—Original Article—

Diurnal and Seasonal Fluctuations in Rectal Temperature, Respiration and Heart Rate of Pack Donkeys in a Tropical Savannah Zone

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The study was designed to determine diurnal and seasonal changes in basic physiologic responses of donkeys adapted to the tropical Savannah. The rectal temperature (RT), respiratory rate (RR) and heart rate (HR) of six male Nubian pack donkeys, and the dry-bulb temperature (DBT), relative humidity and heat index of the experimental site were concurrently recorded hourly, from 06:00 h to 18:00 h (GMT +1), for three days, spread 1 week apart, during the cold-dry (harmattan), hot-dry and humid (rainy) seasons, in an open grazing field. Values of the physiologic parameters recorded during the morning (06:00 h–11:00 h) were lower (P<0.001) than those obtained in the afternoon (12:00 h–15:00 h) and evening (16:00 h–18:00 h) hours in all seasons, but the robustness of the diurnal rhythm differed (P<0.05) between seasons. Many diurnal hourly DBT mean values recorded during the harmattan and hot-dry seasons fell outside the established thermoneutral zone for tropically-adapted donkeys, while those obtained during the rainy season were within the zone, indicating that the dry seasons were more thermally stressful to the donkeys than the humid season. Overall mean RT dropped (P<0.05) during the harmattan season. The RR rose, while HR dropped (P<0.001) during the hot-dry season. In conclusion, daytime and season had profound influence on RT, RR and HR of the donkeys, therefore, diurnal and seasonal variations should be taken into account during clinical evaluation before reaching conclusion on health status and fitness for work in donkeys.

Key words: *clinical evaluation, fitness, physiologic parameters, stress, thermal load*

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Donkeys (*Equus asinus, Perissodactyla*) in the tropics and sub-tropics are used for riding, packing and draught power [13, 21]. They survive, reproduce and produce meat and milk in arid and semi-arid regions under adverse environmental conditions [2, 22]. The donkeys are increasingly recognized by small-holder farmers in their selection of working animals due to the survival advantage they have over cattle [23]. The environment exerts effects on donkeys, but there are wide differences among individuals so that selection and adaptation can greatly improve climatic tolerance [4, 19, 23]. Therefore, it is important to study the physiologic responses of donkeys to changing meteorological conditions, which may form a basis for improving their work output. The harmattan is a cold-dry and dust-laden West African north-east trade wind, which blows southward from the Sahara desert into the Gulf of Guinea, between the end of November and the middle of March [3]. In northern Nigeria, the harmattan season is characterized by a wide range of dry-bulb temperatures (DBTs), from low in the night to high in the afternoon, and very low relative humidity (RH); the hot-dry season, which is a transition period between the dry and rainy seasons, is characterized by a consistently high DBT and low to medium RH; while the rainy season is predominated by medium-level DBTs and high RH [1, 6, 11, 15]. Igono *et al*. [16] suggested that the harmattan season (November–February) may be thermally more stressful than either the hot-dry (March–April) or rainy (May–October) season because of its wider range of

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circadian DBTs. Vathana *et al.* [23], however, indicated that the hot-humid transition period from dry to rainy season was more thermally stressful to animals in the tropics. Recent studies on seasonal changes in physiologic responses and production show that the transition period (hot-dry season) in the Northern Guinea Savannah zone of Nigeria is more thermally stressful to poultry [5] and rodents [10, 12]. However, the physiologic responses of donkeys to seasonal changes in the climatic zone have not been investigated.

Extreme weather has a direct effect on animal health, growth and reproduction [5, 7]. Very often in the tropical and sub-tropical regions, animals are subjected to the influence of extreme thermal environment factors of high DBT and high RH, resulting in heat stress [4, 25]. Hence, the heat index (HI), which is a measure of the combined thermal effects of DBT and RH, is a useful tool in evaluating the actual environmental thermal load on animals in the tropics [10–12, 20]. It has been established that rectal temperature (RT), respiratory rate (RR) and heart rate (HR) are important physiologic indices in determining the responses of donkeys to changes in thermal environmental conditions [4, 19, 21]. The parameters can be measured easily, even in rural areas.

The aim of the present study was to investigate the basic physiologic changes in RT, RR and HR of pack donkeys during the day and across the seasons, prevailing in the Northern Guinea Savannah zone of Nigeria. Inferences derived from the data obtained may be useful in identifying diurnal and/or seasonal changes in the physiologic parameters, which may aid accurate diagnosis during clinical evaluation.

