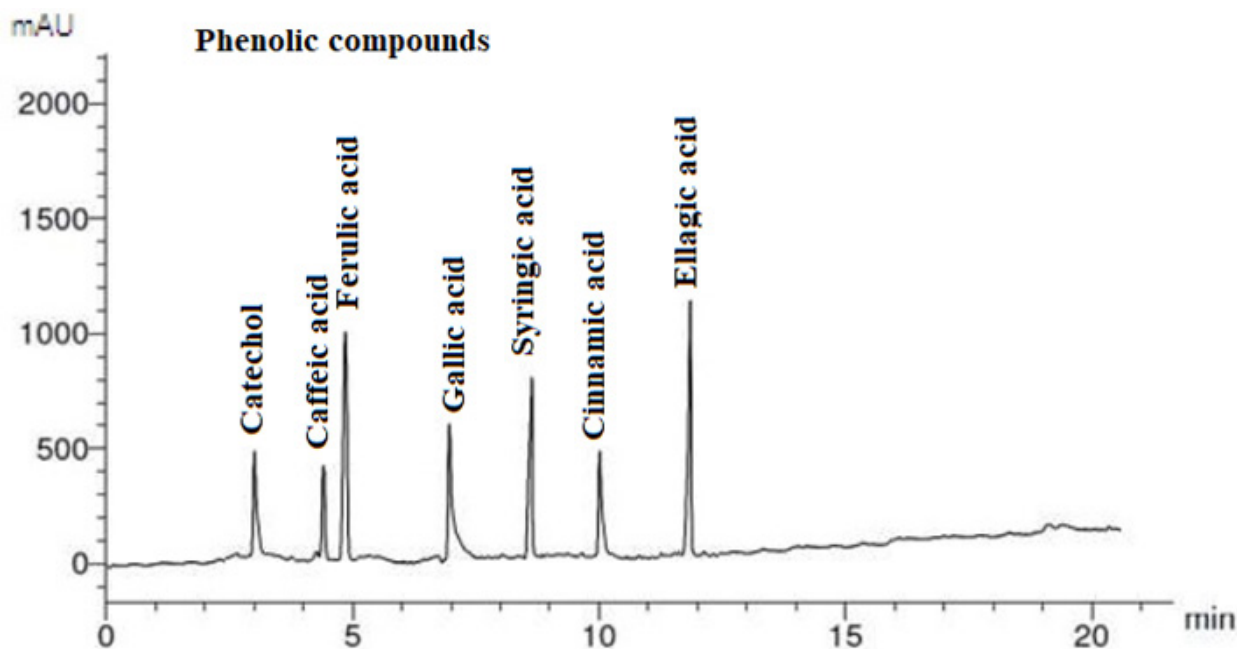
### 3.1. HPLC for extract characterization

Based on the HPLC analysis of *Caesalpinia coriaria* fruits, ferulic acid, ellagic acid, and syringic acid, as phenolic compounds, had the highest concentrations, while caffeic acid, catechol, and cinnamic acid had the lowest concentrations. Flavonoid compounds such as quercetin and hesperidin also had higher concentrations than kaempferol, luteolin, and naringin in the *Caesalpinia coriaria* fruit extract (Table 1 and Table 2; Figures

1 and 2). The phenolic compound showed that ellagic acid, followed by ferulic acid, and then syringic acid with 16.89, 13.25, and 10.41  $\mu\text{g}/\text{mL}$ , respectively. Furthermore, quercetin, hesperidin, rutin, and chrysoeriol were the highest concentrating flavonoid compounds with 16.12, 14.88, 7.69 and 7.14  $\mu\text{g}/\text{mL}$ , respectively. Water-soluble vitamin compounds were also detected and analyzed by HPLC in the *Caesalpinia coriaria* fruit extract, and significant concentrations were detected for vitamin B1 (thiamine), vitamin B5 (pantothenic acid), and vitamin B3 (niacin) (Table 4 and Figure 4). Vitamin B was the highest concentrating water soluble vitamins. Vitamin B3 (Niacin), Vitamin B5 (Pantothenic acid), and Vitamin B1 (thiamine) were 28.36, 18.44, and 13.16  $\mu\text{g}/\text{mL}$ , respectively.

## 2.2. Biological and physiological response to the characterized extract feeding

### Biological response



**Figure 1.** HPLC chromatograms of phenolic compounds from aqueous extract of *Caesalpinia coriaria* Jacq. Willd. fruits

The aqueous extract linearly ( $P < 0.0001$ ) improved the average daily gain of lambs. It further showed that the lambs had the highest daily gain on days 30 and 45 ( $P < 0.0001$ ). Feed and water intake linearly increased ( $P < 0.001$ ) throughout the experiment, with the highest values on day 45 (Table 5). The feed and water intake followed the same pattern throughout the experiment (Figure 6).

Table 6 shows that lambs given 30 mL of aqueous extract (EC30) spent the most time eating ( $P < 0.03$ ) and the least time ruminating, while lambs given no extract spent the shortest time eating and more time ruminating. Furthermore, EC60 lambs spent more time resting. EPG linearly decreased ( $P = 0.048$ ) with increasing dietary aqueous extract. Lambs giv-

en no extract had the highest EPG, while those given extract (EC60) had the least EPG. EPG decreased ( $P = 0.0001$ ) with each day of applying the extract. Day 3 had the highest EPG, while day 0 had the lowest (Table 7).

### Physiological response

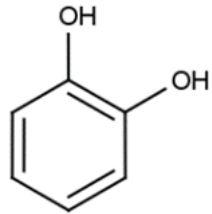
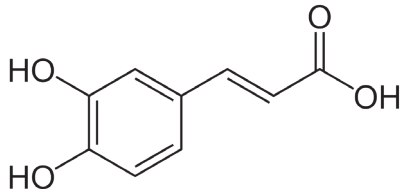
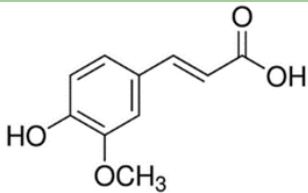
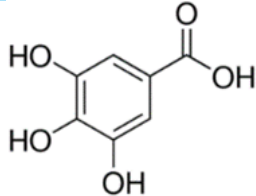
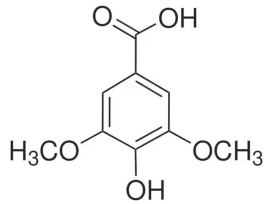
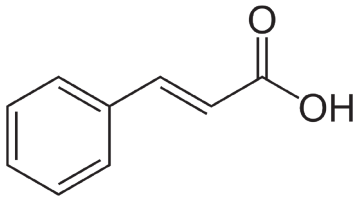
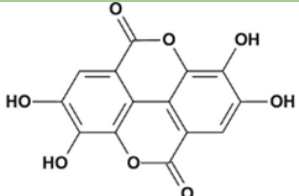
EC60 lambs had lower ( $P < 0.05$ ) head, neck, scapula, flank, abdomen, rump, leg, right testicle, and anus temperatures. All respiratory frequencies and other body site temperatures measured increased ( $P < 0.05$ ) after the first meal (AFM) compared to before the first meal (BFM), were lower in BFM than after the second meal (ASM). The rectal temperature had the lowest excursion range, while the rump had the highest (Table 8).

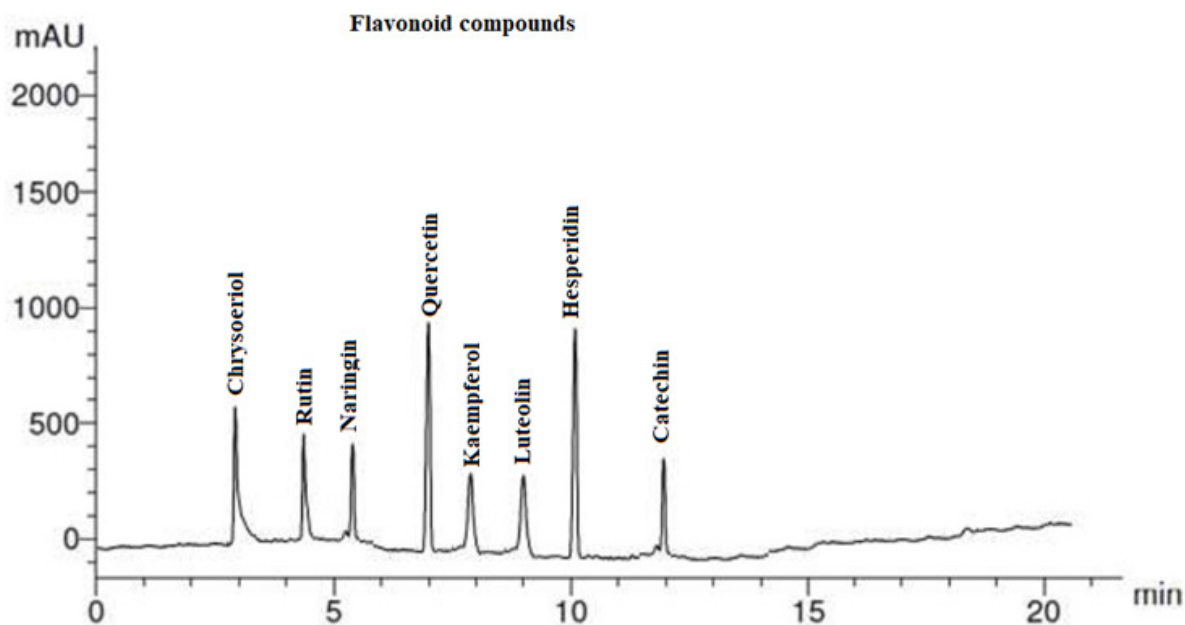
Figure 7 (a to k) shows the daily temperature rhythm of respiratory frequency, head, neck, scapula, flank, abdomen, haunch, leg, right testicle, and anus as influenced by the extract. Figure 2 shows that all lambs, regardless of their group,

had increased ( $P < 0.001$ ) RE, with the EC30 lambs having the lowest value. In other graphs (2c-k), there were rhythms. Of particular interest are those showing a general dip in the head, neck, scapula, flank, rump, and leg temperature on day 7. Similarly, on day 22, another depression in the temperature of the flank, abdomen, rump, leg, right testicles, neck, scapula, head, and anus was observed. It was also noted that in the 30-day trend, lambs with no extract had the highest temperature at these body points (head, neck, scapula, flank, abdomen, leg, right testicles, and anus). Conversely, EC60-supplemented lambs had the lowest head, neck, abdomen, right testicle, and anus temperatures.

