University of Nevada, Reno

Effects of Artificial Light at Night on Zebra Finch Nocturnal Behavior

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Abstract

With more of the world’s population living in urban areas every year, artificial light is becoming an increasingly pervasive pollutant worldwide. It is important to study the potential effects of artificial light at night (ALAN) exposure, particularly on diurnal vertebrates. Previous studies have shown that ALAN suppresses melatonin and increases nighttime activity, however, studies quantifying specific behaviors taking place under exposure to dim ALAN are lacking. I exposed zebra finches (*Taeniopygia guttata*) to dim ALAN (1.5 lux) and used infrared video recording to test how ALAN affects specific nocturnal behaviors. I recorded body movement, hopping, grooming, eating, and drinking at four timepoints in the night (17:00, 21:00, 2:00, and 6:00). After 4 days of ALAN exposure, activity significantly increased for all ALAN-exposed individuals, including increased eating behavior. For both groups, body movement was the most common behavior for birds when active. I provide evidence that suggests dim ALAN is enough to alter overall nocturnal behavior and specific activity levels.

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Table of Contents

Introduction 1

Methods 3
  Experimental Design 3
  Video Recording 4
  Statistical Analyses 4

Results 5

Discussion 9

References 9

List of Tables

Results

Table 1. Individual bird nighttime activity analysis between pre-treatment and treatment 7

List of Figures

Results

Figure 1. Average nighttime activity of control and ALAN birds............................. 7
Figure 2. Average nighttime activity breakdown for control and ALAN birds......... 8
Figure 3. Average nighttime activity during treatment at different timepoints 8

Introduction

It is estimated that over 4.2 billion people currently live in urban areas, and that 6.7 billion people will live in urban areas by 2050, indicating that the world's population is rapidly urbanizing (United Nations et al., 2019). With increased urbanization comes increased light pollution caused by artificial light at night (ALAN). More than 80% of the world's population experiences light pollution (Falchi et al., 2016). This can be harmful to the wellbeing of humans and wildlife, since ALAN is linked to the disruption of circadian rhythms (Dominoni et al., 2016; Gaston et al., 2015). Circadian rhythms are necessary for regulating sleep, metabolism, hormone secretion, and cardiovascular systems, and disrupting them can lead to pathology in the form of chronic diseases, obesity, depression, and other adverse health effects (Potter et al., 2016). ALAN also causes the suppression of nighttime melatonin and sleep loss in diurnal animals, which also have serious health consequences (McCoy & Strecker, 2011; Trivedi et al., 2017). Therefore, it is important to understand how ALAN affects behavior in organisms, so that the negative effects of light pollution can be properly alleviated.

Birds are diurnal and rely on circadian light cycles for biological rhythms, making them an ideal model to observe for artificial light at night effects (Trivedi et al., 2017; Woller & Gonze, 2013). ALAN has been found to reduce nighttime melatonin levels, disrupt rest and eating patterns, and lead to weight gain in birds (Batra et al., 2019). However, effects of ALAN on behavior effects are not well understood.

Light loggers have been placed on free-living birds in previous studies to determine how long birds were exposed to unnatural light intensities at night and how it related to their activity (de Jong, 2016b; Dominoni & Partecke, 2015). Dominoni and

Partecke (2015) used radio-telemetry to determine locomotion activity, and found that activity patterns were related to light exposure at night. Birds were also found to exhibit light-avoidance behaviors by choosing roosting locations with reduced ALAN intensities (de Jong et al., 2013b). It has also been established that free-living songbirds exhibit photoperiodic singing behavior earlier in the morning when exposed to ALAN (da Silva et al., 2016). Still, these studies are on free-living birds, and activities were not video recorded.

In previous studies in labs, activity was monitored using perches or motion detection to know a general idea of when activity was occurring (Alaasam et al., 2018; Batra et al., 2019), but these methods may under-estimate total awake time if individuals are awake but not moving, and overlook potential fine-scale behavioral changes. To-date, little is known about what kinds of behavioral activities are occurring during ALAN-exposure. Furthermore, many studies have investigated the impacts of bright night-light that causes extreme sleep-debt, but relatively few have investigated how dim ALAN, which is comparable to diffuse environmental light, may be impacting nocturnal behavior.

