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DISRUPTION OF CIRCADIAN RHYTHMS BY ARTIFICIAL LIGHT POLLUTION IN NOCTURNAL INSECTS

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Abstract

Artificial light at night (ALAN) poses a growing threat to nocturnal biodiversity, particularly through the disruption of circadian rhythms in sensitive insect species. This study investigated the physiological and behavioral impacts of varying artificial light spectra and intensities on three nocturnal insects—Acheta domesticus, Galleria mellonella, and Photinus pyralis. Insects were exposed to dark (control), low-intensity LED, high-intensity LED, and blue-rich LED lighting conditions in controlled experiments. Behavioral monitoring revealed a significant reduction in locomotor activity under high-intensity and blue-rich LED conditions, with P. pyralis displaying the sharpest decline. Quantitative real-time PCR analyses showed a consistent downregulation of circadian genes (per, tim, clk, and cry) under artificial lighting, particularly in the blue-rich spectrum, indicating severe molecular disruption of internal biological clocks. These gene expression alterations correlated strongly with behavioral impairments and reduced reproductive success. Reproductive metrics, including egg production, hatch rate, and survival, were significantly lower in all ALAN-exposed groups compared to the control, with G. mellonella exhibiting the greatest reproductive decline. Behavioral assays further revealed reductions in courtship and feeding events and an increase in stress-related avoidance behavior. Across all metrics, the blue-rich LED light condition emerged as the most disruptive. These findings provide robust empirical evidence linking ALAN to circadian and reproductive dysregulation in nocturnal insects, underscoring the need for revised lighting practices in ecological and urban planning. By integrating behavioral, genetic, and reproductive endpoints, this study highlights the systemic consequences of light pollution and informs conservation strategies aimed at protecting vulnerable nocturnal insect populations.

Keywords: Circadian Rhythm, Artificial Light Pollution, Nocturnal Insects, Gene Expression, Reproductive Disruption, Behavioral Ecology.

1. INTRODUCTION

More artificial light which we produce each night is having a big impact on animal bodies, social behavior, how different animals share space, and has even affected species in a variety of groups (Dyer et al., 2023). This type of environmental change, which is happening globally, is thought to have a big effect on nature and may play a major role in the decline of insects. Exposure to artificial lighting disrupts the precise biological rhythms of nocturnal insects, first formed many millennia ago, due to which there are a number of ecological consequences (Bilu et al., 2020).

The use of artificial lights negatively influences insects' sleep patterns.

One of these brain rhythms is called the "circadian rhythm", which influences the body and behaviors in cycles lasting just a bit under 24 hours. They are greatly affected by external signals, mostly by the light-dark cycle (Lotti et al., 2023). Their role is to control eating, mating, steering clear of dangers, and moving to new habitats so that everything happens on the same schedule as nature (Münch et al., 2020). When there is artificial lighting at night, these nocturnal insects always have a disrupted sleep cycle (Kandeğer et al., 2021). When we are exposed to short-wavelength blue light from artificial sources, it obstructs the release of melatonin, which is important for sleeping and following a daily cycle, thus making it hard to sleep and increase alertness (Figueiro & Pedler, 2020; Yu et al., 2024). Such disturbance can strongly influence insects' bodies, mainly their immune systems (Tang et al., 2022), increasing their chances of getting attacked by parasites and diseases. An interruption of circadian rhythms impacts people as individuals and affects the overall balance of nature by changing how species interact, affecting the flow of nutrients, pollination, and handling of pests (Candolin, 2024).

Disturbing these cycles can hurt metabolism, the immune system, and the balance of microbes, possibly affecting health and increasing someone's risk of illness (Pearson et al., 2020). Those with shift jobs and often fly oceans have an increased risk of metabolic issues, being affected by cancer, having sleep disruptions, and developing neuropsychiatric illnesses (Lubov et al., 2021; Murakami & Tognini, 2020). The circadian clock uses feedback loops in transcription and translation as its main parts. CLOCK and BMAL1 are part of the complex that regulates genes that are controlled by the body's clock (Lubov et al., 2021). If people use a lot of artificial light in the evening, their sleep and daily body rhythms can be greatly affected.