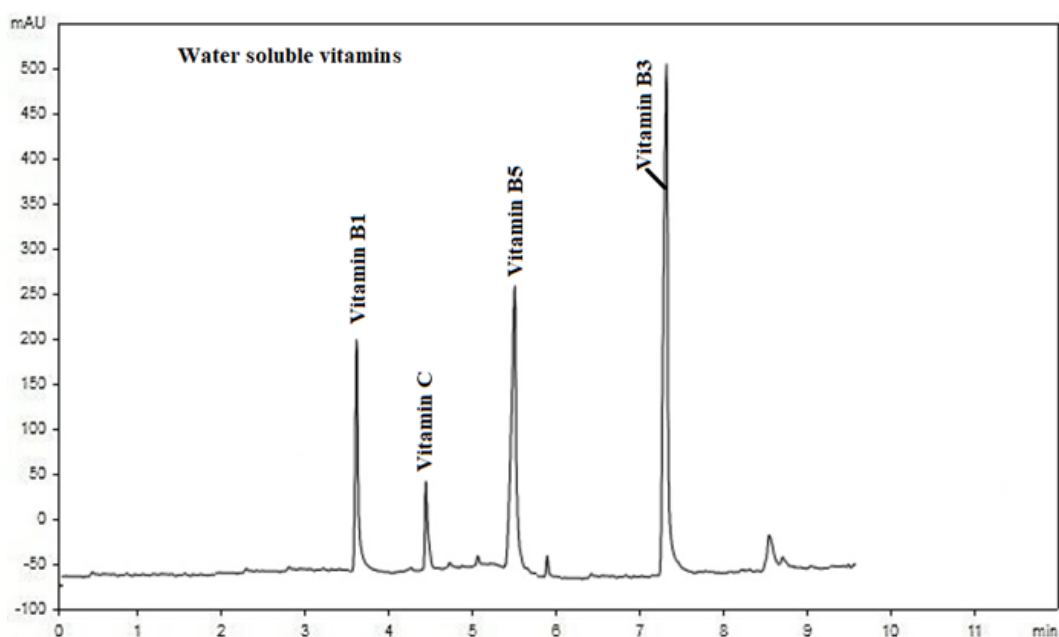
Values of the Pearson correlation for the variables during the

**Table 1.** Concentration of phenolic compounds from water extract of *Caesalpinia coriaria* fruits

| RT (min) | Phenolic compound | Concentration ( $\mu\text{g/mL}$ ) | Chemical structure  |
|----------|-------------------|------------------------------------|---|
| 3.0      | Catechol          | 6.12                               |    |
| 4.4      | Caffeic acid      | 4.09                               |    |
| 5.0      | Ferulic acid      | 13.25                              |   |
| 7.0      | Gallic acid       | 8.36                               |  |
| 8.5      | Syringic acid     | 10.41                              |  |
| 10.0     | Cinnamic acid     | 5.16                               |  |
| 12.0     | Ellagic acid      | 16.89                              |  |



**Figure 2.** HPLC chromatograms of flavonoid compounds from aqueous extract of *Caesalpinia coriaria* Jacq. Willd. fruits

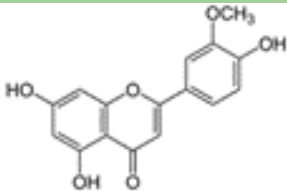
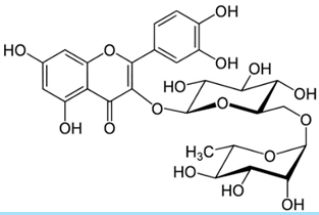
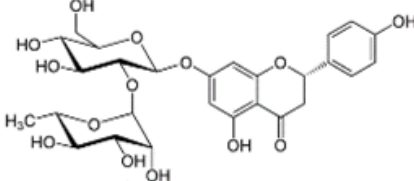
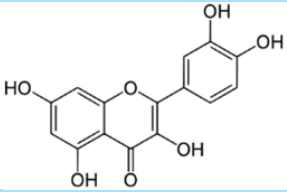
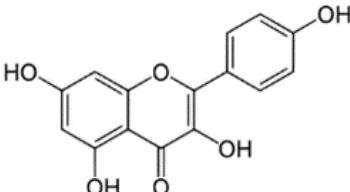
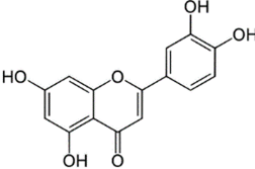
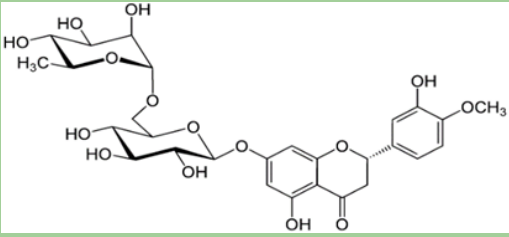
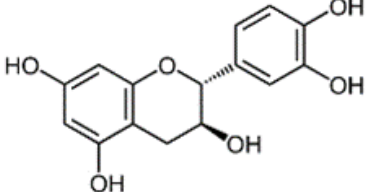


**Figure 4.** HPLC chromatograms of water-soluble vitamin compounds from aqueous extract of *Caesalpinia coriaria* Jacq. Willd. fruits

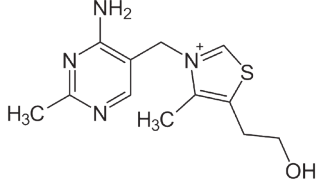
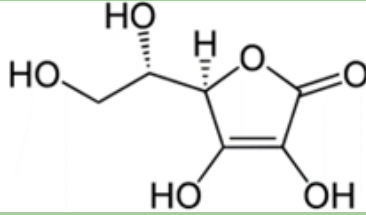
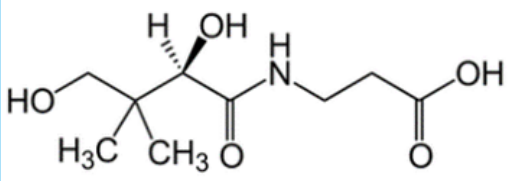
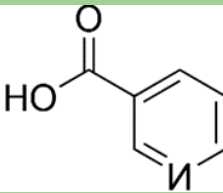
morning shows that RF was moderately correlated with rectal temperature. Surprisingly, we found a strong correlation between RF and neck temperature and a weak correlation between RF and anal temperature. Furthermore, the results showed that rectal temperature strongly correlated with abdomen temperature and had a weak to moderate correlation with temperature of other body parts (head, neck, scapula, flank, rump, leg, right testicle, and anus) (Table 9). During the afternoon, variables of the Pearson correlation presented

changes in the correlation coefficients, which differed from the morning. There was a strong correlation between RF and rectal, haunch, and anal temperatures. A similar pattern was observed between rectal temperature and abdomen, right testicles, and anus temperatures. This correlation can indicate the medium through which temperature is emitted from different body parts at various times of the day (Table 10).

**Table 2.** Concentration of flavonoid compounds from water extract of *Caesalpinia coriaria* fruits

| RT (min) | Phenolic compound | Concentration (µg/mL) | Chemical structure  |
|----------|-------------------|-----------------------|---|
| 3.0      | Chrysoeriol       | 7.14                  |    |
| 4.2      | Rutin             | 7.69                  |    |
| 5.2      | Naringin          | 5.34                  |    |
| 7.0      | Quercetin         | 16.12                 |  |
| 7.9      | Kaempferol        | 3.22                  |  |
| 9.0      | Luteolin          | 3.19                  |  |
| 10.0     | Hesperidin        | 14.88                 |   |
| 12.0     | Catechin          | 5.47                  |  |

