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Full Length Research

Effect of Anthropogenic Light at Night (ALAN) on Condition Factor (CF) and Hepato-somatic Index (HSI) of *Sclerophrys maculata*

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Anthropogenic light at night (ALAN) alters natural day length and has physiological and behavioural effects on living organisms. Here we examine the impact of ALAN on the body condition factor (CF) and hepato-somatic index (HSI) of anurans, *Sclerophrys maculata*. Fifteen specimens were collected from Bakana, Rivers State, Nigeria, for the research. They had initial mean snout-vent length (SVL), body mass (BM) and CF values of 4.45cm, 7.63g and 8.64, respectively. After acclimatization, they had mean SVL, BM and CF of 4.44cm, 11.41g and 13.00, respectively. Among the specimens exposed to ALAN, after 7, 14, and 21 days, the mean CF values were 13.32, 8.91, and 13.98, respectively; the mean HSI values were 3.24, 2.54, and 2.18, respectively. Among the specimens of the control group, after 7, 14, and 21 days, the mean CF values were 8.72, 10.29, and 12.55, respectively; the mean HSI values were 1.95, 1.45 and 1.90, respectively. Student t-test showed no significant differences in the mean condition factor values obtained for both groups of specimens over the experimental period (t_3=0.798, p=0.23). Same test showed no significant differences in the total body mass of the specimens exposed to ALAN and control conditions (t_3=1.06, p=0.18). However, there was a significant difference in the HSI of the specimens exposed to both experimental conditions (t_3=2.54, p=0.04). Significantly higher HSI values were obtained in specimens exposed to ALAN. This research has shown that ALAN poses stress on amphibians and the effect is better revealed by a measure of the HSI.

Key words: ALAN, condition factor, hepato-somatic index, *Sclerophrys maculata*, amphibians.

INTRODUCTION

Artificial light at night (ALAN) has been identified as a type of environmental pollution which primarily leads to perceived increase in day length, with its attendant disruption of the physiological processes of living organisms, including reproduction, feeding and immune response (Robert et al., 2015). Darkened conditions have been reported to reduce stress in wild animals (Beaupre et al., 2004). However, continuous light or dark conditions induce stress (Frye, 1991).

Some researchers have reported on the effect of artificial light at night (ALAN) on living organisms ranging from vegetation (Bennie et al., 2018) to mammals (Robert et al., 2015). Anthropogenic light at night was found to disrupt the timing of reproduction in free-ranging wallabies (Robert et al.,

2015). It has been reported to alter the behaviour of night-time migratory birds, causing an increase in their calling rates (Watson et al., 2016).

The condition factor is an index that indicates the effect of environmental factors on the welfare of animals (Chaves et al., 2017). Weight ratios between structures in an organism also form indices that provide information on the success of organisms in their environment. One of such is the hepato-somatic index (HSI) (Hegde and Krishnamurthy, 2014). Amphibians feature prominently as animal models in physiological research (Burggren and Warburton, 2007), and were selected for this study due to their availability and ease of handling, because stress can lead to abnormal behavioural and physiological responses (Beaupre et al., 2004).

MATERIALS AND METHODS

Study Location

The sample location was Bakana (E6^o 96', N4^o 73'), an Island located within the Niger Delta region of Nigeria. The aquatic bodies were generally marine, however, there were pockets of freshwater in temporary ponds and ditches that provided habitat for amphibians. Wet grass fields were also common and supported amphibian species adapted to altered environments.

Collection of amphibians

Amphibians, *Sclerophrys maculata* (toads), were actively collected, using the simple search and capture technique, between the hours of 7.00pm and 10.00pm. The specimens used for the research were collected during one expedition in June, 2018. They were transported in wet buckets to the Parasitology Laboratory of Department of Animal and Environmental Biology, Rivers State University, Nigeria. They were identified after the protocols of Rödel (2000).

Experimental Set-Up

Eight big plastic bowls were purchased along with mosquito nets and ropes. They were half-filled with sandy loamy soil collected from the vicinity of Department of Animal and Environmental Biology, Rivers State University, Port Harcourt. Grasses were planted in the bowls, and sticks and stones were incorporated to provide hiding places for the amphibians. The bowls were covered with mosquito nets which were tied tightly around the bowls to allow air while preventing the amphibians from escaping. On arrival in the lab, the amphibians were weighed and their snout-vent length taken. They were afterwards organised into two in each bowl and kept in the control environment for acclimatization for five days. At the end of the acclimatization period, the specimens were weighed and their SVL taken again. They were then separated into those exposed to ALAN and the control.