Materials and Methods

Description of experimental site

The experiment was performed in a natural pasture, which was an open grazing field in Samaru (11°10/N, 07°38/E), Zaria, located at an altitude of 686 m above maximum sea level [17] and in the Northern Guinea Savannah zone of Nigeria, during the harmattan, hot-dry and rainy seasons [15].

Animals and management

Six healthy male Nubian pack donkeys, aged between 10 and 14 years, served as subjects of the experiment. The live weights of the donkeys ranged from 103–145 kg. The donkeys were reared under the traditional extensive management system, with no shelter throughout the day. They were fed 2.5 kg of dry cereal milling and cowpea pods with groundnut hay, and given water after feeding in the morning (06:00 h–08:00 h). Thereafter, from 10:00 h–18:00 h, the donkeys were released to graze on natural pasture, comprising mainly Gamba grass (*Andropogon gayanus*) for the remaining part of the day, while the experiment lasted. At the end of the day, the donkeys were kept in an enclosure with no shed to serve as protection in the night. Thus, they were exposed daily to direct sunlight, rainfall, wind and other meteorological factors all the year round. The donkeys were not subjected to packing (carrying load) on the experimental days and no medication was given throughout the experimental period.

Experimental design and measurement

The experiment was conducted during the peak of the harmattan (13th and 20th January, and 10th February), hot-dry (5th, 12th and 19th April) and rainy (16th, 23rd and 30th August) seasons. Measurements of the RT, RR and HR were taken hourly from 06:00 h to 18:00 h (GMT +1) on each experimental day as described by Ayo *et al.* [4]. Briefly, each donkey was restrained lightly and all the measurements from the animals were completed within five minutes. The RT was recorded as an indicator of body temperature using a digital thermometer (COCET, Kangfu Zhejiang Yuequing, China), inserted about 3.5 cm into the rectum via the anus until a beeping (alarm) sound was heard, indicating that the recording was completed. The RR was taken by observing and counting the number of respiratory flank movements for one minute. The HR was recorded through auscultation of the heart by placing a stethoscope (Sprague, Rappaport Type Stethoscope, England) between the fourth and fifth rib on the left side of each donkey and counting the number of heart beats per minute.

The dry- and wet-bulb temperatures were recorded, concurrently with the RT, RR and HR measurements, every hour, at the experimental site using a dry- and wet-bulb hygrometer (Mason's type, Zeal, England). The DBT reading indicated the ambient or air temperature, while RH was deduced from the dry- and wet-bulb temperatures using conversion tables, condensed from the Bulletin of the US Weather Bureau No. 1071. Other meteorological data (wind speed, WS, or velocity; wind direction, WD; and rainfall) from the study period were collated from the Institute for Agricultural Research, Ahmadu Bello University, Samaru, Zaria, Nigeria, located at a distance of about 2 km from the experimental site. The HI was calculated from the DBT and RH values using an HI calculator (National Weather Service Hydrometeorological Prediction Centre, Meteorological Conversions and Calculations, www.hpc.ncep.noaa.gov/html/heatindex.shtml, accessed on April 20, 2012).

Statistical analysis

The data obtained were expressed as mean ± standard deviation of the mean (Mean ± SD). Diurnal and seasonal differences in mean values of DBT, RH and HI were determined by employing repeated-measures analysis of variance (ANOVA), while one-way ANOVA was used for the RT, RR and HR, followed by Tukey's multiple comparison *post-hoc* test in each case. GraphPad Prism version 4.3 for Windows (GraphPad Software, San Diego, California, USA) was used for the analyses. Values of $P<0.05$ were considered significant.

Results

Meteorological conditions

Table 1 shows average meteorological conditions at the study site on experimental days during the harmattan, hot-dry and rainy seasons. The mean WS recorded during the harmattan season was significantly ($P<0.05$) higher than values obtained during the hot-dry and rainy seasons. WD was predominantly north-east (NE) and south-east (SE) during the harmattan season; south-east, south-west and north-west during the hot-dry season and south-west during the rainy season. Rainfall (30.9 mm) was recorded only on the experimental day 3 of the rainy season. The highest seasonal mean values of DBT and HI were recorded during the hot-dry season, while the least were both obtained during the harmattan season ($P<0.001$). The RH was least and highest ($P<0.001$) during the harmattan and rainy seasons, respectively.

