



Rectal temperature and body surface temperature rhythm in Red Sokoto goats infected with *Trypanosoma evansi* during the rainy season

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Abstract

There are limited studies on circadian rhythmicity in farm animals affected by protozoan diseases. The rectal temperature (RT) and body surface temperature (BST) rhythmic patterns in experimental *Trypanosoma evansi*-infected Red Sokoto goats (*T. evansi*-infected RSG) July – September were evaluated. Five goats were assigned to each experimental group, with group I as the negative control (CRSG), while animals in group II were individually infected with about $2.0 \times 10^6/2\text{ml}$ *T. evansi*-inoculum. Temperature reaction patterns were monitored by digital clinical and infrared thermometry. The RT and BST of goats, the ambient temperature (AT) and relative humidity (RH) inside the pen were measured at 06:00, 12:00 and 18:00 hours (GMT+1) three times a week for 12 weeks. The AT had maximum, minimum and mean values of 27.0°C, 24.2°C and 25.6±0.26°C, respectively, while RH had maximum, minimum and mean values of 94.5%, 53.6% and 80.5±11.5%, respectively. Results obtained, via the application of a single Cosinor procedure showed that the RT and BST exhibited daily rhythmicity in both groups with the pattern distorted in *T. evansi*-infected RSG which showed an ascent from the 5th to the 10th week. The rhythmic characteristic of RT of the *T. evansi*-infected RSG showed that the mesor and amplitude were higher ($p < 0.05$) than those recorded for CRSG. Both groups showed no difference ($p > 0.05$) in acrophase. The eye region in both groups showed the highest BST, while the coronary band had the lowest. The result, for the first time, reported rhythmicity of RT and BST in *T. evansi*-infected goats under tropical conditions. The results obtained from our study may be useful for the early screening, diagnosis, treatment and prevention of protozoan diseases.

Keywords: Body surface temperature, Circadian rhythm, Infrared thermometer, Rainy season, Rectal temperature, *Trypanosoma evansi*

Introduction

The domestic goat (*Capra hircus*) commonly referred to as the “poor man’s cow”, is a member of the family Bovidae and the goat-antelope subfamily Caprinae (Mathee & Davis, 2001; Arif et al., 2012) providing constant access to milk, meat, skins, fibre and manure

for 95% of farmers in Nigeria’s rural areas (Ukpabi et al., 2000; Peacock, 2005). Goats are often called the “village bank” (Maxwell, 1990; Oluwatayo & Oluwatayo, 2012), as they provide cash and ready

credit access to meet immediate social and financial needs (Oluwatayo & Oluwatayo, 2012)

The world population of goats currently stands at 921 million, of which approximately 96% are found in developing countries in Asia and Africa (FAOSTAT, 2008; Mataveia *et al.*, 2021) with Africa accounting for 35% after Asia while Nigeria, Sudan and Kenya have the largest population (Skapetas & Bampidis, 2016).

Major challenges in goat production include nutrition, diseases and current climate change.

Among the protozoan parasites of goats is *Trypanosoma evansi*, the causative agent of Surra, causing debility and loss of production. *T. evansi* is widely distributed in both wild and domestic animals (Dargantes *et al.*, 2009) and has been found in humans in India, Vietnam and Egypt (Josh *et al.*, 2006; Haridy *et al.*, 2011; Vinh Chau *et al.*, 2016).

Surra is a disease that is severely neglected in terms of awareness, control interventions and research into improved control methods, despite over 100 years of study (Birhanu *et al.*, 2016).

Recent chronophysiology or biological rhythm is used in assessing the physiological responses of animals to environmental changes, diseases, nutrition and medication (Minka & Ayo, 2016). Circadian rhythmicity is a common feature of mammalian physiology and behaviour (Refinetti, 2006). The multi-oscillatory network in all levels of cells of the body is controlled by the master pacemaker clock in the suprachiasmatic nucleus (SCN) of the hypothalamus, directing an animal's rhythmic expression of the physiological functions and behaviour via a hierarchical system (Matsui *et al.*, 2016; Gianetto *et al.*, 2016).

Biological rhythm is a new field of study in livestock production with promising results and could be used to assess the responses of body temperature and body surface temperature of goats infected with *Trypanosoma evansi*. Results from such studies will shed more light on the response of the biological clock during fever in goats infected with *Trypanosoma evansi* infection, which is currently lacking in the available literature.

Materials and Methods

Experimental site

The experiment was conducted at the Livestock Farm of the Ahmadu Bello University College of Agriculture and Animal Science, Mando Road, Kaduna. The experimental site was predominantly an agricultural farming area located in the Northern Guinea Savannah of Nigeria.

Experimental animals and management

Fifteen apparently healthy male RSG aged between 10 - 12 months were purchased from a nearby village. They were acclimatized and conditioned for two weeks before the experimental day. The study was approved by The Nigerian Defence Academy's Post Graduate School on Animal Welfare and Use through approval number NDA/PGS/FS/P/0028/18/VCN1886. All experimental animals were treated humanely as recommended by International Research Organizations (CRWL, 2003).

The RSG were housed and stocked in fly and tick-proof pens (2.5m high) at a rate of 1 m² per goat (Kannan *et al.*, 2002). The RSG were zero grazed and placed on guinea corn residue, hay, legume hay (beans and groundnut) supplemented with maize/wheat offal, *Digitaria smutsii* and molasses. Water and blocks of commercial salt lick were provided *ad libitum*.

During the 14-day acclimatization (Wolfensohn & Lloyd, 2013) and before the commencement of the experiment, a 5g faecal sample was collected from the rectum of each goat and screened for gastrointestinal parasites using flotation and sedimentation techniques (Cole, 1986).

All the goats were prophylactically treated with Amprolium 250 WSP® (KEPRO B. V., Holland) against coccidiosis at a dose of 3g/20kg orally, dewormed using Fenbendazole bolus (Fenacure®, Ashish Life Science PVT Ltd, India) once, orally at the dosage rate of 10mg/kg body weight and they were treated for ectoparasites with cypermethrin (Cypermethrin Pour On, Ourofino, Brazil) at the dosage rate of 1ml/kg body weight.