In this study, the effects of ALAN on zebra finch (*Taeniopygia guttata*) nocturnal behavior are observed. Zebra finches were exposed to LED night light at 1.5 lux, as this intensity is in a realistic range experienced by wild birds who live in the vicinity of ALAN (Dominoni et al., 2013). This level was also shown to alter passerine nighttime behavior (de Jong et al., 2016a) and suppress melatonin levels in rodents (Brainard et al., 1982). I hypothesized that individuals exposed to ALAN would have increased levels of nighttime activity, in accordance with previous studies (Alaasam et al., 2018; Batra et al., 2019; de Jong et al., 2016a; Dominoni et al., 2013; Ouyang et al., 2017). I also expected an increase of eating behavior to occur in ALAN individuals, as weight gain in birds was an outcome seen in a previous study (Batra et al., 2019).

Methods

Experimental Design: All procedures were carried out in accordance with National Institute of Health guidelines and were approved by the University of Nevada, Reno Institutional Animal Care and Use Committee. Six male zebra finches, that had been raised in open-air aviaries, were brought to the University of Nevada, Reno for the experiment. The birds were not previously exposed to artificial light at night. The birds were housed individually and habituated for 4 weeks to a photoperiod of 10 hours of light and 14 hours of darkness (10L:14D). This was chosen to mimic the natural photoperiod during late fall, when the experiment was carried out. Day lights (circular 1.4-Watt 5000K LED aluminum fixtures rated at 95 Lumens) turned on at 7:00 and turned off at 17:00. Blackout shades were attached to each cage so that outside light could not enter. White noise was played throughout the bird housing to eliminate extraneous noise and deter communication between the birds. Birds were free-fed and provided with as much water as necessary.

After the habituation period, the birds were divided into control (n=3) and ALAN treatment (n=3) groups. For the ALAN treatment group, night lights (20cm X 1.5cm 5000K LED strips) turned on at 17:00 and turned off at 7:00. ALAN exposure lasted for ten days. The mean illuminance of these lights was calculated to be 1.5 lux ±0.01 with an Extech Easyview Digital Light Meter (model EA13). For more information on the spectral composition of light sources, see Alaasam et al. (2018). The control group experienced the same conditions as the habituation period, with a 10L:14D photoperiod.

Video Recording: Bird activity was recorded by ELP wide angle fisheye 170 degree LED infrared cameras affixed to the top of each cage via iSpy Video Surveillance Software 64 (v7.2.1.0). Activity was recorded during the hours of 17:00, 21:00, 2:00, and 6:00. Birds were recorded one at a time at one-minute intervals during the recording hours. Recording took place the four days prior to treatment and the first four days of treatment. Videos were then watched and behavior was noted by an observer blind to the treatment. For every one second of video, behavior was categorized into one of 6 categories: nothing, body movement, hopping, grooming, eating or drinking. “Nothing” was defined as no activity observed aside from breathing. “Body movement” was defined as head and body movement where foot position was maintained, and did not include grooming, eating, or drinking. “Hopping” was defined as bird movement around the cage or perches, and a change in foot position. “Grooming” was defined as having head turned and actively preening feathers, or using foot to scratch face. “Eating” was defined as pecking at seeds, or consuming seeds. “Drinking” was defined as drinking water from the water dish. Activities were then summed into broader categories of “active” or “inactive” for further analysis, where “active” activities included all behaviors except for “nothing” which was considered “inactive.”

Statistical Analyses: A paired t-test was run on the mean nightly activity level of each group pre-treatment and during treatment to determine if there was a significant increase in nighttime activity. A Student’s t-test was also run on each bird individually pre-treatment and during treatment to investigate possible individual variation. Significance was set at $p<0.05$ for t-tests.