Results of the diurnal variations in mean DBT, RH and HI values of the experimental site during the harmattan, hot-dry and rainy seasons are represented in Fig. 1. In all seasons, the DBT and HI values were least ($P<0.05$) during the morning (06:00 h–11:00 h) compared with the afternoon (12:00 h–15:00 h) and evening (16:00 h–18:00 h) hours, while the reverse was the case with RH. However, the robustness of the diurnal slope differed ($P<0.05$) between the seasons. The diurnal slopes of DBT and HI values were bottommost (and steeper), moderate and topmost during the harmattan, rainy and hot-dry seasons, respectively; but the afternoon and evening DBT values during the harmattan and rainy seasons did not differ ($P>0.05$). The diurnal slope of RH values during the hot-dry season was the steepest ($P<0.05$), while those of the harmattan and rainy seasons were bottommost and topmost, respectively. The diurnal DBT values (Fig. 1a) were consistently higher ($P<0.0001$) during the hot-dry season, while the lowest values ($P<0.001$) were recorded during the morning hours of the harmattan season. The lowest diurnal mean DBT value of 15.2 ± 2.9°C was recorded at 07:00 h, during the harmattan season; while the highest value of 37.7 ± 2.3°C was obtained at 15:00 h, during the hot-dry season. Afternoon DBT values differed only slightly ($P>0.05$) between the harmattan and rainy seasons. Diurnal mean RH values (Fig. 1b) were low (<22%), low to moderate (19–65%) and high (>79%) during the harmattan, hot-dry and rainy seasons, respectively ($P<0.0001$); the RH diurnal slope was more robust during the hot-dry season. The diurnal slope of the HI (Fig. 1c) followed the same pattern with that of the DBT, except that afternoon and evening HI values differed ($P<0.05$) between the harmattan and rainy seasons. Diurnal HI mean values throughout the day were lowest and highest ($P<0.001$) during the harmattan and hot-dry seasons, respectively.

Physiological parameters

Results on diurnal variations in mean values of RT, RR and HR of the pack donkeys during the harmattan, hot-dry and rainy seasons are illustrated in Fig. 2. In all the seasons, values of the RT, RR and HR recorded during the morning were lower ($P<0.001$) than those obtained in the afternoon and evening hours, but the robustness of the diurnal slope differed ($P<0.05$) between the seasons. The least and highest diurnal mean RT values of 34.8 ± 1.0°C and 38.2 ± 0.6°C were obtained at 09:00 h and 16:00 h, respectively, all during the harmattan season (Fig. 2a). Diurnal mean RR values (Fig. 2b) rose from a nadir of 11.1 ± 1.8 breaths/min at 06:00 h during the rainy season, to a zenith value of 52.7 ± 9.4 breaths/min at 12:00 h during the hot-dry season. The least diurnal mean HR value of 36.5 ± 6.2 beats/min was recorded at 09:00 h during the hot-dry season, while the peak value of 58.8 ± 15.8 beats/min was obtained at 16:00 h during the rainy season.

Results of seasonal variations in physiological parameters of the donkeys are presented in Fig. 3. The seasonal nadir and zenith RT mean values were obtained during the harmattan and rainy seasons, respectively ($P<0.05$); the overall RR mean value was least during the rainy season and peaked during the hot-dry season ($P<0.001$); and the HR mean value was lowest and highest during the hot-dry and rainy seasons, respectively ($P<0.001$).

Behavioural observation

The donkeys were observed to shiver during the early morning hours of the harmattan season, when DBT values fell below 20°C.

Discussion

The results showed that the diurnal pattern of physiologic parameters of the donkeys followed that of the DBT and HI during all the seasons, with higher values in the afternoon and evening than morning hours, suggestive of positive influence from the thermal environmental factors. Although diurnal variations in physiologic parameters of homeotherms are internally driven, they are adjusted (entrained) to the environment by external cues, particularly photoperiod and ambient temperature [8]. The monthly mean photoperiod of the study area ranges from a nadir of 11.49 hr in December (harmattan) to a peak value of 12.76 hr in June (rainy

Table 1. Average meteorological conditions at the study site on experimental days during the harmattan, hot-dry and rainy seasons