Experimental design

Using a sterile disposable 5ml syringe and 21G 1¹/₂inch needle (DELEJECT®, HMA Medical Ltd., Nigeria), two millilitres of whole blood were collected from each goat through the jugular vein, into sample bottles containing liquid Ethylene Diamine Tetra-acetate (K₃EDTA). One millilitre was used to examine for haemoparasites especially *Trypanosoma species* via wet blood film x40 objective, HCT and Giemsa stained thin blood smear at x100 objectives (Weiser, 2012). An average of 20 microscopic fields of each preparation was examined, this is to ensure that the goats were free from haemoparasites infection. A rat inoculation test was carried out using the remaining 1 ml of blood (out of the 2ml taken). A 2 ml sterile disposable syringe with 23G x 1¹/₄inch

needle (CENT-JECT, CenturyCare Ltd., Nigeria) was used to inoculate 2 representative rats (OIE, 2010) for each goat half a millilitre (0.5ml) of blood intraperitoneally (Wolfensohn & Lloyd, 2013). The inoculated rats were monitored for haemoparasites on HCT and wet film using tail tip blood every 48 hours for 14 days (Monzon *et al.*, 1990) to ensure that the experimental goats were not latent carriers of haemoparasites from field challenge. The rats were further observed for 60 days post-inoculation.

Grouping of the animals

On experimental day, ten goats that were found apparently healthy after screening were ear-tagged at random for easy identification and the animals were randomly assigned into two groups of five goats each. Group I was control (CRSG) and Group II *Trypanosoma evansi*-infected Red Sokoto goat (*T. evansi*-infected RSG).

Collecting baseline parameters of the experimental goats

Rectal temperature (RT) was taken using a digital clinical thermometer (COCET®, Kangfu Medical Equipment, China) inserted into the rectum (Minka & Ayo, 2016) till the sound of an alarm was heard, which indicated the end of reading, while the body surface temperature (BST) was taken with the aid of an infrared thermometer (Model AR330, China), the temperatures of four locations (mid-head, nose, eye and coronary band) on each goat were taken. These locations are excellent thermoregulatory endpoints giving a fair accurate reading of body temperatures as the most sensitive to body temperature. The infrared thermometer was located at a distance of 12 inches from the animal, this distance was maintained throughout the measurement period. The infrared thermometer has an up to 12:1 distance to spot which measures the temperature of a 1-inch diameter circle of surface area from 12 inches away (Ramey *et al.*, 2011; Rushton *et al.*, 2015; Minka & Ayo, 2016).

Trypanosoma evansi and infection of experimental goats

The *Trypanosoma evansi* used in this experiment was obtained from the stabulates kept in the Vector and Parasitology Department of the National Institute for Trypanosomiasis Research (NITR), Kaduna, Nigeria. The parasite was inoculated intraperitoneally into two clean rats serving as donor animals (Monzon *et al.*, 1990; Wolfensohn & Lloyd,

2013). Using HCT the rats were monitored every 48 hours for rising parasitaemia of +2 (6-10 parasites/field) to +4 (>20 parasites/field) parasitaemia levels (Woo, 1977). At peak parasitaemia of day 10 post-infection (DPI) all the rats were bled and about 8ml of parasitaemic blood was pooled into a sterile conical flask containing heparin as an anticoagulant. The parasite concentration was adjusted by the addition of phosphate-buffered saline solution (Monzon *et al.*, 1990) the number of parasites in the blood was determined through the method described by Herbert & Lumsden (1976) and a volume containing approximately 2×10^6 /2ml inoculum was injected via jugular venipuncture of each goat in group II and the day was considered day 0.

Monitoring the level of parasitaemia in the RSG post-infection

After the goats were inoculated with *T. evansi* (*T. evansi*-infected RSG), the level of parasitaemia was determined using a sterile needle prick on the ear vein. The evaluation was carried out daily from day 1 post-infection to day 5 when patency was attained. Two prepared microhaematocrit tubes and two Giemsa-stained thin blood smear slides per blood sample were examined for parasitaemia as per OIE (2010) and FAO (2014) recommendations. Evaluation for the level of parasitaemia was carried out and scored as +1 [1-5 parasites (p)], 2+ (6-10p), 3+ (11-20p) and +4 (>20p) per microscopic field (Woo, 1977).

Measurements of temperature

The body temperature of all the animals in all groups was measured three times a day at 06:00, 12:00 and 18:00hrs three times a week (Mondays, Wednesdays and Saturdays) for 12 weeks. Each animal was gently restrained and a digital thermometer with a maximum gauge of 42°C (COCET®, Kangfu Medical Equipment, China accuracy +/- 0.1°C) was inserted into the colorectum of each animal and kept as such till the sound of a beep was heard and the values obtained recorded accordingly (Minka & Ayo, 2016).

Body surface temperature (BST) was taken with the aid of an infrared thermometer (Model AR330, China) at four locations; mid-head, nose, eye and coronary band. The BST was recorded at the same time that the RT was taken.

Thermo-hygrometric readings were recorded at eight-hour intervals throughout the study period using a dry- and wet-bulb thermometer (DTH 1;

Clarke International Ltd, Essex, UK), and the relative humidity (RH) was obtained using Osmond’s hygrometric table (Narindra Scientific Industries, Haryana, India). In the pen, the dry- and wet-bulb was placed in the middle of the room at 1.5m from the floor. The heat load on the goats was obtained using the thermal humidity index (THI) and calculated during the experimental period according to the method of Yousef (1985).

$$THI = (0.35t_d + 0.65t_{wb}) \times 1.8 + 32$$

where t_d is the dry-bulb temperature (°C) and t_{wb} is the wet-bulb temperature (°C).

Statistical analyses

The trigonometric statistical model of each time series was evaluated for rhythmicity by repeated measures of analysis of variance (ANOVA MODEL-3) and by the Cosinor procedure (Refinetti *et al.*, 2007; Piccione *et al.*, 2013). Four rhythmic parameters were determined; mean level (mesor), amplitude, acrophase (time at which the peak of rhythm occurs) and robustness (strength of rhythmicity). For each goat, the mean level of the rhythm was computed as the arithmetic mean of all values in the data set. The amplitude of the value rhythm was calculated as half the max-min range of the oscillation which in turn was computed as the difference between the peak and trough. The acrophase of a rhythm was determined by an iterative curve-fitting procedure based on the single Cosinor procedure. Rhythm robustness was computed as a percentage of the maximal score attained by the Chi-square periodogram. The

difference between times of measurement was tested by Student’s paired t-test, and values of $p < 0.05$ was considered significant.

