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Effects of Photoperiodism on the Histomorphometry of the Thyroid Gland of the African Giant Rat (*Cricetomys gambianus*)

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ABSTRACT

The effects of photoperiodism on the histomorphometry of the thyroid gland of African giant rats (AGR) waa investigated. A total of 30 captive AGR of both sexes were randomly divided into 3 different photoperiodic groups of 10 rats each (5 males and 5 females). Group 1 rats had 12 hours darkness and 12 hours lighting for 8 weeks, Group 2 rats had total darkness (24 hours) for 8 weeks while, Group 3 rats had total lighting (24 hours) for 8 weeks. Each rat was euthanized using sodium pentobarbital at 86mg/kg BW intraperitoneally. There was no significant increase ($P>0.05$) in the body weight of the rats in exposed to total darkness (Group 2). However, there was a significant decrease in the body weight of rats exposed to light for 8 weeks ($P<0.05$) when compared to rats in control group. Continuous lighting decreases the adipose tissue of thyroid glands with increase number of follicular cells, while, continuous darkness decreases follicular cells with increased adipose tissue. The thyroid weights increased significantly ($P<0.05$) in Group 3 and Group 2 rats when compared to Group 1. Exposure of AGR to continuous light significantly decreased the body weight and increased the weight and width of the thyroid gland. It is therefore recommended to house AGR under controlled conditions with at least 12 hours each of light and darkness

Keywords: African giant rat; Darkness; Light; Photoperiodism; Thyroid gland**.**

INTRODUCTION

The thyroid gland, or simply the thyroid, is an endocrine gland in the neck, consisting of two lobes connected by an isthmus. In the grasscutter, the gland is oval in shape, it is a compact mass with smooth surface which is reddish in colour (Igbokwe, 2010). Previous studies showed that the thyroid gland is reddish brown in colour and located close to the tracheal first ring. It consisted of two lobes on both side and an isthmus connecting these lobes, in large animals such as cattle, buffaloes and camels (Getty *et al*., 1986; Schwartz and Dioli, 1992). The thyroid gland secretes thyroid hormones, which primarily influence the metabolic rate and protein synthesis, as well as on development. The thyroid hormones which include triiodothyronine (T3) and thyroxine (T4) are synthesized from iodine and tyrosine. Calcitonin is also a hormone produced by the thyroid and this plays a significant role in calcium homeostasis (Boron and Boulapep, 2012).

Photoperiodism is the physiological reaction of organisms to the length of night or a dark period (Casal, 2014). It occurs in plants and animals. Photoperiodism is classified into three groups according to the photoperiods; total light, total darkness and normal day/night regime (Mauseth, 2003). A number of biological and behavioural changes are photoperiodism dependent. Together with temperature changes, photoperiod provokes changes in the color of fur and feathers, migration, entry into hibernation, sexual behaviour, and even the resizing of sexual organs (Nelson, 2005). Many animals use day length (photoperiod) to predict and adjust seasonal changes in the environment through predictive changes in physiology and behaviour (Pyter *et. al.,*2007). Most physiological processes in mammals exhibit daily rhythms generated by a system of cell autonomous circadian oscillators located in the brain and in peripheral organs and tissues. A master clock in the hypothalamic suprachiasmatic nucleus provides circadian output signals that are essential for maintaining synchrony of oscillators within organs and between organ systems and for coupling circadian physiology to environmental light-dark cycles (Mistlberger *et.al*., 2013). The effects of season on some organs such as reproductive organs of AGR have been previously reported (Oke, 2008) and gastro-intestinal organs on (Ali *et al*., 2017). Therefore, the present work was aimed at studying the effect of different photoperiods on the gross morphology, morphometry, histology and histomorphometry of thyroid gland in adult rats. The result of this studies would highlight the increased complexities of the internal structural anatomic organization of the thyroid during different photoperiods in AGR.