They were left for seven days after which amphibian specimens in one bowl from both ALAN and control groups were sacrificed, measured and weighed and dissected for parasites. This was repeated after fourteen days and again after twenty-one days. Throughout the experimental period, the amphibians were fed twice weekly and water was sprinkled on them (depicting rainfall) twice weekly. Care was taken to ensure that the soil was not soaked with water.

Care of the amphibians

The amphibians were fed with insects and gastropods ad libitum twice weekly. Water was provided in plastic plates inside the enclosure. This plate was placed inside the soil in such a way that its top surface was flush with the soil surface, and each plate was deep enough to allow each specimen to be completely submerged. The initial water used was tap water fetched and kept overnight in an open bucket (Beaupre et al., 2004). Subsequently, the water was partially exchanged twice weekly. Water was sprinkled on the amphibians twice every week mimicking light rainfall.

Morphometric Measurements and Computation of CF and HIS

The body mass (BM) of the specimens was taken using an electronic weighing balance (Denver Instrument, Model TP-512A) while snout to vent length (SVL) was measured using a meter rule.

Condition factor (CF) and hepato somatic index (HSI) were computed according to Zhelev et al. (2015) as follows:

$CF = BM / SVL^3 \times 10^2$

Table 1. Morphometric measurements and CF of *S. maculata* from Bakana (Rivers State, Nigeria) before and after acclimatization.

	SVL (cm)		BM (g)		CF	
	Initial	After acclimatization	Initial	After acclimatization	Initial	After acclimatization
Range	4.2 - 4.5	4.3-4.5	5.48 – 9.52	8.39 – 14.49	6.77 – 10.45	10.55 – 15.90
Mean	4.45	4.44	7.63	11.41	8.64	13.00

Key: SVL= snout-vent length, BM= body mass, CF=condition factor.

HSI= (LM/BM) X 10^2.

Parasitic Examination and Computation of Prevalence and Mean Intensity

The toads were dissected to reveal their organs (such as, lungs, liver, gastro-intestinal tract and urinary bladder). In order to examine them for parasites, the organs were excised into Petri dishes filled with 0.72% laboratory saline, and cut open using dissecting scissors to reveal their contents. Parasites, when present, were picked up using pipettes and fixed appropriately as described by Amuzie and Ekerette (2017), and identified following the keys of Prudhoe and Bray (1982).

The prevalence and mean intensity of infection was computed for each parasite species according to Bush et al. (1997) as follows:

$$\text{Prevalence (P \%)} = \frac{\text{(Number of infected host)}}{\text{(Total number of hosts examined)}} \times 100.$$

$$\text{Mean intensity (MI)} = \frac{\text{(Total number of parasites in all infected hosts)}}{\text{(Total number of infected hosts)}}$$

STATISTICAL ANALYSIS

Student-t tests were used to test for differences in the total body weight, condition factor and hepato-somatic index values of the specimens exposed to ALAN and control conditions. These were done using MS-Excel.

Ethics

The research was approved by the departmental ethics committee of Department of Animal and Environmental Biology, Rivers State University, Port Harcourt, Nigeria.

RESULTS AND DISCUSSION

Effects of ALAN on condition factor and hepato-somatic index

Fifteen specimens of *S. maculata* were received on June 1, 2018, from collectors in Bakana, Degema Local Government Area of Rivers State, Nigeria. The SVL, BM and condition factor values of the specimens before and after acclimatization are presented in Table 1.

After acclimatization, they were separated into the experimental units and either exposed to ALAN or kept as control specimens. The specimens exposed to ALAN and the control group were examined in intervals of 7days and the results of their morphometric measurements, condition factors and hepato-somatic indices are presented in Table 2.

The mean values of the body mass, condition factor and liver mass of specimens exposed to ALAN were high after 7days, reduced after 14 days and increased further after 21 days. However, the hepato-somatic index was higher after 7days, but reduced after 14 and 21days.

In the specimens kept under control conditions, mean values of both the body mass and condition factor were at the lowest after 7days but subsequently increased after 14 and 21 days. However, mean liver mass and hepato-somatic index reduced after 14days but increases were recorded after 21days. A comparison of the mean values of the body mass, condition factor and hepato-somatic index over the period of the experiment was plotted and is shown in Figure 1. Student-t test was used to test for significant differences between the mean condition factor

Table 2. Morphometric measurements, CF and HSI of *S. maculata* under ALAN and Control Conditions.