**Table 4.** Concentration of water- soluble vitamin compounds from water extract of *Caesalpinia coriaria* fruits.

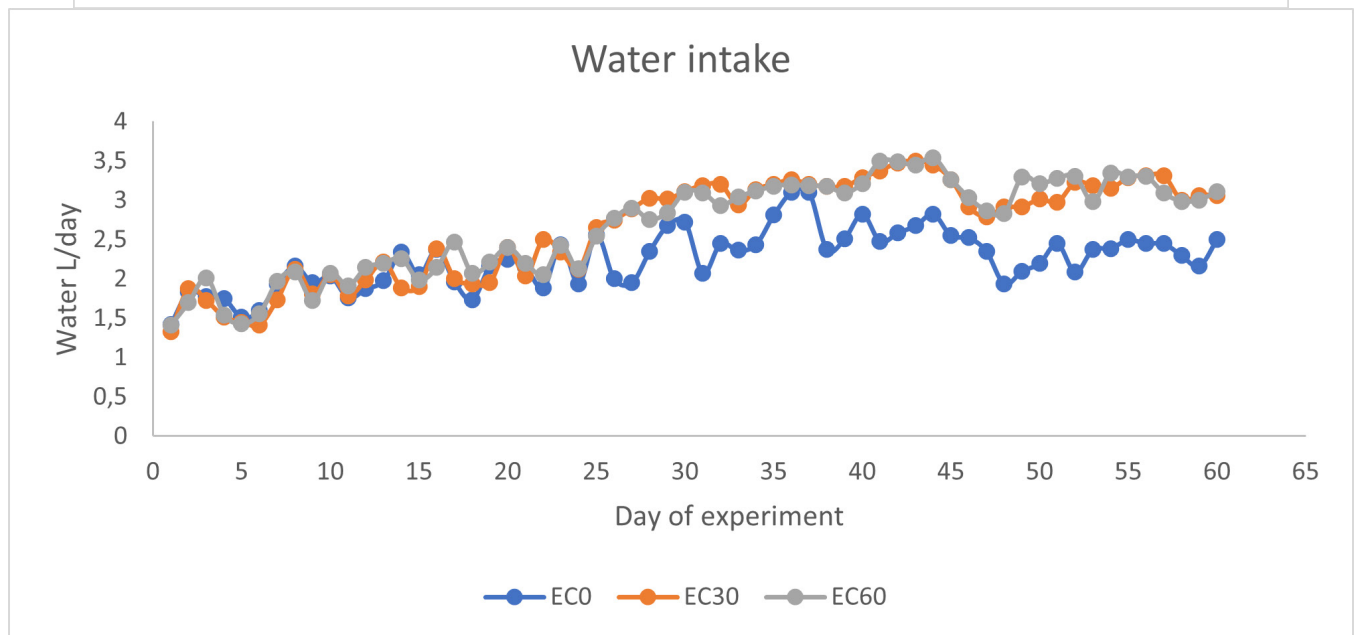
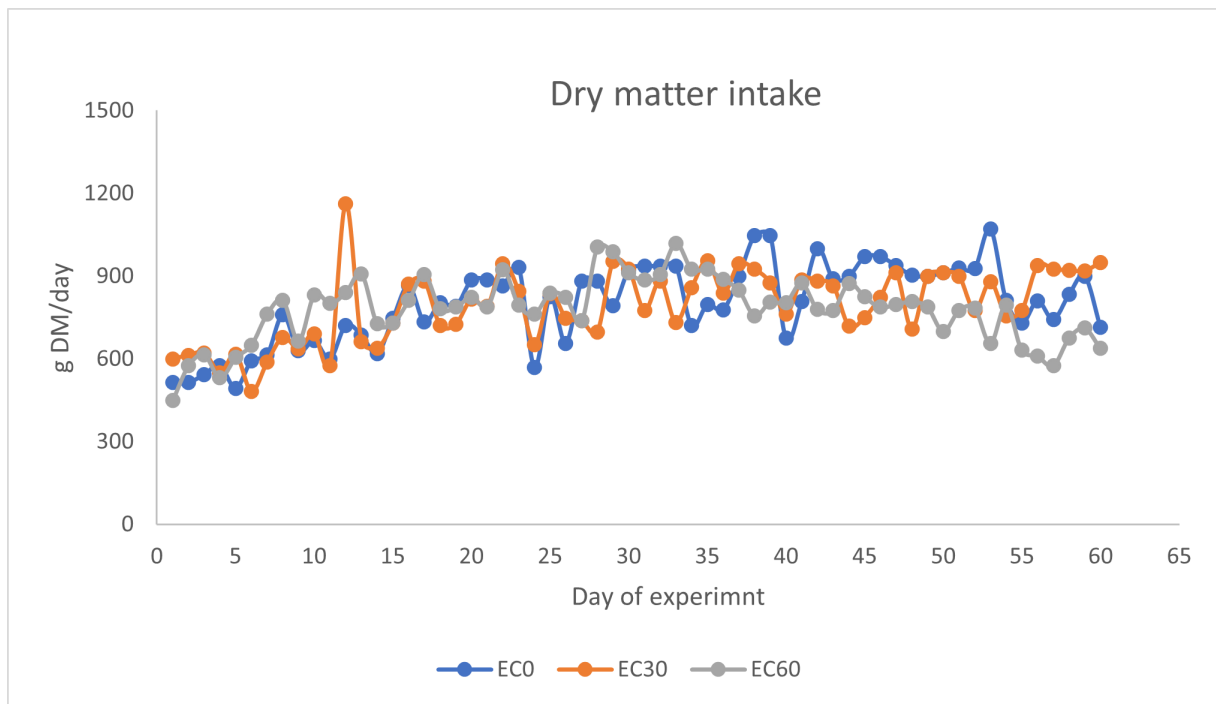
| RT (min)               | Phenolic compound             | Concentration ( $\mu\text{g/mL}$ ) | Chemical structure  |
|------------------------|-------------------------------|------------------------------------|---|
| Water-soluble vitamins |                               |                                    |   |
| 3.7                    | Vitamin B1 (thiamine)         | 13.16                              |    |
| 4.5                    | Vitamin C (ascorbic acid)     | 9.08                               |    |
| 5.6                    | vitamin B5 (Pantothenic acid) | 18.44                              |    |
| 7.5                    | Vitamin B3 (Niacin)           | 28.36                              |  |

## Discussion

Production and efficiency are crucial parameters in livestock production. Feed and water intake play significant roles in these processes. The increased feed and water intake in this study may be attributed to the increased growth rate or body weight of lambs, which also required increased nutritional demand. The simultaneous water and feed intake increase suggests a positive relationship between both parameters. This increased feed intake increased average daily gain (ADG), indicating that the lambs efficiently utilized the feed. The dose-dependent increase in ADG with increasing extract levels may be attributed to its ability to stimulate digestion, thereby improving feed intake and utilization. Plants rich in bioactive ingredients can help improve nutrient digestion and availability because their metabolites can enhance rumen activity and improve amino acid supply to the intestine [17]. However, at high concentrations, rumen microbe activities and digestive enzymes could be decreased and inhibited [18]. The dose-dependent increase in ADG suggests that EC60 did not hamper lamb growth. A comprehensive understanding of livestock feeding behavior is crucial for planning feeding strategies and

ensuring welfare [19]. The increased eating activity correlates well with increased feed intake, indicating that EC30 and EC60 extract stimulated lambs' appetite, leading them to spend more time eating. Another possibility is that increased feed digestion improved feed transit time or gut emptying, encouraging them to eat more. Studies have shown that rumination activity decreases as temperature rises, and a lower metabolic heat rate results in more rumination [20,21]. Therefore, in this study, the lower rumination time observed in EC30 lambs may be due to the higher body temperature among the lambs. It was also noted that lambs with less eating time spent more time ruminating, which reduced the metabolic heat generated during digestion. This suggests that ruminating became the preferred option as an alternative to eating time. Additionally, Itavo et al. [22] observed increased resting time with decreased rumination time. This implies that the increased rumination time observed in EC0 lambs could be the reason for less resting time. Gastrointestinal parasites contribute significantly to economic losses in livestock production as they compete for nutrients with livestock and can impair growth rate of animals. Hence, using herbs containing potent secondary metabolites can help reduce reliance on synthetic drugs in ruminant





**Figure 6.** Feed dry matter and water intake of growing lambs fed on diets with 0 (EC0), 30 (EC30) and 60 (EC60) mL of Casca-lote (*Caesalpinia coriaria* Jacq. Willd.) fruits aqueous extract during the experimental period of 60 days.

production [6].

Studies by Jesus-Martinez et al. [4] and García-Hernández et al. [23] demonstrated the *in vitro* and *in vivo* effects (78.6% goats under grazing conditions) of *Caesalpinia coriaria* against intestinal parasites. The dose-dependent decrease in EPG may be attributed to the presence of polyphenols such as tannins and methyl gallate in the plant [24]. Therefore, this reduction in egg count contributed to the observed growth-promoting effects or average daily gain in this study [25]. The decrease in EPG helped reduce competition for nutrients, which diverted the nutrients for growth promotion, resulting in increased

average daily gain. The protein-binding ability of tannin [26] could have also resulted in a synergistic effect responsible for reducing fecal egg count. Dan iel et al. [27] reported that egg reduction was a function of physiological distortion by the precipitation of its protein content by tannin. This view of the ability of plants to reduce egg count resulting in increased growth was also reported by Jack et al. [6] when they used need seed in lambs. The ability of *C. coriaria* to reduce fecal egg count is vital in a world that is trying to reduce its dependence on synthetic drugs and go “clean”. This shows the nutraceutical tendencies of the extract, and when combined with manage-