MATERIALS AND METHODS

Study Area

The research was conducted in Zaria, located in the Northern Guinea Savannah zone of Nigeria, an altitude of 686 m above sea level, and a mean (± Standard Error) monthly photoperiod of 12.13 ± 0.13 h (Kowal and Knabe, 1972, Dzenda *et al*., 2011). The zone is characterized by three seasons: Wet Season (June-October), Cold dry Season (November-February) and Heat Season (March-May) (Ayo *et al*., 1999; Dzenda *et al*., 2011).

Experimental Animals

A total of 30 matured African Giant rats (AGR) (*Cricetomys gambianus*) were sourced from surrounding villages of Zaria Local Government, Kaduna State-Nigeria during rainy season. They were transported live in standard laboratory cages to the experimental animal unit of the Department of Veterinary Public Health, Faculty of Veterinary Medicine, Ahmadu Bello University, Zaria, Nigeria. The rats were fed with groundnuts, carrots, cucumber, sweet potatoes and onions with water provided *ad libitum*.

Experimental Procedure

The rats were weighed and grouped into three, each having 10 rats per group. Group 1- control rats (12 h light/ 12h darkness); Group 2-rats exposed to continuous darkness (24h); Group 3- rats exposed to continuous lighting (24h) for 8weeks. The rats in group 2 were provided an uninterrupted power source through a solar inverter, whereas the rats in absolute continuous darkness had their windows and doors covered with dark curtains. The duration of the experiment was eight weeks.

After exposure to varying photoperiods, the body weights of each rat were measured using a Mettler balance P 1261 (Mettler Instrument AG. Greinfense, Switzerland) with a sensitivity of 0.01g. The rats were humanely euthanized using sodium pentobarbital at 86mg/kg I.P.

Extraction of Thyroid Gland

A ventral mid-line incision was made across the length of the neck to expose the thyroid glands. The thyroid glands were examined in situ to rule out any pathology before removal for gross morphological investigation. The thyroids were trimmed of fat and measured using Vernier Caliper MG6001DC (General Tools and Instruments Co., New York) and weighed using Mettler balance. The measurements recorded were used to calculate Organo-somatic Index (OSI) of the thyroid, by dividing thyroid weight with body weight multiplied by 100 (weight of thyroid (g)/ weight of body (kg) X 100) (Radhiah and Azhar, 2020).

Histological Evaluation

The extracted gland was fixed in 10% formalin for 24hour, dehydrated through series of ascending concentrations of ethanol (70%, 90%, 95%, 100%) for 2 hours at room temperature and cleared in xylene, embedded in paraffin wax and sectioned at 5µm. Hematoxylin & Eosin stain was used for general histological evaluation (Kiernan, 2007).

Data Analysis

All recorded weights and dimensions of the thyroids were expressed as mean (± standard error of the mean), and were subjected to statistical analysis using Statistical Package for Social Science, Version 20.0 Variation in parameters in groups were analysed using one- way analysis of variance (ANOVA). Value of $P< 0.05$ was considered significant.

Ethical statement: The experimental procedures were approved by the Institutional Animal Ethics Committee of Ahmadu Bello University Zaria-Nigeria with approval number ABUAUC/2021/043.

RESULTS

Gross Morphology

The thyroid gland of the AGR in all the three groups were observed to be bi-lobed, elongated from the third to seventh tracheal ring and red in colour. A connective tissue attached the glands to the larynx and trachea. Each gland was seen to be oval shaped with an apex that pointed upward towards the oblique line on the laminae of the thyroid cartilage. The base was towards the fifth tracheal ring. The medial side was seen to be attached to the trachea while the lateral side was convex. The lobes were connected by a thin isthmus located along the ventral surface of the second tracheal rings. There were no obvious differences grossly across the groups. (Figures 1, 2 and 3).

Morphometric Results

The onset of the experiment (day 0), mean initial body weight of AGR in control, total light and total darkness groups were 1616.00 ± 551.97 g, 1460.00 ± 485.80 g and 1398.00 ± 545.01 g respectively. No statistical ($P > 0.05$) difference in body weight across the group at day 0. At 4 weeks, there was a significant decrease (1237.50 ± 438.51 g) in body weight of the group exposed to continuous light when compared to the control group (1550.00 ± 465.48 g), While, a significant ($P<0.05$) increase in the body weight of the group exposed to continuous darkness (1737.50 ± 612.88 g) in comparison to control group. At 8 weeks, there was an insignificant increase in the body weight of the group exposed to total darkness (1400.00 ± 451.66 g) and a significant decrease in the group exposed to total light (1058.33 ± 372.04 g) when compared to the control (1350.00 ± 649.62 g) (Table 1).