Results

The AT and RH are shown in Figure 1. The maximum, minimum and mean AT values of 27.0°C, 24.2°C and 25.6±0.27°C, respectively were recorded during the rainy season of June – September, while the RH had maximum, minimum and mean values of 94.5%, 53.6% and 80.3±11.5%, respectively.

The RT of the CRSG during the study period had a minimum value of 37.5°C during the early morning hours (06:00h) and a maximum value of 39.3°C recorded during the evening hours (18:00h). The overall mean RT value of the CRSG was 38.3±0.3°C. In *T. evansi*-infected RSG, a similar pattern of RT fluctuation between the minimum and maximum was observed with maximum, minimum and mean values of 40.6°C, 37.34°C and 38.6±0.3°C, respectively. However, the RT in the infected goats were significantly higher ($P < 0.05$) than the control from the 4th week to the 12th week of the experimental period (Figure 2a).

The RT in both CRSG and *T. evansi*-infected RSG clearly displayed circadian rhythmicity throughout the experimental period. However, the pattern of rhythmicity was distorted in the infected goats from the 5th week of the experimental period (Figure 2). The result showed a descent in RT value during the early morning hours (06:00h) and an ascent during the evening hours (18:00h) in both CRSG (06:00h,

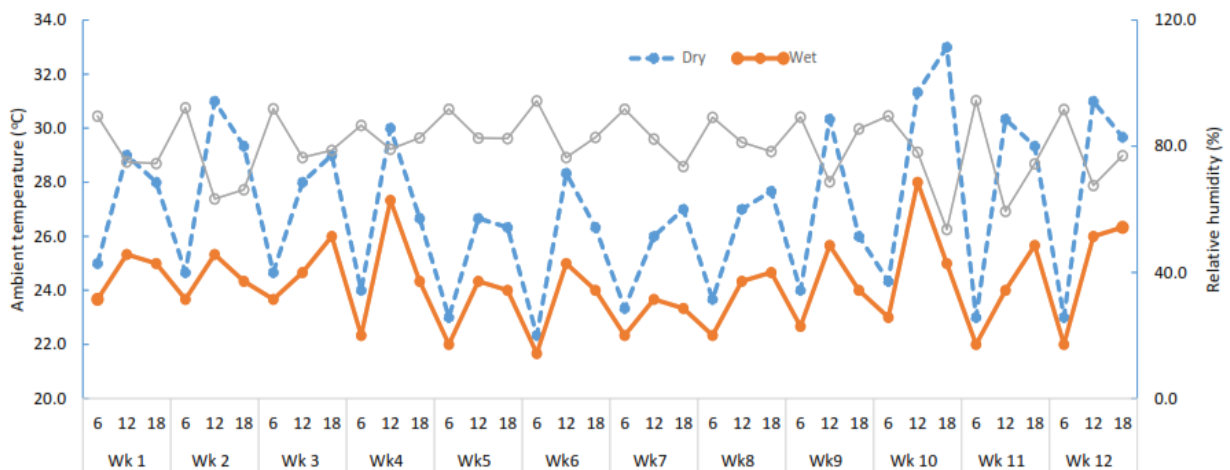


Figure 1: Hourly fluctuations of dry and wet bulb temperature and relative humidity inside the goat’s pen during 12 weeks of the experimental period. For each week measurements were done three times at 6, 12 and 18 (06:00h, 12:00h and 18:00h), respectively. Dry= dry bulb temperature; Wet= wet bulb temperature; RH= relative humidity; WK= week

37.9±0.14°C; 18:00h, 38.7±0.14°C) and *T. evansi*-infected RSG (06:00h, 39.0±0.27°C; 18:00h, 39.7±0.20°C). Figures 2a and b showed the daily rhythm and weekly fluctuation of RT of the goats. The result showed that the CRSG displayed a relatively rhythmic pattern, while in the *T. evansi*-infected RSG weekly rhythmicity was also distorted with ascent from the 5th to the 10th week of the study period. Although there is no significant difference (p>0.05) in the mean RT between CRSG and *T. evansi*-infected RSG, the RT of *T. evansi*-infected RSG from the 5th to the 10th week of the experiment was higher compared to that of CRSG.

Figure 3 shows the RT rhythmic characteristics of mesor (mean level), amplitude (half the range of excursion or extent of predictable changes) and

acrophase (time of peak). In the CRSG, the mesor (38.0°C) and amplitude (0.85°C) were lower than the *T. evansi*-infected RSG which had higher (p<0.05) mesor (38.8°C) and amplitude (1.7°C). The CRSG and *T. evansi*-infected RSG showed no difference (p>0.05) in acrophase, which was recorded at 18:00hrs (Figure 3c).

Figures 4 to 7 show the general BST of CRSG and the *T. evansi*-infected RSG. The eye region had the highest BST (37.5°C and 37.8°C for CRSG and *T. evansi*-infected RSG, respectively) while the coronary band had the lowest (32.7°C and 32.3°C for CRSG and *T. evansi*-infected RSG, respectively). In all the regions of measurement, BST exhibited clear rhythmicity. The lowest BST was recorded at 06:00hrs in both CRSG and