Meteorological Parameter	Season		
	Harmattan (January)	Hot-dry (April)	Rainy (August)
Wind speed (km/day)*	240.5 ± 31.8[a]	151.5 ± 24.9[b]	122.8 ± 21.9[b]
Wind direction*	North-east & south-east	South-east, south-west & north-west	South-west
Rainfall (mm)*	0.0 ± 0.0	0.0 ± 0.0	10.3 ± 17.8
Dry-bulb temperature (°C)	23.3 ± 6.3[a]	33.2 ± 3.9[b]	25.6 ± 2.8[c]
Relative humidity (%)	16.5 ± 4.4[a]	34.5 ± 19.9[b]	88.4 ± 5.7[c]
Heat index (°C)	21.6 ± 6.1[a]	33.5 ± 4.4[b]	27.7 ± 6.2[c]

*Data collated from the Meteorological Section, Institute for Agricultural Research, Ahmadu Bello University, Zaria, located 2 km from the experimental site. Values are means ± standard deviation. Means (±SD) with different superscript letters ([a, b] and/or [c]) are significantly different ($P<0.05$).

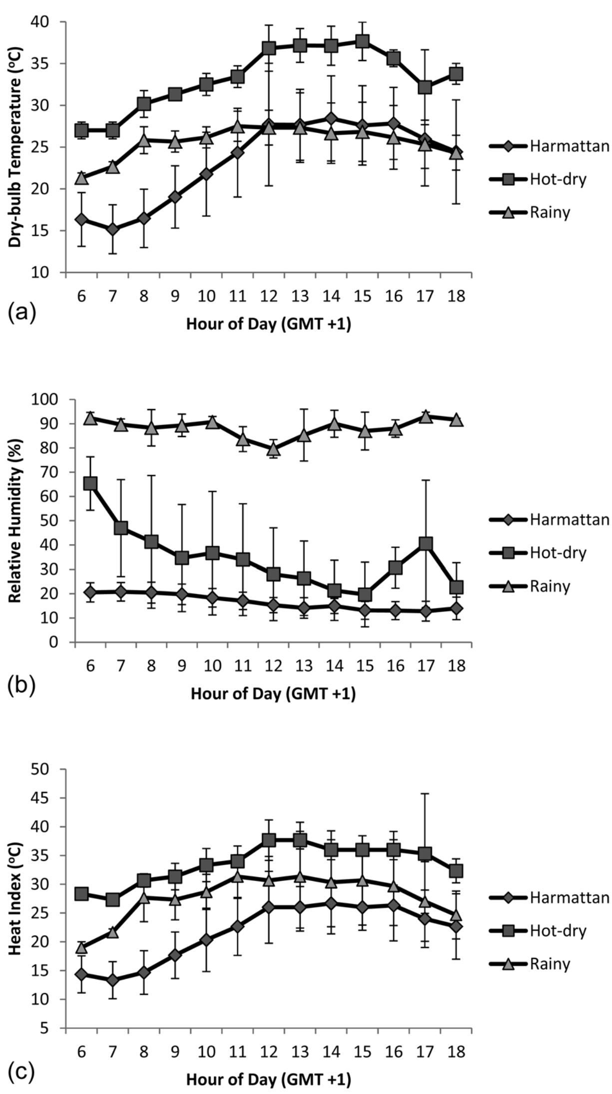

Fig. 1. Diurnal variations (Mean ± SD) in (a) dry-bulb temperature, (b) relative humidity and (c) heat index at the experimental site during the harmattan, hot-dry and rainy seasons.