In AGR of control groups, the body weight decreased by 4.08% at 4 weeks and by 16.46% at 8 weeks. In the total light group, decrease in body weight was by 15.24% at 4 weeks and by 27.51% at 8 weeks. There was an increase in body weight by 24.14% at 4 weeks and by 0.14% at 8 weeks in AGR of total darkness group (Table 2).

Weight and Relative Weight of the Thyroid Gland

The mean weight of the thyroid gland at 4 weeks recorded in AGR of control, total light, and total darkness groups were 32.00 ± 2.00 g, 43.00 ± 3.00 g, and 42.00 ± 1.00g respectively. At 8 weeks, the weight of the thyroid glands in

AGR of control, total light, and total darkness were 33.00 ± 2.00 g, 44.00 ± 2.00 g and 43.00 ± 2.00 g, respectively.

The relative weights (RW) of thyroid gland was insignificantly ($P > 0.05$) higher at 4 weeks in AGR of total light ($3.58 \pm 1.35 \times 10^{-3}$) when compared to control ($2.18 \pm 0.51 \times 10^{-3}$) and total darkness ($2.65 \pm 0.96 \times 10^{-3}$) groups. At 8 weeks, there was also an insignificant increase in relative weight of AGR from total light group ($4.62 \pm 1.66 \times 10^{-3}$) compared to control ($3.07 \pm 1.58 \times 10^{-3}$) and total darkness group ($3.39 \pm 1.09 \times 10^{-3}$).

Length and Width of the Thyroid Gland

The mean length of thyroid gland from the control group (7.27 ± 0.01 mm) showed insignificant ($P > 0.05$) variation across both the total darkness (7.10 ± 0.14 mm) and total light (7.27 ± 0.01 mm) groups at 4 weeks. While, at 8 weeks, the length of the thyroid gland from the group exposed to total light showed a significant (6.98 ± 0.02 mm) decrease when compared to those from control (7.25 ± 0.04 mm) and total darkness (7.17 ± 0.06 mm) groups. The width of the thyroid gland was significantly ($P < 0.05$) higher at 4 and 8 weeks in the group exposed to total light (4.36 ± 0.01 mm; 4.22 ± 0.02 mm) when compared to the total darkness group (4.13 ± 0.02 mm; 4.15 ± 0.03 mm) and control group (4.04 ± 0.01 mm; 4.04 ± 0.02 mm) respectively. (Table 3).