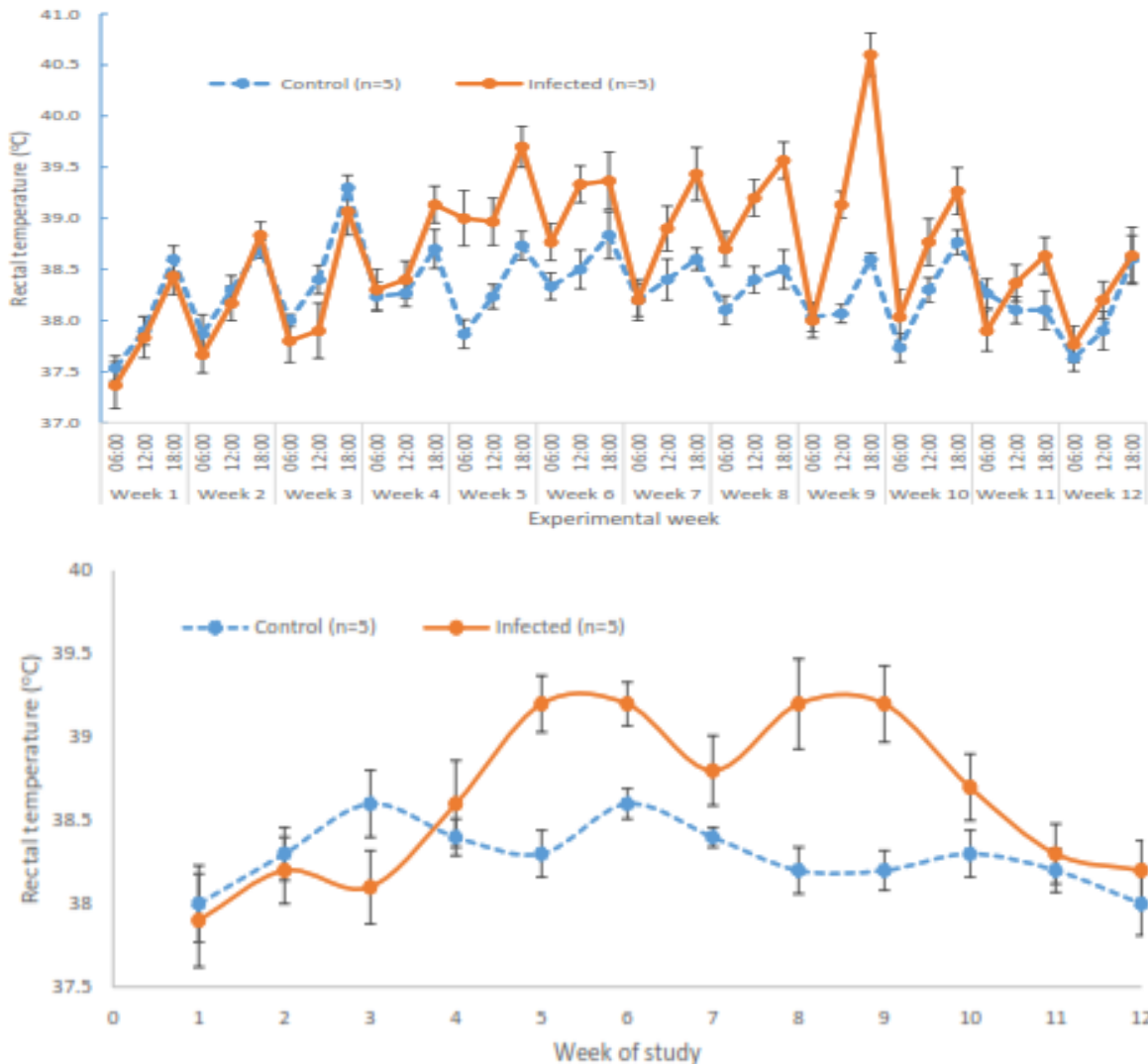


Figure 2: Daily rhythm (a) and weekly fluctuation (b) of the rectal temperature of control (n=5) and *T. evansi*-infected (n=5) goats during 12 weeks of the experimental period

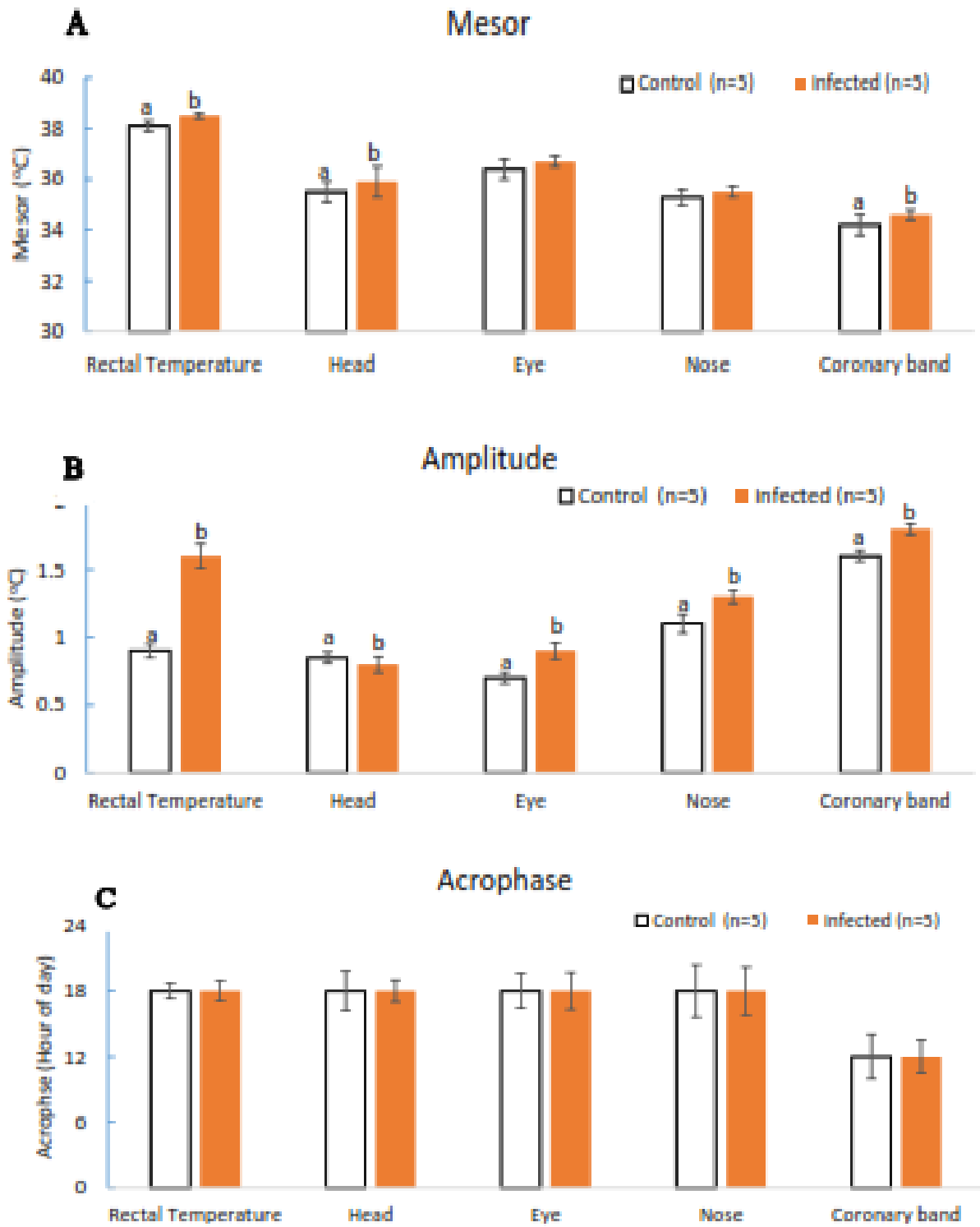


Figure 3: Characteristics of rectal temperature and body surface temperature daily rhythm in control and *T. evansi*-infected male goats. Measurements were made three times a day for a period of 12 weeks. Mean values with different superscript alphabets are significantly different at $p < 0.05$.