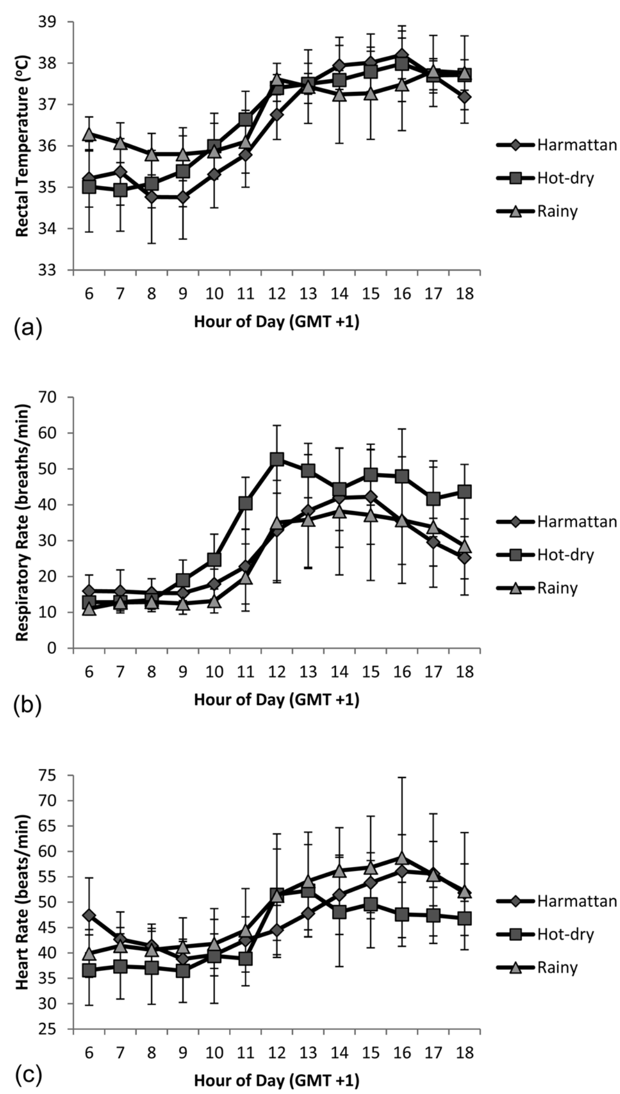

Fig. 2. Diurnal variations (Mean ± SD) in (a) rectal temperature, (b) respiratory rate and (c) heart rate of pack donkeys (n=6) during the harmattan, hot-dry and rainy seasons.

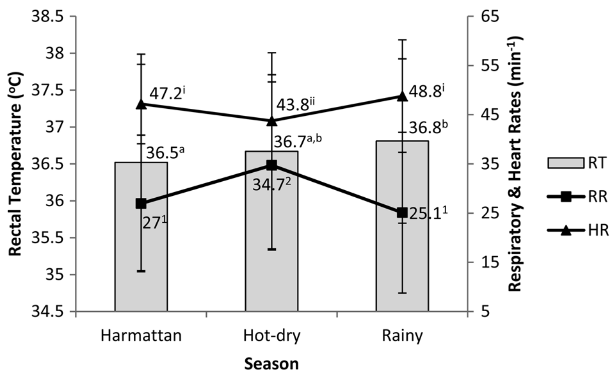

Fig. 3. Seasonal variations (Mean ± SD) in rectal temperature (RT), respiratory rate (RR) and heart rate (HR) of pack donkeys (n=6). Mean values in the same series with different and/or not sharing superscript letters (^{a,b,c}) or numbers ($^{i,ii,iii/1,2,3}$) are significantly different (P<0.05).

season) [17]. Thus, seasonal changes in photoperiod and DBT values may explain the differences in the steepness and positioning of the diurnal slopes of the parameters between the seasons, observed in the present study. For instance, the diurnal slopes of both DBT (Fig. 1a) and RT (Fig. 2a) were more flat during the rainy season compared with the dry seasons. Although the diurnal slopes of the DBT, RH and HI were largely wide apart across seasons (Fig. 1), those of the RT (Fig. 2a) were largely interwoven, indicating that the donkeys employed thermoregulatory mechanisms to achieve homeothermy during the day across the seasons. Despite the fact that many mean diurnal DBT values fell outside the established thermoneutral zone (22–32°C) for tropically-adapted donkeys [13] during the dry seasons (harmattan and hot-dry), the corresponding hourly mean RT values were all within the normal range (35–39°C) [13] throughout the day during all the seasons, indicating that the donkeys have successfully adapted to the thermal stress of the seasons in the Northern Guinea Savannah climate.

Mean DBT values below the lower critical temperature for tropically-adapted donkeys (22°C) were recorded during the early morning hours (06:00–09:00 h) of the harmattan season, while values above the upper critical temperature (32°C) were obtained from the late morning till evening hours (10:00–18:00 h) of the hot-dry season, indicating cold and heat stresses, respectively. In addition, the donkeys were observed to shiver during the morning hours of the harmattan season, when DBT values fell below the lower critical temperature, indicating that they were cold-stressed and employed shivering thermogenesis to maintain homeothermy [14]. The consistently higher diurnal RR mean values obtained during the hot-dry season signified that the donkeys employed respiratory evaporative heat loss to maintain homeothermy during the hot season. The results demonstrated that the dry seasons were more thermally stressful to the pack donkeys than the humid season, and that the donkeys have successfully adapted to the seasons. Igono *et al*. [16] compared the dry seasons and concluded that the harmattan was more thermally stressful than the hot-dry season in goats because of the higher range of DBT and RT values recorded in the former. Similar findings were observed in the present study, but the cold spell experienced during the harmattan season was opined to be less stressful compared to the heat of the hot-dry season, based on physiologic [9, 10], feed [12] and performance [5, 25] indices in some homeotherms in the same zone.