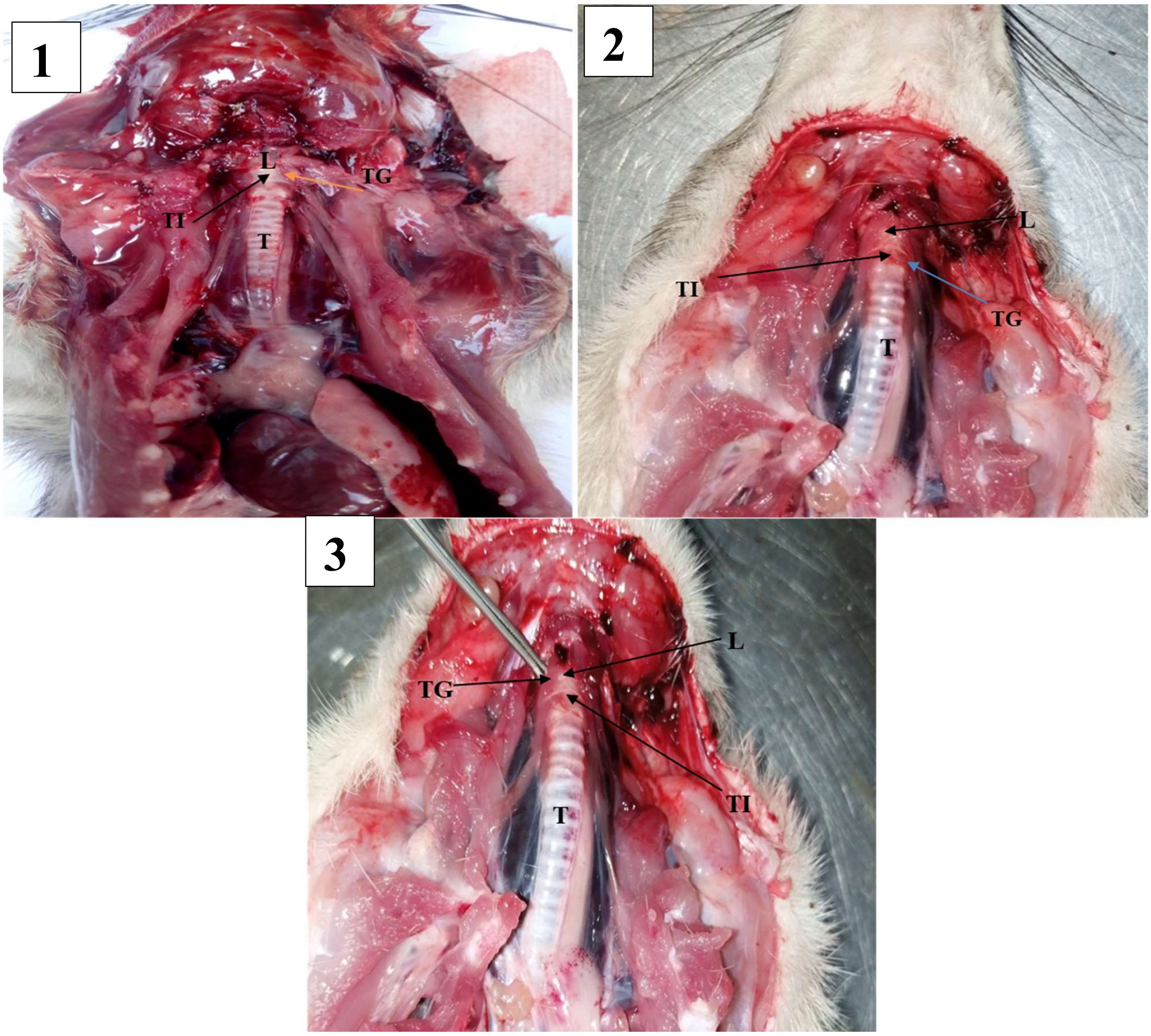


Figure I: Photograph of thyroid gland (TG) and other related structures *in situ* from the control group showing the larynx (L), thyroid isthmus (TI) and trachea (T). **Figure 2**: Photograph of Thyroid gland (TG) and other related structures *in situ* from group exposed to total light showing larynx (L), thymus isthmus (TI) and trachea (T). **Figure 3:** Photograph of Thyroid gland (TG) and other related structures *in situ* from group exposed to total darkness showing larynx (L), thymus isthmus (TI) and trachea (T).

Table 1: Mean values of Body Weights of different groups of African Giant Rats

Parameter	Group 1 Control	Group 2 Total Darkness	Group 3 Total Light
Initial body weight (g)	1616.00 ± 551.97	1398.00 ± 545.01	1460.00 ± 485.80
Body weight at 4 weeks (g)	1550.00 ± 465.48	1737.50 ± 612.88	1237.50 ± 438.51
Body weight at 8 weeks (g)	1350.00 ± 649.62	1400.00 ± 451.66	1058.33 ± 372.04
Body weight change at 4 weeks (%)	4.08^{a}	24.14^{b}	15.24^{a}
Body weight change at 8 weeks (%)	16.46^{a}	0.14^{b}	27.51^{a}

Values with (^{a})shows decrease while values with (^{b}) shows increase in recorded parameters.

Table 2: Mean values of Thyroid weight and Relative weights of the Thyroid Gland of African Giant Rats in different groups.