**Table 6.** Feed dry matter and water intake and average daily gain (ADG) of growing lambs fed on diets with 0 (EC0), 30 (EC30), and 60 (EC60) mL of Cascalote (*Caesalpinia coriaria* JACQ. WILLD.) fruits extracts for 60 days

| Extract (EC, mL/day) | Day | DM intake (g/day) | Water intake (L/day) | ADG (g/day) |
|----------------------|-----|-------------------|----------------------|-------------|
| EC0                  | 1   | 514.3             | 1.46                 | 17.1        |
|                      | 15  | 618.3             | 1.98                 | 110.7       |
|                      | 30  | 819.1             | 2.53                 | 122.9       |
|                      | 45  | 889.1             | 2.99                 | 137.1       |
|                      | 60  | 872.7             | 2.65                 | 135.0       |
| EC30                 | 1   | 600.0             | 1.33                 | 16.8        |
|                      | 15  | 656.0             | 1.79                 | 140.3       |
|                      | 30  | 809.2             | 2.47                 | 151.4       |
|                      | 45  | 843.3             | 3.26                 | 141.0       |
|                      | 60  | 866.3             | 3.07                 | 152.1       |
| EC60                 | 1   | 457.1             | 1.41                 | 16.1        |
|                      | 15  | 695.9             | 1.87                 | 168.6       |
|                      | 30  | 832.3             | 2.47                 | 181.0       |
|                      | 45  | 926.3             | 3.23                 | 168.9       |
|                      | 60  | 817.5             | 3.13                 | 166.9       |
| SEM                  |     | 64.940            | 0.119                | 11.38       |
| P value              |     |                   |                      |             |
| Extract (EC)         |     | 0.9559            | 0.5033               | 0.0002      |
| Linear               |     | 0.9437            | 0.247                | <.0001      |
| Quadratic            |     | 0.7708            | 0.8719               | 0.7604      |
| Day                  |     | <.0001            | <.0001               | <.0001      |
| Linear               |     | <.0001            | <.0001               | <.0001      |
| Quadratic            |     | 0.7524            | 0.4935               | <.0001      |
| EC X day             |     | 0.8524            | 0.2719               | 0.4903      |

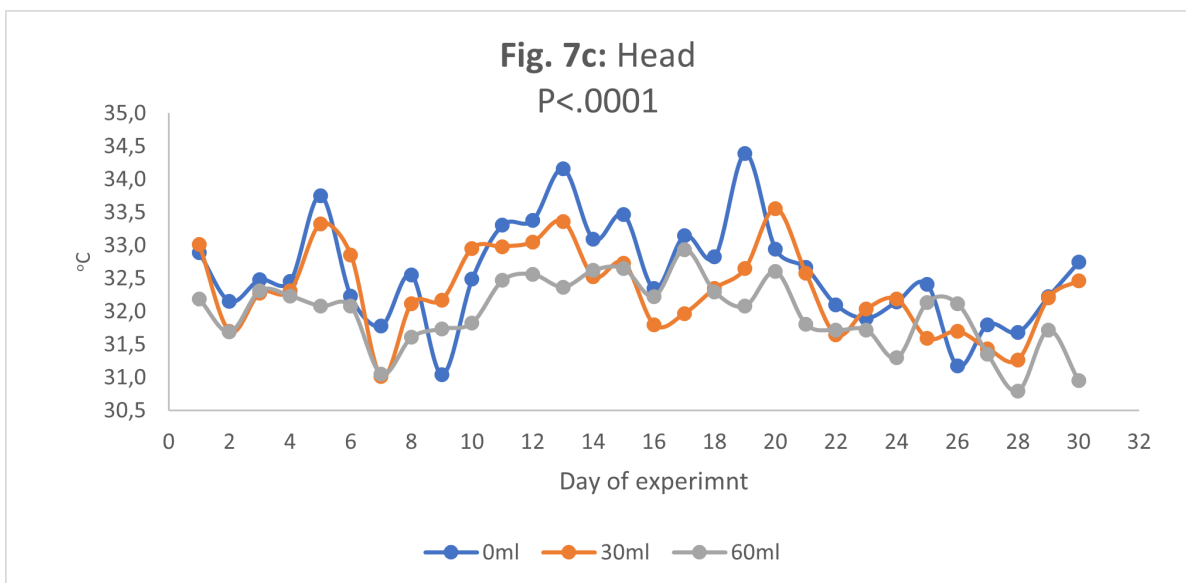
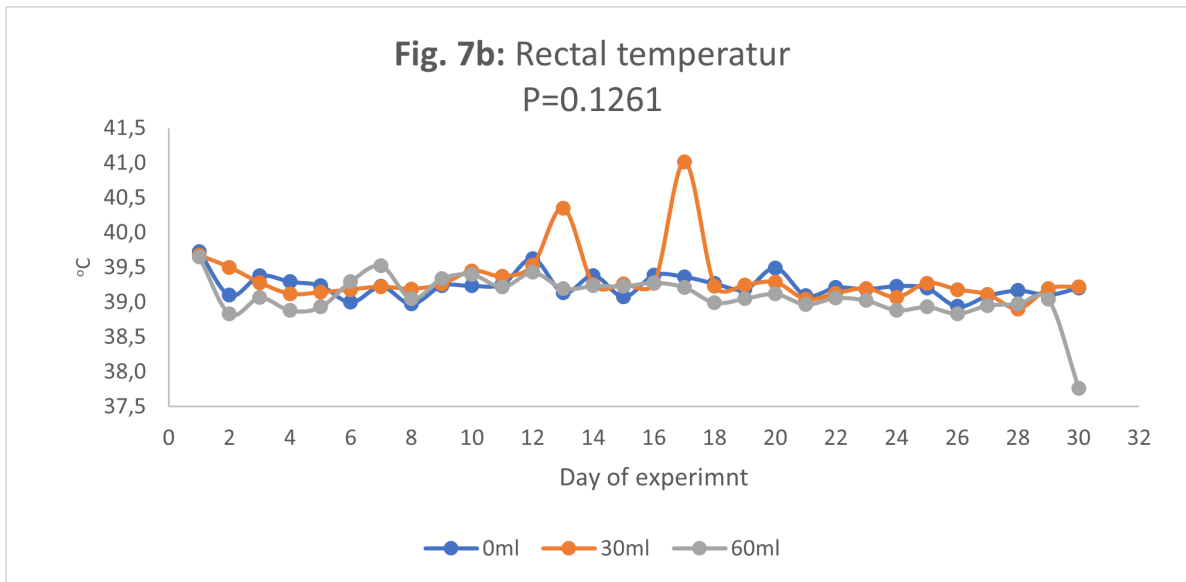
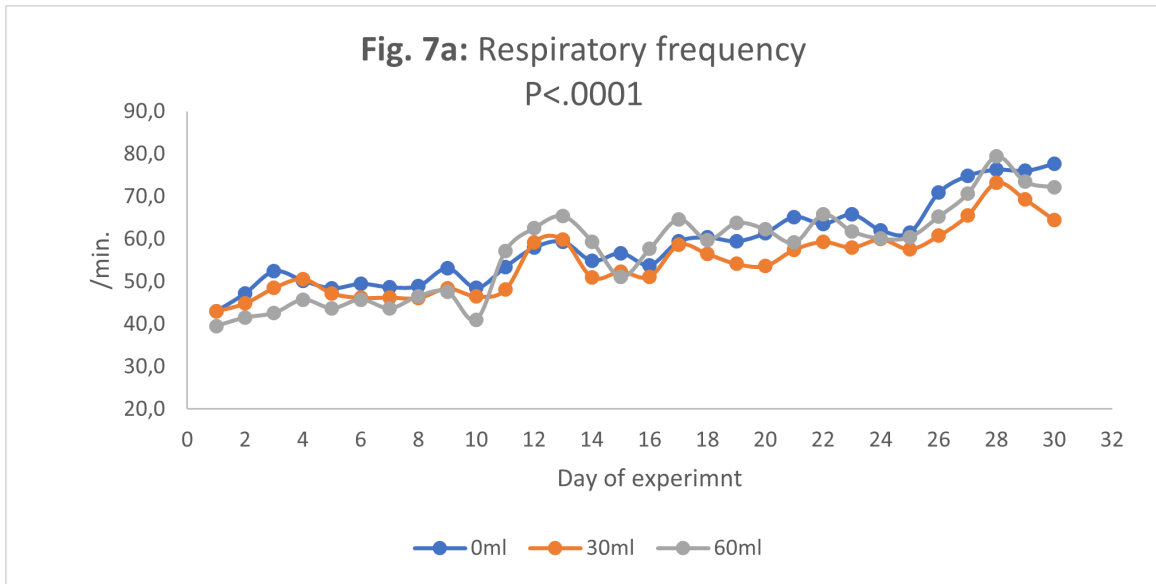
ment, it will improve animal production. The implication of this is that local farmers can use this as alternative to drugs and might lead to herbal production development. The only caveat is that companies should avoid the extraction of the active ingredients in these herbs alone, as it might lead to resistance over time. Herbs for many years have been used by animals in the wild for millennia without resistance because many phytochemicals in plants work synergistically to aid one another.