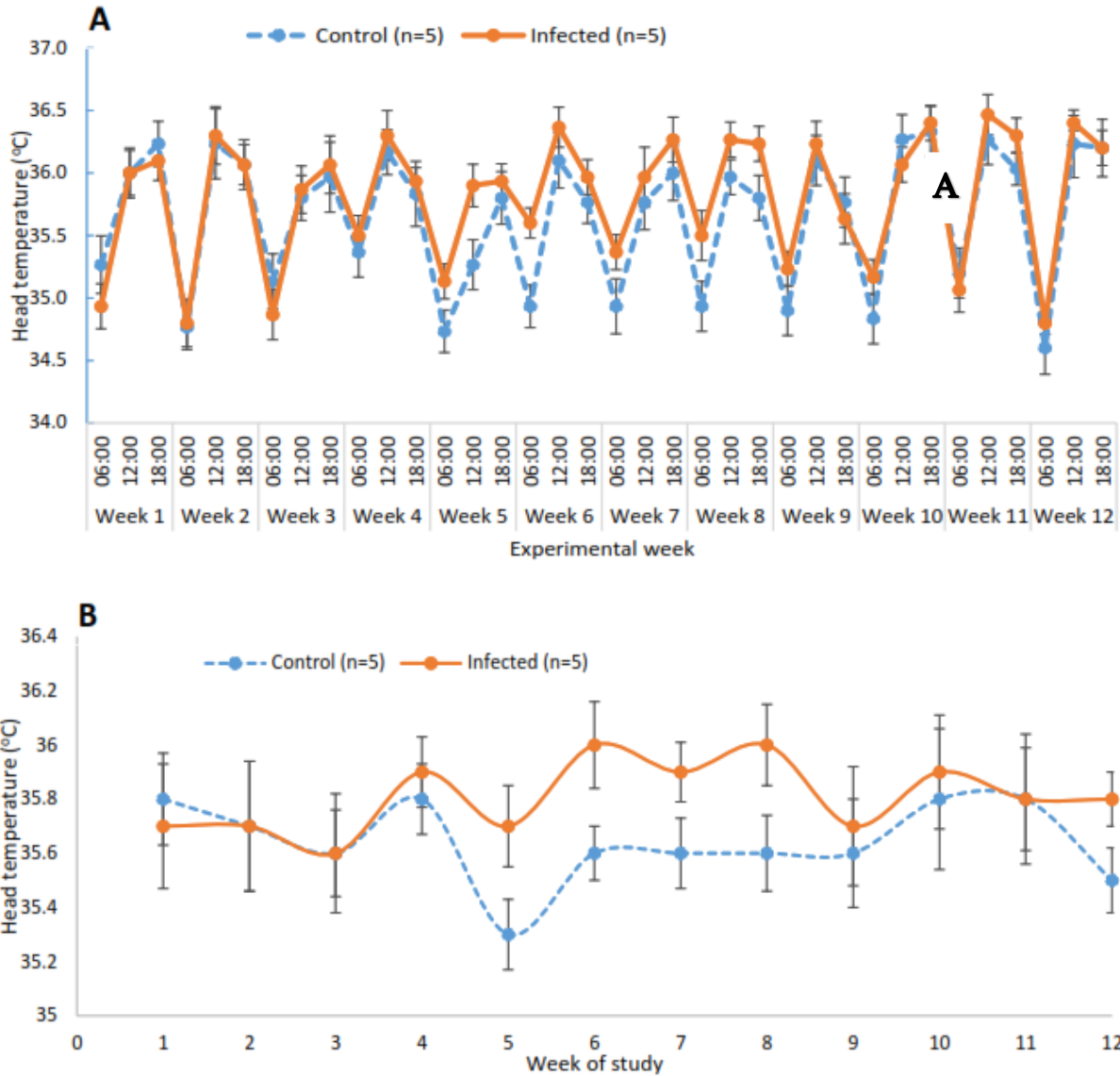


Figure 4: Daily rhythm (a) and weekly fluctuations (b) of head temperature in control (n=5) and *T. evansi*-infected (n=5) goats during 12 weeks of the experimental period

the *T. evansi*-infected RSG, while the highest BST was recorded at 12:00hrs and 18:00hrs depending on the area of measurement, especially in the *T. evansi*-infected RSG.

The mean temperature at the head region in *T. evansi*-infected RSG ($35.9 \pm 0.1^\circ\text{C}$) was higher ($p < 0.05$) than $35.4 \pm 0.4^\circ\text{C}$ recorded in CRSG.

The eye, nose and coronary band temperatures in CRSG had mean values of $36.7 \pm 0.1^\circ\text{C}$, $35.5 \pm 0.2^\circ\text{C}$ and $34.3 \pm 0.2^\circ\text{C}$, respectively, which were lower than the mean values of $36.8 \pm 0.4^\circ\text{C}$, $35.7 \pm 0.3^\circ\text{C}$ and $34.6 \pm 0.3^\circ\text{C}$ recorded in *T. evansi*-infected RSG, respectively.

The mesors of BST in all the regions recorded in the *T. evansi*-infected RSG were significantly higher ($p < 0.05$) than those of the CRSG (Figure 2a) during the 12 weeks of the study period.

Except the head region, the amplitude of the eyes, nose and coronary band was significantly higher ($p < 0.05$) in *T. evansi*-infected RSG as compared to the CRSG (Figure 2b).

The acrophase (Figure 2c) for the head, eye and nose regions was not different between the CRSG and *T. evansi*-infected RSG, which was recorded at 18:00hrs. However, the acrophase of the coronary band was at 12:00hrs for both the CRSG and the *T. evansi*-infected RSG (Figure 2c).