Parameter	Time of exposure (Weeks)	Group 1 Control	Group 2 Total Darkness	Group 3 Total Light
Weight (g)	4	32.00 ± 2.00^{a}	42.00 ± 1.00^{b}	43.00 ± 3.00^{b}
	8	33.00 ± 2.00^{a}	43.00 ± 2.00^{b}	44.00 ± 2.00^{b}
Relative weight (× 10^{-3})	4	2.18 ± 0.51^{a}	2.65 ± 0.96^{a}	3.58 ± 1.35^{a}
	8	3.07 ± 1.58^{a}	3.39 ± 1.09^{a}	4.62 ± 1.66^{a}

Values with the same superscript alphabets in the same row do not differ significantly at P > 0.05.

Table 3: Mean Length and Width of the Thyroid Gland of African Giant Rats in different Photoperiods

Parameter	Time of exposure (Weeks)	Group 1 Control	Group 3 Total darkness	Group 2 Total light
Length (mm)	4	7.27 ± 0.01^{a}	7.10 ± 0.14^{a}	7.27 ± 0.01^{a}
	8	7.25 ± 0.04^{a}	7.17 ± 0.06^{a}	6.98 ± 0.02^{b}
Width (mm)	4	4.04 ± 0.01^{a}	4.13 ± 0.02^{c}	4.36 ± 0.01^{b}
	8	4.04 ± 0.02^{a}	4.15 ± 0.03^{c}	4.22 ± 0.02^{b}

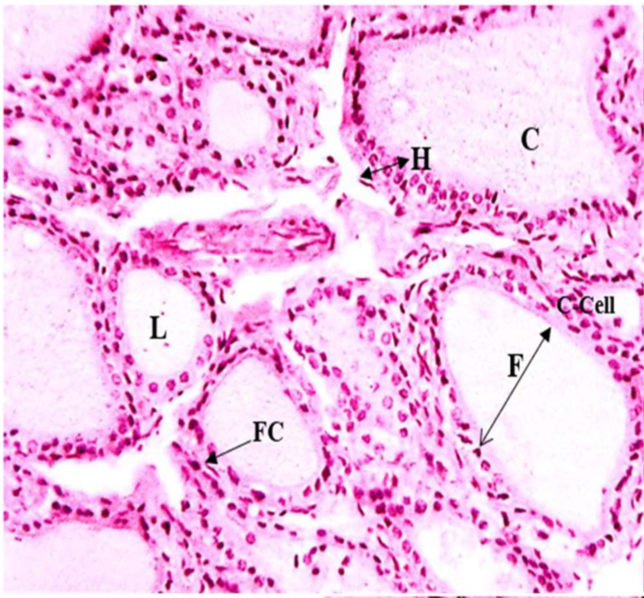
Values with the same superscript alphabets in the same row do not differ significantly at P > 0.05.

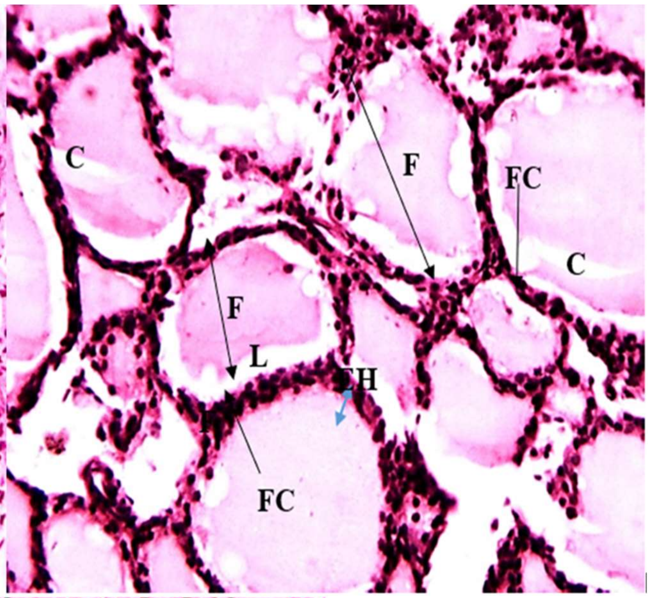
Histology Results

Histologically, the thyroid gland of AGR is composed of peripherally located follicles which are distended with colloid and lined by flat follicular cells containing flat dark nuclei of myoepithelial cells (Figure 4). In rats exposed to total lighting, the thyroid gland showed numerous follicular cells with absence of reactive colloids (Figure 5). While, those exposed to total darkness showed fewer follicular cells with reactive colloids and s5canty stroma (Figure 6).