Results

ALAN exposure had a significant effect on activity, where birds exposed to ALAN were more active at night than control birds (Figure 1). For the ALAN-treatment group there was a significant difference between the nighttime activity before treatment and during treatment ($t=8.458$, $df=2$, $p=0.007$; Figure 1). For control birds, there was no significant difference between the nighttime activity before treatment and during treatment ($t=0.752$, $df=2$, $p=0.265$; Figure 1). Nighttime activity also increased during treatment for each ALAN bird individually (Table 1). All three ALAN birds exhibited a significant increase in activity, while all three control birds exhibited no significant increase in activity (Table 1), indicating treatment had a significant effect on nighttime activity.

Body movement made up the largest percentage of activity for both groups, both before and during treatment (Figure 2). During treatment, body movement and grooming occurred at higher rates for ALAN birds than control birds, while hopping and eating occurred at lower rates (Figure 2B). This differs from activity observed before treatment, where grooming occurred at a higher rate for control birds than ALAN birds (Figure 2A). ALAN birds ate during treatment, but not during pre-treatment, while eating activity did not change for control birds (Figure 2). ALAN birds were more active than control birds at all recorded timepoints during treatment (Figure 3). The greatest differences in activity between control and ALAN birds occurred at 2:00 and 6:00 (Figure 3). In both groups, the birds were most active at 6:00 and least active at 2:00 (Figure 3).

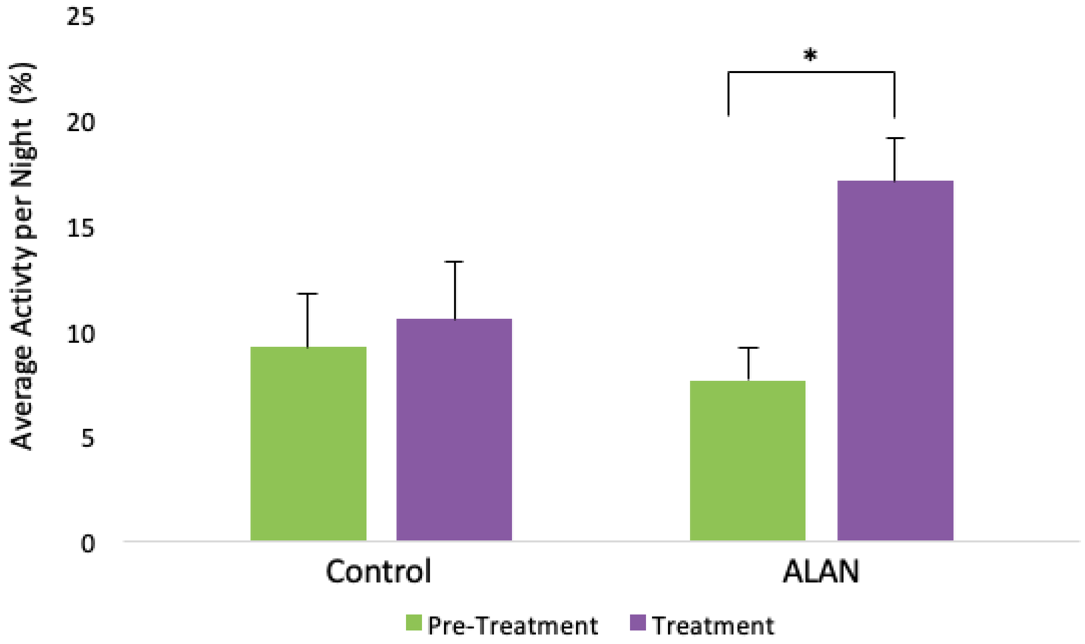

Figure 1. Average nighttime activity of control and ALAN birds. During treatment, ALAN birds had increased nighttime activity as compared to control birds. Activity also increased for ALAN birds from pre-treatment to during treatment, while activity did not increase for control birds.

Table 1. Individual bird nighttime activity analysis between pre-treatment and treatment. Each bird was analyzed individually using a student's t-test. For all ALAN birds, nighttime activity increased significantly from pre-treatment to treatment. There was no significant nighttime activity increase for any of the control birds. Activity change is based on average activity percentages during pre-treatment and treatment. P-values <0.05 are in bold.