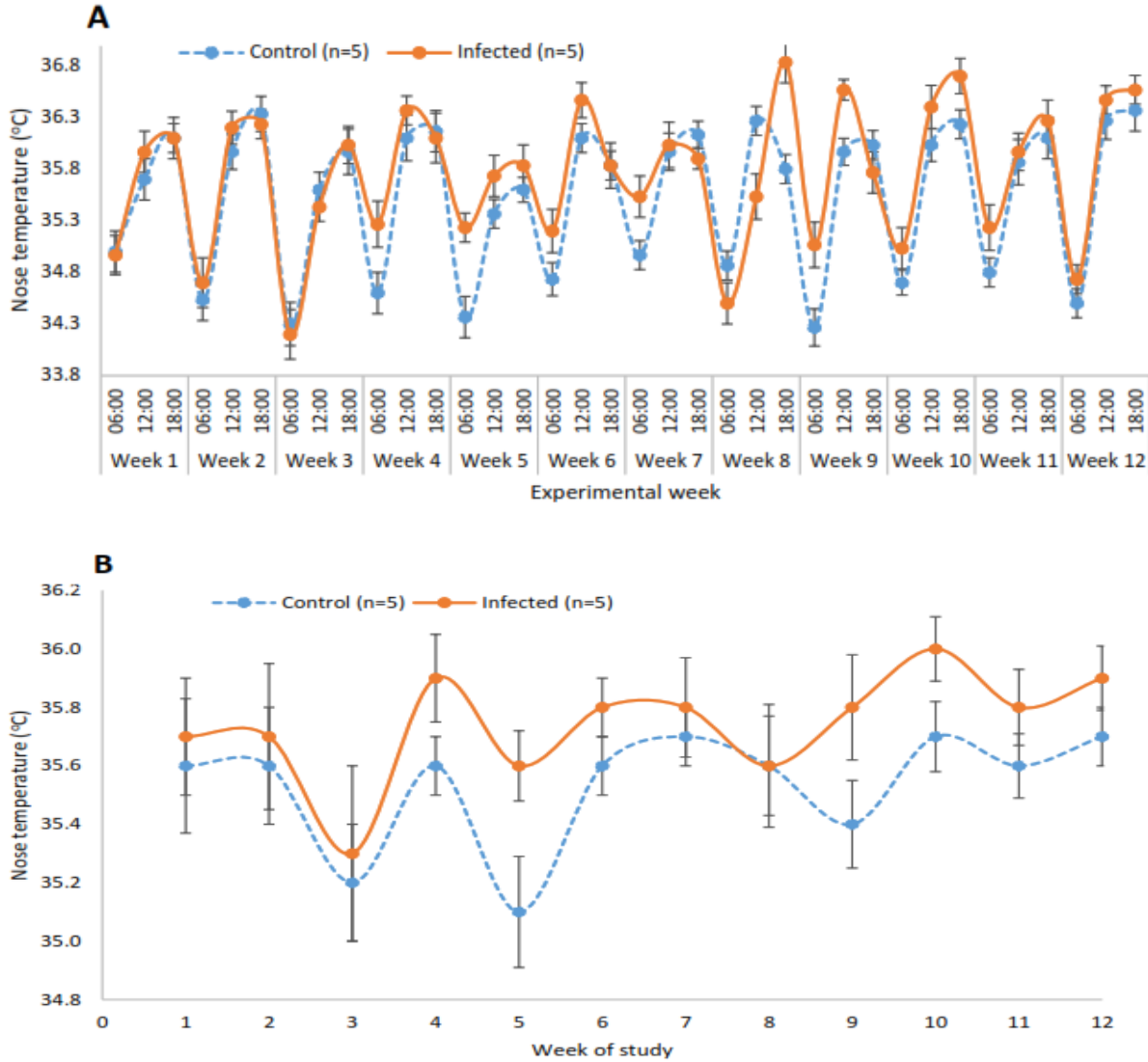


Figure 5: Daily rhythm (a) and weekly fluctuations (b) of nose temperature in control (n=5) and *T. evansi*-infected (n=5) goats during 12 weeks of the experimental period

Discussion

The mean AT recorded in this study was within the thermoneutral zone of 22 – 35°C established for sheep and goats in the region of study (Ayo *et al.*, 1998; Silanikove, 2000; Richardson, 2002; Minka & Ayo, 2016), while the mean RH of 80.3%±11.5 recorded was outside the thermoneutral zone established for sheep and goats in the tropical regions, this is because the present study was conducted during the rainy season as compared to afore-mentioned studies done during the hot-dry and Harmattan seasons.

The mean RT (38.3±0.3°C) for the CRSG and 38.6±0.3°C for *T. evansi*-infected RSG were within the normal reference values of 38-40°C for goats in the

tropics (Igono *et al.*, 1982; Ayo *et al.*, 1998; Minka & Ayo, 2016).

By the application of a periodic Cosinor model, the RT obtained under experimental conditions indicated that clear circadian rhythmicity does exist in both the CRSG and *T. evansi*-infected RSG. Previous studies conducted on sheep and goats’ body temperature reveal a daily rhythm with an ascent phase during the day and a descend phase during the night (Piccione *et al.*, 2002a; Piccione *et al.*, 2002b; Piccione & Refinette, 2003; D’Alterio *et al.*, 2012; Minka & Ayo, 2016), which was similar to the results of the present study. The present study, for the first time, established the fact that circadian rhythmicity of body temperature is not abolished under *T. evansi*