It was also observed in the present study that, whereas diurnal variations in RT, RR and HR followed the same trend as DBT and HI, seasonal variations in RT followed the pattern of the RH, being lowest and highest (P<0.05) during the harmattan and rainy seasons, respectively. There are two possible reasons for this. First, the relatively dry atmosphere of the harmattan season (mean RH, 16.49 ± 4.44%) favoured rapid evaporation of water with accompanying latent heat of vaporisation [10] from the donkeys. Second, the relatively lower DBT and HI during the harmattan season increased the temperature gradient between the donkeys and the environment, causing rapid increase in sensible heat loss via radiation, convection and conduction [10, 16], thus, lowering the RT.

The results also suggest that the donkeys were efficiently adapted to the high ambient temperatures of the hot-dry season, since there was no corresponding rise in RT during the season. The very highly significant (P<0.001) rise in RR during the hot-dry season was, apparently, a respiratory evaporative heat loss mechanism, which prevented a rise in RT. The findings support reports [4, 13] that increased respiratory rate, which increases evaporative heat loss from the lungs, and sweating are the most important means by which donkeys lose heat and maintain normal body temperature. No sweating was observed in the present study probably due to the dry nature of the hot season, which encouraged swift evaporation of sweat, as well as the fact that the donkeys were experimented upon under resting conditions. The HR dropped to a nadir during the hot-dry season (Fig. 3), when seasonal ambient temperatures rose to a peak (Table 1). The result agreed with the findings of Steven *et al*. [24] that seasonal ambient temperatures and HR have an inverse relationship, which may be attributed to seasonal decrease in feed intake [12, 18].

It is concluded that day time and season of the year have profound influence on the RT, RR and HR of donkeys in the Northern Guinea Savannah zone of Nigeria. It is, therefore, recommended that diurnal and seasonal variations in the physiologic parameters should be considered during clinical evaluation before reaching conclusion on health status and fitness for work in pack donkeys.