Bird ID	**Treatment**	**Activity Change**	**t-value**	**p-value**
5	Control	-0.566%	0.214	0.419
6	Control	5.574%	1.664	0.074
13	Control	-1.075%	0.276	0.396
2	ALAN	6.707%	1.966	**0.048**
3	ALAN	10.482%	2.244	**0.033**
14	ALAN	11.052%	7.097	**0.000**

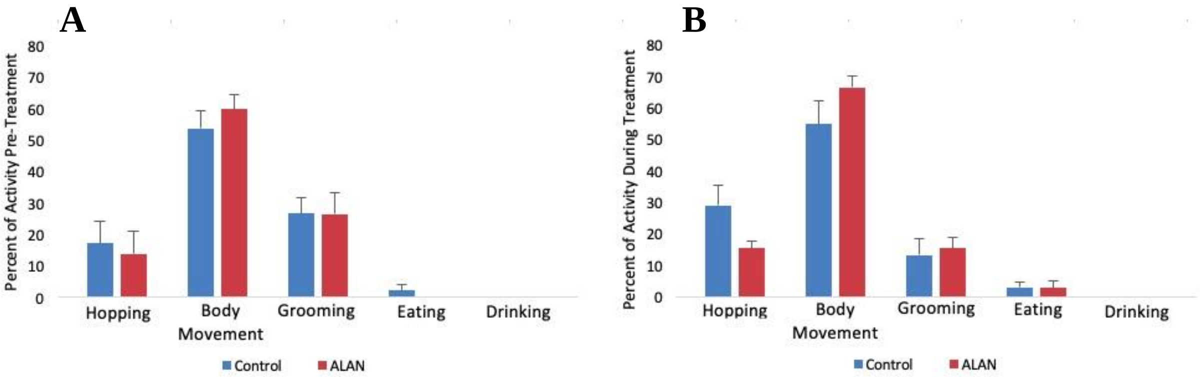

Figure 2. Average nighttime activity breakdown for control and ALAN birds. A) Pre-treatment activity. Body movement made up the largest portion of activity for both groups, followed by grooming and hopping. B) Treatment activity. Body movement still made up the largest portion of activity for both groups. An increased portion of activity was made of up of body movement for ALAN birds.

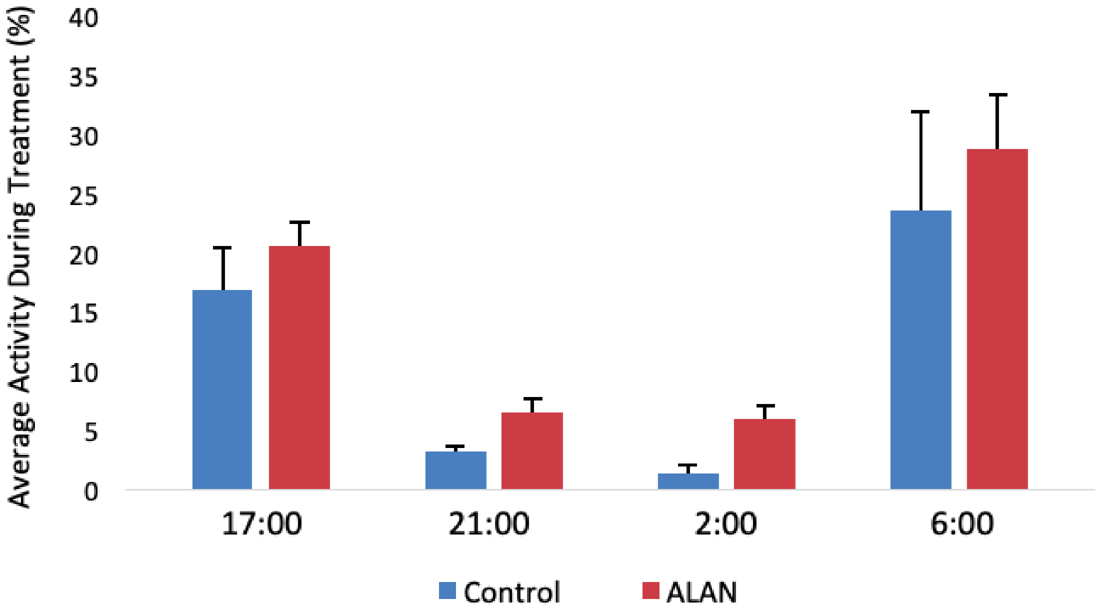

Figure 3. Average nighttime activity during treatment at different timepoints. Nighttime activity was recorded at these four timepoints. ALAN birds were more active than control birds at all timepoints.