DISCUSSION

In this study, the differences in the body weight, thyroid weight, width and length of African giant rats were evaluated upon exposure to varying photoperiodic conditions. There was no significant difference in body weight of AGR in control, total light and total darkness groups at both 4 and 8 weeks, this might be due to the insignificant growth observed in adult animals. This is in agreement with the findings of Kenfack *et al*. (2020) who reported an insignificant difference in body weight of AGR exposed to 4 weeks of varying photoperiods. In contrast, there was a significant increase in the body weight of laboratory rats (Tavolaro *et al*., 2015) with increased photoperiod. At 4 weeks, there was decreased body weight change in rats exposed to total light while there was increase in those exposed to total darkness. The decreased body weight change of rats exposed to total light might be due to decreased food intake. In rats exposed to total darkness, the increased body weight change might be associated with increased food intake since the AGR is a nocturnal animal (Ali *et al*., 2017). However, in rats exposed to total darkness, the weight change was decreased at 8 weeks compared to at 4 weeks and this might be associated with decreased feed intake resulting from prolonged captivity and restriction. The thyroid gland of the AGR was bi-lobed, reddish colour, oval and located around the bilateral portions of the trachea. This is in line with the findings of Enemali *et al*. (2016) in AGR and is similar to those reported in Grass cutter (Igbokwe, 2010), Wistar rat and humans (Dalley and Moore, 2006). There was increase in the weight of the thyroid gland in AGR exposed to continuous light and darkness compared to those in control. Increase in thyroid gland weight in AGR exposed to continuous light in this study is consistent with the findings of Olatunji-Bello and Sofola (2001) who reported increased weight of thyroid gland in male rats exposed to continuous light for 4 weeks. In contrast, Olatunji-Bello and Sofola (2001) reported decreased weight of thyroid gland in rats exposed to continuous darkness for 4 weeks. The study by Eleiwe *et al*. (2014) demonstrated increased thyroid gland weight in rats exposed to continuous darkness. Variation in the age, geographical locations and type of feed might be responsible for the disparity in these sets of observations. The increase in weight and relative weight of the thyroid gland in rats exposed to total darkness in this study might be due to hibernal latency of the AGR. The length and width of the thyroid gland in rats at 4 and 8 weeks in this study were lower than that reported by Enemali *et al*. (2016). At 4 weeks, there was no significant difference in the length of the thyroid gland in all rats and this indicated that photoperiod had no effects on the length of the thyroid gland.