		7 days		14 days		21 days	
		Range	**Mean**	**Range**	**Mean**	**Range**	**Mean**
SVL (cm)	ALAN	4.5	4.5	4.5-4.6	4.55	4.3-4.5	4.4
	Control	4.5	4.5	4.5-4.6	4.55	4.3-4.4	4.35
BM (g)	ALAN	9.58-14.7	12.14	6.76-10.12	8.44	11.5-12.3	11.9
	Control	7.87-8.03	7.95	9.2-10.21	9.71	8.5-12.28	10.39
CF	ALAN	10.51-16.13	13.32	7.42-10.40	8.91	13.5-14.46	13.98
	Control	8.64-8.81	8.72	10.1-10.50	10.29	10.69-14.42	12.55
LM (g)	ALAN	0.19-0.66	0.43	0.19-0.23	0.21	0.25-0.27	0.26
	Control	0.13-0.18	0.16	0.13-0.15	0.14	0.15-0.25	0.20
HSI	ALAN	1.98-4.49	3.24	2.27-2.81	2.54	2.17-2.19	2.18
	Control	1.62-2.29	1.95	1.27-1.63	1.45	1.76-2.04	1.90

Key: SVL= snout-vent length; BM= body mass; CF= condition factor; LM= liver mass; HIS= hepato somatic index.

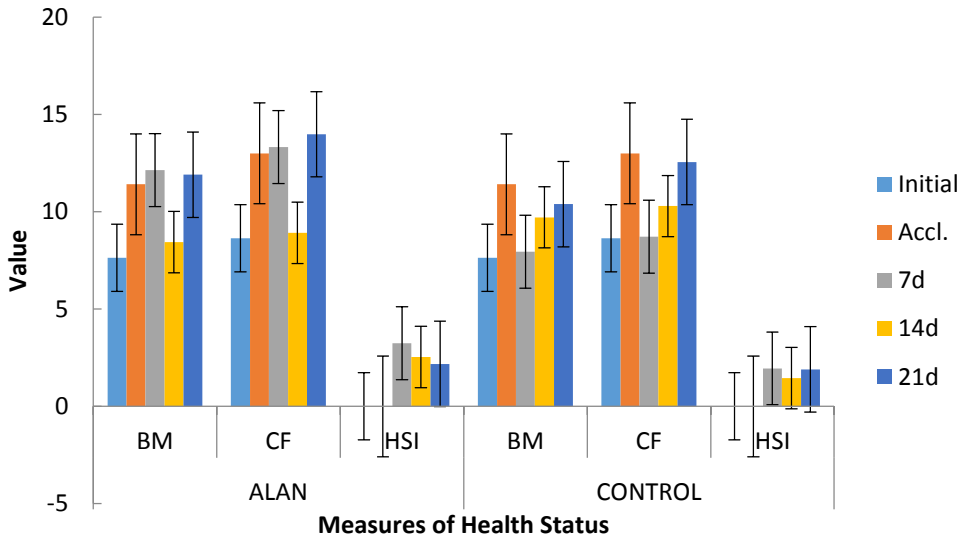

Figure 1. Comparison of the BM, CF and HSI of *S. maculata* exposed to ALAN and Control Conditions.

values obtained for both groups of specimens over the experimental period and the test showed no significant differences (t_3=0.798, p=0.23). Same test showed no significant differences in the body mass of the specimens exposed to ALAN and control conditions (t_3=1.06, p=0.18). However, there was a significant difference in the HSI of the specimens exposed to both experimental conditions (t_3=2.54,

Table 3. Prevalence (%) and Mean Intensity (MI) of Parasite Infections from *S. maculata* from Bakana after exposure to ALAN and Control Conditions.

			ALAN			CONTROL	
		No. of Infected Hosts	%	MI	No. of Infected Hosts	%	MI
Trematoda	PS						
M. monodi	SI	1	16.67	4.0	3	50.00	2.0
Nematoda							
A. africanum	SI	2	33.33	2.0	1	16.67	1.0
C. ornata	LI	4	66.67	2.75	4	66.67	2.25
R. africanus	L	4	66.67	5.75	5	83.33	13.8

Key: PD= Predilection site; SI= Small intestine; LI= Large intestine; L= Lungs

p=0.04). Significantly higher values of HSI were obtained in specimens exposed to ALAN, although the values reduced as the experiment progressed.