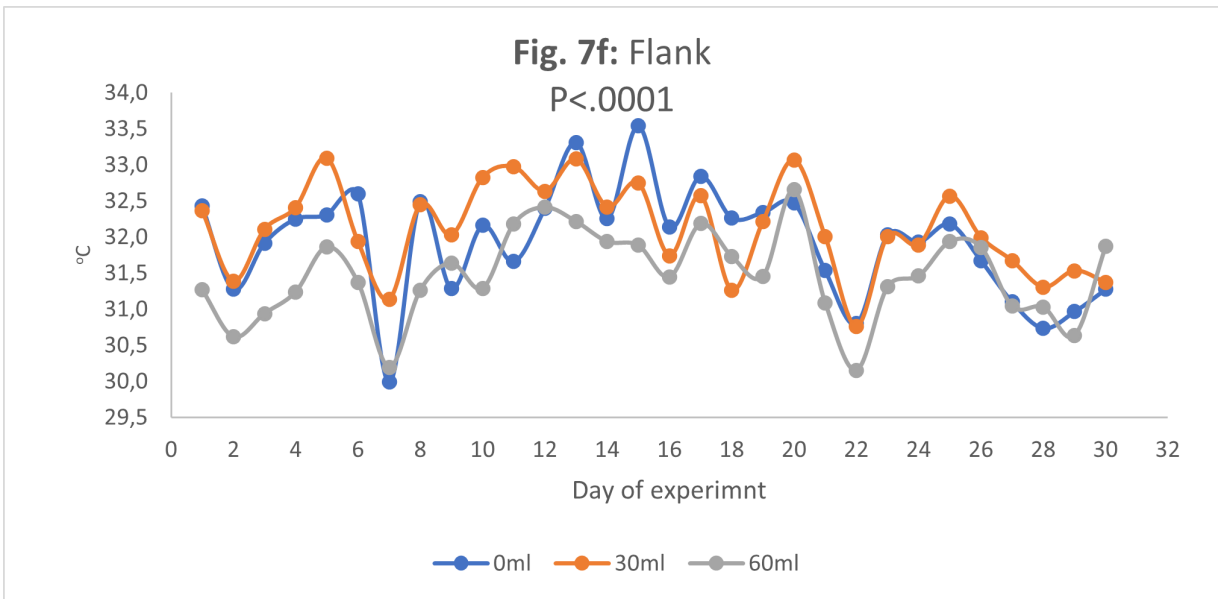
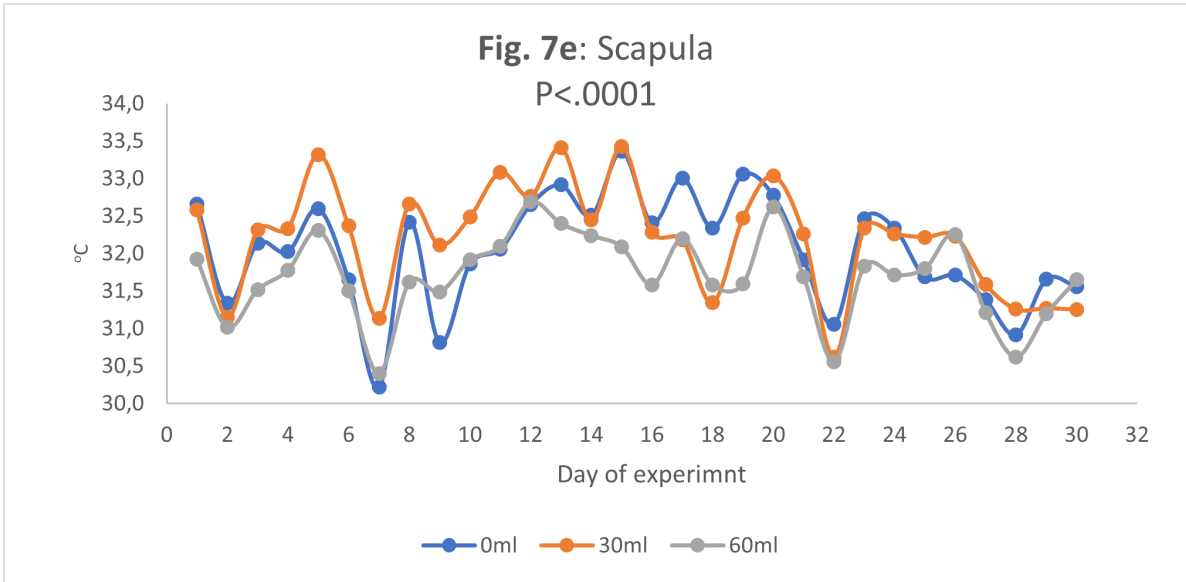
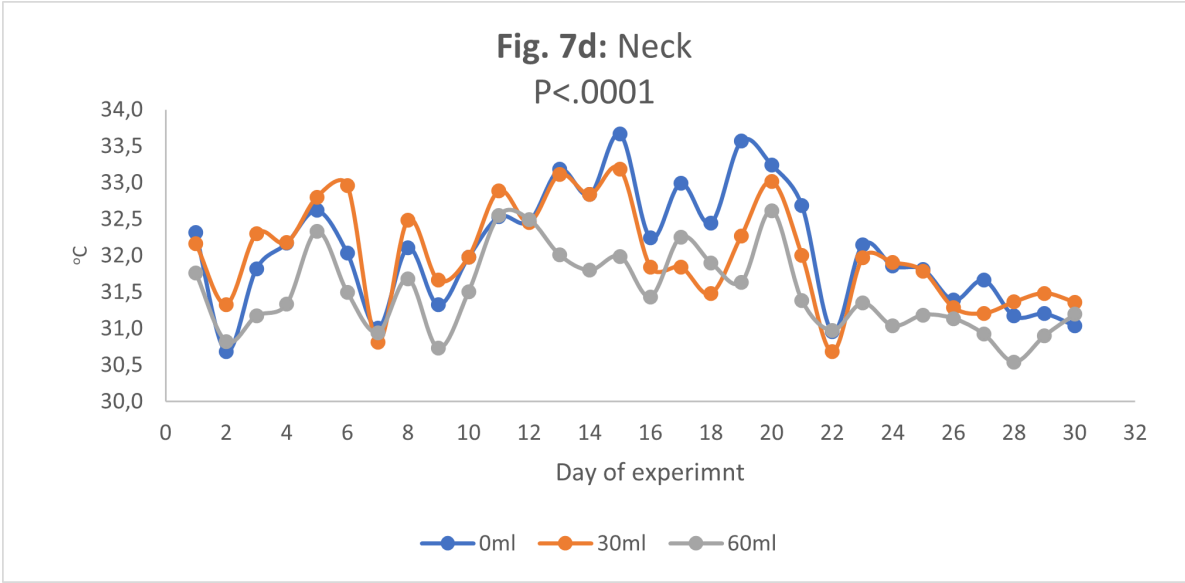
RF indicates heat stress in sheep and may act as a modulator between heat dissipation and heat production. An increased RF represents an effort to enhance respiratory evaporation and lower skin temperature [28]. Hence, the dose-dependent decrease in respiratory rate and anal temperature indicates that the extract could manage body temperature. On the other hand, the higher respiratory rate might be due to increased heat generation during rumination, leading to a need for higher respiration frequency to dissipate the heat generated during rumination.