Discussion

Artificial light at night is currently a major worldwide pollutant and potential health risks caused by light pollution is an active area of research. In this study, I observed the effect of dim artificial light at night on the nighttime activity of zebra finches. Nighttime activity significantly increased for all individuals exposed to ALAN. A night-light level at 1.5 lux was enough to produce this change. This is noteworthy because most of what is known about ALAN effects on behavior comes from research using much brighter light (5-180 lux; Batra et al., 2019; Bedrosian et al., 2011; Coomans et al., 2013; Ouyang et al., 2017). Dim night-light levels used here are ecologically relevant, because previous studies have shown that free-living birds are exposed to a wide range of light intensities (de Jong, 2016b; Dominoni & Partecke, 2015).

It has previously been shown that general activity levels have been affected when birds are exposed to ALAN (Alaasam et al., 2018; Batra et al., 2019; de Jong et al., 2016a; Dominoni et al., 2013; Ouyang et al., 2017), which is consistent with what I found in this study. Here, activity under ALAN increased at all four timepoints throughout the night. This indicates ALAN-exposure affects behavior throughout the entire night, and that ALAN-exposed birds were sleeping for shorter durations than control birds. However, the greatest amount of activity occurred at 17:00 and 6:00, and less activity occurred at 21:00 and 2:00. This suggests that ALAN-exposed birds went to bed later and woke up earlier and that sleep loss was not consistent throughout the night, since activity time varied throughout all recorded hours of the night. Therefore, this might also suggest that circadian shifts may happen at activity onset and offset (Potter et al., 2016). A limitation to this study is that only four timepoints were observed, and it is possible for discrepancies to occur if the entire night was monitored.

In this study I present novel findings about how ALAN changes specific activities in individuals while active at night. In individuals exposed to ALAN, body movement and grooming activities occurred at higher rates than control individuals. Increased body movement may be attributed to overall restlessness while perched that has been reported in other research (Ouyang et al., 2017). It has been found that grooming behaviors are increased in stressful situations in rodents, as an outlet of stress-relief (Kalueff & Tuohimaa, 2004). Therefore, the increased grooming behavior in ALAN-exposed birds may be indicative of increased stress caused by ALAN exposure (Alaasam et al., 2018).

Eating behavior also increased for birds under ALAN exposure, but not for controls. This could explain why weight-gain was found in birds exposed to ALAN in a previous study (Batra et al., 2019). Photic desynchronization can lead to alterations in metabolism-regulators such as glucocorticoids and endocrine and autonomic pathways, which can ultimately lead to weight-gain and obesity (Plano et al., 2017). In another study, increased food intake and weight-gain were not found (Alaasam et al., 2018), which may indicate ALAN-exposed birds are not increasing their caloric intake, but eating at different times relative to normal. The increased eating behavior in ALAN-exposed birds at night is likely due to a disruption in circadian feeding rhythms. This eating behavior occurred during the 6:00 hour, indicating the ALAN-exposed birds were waking up earlier and eating earlier than control individuals.

In future studies, it would be interesting to look at how daytime behavior compares to nighttime behavior with ALAN, especially regarding specific activities. It would also be interesting to look at how color temperatures of light affect different activity rates, as different night-light colors have been shown to have different effects on behavior (Alaasam et al., 2018). Since ALAN-exposed individuals exhibited an increase in eating behavior in this study, it would be of interest to further monitor eating behaviors under ALAN. This study was limited in sample size, and so only adult males were used in order to reduce possible effects of individual variation. Follow up research could use larger sample sizes and test for differential effects across sexes and age classes.

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