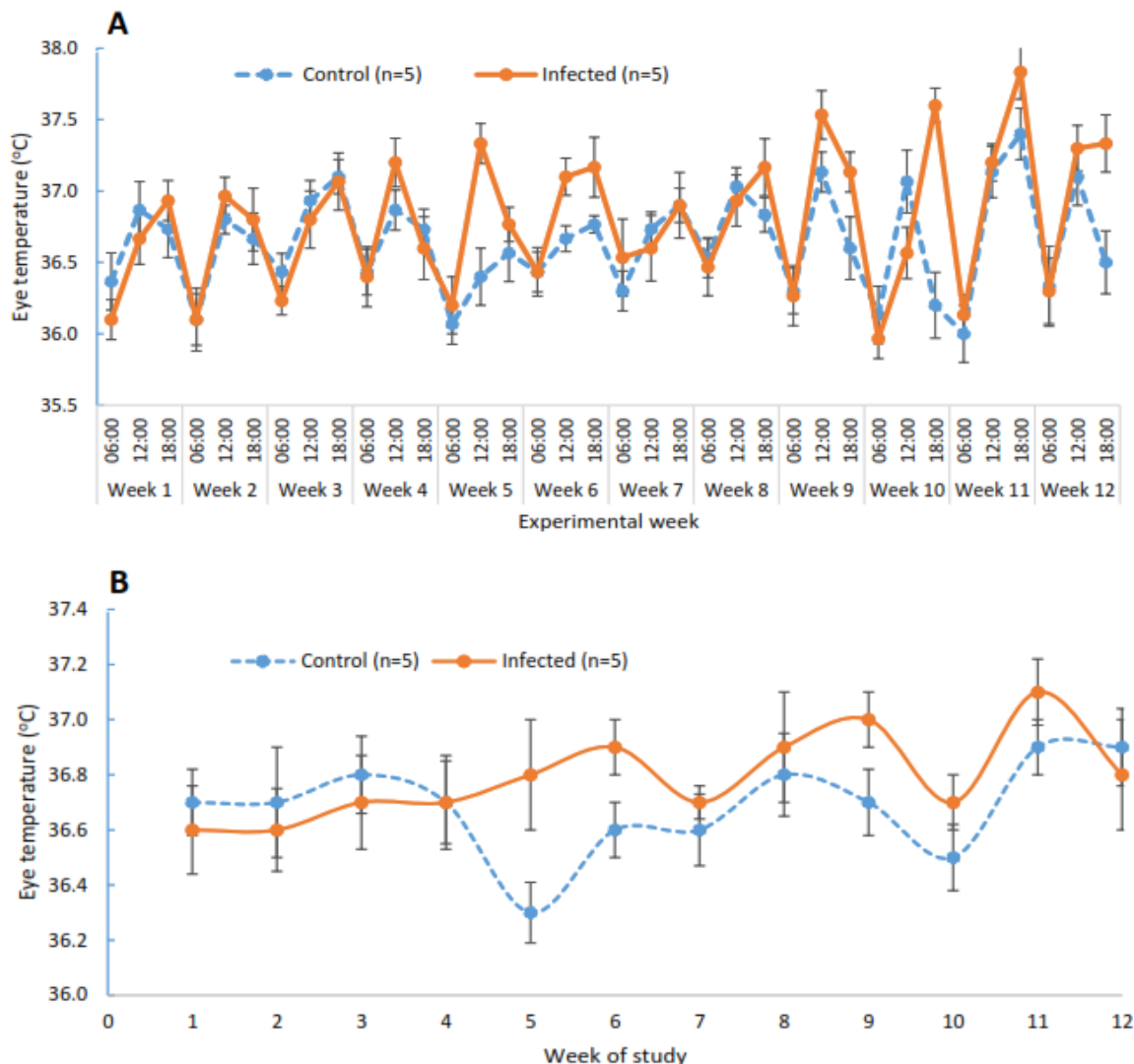


Figure 6: Daily rhythm (a) and weekly fluctuations (b) of eye temperature in control (n=5) and *T. evansi*-infected (n=5) goats during 12 weeks of the experimental period

infection. Although the overall mean RT of both CRSG and *T. evansi*-infected RSG were not different during the study period, the maximum RT value of 40.6°C recorded in *T. evansi*-infected RSG was higher than the value of 39.3°C recorded in CRSG and also higher than the normal RT value of 38.0 to 40°C recorded in Red Sokoto goats (Ayo *et al.*, 1998; Minka & Ayo, 2016). This observation indicated that infection with *T. evansi* was responsible for the increase in RT value especially from the 5th week of the experimental study. The RT in the *T. evansi*-infected RSG were significantly higher ($p < 0.05$) than the control from the 4th to the 10th week of the experimental period. The RT and infrared thermometry of BST in both control RSG and *T. evansi*-infected RSG exhibited individual variations. The RT and BST data of the

control RSG is in agreement with previous studies (Piccione *et al.*, 2013; Minka & Ayo, 2016), while there is a paucity of information to support the *T. evansi*-infected RSG. Although, D’Alterio *et al.*, 2012 showed a distortion in daily rhythmic pattern in sheep with foot rot, no data was available for BST studies nor on animals with systemic infections. The application of the periodic model and fitting of the Cosinor procedure showed that the RT exhibited a strong daily and weekly rhythmicity while the BST exhibited a weak daily and weekly rhythmic pattern in both the CRSG and *T. evansi*-infected RSG. The results recorded in the CRSG agree with previous studies in horses, sheep and goats (D’Alterio *et al.*, 2012; Minka & Ayo, 2016; Piccione *et al.*, 2013) and in Red Sokoto goats recorded only during the day time