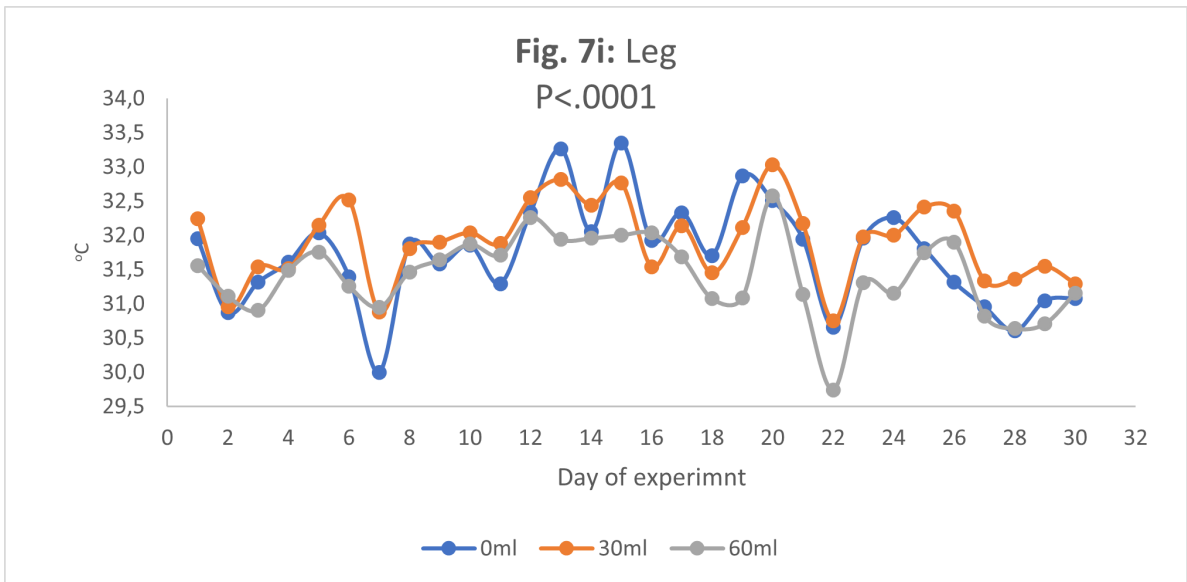
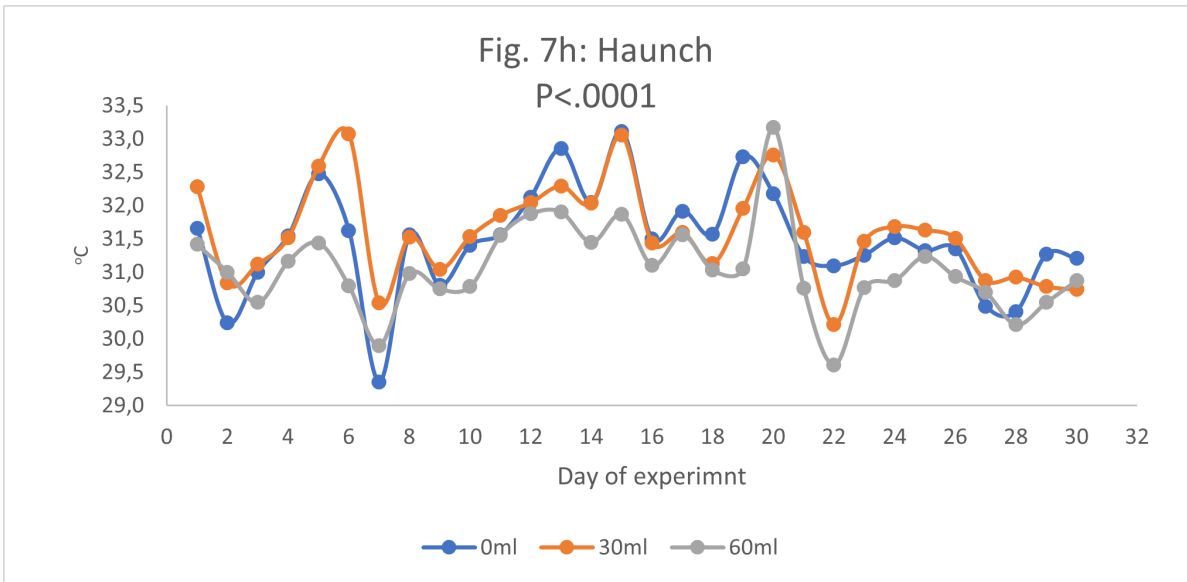
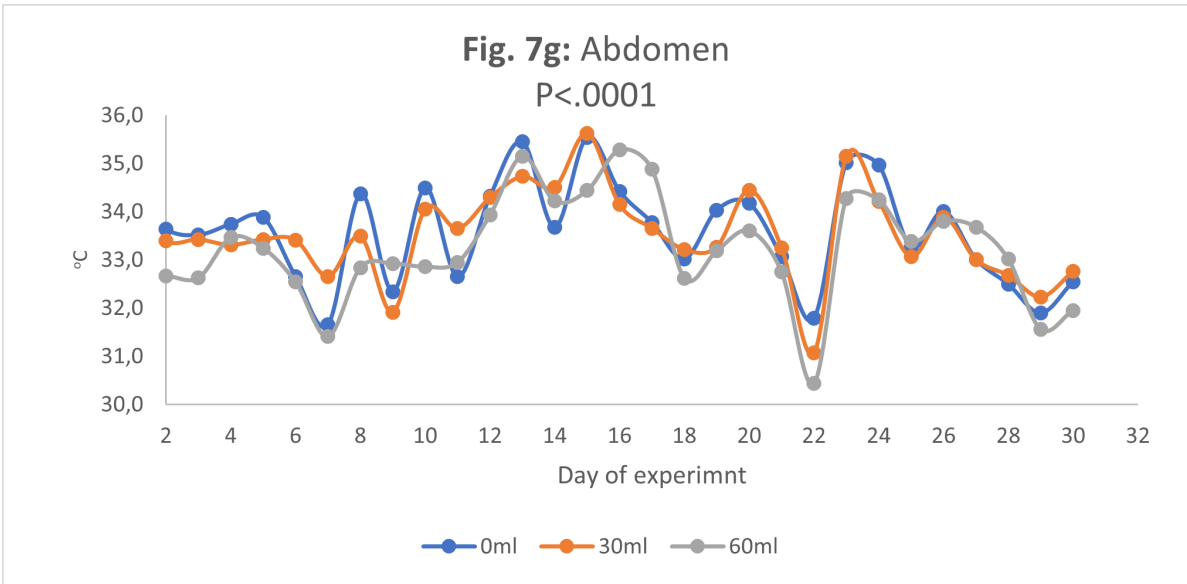
As a means of heat dissipation, animals utilize peripheral

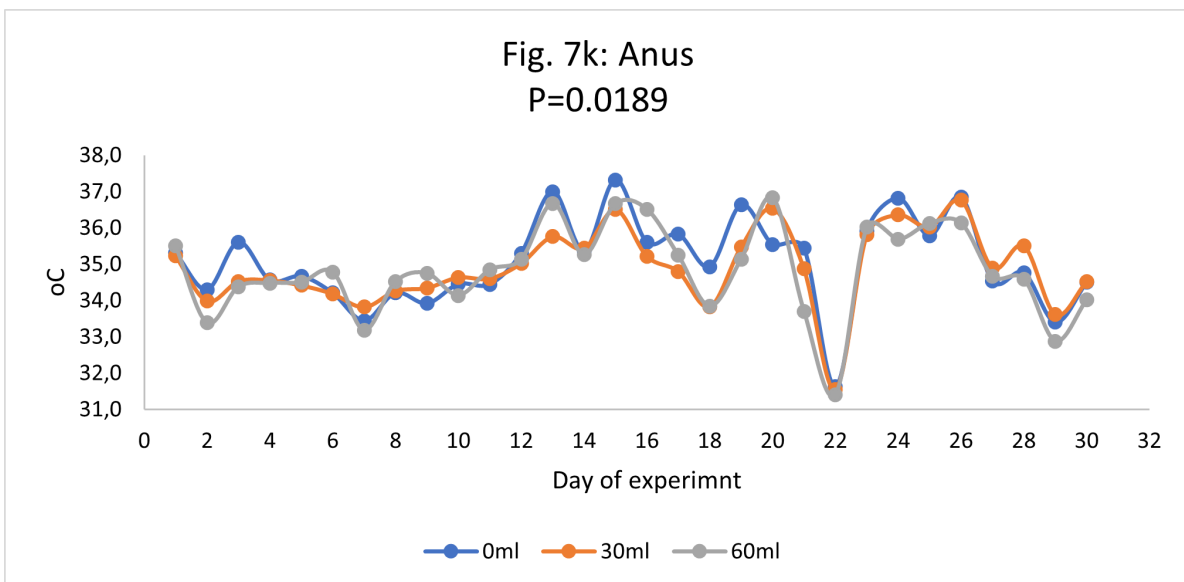
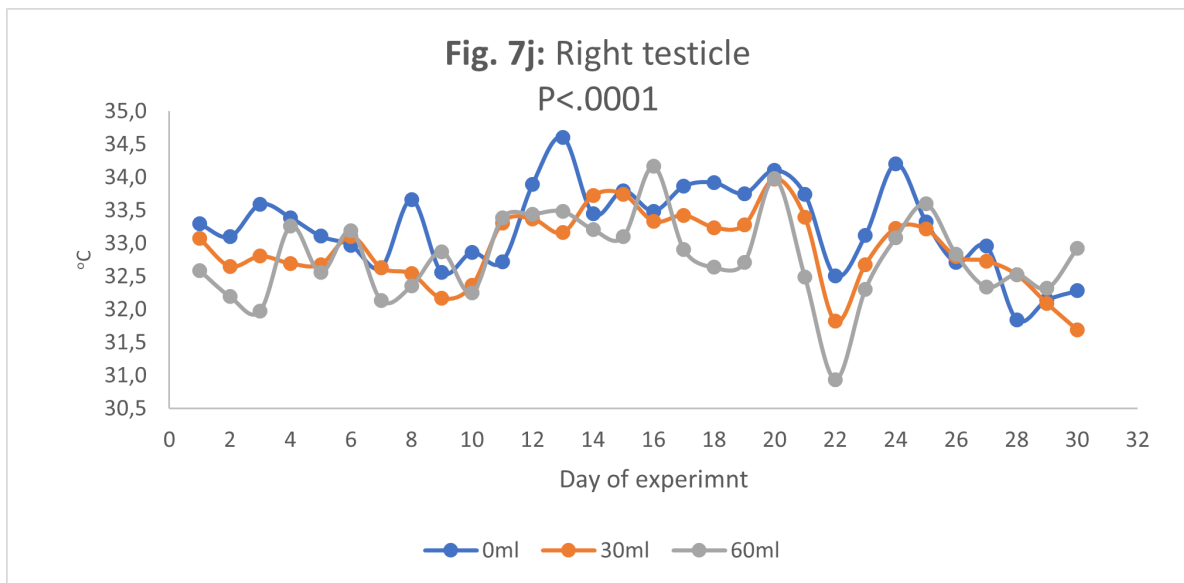
vasodilation, elevate respiratory and heart rates, increase skin temperature, and consequently reduce feed intake [29]. The observed increase in RF and body part temperature after feeding (AFM) compared to before feeding (BFM) in this study can be attributed to diet-induced thermogenesis. The elevation in body temperature post-feeding results from increased energy expenditure, known as diet induced thermogenesis or the thermic effect of feeding [8].

