



Melatonin modulates tonic immobility and vigilance behavioural responses of broiler chickens to lighting regimens during the hot-dry season



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HIGHLIGHTS

- Ambient temperature fluctuated significantly as the hours of the day increased.
- Induction trial attempts lowest in broiler chickens administered with melatonin.
- Tonic immobility duration longest in broiler chickens administered with melatonin.
- Overall mean vigilance behavioural values lowest in melatonin-treated broiler chickens.

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ABSTRACT

Experiments were conducted with the aim of determining the influence of melatonin administration on vigilance and tonic immobility (TI) responses of Marshall broiler chickens. The broiler chickens were reared on different lighting regimens and subjected to heat stress during the hot-dry season. Simple random sampling was used to assign 300 broiler chicks into three groups, comprising 100 broiler chicks each. Group I (12D:12L cycle) was raised under natural photoperiod of 12-h light and 12-h darkness, without melatonin supplementation. Group II (CL) was kept under 24-h continuous lighting, without melatonin administration. Group III (CL + MEL) was raised under 24-h continuous lighting; with melatonin supplementation at 0.5 mg/kg per os, via drinking water using a syringe. Beginning from day-old, broiler chickens in group III were individually administered with melatonin once daily for 8 weeks at 17:00 h. TI was induced by manual restraint, and vigilance elicited at self-righting graded for three days, two weeks apart, in 15 labeled broiler chickens from each of the three groups; at 06:00 h, 13:00 h and 18:00 h, starting from week 4–8. Each broiler chicken was laid on its back in a U-shaped cradle, covered with cloth. Thermal microenvironment parameters of dry bulb temperature (DBT) and relative humidity (RH) were recorded at the experimental site, concurrently during the vigilance and TI tests. Inside the broiler chickens' house, the weekly temperature-humidity index (THI) was lowest at week 4 of the study, with the value of 48.60 ± 0.08 °C. At week 4, the relationship between the THI and TI induction attempts was stronger in 12D:12L cycle ($r = 0.589$, $P < 0.001$) than CL ($r = 0.264$, $P > 0.05$) or CL + MEL ($r = 0.096$, $P > 0.05$) broiler chickens. This indicated that the broiler chickens on 12D:12L cycle were more active compared to their melatonin-treated counterparts, apparently due to adverse effects of high DBT and high RH on the broiler chickens during the hot-dry season. The highest numbers of TI induction trial attempts were recorded at 13:00 h in 12D:12L cycle and CL groups (2.13 ± 0.34 and 2.15 ± 0.22 , respectively), when the broiler chickens were at week 8. The overall mean values of induction trial attempts differed significantly ($P < 0.0001$) between the groups; with the lowest mean values of 1.22 ± 0.4 recorded in CL + MEL broiler chickens. At day 42, the lowest mean TI duration of 101.87 ± 10.24 s in the CL group, recorded at 06:00 h rose ($P < 0.001$) to 184.07 ± 23.69 s at 13:00 h. The overall mean duration of TI differed significantly ($P < 0.0001$) again between the groups; with the highest mean duration of 167.82 ± 8.35 s, recorded in CL + MEL broiler chickens administered with melatonin. The overall mean vigilance behavioural ranking values of $1.85 + 0.07$ and $1.70 + 0.08$, obtained in 12D:12L cycle

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and CL broiler chickens, respectively were higher ($P < 0.0001$) than the value of 1.44 ± 0.05 recorded in melatonin-treated broiler chickens. The results indicated that broiler chickens belonging to both 12D:12L cycle and CL groups were more emotional, fearful or anxious, compared to CL + MEL broiler chickens. It was concluded that melatonin administration elicits boldness and confidence by suppressing freezing behaviour in broiler chickens, and it may improve their welfare and productivity.