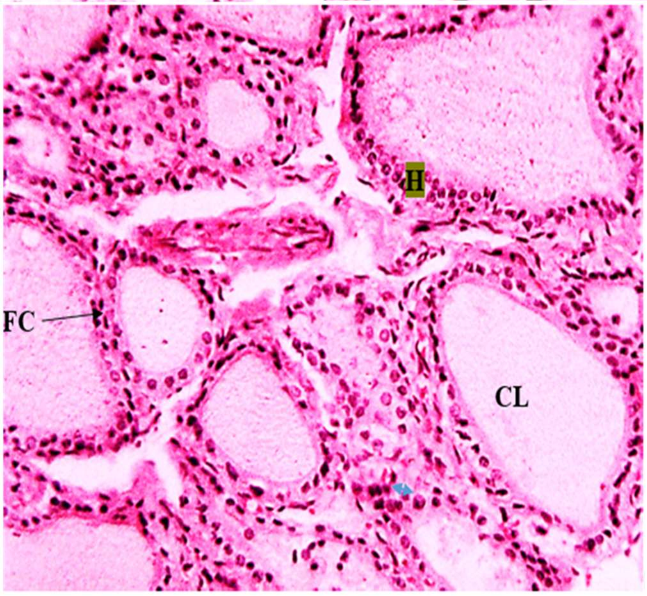


Figure 4: Photomicrograph of the thyroid gland of African giant rat from the control Group showing follicular diameter (F), follicular cells (FC), C- Cell, lumen (L) with colloid(C) and epithelial heights (H). H & E (× 100). **Figure 5:** Photomicrograph of the thyroid gland of African giant rat from the group exposed to total darkness showing lumen (L) with reactive and active follicles showing active uptake of colloids, follicular cells (FC), follicular diameter (F) and epithelial heights (EH). H & E (× 200). **Figure 6:** Photomicrograph of the thyroid gland of African giant rat from the group exposed to total light showing inactive follicular activity by absence of reactive (active) colloid in lumen (CL), no white areas on the periphery of the colloid, follicular cells (FC) and epithelial heights (H). H & E (x 200).

At 8 weeks, the decrease in length of the thyroid gland in rats exposed to total light compared to control indicated that prolonged photoperiod had effect on thyroid gland length. The width of the thyroid gland was increased in rats exposed to total light and darkness compared to control at 4 and 8 weeks. Histologically, the thyroid gland of AGR is composed of peripherally located follicles which are distended with colloid and lined by flat follicular cells containing flat dark nuclei of myoepithelial cells. This is in agreement with the findings of El-Rouby, (2010), who reported that the thyroid gland of albino rats is composed of peripherally located follicles with colloids and flat follicular cells. In rats exposed to total light, showed numerous follicular cells with absence of reactive colloids while those exposed to total darkness showed fewer follicular cells with reactive colloids and scanty stroma.

Conclusions

This study has shown that, exposure of AGR to continuous light significantly decreases the body weight and increases the weight, width and follicular cells of the thyroid gland. While, the continuous exposure of AGR to darkness significantly increases both the body weight, weight of the thyroid gland, and decreases thyroid follicular cells. It is therefore recommended to house AGR under controlled conditions of at least 12 hours of light and darkness.

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Conflict of Interest

The authors declare that they have no conflict of interest.

Author's Contributions

ZM, AMN and UAD designed the experiment. ZM did the laboratory work. ZM and MTA analyzed the experimental results. MTA and ASM wrote the manuscript. BA and UMD proof read the manuscript. All authors have read and approved the final manuscript.

REFERENCES

Ali, M. N., Rashid, S., Nafees, S., Hasan, S.K., Shahid, A., Majed, F. and Sultana, S. (2017). Protective effect of Chlorogenic acid against methotrexate induced oxidative stress, inflammation and apoptosis in rat liver: An experimental approach. *Chemico-Biological Interactions*, 272: 80-91. Doi.org/10.1016/j.cbi.2017.05.002.

Ayo, J. O., Oladele, S. B., Ngam, S., Fayomi, A. and Afolayan, S. B. (1999). Diurnal fluctuations in rectal temperature of the Red Sokoto Goat during the harmattan season. *Research in Veterinary Science*, 66 (1): 7-9. Doi.org/10.1053/rvsc.1998.0231.

Boron, W.F. and Boulapep, E.L. (2012). Medical Physiology (2nd ed.). Philadelphia: Saunders. Pp. 1052. ISBN 978-1-4377-1753-2.

Casal, J. J. (2014). Light perception and signaling by phytochrome A. *Journal of Experimental Biology,* 65 (11): 2835-2845. Doi:10.1093/jxb/ert379

Dalley, A.F. and Moore, K. L. (2006). Embryological and Surgical Anatomy of the Intrahepatic and Extrahepatic Biliary Tree. In Liver and Biliary Tract Surgery, *Springer,* Pp. 3-16. Doi.org/10.1007/978-3-211-49277-21.

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). *Journal of Thermal Biology*, 36: 255-263. Doi.org/10.1016/j.therbio.2011.03.010.

Eleiwe, S. E., Nibras, H. K. and Shilan, H. K. (2014). Continuous darkness induces structural changes in thyroid gland of adult male rats. *Journal of Science and Medicine in Sports*, 4 (1): 1 - 9. Doi.org/10.17656/jsmc.10043.