Diet-induced thermogenesis is influenced by feed digestion and increased energy utilization, causing an acute rise in body temperature of various species [30]. The overall increase in temperature at different body sites and respiratory rate between BFM and ASM may be attributed to the circadian rhythm of body temperature, which is low in the morning and high in the evening and can be influenced by environmental temperature and feeding-induced thermogenesis [31]. Furthermore, the increased temperature in ASM may result from the additive effect of food anticipation activities, diet-induced metabolic heat production, and the rhythmically









**Figure 7 (a to k).** Daily measurements of the physiological constants, along the first 30 days of the experiment of growing lambs fed diets with 0 (EC0), 30 (EC30), and 60 (EC60) mL of Cascalote (*Caesalpinia coriaria* Jacq. Willd.) fruits aqueous extract.

increasing body temperature [32]. However, regardless of the cause, the temperatures obtained in this study were within the range reported by Adebeye et al. [8] for small ruminants. In all body sites, the increased temperature can be attributed to blood flow to various body regions, which leads to vasomotor heat loss and plays a crucial role in temperature regulation [33]. The temperature excursion range for the body sites in this study was 0.5 - 4.93°C. This suggests that the head, neck, scapula, flank, abdomen, rump, and leg are important for heat elimination/dissipation, with the rump having the highest temperature excursion. The various excursion ranges observed in this study for each extract suggest that the dose of extract/herb intake has varying effects on the blood flow rate carrying

heat from core body temperature to peripheral or other body sites. Cuesta et al. [34] supports this view who found that an increase in skin temperature is triggered by increased skin blood flow, resulting in heat loss that down-regulates core body temperature. Correlation is often used to indicate the degree of relationship between two variables, whether positive or negative, and to what extent the correlation exists as strong, medium, or weak. In many physiological studies, RF can be used to assess whether an animal is heat-stressed, among other factors, while rectal temperature can be used to determine core body temperature and indicate whether an animal is heat-stressed.

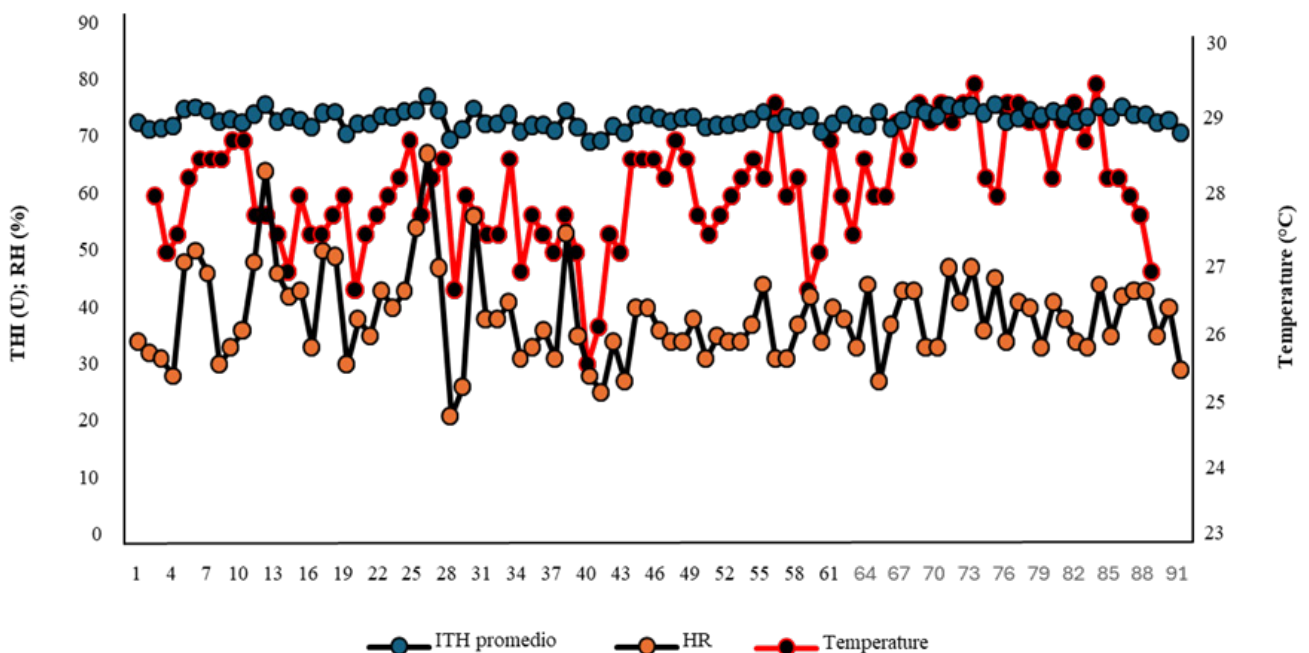
Therefore, comparing the variation in body part temperature

**Table 7.** Feeding behavior (time spent eating, ruminating, resting, and other activities, min/day) of growing lambs fed on diets with 0 (EC0), 30 (EC30), and 60 (EC60) mL of Cascalote (*Caesalpinia coriaria* JACQ. WILLD.) fruits extracts for 60 days.

| Extract (EC, mL/day) | Day <sup>1</sup> | Eating | Rumination | Resting | Other activities <sup>2</sup> |
|----------------------|------------------|--------|------------|---------|-------------------------------|
| EC0                  | 1                | 295.5  | 456.3      | 529.1   | 159.1                         |
|                      | 60               | 292.8  | 459.7      | 535.0   | 152.6                         |
| EC30                 | 1                | 304.4  | 433.6      | 542.3   | 159.8                         |
|                      | 60               | 303.1  | 437.1      | 543.8   | 156.1                         |
| EC60                 | 1                | 302.4  | 445.2      | 547.2   | 145.2                         |
|                      | 60               | 300.1  | 443.8      | 550.8   | 145.3                         |
| SEM                  |                  | 3.09   | 3.93       | 3.36    | 5.00                          |
| P value              |                  |        |            |         |                               |
| Extract (EC)         |                  | 0.0246 | <.0001     | <.0001  | 0.1355                        |
| Linear               |                  | 0.0505 | 0.0027     | <.0001  | 0.322                         |
| Quadratic            |                  | 0.0545 | <.0001     | 0.4012  | 0.0819                        |
| Day                  |                  | 0.4622 | 0.6045     | 0.1903  | 0.2536                        |
| EC X day             |                  | 0.9806 | 0.8152     | 0.8203  | 0.5508                        |

<sup>1</sup>(n=8 animals per treatment, each value was an average of two consecutive days per each experimental period (day 1 and day 60)).

<sup>2</sup>spent time during waters, sleeping, other unexpected movements within the day.



**Figure 8.** Behavior of the temperature and humidity index for the 60-day experimental period.

taken in the morning to the afternoon, an observed increase in the morning showed a strong positive correlation between RF and neck temperature, and between rectal temperature and abdominal temperature. However, in the afternoon, there was a shift in the correlation pattern, with a strong correlation

emerging between RF and rectal, haunch, and anal temperature. Furthermore, a strong correlation was found between rectal temperature and the abdomen, right testicle, and anal temperature. The only consistent correlation between morning and afternoon temperatures was a strong positive correlation

**Table 8.** Parasites eggs counts (EPG) of growing lambs fed diets with 0 (EC0), 30 (EC30), and 60 (EC60) mL of Cascalote (*Caesalpinia coriaria* JACQ. WILLD.) fruits extracts for 60 days.

| Extract (EC, mL/day) | Sample day | Eggs/ g feces |
|----------------------|------------|---------------|
| EC0                  | 1          | 3323          |
|                      | 3          | 6414          |
|                      | 6          | 3233          |
|                      | 25         | 6830          |
|                      | 30         | 4616          |
|                      | 35         | 2940          |
|                      | 50         | 2050          |
|                      | 55         | 1437          |
|                      | 60         | 1480          |
| EC30                 | 1          | 3060          |
|                      | 3          | 4860          |
|                      | 6          | 3680          |
|                      | 25         | 4430          |
|                      | 30         | 4960          |
|                      | 35         | 5986          |
|                      | 50         | 1475          |
|                      | 55         | 990           |
|                      | 60         | 670           |
| EC60                 | 1          | 2150          |
|                      | 3          | 4140          |
|                      | 6          | 2390          |
|                      | 25         | 3390          |
|                      | 30         | 2500          |
|                      | 35         | 2400          |
|                      | 50         | 1990          |
|                      | 55         | 2000          |
|                      | 60         | 1163          |
| SEM                  |            | 1018.63       |
| P value              |            |               |
| Extract (EC)         |            | 0.1155        |
| Linear               |            | 0.0486        |
| Quadratic            |            | 0.5159        |
| Day                  |            | 0.0001        |
| Linear               |            | 0.0217        |
| Quadratic            |            | 0.2977        |
| EC X day             |            | 0.7912        |

between rectal temperature and abdominal temperature. This suggests that there are multiple body points for heat dissipation as the time-of-day changes from morning to afternoon. It also indicates a heat transition from the abdominal region to rectal area.

## Conclusions

Based on chromatographic analysis, the aqueous extract of *C. coriaria* fruits contained biologically effective phenolic and flavonoid compounds and vitamins, which improved lambs' average daily gain, reduced EPG, and enhanced feed and water intake. *Caesalpinia coriaria* extract influences feeding behaviors such as time spent eating and ruminating.