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References

1. Abdulhamid, G. 2011. Relationship between some meteorological parameters in Ilorin, North Central Nigeria. *Comp. Eng. Intell. Sys.* **2**: 46–52.
2. Aganga, A.A., and Aganga, A.O. 2003. Carcass analysis and meat composition of the donkey. *Pak. J. Nutr.* **2**: 138–147. [CrossRef]
3. Anuforom, A.C., Akeh, L.E., Okeke, P.N., and Opara, F.E. 2007. Inter-annual variability and long-term trend of UV-absorbing aerosols during harmattan season in sub-Saharan West Africa. *Atmos. Environ.* **41**: 1550–1559. [CrossRef]
4. Ayo, J.O., Dzenda, T., and Zakari, F.O. 2008. Individual and diurnal variations in rectal temperature, respiration and heart rate of pack donkeys during the early rainy season. *J. Equine Vet. Sci.* **28**: 281–288. [CrossRef]
5. Ayo, J.O., Obidi, J.A., and Rekwot, P.I. 2011. Effects of heat stress on the well-being, fertility and hatchability of chickens in the Northern Guinea Savannah zone of Nigeria: a review. *ISRN Vet. Sci.* doi:2011838606.
6. Ayo, J.O., Oladele, S.B., and Fayomi, A. 1996. Effects of heat stress on livestock production: a review. *Nig. Vet. J.* (Special Edition). **1**: 58–68.
7. Bianca, W. 1976. The signifiance of meterology in animal production. *Int. J. Biometeorol.* **20**: 139–156. [Medline] [CrossRef]
8. Cable, N.T., Drust, B., and Gregson, W.A. 2007. The impact of altered climatic conditions and altitude on circadian physiology. *Physiol. Behav.* **90**: 267–273. [Medline] [CrossRef]
9. Djordjević, J., Djurašević, S., Vučković, T., Jasnić, N., and Cvijić, G. 2006. Effect of cold and heat stress on rat adrenal, serum and liver ascorbic acid concentration. *Arch. Biol. Sci. Belgrade* **58**: 161–164. [CrossRef]
10. Dzenda, T., Ayo, J.O., Lakpini, C.A.M., and Adelaiye, A.B. 2011. Diurnal, seasonal and sex variations in rectal temperature of African Giant rats (*Cricetomys gambianus*, Waterhouse). *J. Therm. Biol.* **36**: 255–263. [CrossRef]
11. Dzenda, T., Ayo, J.O., Lakpini, C.A.M., and Adelaiye, A.B. 2011. Seasonal and sex variations in live weights of captive African Giant rats (*Cricetomys gambianus*, Waterhouse) in the Northern Guinea Savannah zone of Nigeria. *Int. J. Zool. Res.* **7**: 49–58. [CrossRef]
12. Dzenda, T., Ayo, J.O., Lakpini, C.A.M., and Adelaiye, A.B. 2013. Seasonal, sex and live weight variations in feed and water consumptions of adult captive African Giant rats (*Cricetomys gambianus*, Waterhouse - 1840) kept individually in cages. *J. Anim. Physiol. Anim. Nutr. (Berl)* **97**: 465–474. [Medline] [CrossRef]
13. Fielding, D., and Krause, P. 1998. Donkeys. p. 119. *In:* The Tropical Agriculturist. Macmillan Education Ltd., London and Basingstoke.
14. Gordon, C.J. 2005. Principles of temperature regulation. pp. 13–50. *In*: Temperature and Toxicology: An Integrative, Comparative, and Environmental Approach. Retrieved July 12, 2012, from www.crcnetbase.com/doi/abs/10.1201/9781420037906.ch2.
15. Igono, M.O., and Aliu, Y.O. 1982. Environmental profile and milk production of Friesian-Zebu crosses in Nigerian Guinea Savanna. *Int. J. Biometeorol.* **26**: 115–120. [CrossRef]
16. Igono, M.O., Molokwu, E.C.I., and Aliu, Y.O. 1982. Body temperature responses of Savanna Brown goat to the harmattan and hot-dry season. *Int. J. Biometeorol.* **26**: 225–230. [CrossRef]
17. Kowal, J.M., and Knabe, D.T. 1972. An Agroclimatological Atlas of the Northern States of Nigeria. p.139. Ahmadu Bello University Press, Zaria.
18. Mesteig, K., Tyler, N.J.C., and Blix, A.S. 2000. Seasonal changes in heart rate and food intake in reindeer (*Rangifer tarandus tarandus*). *Acta. Physiol. Scand.* **170**: 145–151. [Medline] [CrossRef]
19. Minka, N.S., and Ayo, J.O. 2007. Effects of shade provision on some physiological parameters, behaviour and performance of pack donkeys (*Equinus asinus*) during the hot-dry season. *J. Equine Sci.* **18**: 39–46. [CrossRef]
20. Minka, N.S., and Ayo, J.O. 2012. Assessment of thermal load on transported goats administered with ascorbic acid during the hot-dry conditions. *Int. J. Biometeorol.* **56**: 333–341. [Medline] [CrossRef]
21. Pal, Y., Kumar, S., and Gupta, A.K. 2002. Blood gases, acid-base and physiological indices in donkeys as pack animals. *Draft Anim. News.* **37**: 27–33.
22. Polidori, P., Vincenzetti, S., Cavallucci, C., and Beghelli, D. 2008. Quality of donkey meat and carcass characteristics. *Meat Sci.* **80**: 1222–1224. [Medline] [CrossRef]
23. Smith, D.G., and Pearson, R.A. 2005. A review of the factors affecting the survival of donkeys in semi-arid regions of sub-Saharan Africa. *Trop. Anim. Health Prod.* **37**(Suppl 1): 1–19. [Medline] [CrossRef]
24. Swoap, S.J., Overton, J.M., and Garber, G. 2004. Effect of ambient temperature on cardiovascular parameters in rats and mice: a comparative approach. *Am. J. Physiol. Regul. Integr. Comp. Physiol.* **287**: R391–R396. [Medline] [CrossRef]
25. Vathana, S., Kang, K., Loan, C.P., Thinggaard, G., Kabasa, J.D., and ter Meulen, U. 2002. Effect of vitamin C supplementation on performance of broiler chickens in Cambodia. Conference on International Agricultural Research for Development, Witzenhausen: Deutcher Tropentag, October 9–11. Retrieved January 6, 2012, from http://www.tropentag.de/2002/abstracts/full/211.pdf.