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1. Introduction

Thermal environment parameters may adversely affect the health and productivity of chickens and they constitute a major economic threat to poultry farmers in tropical zones of the world [1–3]. High ambient temperature (AT) and high relative humidity (RH) are the most important thermal environment parameters, causing heat stress in the tropics [4–7]. Tonic immobility (TI) and vigilance in birds are anti-predator behaviours [8,9], used to measure the level of fear or stress [10–12]. They are also important in the evaluation of adaptability of poultry to environmental stress factors, including climate variability [13,14]. Fear reactions are elicited in stressful situations, including those related to predator defence [8,15]. Predator-prey interactions are regulated by the ability of individuals to detect, and then approach or avoid one another [16–18]. TI is the last in a series of defensive behaviours displayed in response to attack by a predator, thought to function by reducing stimuli leading to further attack [19]. Fear and anxiety are similar, but technically different emotional terms. Fear is a reaction to the feeling of real danger, whereas anxiety is a reaction to stimuli that can cause danger and exert negative effects on individual's survival [13]. Both anxiety and fear are emotional status, which increase with age in broiler chickens [20,21]. They are damaging stressors, resulting in impaired animal welfare [22,23], poor production [24], involved in regulation of the hypothalamic-pituitary-adrenal axis [25–27], and they predispose birds to depressive-like behaviour [28,29]. TI is a fear response because it is attenuated by procedures that reduce fear, and enhanced by those that increase it [30]. Some characteristics of TI described by [19] include temporary suppression of the righting response, reduced vocalisation, intermittent eye closure, rigidity, Parkinsonian-like muscle tremors in the extremities, altered electroencephalographic patterns and changes in heart rate, respiration and core body temperature. For many years, broiler chickens have been reared under continuous or near continuous (23L:1D) photoperiods in order to maximize feed consumption and growth rate [31]. However, several investigations showed that adopting continuous lighting (CL) programmes induce sleep deprivation and severe stress responses [32–34]. The resultant effect of raising chickens under unfavourable conditions is oxidative stress, involving excessive free-radical generation and propagation, and causing cell destruction and the associated production losses [1,35,36]. Hence, [37] showed, for the first time, that the administration of the antioxidant, ascorbic acid (vitamin C) to chickens ameliorated the heat-stress induced hyperthermia during the hot-dry season.

Melatonin is a potent antioxidant and hormone [38–40], responsible for synchronising the circadian rhythms of vital parameters in animals [41–43], and possessing antibiotic, antiviral and antiparasitic activities [42]. Besides its antioxidant action, it potentiates the effects of other antioxidants such as ascorbate, trolox (a tocopherol analogue), reduced glutathione, and NADH [38,44]. Melatonin is superior to other antioxidants, such as vitamins C and E, because it is not subjected to redox cycling [45], and its metabolites are ROS scavengers. For this reason, melatonin is regarded as a suicidal or terminal antioxidant [46,47]. Data on the modulating effects of exogenous melatonin administration on TI and vigilance behavioural responses of broilers, reared on different lighting regimens during the hot-dry season in the hot-humid zones of the world, are currently lacking in the available literature.

The aim of this study was to evaluate the impact of different lighting regimens and heat stress on TI and vigilance behavioural responses in

broiler chickens, administered with melatonin during the hot-dry season.

2. Materials and methods

2.1. Experimental site, location, and period

The experiment was performed in the broiler chickens' house of the Department of Veterinary Physiology, Ahmadu Bello University, Zaria ($11^{\circ} 10' N$, $07^{\circ} 38' E$), Nigeria, located in the Northern Guinea Savannah zone of Nigeria from April to May during the hot-dry season, prevailing in the zone. The width of the broiler chickens' house was 8.5×7.2 m, while the height was 2.5 m. The broiler chickens' house, from the ground to a height of about 0.80 m was made of cement blocks, while wire mesh covered from that point to the zinc roof to allow for proper ventilation. The house had three compartments used for the three treatment groups and each comprising 100 broiler chickens.

2.2. Flock management

Three hundred broiler chicks, belonging to the Marshall breed were obtained at day-old from Fidan Farms, Fidan Investments Nigeria Limited, Ibadan ($07^{\circ} 22' N$, $03^{\circ} 58' E$), Nigeria. The Indian Poultry Equipment Manufacturer's Association [48] reported that the Marshall breed was developed in Nashik, India for high performance.

Complete routine vaccinations and appropriate medications were administered to the broiler chickens according to standard veterinary procedures, from day-old up to the end of the experiment. Deworming was carried out with piperazine solution, dissolved in drinking water at a single oral dose of 160 mg/kg [49]. The broiler chickens were raised on deep litter system and maintained on commercial feeds, obtained from Vital Feeds, Grand Cereals and Oil Mills Limited, Jos, Nigeria (Table 1.0). Starter diets were fed to the broiler chicks from day-old to 21 d of life, after which they were placed on finisher diet up to the end of the experiment. The broiler chicks were given access to feed and water ad libitum.

2.3. Experimental design

A total of 300 broiler chicks were divided into three groups by simple random sampling. Each group consisted of 100 broiler chicks. Broiler

Table 1
Nutrient contents of the ration fed to the broiler chickens.

Nutrient contents	Amount in % by weight	
	Pelletised starter feed (1–21 d)	Pelletised finisher feed (22–56 d)
Calcium (%)	01.20	01.00
Available phosphorous (%)	00.45	00.40
Metabolisable energy (kcal/kg)	2800	2900
Proximate analysis (%) ^a		
a. Dry matter	93.72	95.32
b. Crude protein	22.00	18.56
c. Crude fibre	03.94	04.27
d. Oil	05.09	04.23
e. Ash	10.47	07.91
f. Nitrogen free extract	58.50	65.03

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