El-Rouby, N. M. (2010). A Histological Study on the Effect of Diclofenac Sodium (Declophen) Administration on Thyroid Follicular Cells of Albino Rats. *Egyptian Journal of Histology*, 33: 213 - 223.

Enemali, F.U., Hambolu, J.O., Alawa, J.N. and Anosike, I.V. (2016). Gross Anatomical, Histological and Histochemical Studies of Thyroid Glands of African Giant Rat (*Cricetomys gambianus* Waterhouse, 1840). *Journal of Pharmacy and Biological. Sciences*, 11(4): 40-43. Doi.org/10.9790/3008-1104024043.

Getty, R., Sission, S. and Grossman, J. D. (1986). The Anatomy of Domestic Animals. 5th Ed., W.B. Saunders Company. Philadelphia, USA.

Igbokwe, C.O. (2010). Gross and microscopic anatomy of thyroid gland of the wild African grasscutter (*Thryonomys swinderianus*, Temminck) in Southeast Nigeria. *European Journal of Anatomy*, 14: 5-10.

Kenfack, A., Fonou, L.T. and Fopa, C.F. (2020). Effects of photoperiod on growth performances in female African Giant Rat (*Cricetomys gambianus*). *International Journal Agronomy and Agricultural Research*, 17(4): 11-16.

Kiernan, J.A. (2007). Histochemistry of staining methods for normal and degenerating myelin in the central and peripheral nervous systems. *Journal of Histotechnology*; 30(2): 87-91. Doi.org/10.1179/his.2007.30.2.87.

Kowal, J. M. and Knabe, D. T. (1972). An Agro-Climatic Atlas of the northern states of Nigeria. Ahmadu Bello University Press, Zaria, Nigeria.

Mauseth, J. D. (2003). Botany: An Introduction to Plant Biology (3rd ed.). Sudbury, MA: Jones and Bartlett Learning. Pp. 422–27. ISBN 978 – 0 – 7637 – 2134-3.

Mistlberger, R. E., Refinetti R. and Kriegsfeld, L. J. (2013). Circadian Rhythms and Physiological Processes. *Encyclopedia of Sleep*, *Elsevier Incorporated*, Pp.418-425. Doi.org/10.1016/B978-0-12-378610-4.00093-0.

Nelson R. J. (2005). An Introduction to Behavioral Endocrinology (p.189). Sunderland, MA: Sinauer Associates.

Oke, O.A. (2008). Evaluation of the Effectiveness of Three Insecticides to control Diamond Back moth (*Plutellaxylostella*) in cabbage (*Brassica oleracea* Var. Capital L). *European Journal of Scientific Research*, 22(3): 391-395.

Olatunji-Bello, I. and Sofola, O. A. (2001). Effect of Continuous Light and Darkness Exposure on the Pituitary-Gonadal Axis and thyroid activity in male rats. *African Journal of Biomedical Research*, 4: 119 - 122. Doi.org/10.4314/ajbr. v4i3.53888.

Pyter, L. M., Adelson, J. D. and Nelson, R. J. (2007). Short days increase hypothalamic pituitary adrenal axis responsiveness. *Endocrinology,* 148: 3402-3409. Doi.org/10.1210/en.2006-1432.

Radhiah, N. A. and Azhar, L. J. (2020). Effect of Evisect on Organo-Somatic Index and Pathohistological changes of some vital organs in white Mice. *Systematic Reviews in Pharmacy,* 11 (11): 1910-1914.

Schwartz, H.J. and Dioli, M. (1992). The one-humped camel in Eastern Africa. A pictorial guide to diseases, health care and management. Schonwald Druck, Berlin. F.R. Germany, Pp. 228-229.

Tavolaro, F.M., Thomson, L.M., Ross, A.W., Morgan, P.J. and Helfer, G. (2015). Photoperiodic Effects on Seasonal Physiology, Reproductive Status and Hypothalamic Gene Expression in Young Male F 344 Rats. *Journal of Neuroendocrinology*, 27(2): 79-87. Doi.org/10.1111/jne.12241.