**Table 9.** Physiological constants measured before and after first (BFM, AFM) and second (BSM, ASM) meals feed to growing lambs supplemented with different doses of Cascalote (*Caesalpinia coriaria* JACQ. WILLD.) fruits extracts.

| Extract (EC, mL/day) | Time of Measure (Tm) | Respiratory frequency (/min) | Rectal temperature (°C) | Head (°C) | Neck (°C) | Scapula (°C) | Flank (°C) | Abdomen (°C) | Haunch (°C) | Leg (°C) | Right testicle (°C) | Anus (°C) |
|----------------------|----------------------|------------------------------|-------------------------|-----------|-----------|--------------|------------|--------------|-------------|----------|---------------------|-----------|
| EC0                  | BFM                  | 48.9                         | 39.0                    | 30.4      | 29.7      | 29.7         | 29.5       | 31.8         | 28.8        | 29.2     | 31.6                | 33.8      |
|                      | AFM                  | 50.9                         | 39.1                    | 31.3      | 30.8      | 30.8         | 30.6       | 32.4         | 30.1        | 30.5     | 32.2                | 34.1      |
|                      | BSM                  | 66.1                         | 39.6                    | 34.5      | 34.1      | 34.0         | 34.0       | 35.1         | 33.7        | 33.9     | 35.0                | 36.1      |
|                      | ASM                  | 67.3                         | 39.4                    | 34.2      | 33.8      | 33.6         | 33.6       | 35.0         | 33.4        | 33.4     | 34.2                | 36.2      |
| EC30                 | Excursion range      | 18.4                         | 0.4                     | 3.8       | 4.1       | 3.9          | 4.1        | 3.2          | 4.6         | 4.2      | 2.6                 | 2.4       |
|                      | BFM                  | 45.1                         | 39.0                    | 29.9      | 29.4      | 29.5         | 29.4       | 31.5         | 28.5        | 29.3     | 31.1                | 33.4      |
|                      | AFM                  | 46.1                         | 39.1                    | 31.3      | 31.1      | 31.3         | 31.2       | 32.5         | 30.6        | 30.8     | 31.9                | 34.0      |
|                      | BSM                  | 63.9                         | 41.0                    | 34.1      | 33.8      | 33.9         | 33.9       | 34.9         | 33.5        | 33.8     | 34.6                | 36.0      |
| EC60                 | ASM                  | 63.2                         | 39.7                    | 34.0      | 33.7      | 34.1         | 34.0       | 35.1         | 33.7        | 33.7     | 34.1                | 36.3      |
|                      | Excursion range      | 18.1                         | 0.7                     | 4.1       | 4.3       | 4.6          | 4.6        | 3.6          | 5.2         | 4.4      | 3                   | 2.9       |
|                      | BFM                  | 47.4                         | 38.7                    | 29.5      | 29.0      | 29.1         | 28.8       | 31.6         | 28.2        | 28.7     | 30.9                | 33.6      |
|                      | AFM                  | 50.9                         | 39.0                    | 31.0      | 30.5      | 30.7         | 30.4       | 32.1         | 30.0        | 30.6     | 32.1                | 34.2      |
| SEM                  | BSM                  | 63.3                         | 39.5                    | 33.5      | 33.2      | 33.4         | 33.3       | 34.4         | 33.1        | 33.1     | 34.0                | 35.5      |
|                      | ASM                  | 64.0                         | 39.1                    | 34.0      | 33.5      | 33.7         | 33.5       | 34.9         | 33.2        | 33.4     | 34.2                | 36.1      |
|                      | Excursion range      | 16.6                         | 0.4                     | 4.5       | 4.5       | 4.6          | 4.7        | 3.3          | 5           | 4.7      | 3.3                 | 2.5       |
|                      | SEM                  | 1.07                         | 0.19                    | 0.13      | 0.13      | 0.12         | 0.12       | 0.17         | 0.12        | 0.11     | 0.14                | 0.15      |
| P value              |                      |                              |                         |           |           |              |            |              |             |          |                     |           |
| Extract (EC)         |                      | <.0001                       | 0.1261                  | <.0001    | <.0001    | <.0001       | <.0001     | 0.0011       | <.0001      | <.0001   | <.0001              | 0.0189    |
| Linear               |                      | 0.0108                       | 0.6354                  | <.0001    | <.0001    | <.0001       | <.0001     | 0.0006       | <.0001      | <.0001   | <.0001              | 0.0062    |
| Quadratic            |                      | <.0001                       | 0.0485                  | 0.581     | 0.0028    | <.0001       | <.0001     | 0.1901       | <.0001      | <.0001   | 0.143               | 0.4841    |
| Time of measure (Tm) |                      | <.0001                       | 0.0145                  | <.0001    | <.0001    | <.0001       | <.0001     | <.0001       | <.0001      | <.0001   | <.0001              | <.0001    |
| EC X Tm              |                      | 0.1018                       | 0.5771                  | 0.0011    | 0.0114    | 0.0002       | 0.0046     | 0.2928       | 0.0038      | <.0001   | 0.0023              | 0.0049    |

**Table 10.** Pearson correlation for the variables of respiratory frequency (RF) and temperatures in different body parts (rectal, head, neck, scapula, flank, abdomen, rump, leg, right testicle and anus) during the morning of growing lambs fed diets with 0 (EC0), 30 (EC30) and 60 (EC60) mL of Cascalote (*Caesalpinia coriaria* JACQ. WILLD.) fruits extracts during the experimental period of 60 days.

|                       | Respiratory frequency | Rectal temperature | Head | Neck | Scapula | Flank | Abdomen | Haunch | Leg | Right testicle | Anus |
|-----------------------|-----------------------|--------------------|------|------|---------|-------|---------|--------|-----|----------------|------|
| Respiratory frequency |                       | ns                 | ns   | ns   | ns      | ns    | *       | ns     | ns  | ns             | ***  |
| Rectal temperature    |                       |                    | ns   | ns   | ns      | ns    | ns      | ns     | ns  | ns             | ns   |
| Head                  |                       |                    |      | ***  | ***     | ***   | ***     | ***    | *** | ***            | ***  |
| Neck                  |                       |                    |      |      | ***     | ***   | ***     | ***    | *** | ***            | ***  |
| Scapula               |                       |                    |      |      |         | ***   | ***     | ***    | *** | ***            | ***  |
| Flank                 |                       |                    |      |      |         |       | ***     | ***    | *** | ***            | ***  |
| Abdomen               |                       |                    |      |      |         |       |         | ***    | *** | ***            | ***  |
| Haunch                |                       |                    |      |      |         |       |         |        | *** | ***            | ***  |
| Leg                   |                       |                    |      |      |         |       |         |        |     | ***            | ***  |
| Right testicle        |                       |                    |      |      |         |       |         |        |     |                | ***  |
| Anus                  |                       |                    |      |      |         |       |         |        |     |                |      |

\*\*\*: highly significant difference (P<0.0001), \*\*: significant difference (P<0.05), ns: no significant

**Table 11.** Pearson correlation for the variables of respiratory frequency (RF) and temperatures in different body parts (rectal, head, neck, scapula, flank, abdomen, rump, leg, right testicle and anus) during the afternoon of growing lambs fed on diets 0 (EC0), 30 (EC30) and 60 (EC60) mL of Cascalote (*Caesalpinia coriaria* JACQ. WILLD.) fruits extracts during the experimental period of 60 days.

|                       | Respiratory frequency | Rectal temperature | Head | Neck | Scapula | Flank | Abdomen | Haunch | Leg | Right testicle | Anus |
|-----------------------|-----------------------|--------------------|------|------|---------|-------|---------|--------|-----|----------------|------|
| Respiratory frequency |                       | ns                 | ns   | ns   | ns      | ns    | ns      | ns     | ns  | ns             | ns   |
| Rectal temperature    |                       |                    | *    | ns   | ns      | ns    | ns      | ns     | ns  | ns             | ns   |
| Head                  |                       |                    |      | ***  | ***     | ***   | ***     | ***    | *** | ***            | ***  |
| Neck                  |                       |                    |      |      | ***     | ***   | ***     | ***    | *** | ***            | ***  |
| Scapula               |                       |                    |      |      |         | ***   | ***     | ***    | *** | ***            | ***  |
| Flank                 |                       |                    |      |      |         |       | ***     | ***    | *** | ***            | ***  |
| Abdomen               |                       |                    |      |      |         |       |         | ***    | *** | ***            | ***  |
| Haunch                |                       |                    |      |      |         |       |         |        | *** | ***            | ***  |
| Leg                   |                       |                    |      |      |         |       |         |        |     | ***            | ***  |
| Right testicle        |                       |                    |      |      |         |       |         |        |     |                | ***  |
| Anus                  |                       |                    |      |      |         |       |         |        |     |                |      |

\*\*\*: highly significant difference (P<0.0001), \*\*: significant difference (P<0.05), ns: no significant

*Caesalpinia coriaria* fruits extract helps to reduce respiratory frequency in lamb. Therefore, use of the aqueous extract of *Caesalpinia coriaria* fruits, up to 60 mL (EC60), demonstrated beneficial effects on lambs, including improved productivity, anti-helmintic activity, and alleviation of body temperature. Further studies need to be carried out on the individual components of *Caesalpinia coriaria* fruits to ascertain their effect on body temperature/physiology and determine if the effects are individualistic or synergistic. This should also be done on extensive or semi-intensive system, to ascertain their effectiveness under all management conditions. Also, there should be studies on long term effects, on different breeds, and different environmental conditions to ascertain the reliability and integrity of this extract.

## Funding

There was no external source of funding for this work.

## Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

## Ethics approval

Animal studies were approved by the ethical committee of the Faculty of Veterinary Medicine and Animal Science, Autonomous University of the State of Mexico.

## Consent to participate

All authors agree to participate in the current work.

## Consent to for publication

All authors agree to publish the findings of the current research.

## Availability of data and material

Not applicable

## Code availability

Not applicable

## Authors' contributions

PEHR, MM, MMMYG, and AZMS conceived and designed the experiment; PEHR, MMMYG, AZMS, JLPC, and BCBP conducted the experiment; PH, MM, MMMYG, AZMS, MJA and MSMS prepared the manuscript. All authors approved the manuscript.

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