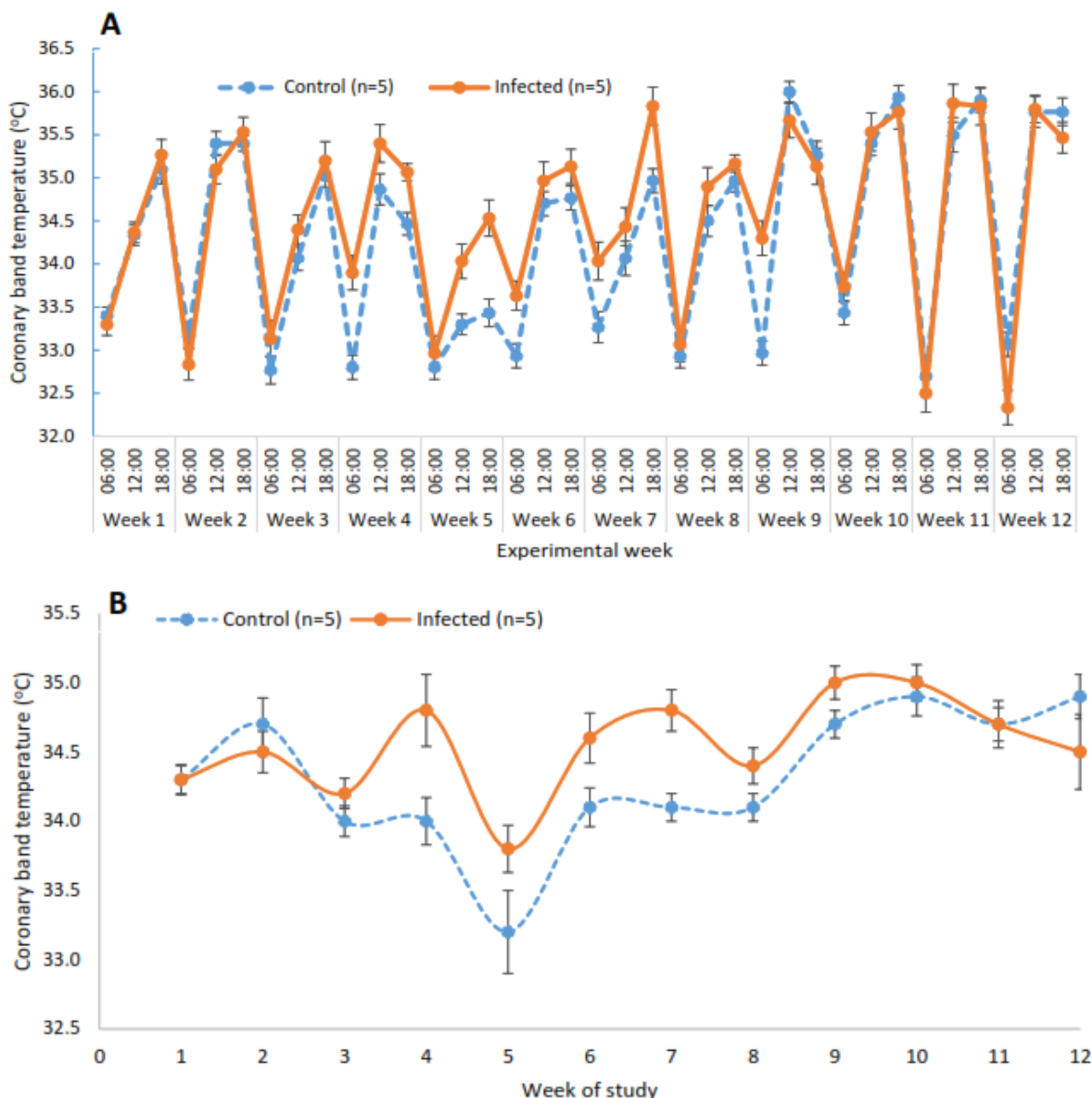


Figure 7: Daily rhythm (a) and weekly fluctuations (b) of coronary band temperature in control (n=5) and *T. evansi*-infected (n=5) goats during 12 weeks of experimental period

(Ayo *et al.*, 1998). However, there is a paucity of data on the RT and BST of *T. evansi*-infected RSG. The oscillatory pattern in RT and BST differed in both the groups under study.

The result for the first time showcased the importance of rhythmicity study, which clearly showed at what period of the day the effect of *T. evansi* infection on RT was at its peak. The result indicates that it will be misleading to rely on mean values of RT when diagnosing *T. evansi* infection. In addition, the present result showed that *T. evansi* infection in goats takes about 4 weeks before the

body temperature is elevated above normal, with a relapse between the 11th and 12th week. These circadian rhythm changes are probably due to a rhythmic input from the suprachiasmatic nucleus acting upon the hypothalamic thermoregulatory centres modulating the set point (Refinetti, 2006). This set point, in thermal physiological processes involved in diseases producing temperature changes. As body temperature rises, various thermoregulatory responses are activated to increase body heat production and reduce heat loss, leading to a temperature elevation. Conversely, various

responses are also later activated to help reduce heat gain and increase heat loss, leading to a return of temperature to its initial nonfebrile state.

The change in body temperature of the *T. evansi*-infected RSG can simply be explained thus, the set point is elevated during the rising phase and returned to normal during the later phase (Tornatzky and Miczek, 1993; Refinetti, 2006; D'Alterio *et al.*, 2012). The mesor of RT and BST recorded in *T. evansi*-infected RSG were higher than those of CRSG, except for the eye and nose. For the coronary band the thermoregulation of the foot was compromised during *T. evansi* infection which was similar to the higher coronary band temperature observed in sheep and cattle infected with foot rot and foot – and – mouth disease respectively (Gloster *et al.*, 2011; D'Alterio *et al.*, 2012). The high temperature seen in the head may be due to the disease increasing brain activities. The higher mesor demonstrated the effect of fever on the thermoregulatory processes in Red Sokoto goats suffering from *T. evansi* infection. The coronary band temperature when compared to RT of CRSG and *T. evansi*-infected RSG were cooler and the same when compared to temperatures in other parts of the body recorded. The lower coronary band temperature recorded can be attributed to the fact that the feet play a more crucial role in thermoregulation than other body parts, as hoof temperature is affected by ambient temperature and posture/activity (Gloster *et al.*, 2011), thereby making it unreliable as an organ to be measured for a changing body temperature in a haemoparasite infection.

The greater amplitude observed in RT, eye, nose and coronary band in *T. evansi*-infected RSG suggests that *T. evansi* infection increases the oscillatory pattern of this parameter. This may be a response of the RT and BST to the infection. Similar increases in amplitude, although not under infection, were reported in mammals under extreme cold or hot environmental conditions (Piccione and Caola, 2002; Minka & Salka, 2016). The RT amplitude values of 0.8 – 1.7°C recorded in the present study were similar to the values of 0.4 – 1.9°C reported in goats (Ayo *et al.*, 1998). In homeothermy, the limit of the variability of RT is +/-2°C (Cabanac & Simon, 1987), which is wider than the amplitude of the present study.

The acrophase, of RT and BST observed at 18:00hrs were not different between the groups except the coronary band, whose acrophase in both groups was at 12:00hrs. The result suggests that *T. evansi* infection did not delay or hasten the peak rise in RT and BST. This result differs from those recorded in non-infected sheep and goats during the Harmattan

and hot-dry seasons (Minka & Ayo, 2016), where the acrophases were between 17-21hrs. The difference in the present result may be because the present experiment was conducted under a thermoneutral AT. Thus, it can be assumed that AT had a more devastating effect on acrophases of RT and BST than infection. The lower coronary band acrophase at 1200hrs confirms the fact that feet temperature peak several hours before RT, suggesting that the mechanism of feet thermoregulation may be involved in the production of the oscillation in RT as earlier suggested (Kraüchi & Wirz-Justice, 1994; Shechter *et al.*, 2011)

In conclusion, the results of the present study show that RT and BST exhibits circadian rhythm, around a set point in both control RSG and *T. evansi*-infected RSG. The oscillatory pattern, mesor and amplitude in *T. evansi*-infected RSG were higher than in control. The results obtained from our study may be useful for the early screening, diagnosis and targeted treatment and prevention of protozoan diseases.

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Conflict of Interest

The authors declare that there is no conflict of interest.

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