Parasites Recovered

Four parasite species were recovered from the host specimens. They were comprised of one trematode (*Mesocoelium monodi*), and three nematodes (*Amplicaecum africanum, Cosmocerca ornata,* and *Rhabdias africanus*). The prevalence and mean intensities of parasite infections are presented in Table 3.

The body mass of the specimens relatively increased over the experimental period and survivability was good. Only three specimens died during acclimatization possibly due to stress of capture and transportation from Bakana to Rivers State University. Those that survived that period were alive until the end of the experiment. This suggests that *S. maculata* are hardy amphibians and are good species to use for laboratory based experiments.

There were no significant differences in the total body mass of the specimens exposed to both ALAN and control conditions. However, the mass of specimens exposed to ALAN were generally higher. This could be because, with increased light, the specimens rested for longer period thereby expending less energy for metabolic activities. It is thought that longer periods of exposure is required for the full effects of ALAN to manifest. For instance, Bennie et al., (2018) found that the effect of ALAN on grassland vegetation was not obvious until the third and fourth year of experimentation.

Among the specimens exposed to ALAN, the CF increased slightly above that value recorded after the period of acclimatization, then reduced by the 14th day but increased again by the 21st day. The HSI was initially high, probably as initial response to the altered lighting but reduced over the rest of the experimental period indicating probably that the specimens adapted to the light pollution via some form of behavioural adaptations. For instance, they were always found buried below the soil surface. Mazeroll (2004) has also noted that ALAN could be beneficial to amphibians because light attracts insects which means more availability of food for the amphibians.

The CF of the control group reduced by the 7th day as compared with the value recorded after acclimatization. The CF however, increased by the 14th and 21st days. The initial drop could be because of the increase in the length of night-time, though they were acclimatized under same condition. Being kept in a room away from the reach of moon light or stars could have lengthened the night-time period and had some behavioural and or physiological impact on the specimens. The HSI values of the specimens in the control group was generally lower than that recorded for specimens exposed to ALAN, indicating that ALAN actually posed stress to the specimens. The abrupt drop, reported on the 14th day, in the total body mass and condition factor of specimens exposed to ALAN was not noticed in specimens of the control group. Though we do not have explanation for that at the moment, it points to the fact that the specimens in the control group were more stable than those exposed to ALAN.

Statistical tests showed that there was a

significant difference in the HSI of specimens exposed to ALAN and those kept under control conditions. The values were higher in those exposed to ALAN. Hedge and Krishnamurthy (2014) found that the body condition factor, *Hepato somatic* index (HSI) and the activity of Acetyl cholinesterase (AChE) could be used as indicators of the health status of amphibians. They reported that healthier amphibians had higher body condition factor and AChE activity in the brain and lower HSI levels on their liver when compared to unhealthy ones from contaminated environments. Changes in HSI generally indicate an effect of chemical exposure on liver function (Paunescu and Ponepal, 2011). This work has shown that HSI could also be used to detect the effect of stress caused by ALAN on amphibians.

There was no significant difference in the mean CF of specimens from both experimental groups. Although CF is an important measure used to establish the health condition of amphibians (Thammachoti et al., 2012), the results obtained in this research highlights the HSI as a more sensitive measure than the CF. Though the period of exposure was short, the HSI could detect physiological responses by the liver to the external stressor.

Only four parasites were encountered in this experiment; they were composed of one trematode and three nematodes. This varied from the finding of Amuzie and Ekerette (2017) on *Sclerophrys* spp. (syn. *Amietophrynus* spp.) in locations in Rivers State, namely, Mgbuoba and Choba. They found one trematode (*M. monodi*), one pentastomid (*Raillietiella* sp.), and five nematodes *(A. africanum, R. africanus and C. ornata, Oswaldocruzia hoepplii* and *Chabaudus leberrei*). Generally, it is known that amphibians lose some parasites, especially intestinal nematode parasites when kept in captivity; these may wander away through the rectum.

The prevalence of *C. ornata* was same in both experimental groups but prevalence of *M. monodi* and *R. africanus* were higher in the control group while that of *A. africanum* was higher in the ALAN exposed group. These differences are not thought to have been contributed to by the experimental treatments. If other wise, more detailed future studies will reveal such contributions.

CONCLUSION

This research has shown that artificial light at night poses stress on amphibians. It has also shown that for short periods of exposure, the effect is better revealed by a measure of the hepato somatic index (HSI), than by the condition factor (CF).

In future similar experiments, the set up should be made under natural conditions of day and night time except for the introduction of the artificial light source to aid in better understanding of the effect of ALAN.

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