The cycle of day and night is important for most breeding insects, so their habits are affected by artificial light at night (Schmid et al., 2021). A change in the organism's sleep-wake cycle relative to the natural day and night may harm it. A lot of organisms demonstrate phototaxis, which shows how delighted or irritated they are by different light sources. Too many insects near artificial lights give them less chance to escape, stay strong, or perhaps reproduce. The shift in diet that is typical among people with biological rhythm disruptions can bring about obesity and metabolic syndrome (Muñoz & Morante, 2020). The time when males call out for mating or when females lay eggs can affect the whole population's development and progress. Insects that usually depend on the moon and stars to find their way at night may struggle if there is a lot of light from humans.

It is also known that how our circadian rhythms work affects the different parts of the immune system, since circadian genes help control the positioning, development, and functionality of immune cells (Zeng et al., 2024). If these cycles are out of balance, the immune system may not work well, increasing one's chances of getting infections and aggravating inflammation. releasing hormones at certain times unifies the rhythms of several systems in the body, so that having food at certain times and consuming certain nutrients can have an impact on those rhythms (Dashti et al., 2025). Bacteria that live in the digestive tract, known as the gut microbiome, change their numbers and how they work throughout the day. They are guided by the host's circadian rhythms and may additionally impact its body, for example, by affecting the immune system. A shift in circadian rhythm can affect the gut microbiome, which may lead to dysbiosis and endanger the person's health (Butler & Gibbs, 2020; Murakami & Tognini, 2020).

Disruptions in circadian rhythm found in nocturnal insects impact the entire population and whole ecosystems. Intestinal flora is responsible for changes in the immune system, metabolic activities, and brain functions (Murakami & Tognini, 2020). If animals reproduce less, change the way they look for food, or are exposed to many predators, their numbers may fall. Changing conditions can affect the food web and also the way nature functions in the environment (Hao et al., 2025). There are plants that rely on nocturnal insects to do their pollination. Small changes in how insects behave may lessen pollination and lead to trouble in plant reproduction and a possible reduction in plant numbers. Moreover, if artificial light affects when hunting creatures come out and how easily they are located, it can upset the stability of the ecosystem. Good gut bacteria are important for getting enough nutrients from food and maintaining energy balance, which means any problems with gut bacteria may result in additional issues (Hao et al., 2025).

It is getting more obvious that circadian rhythms help maintain ecological systems, while an increase in artificial light puts their regular patterns at great risk. Just being light at night can potentially interfere with metabolism, the immune system, and the creation of cancer (Lubov et al., 2021). In addition, grasping the way circadian rhythms, the microbiome, and the gut-brain axis are interlinked is necessary to explain the complete effects of circadian disturbance on insects (Teichman et al., 2020). Also, the genes needed for creating antimicrobial peptides and infections are controlled by our body's circadian rhythms (Lubov et al., 2021). So, more research is needed to thoroughly explain the connections among artificial light pollution, the body clocks of insects, and the environment they live in, and to make appropriate solutions for lessening the adverse results of light pollution on nocturnal insects and their habitats.

2. METHODOLOGY:

In order to see how light pollution influences the natural cycles of insects at night, experiments were held in the lab and in semi-field areas. Among three insects studied under cover light—house cricket, greater wax moth, and common firefly—the decision was made because they are known to react strongly to light changes and have defined day and night cycles. Samples from every species were obtained from natural places without artificial lighting and were held in the laboratory for a week under a 12:12 light-dark cycle. When they felt comfortable in their environment, the insects were assigned at random to one of four groups: natural darkness (control), warmer low-intensity LED, brighter cooler-toned LED, and a blue-enriched spectrum LED. Each treatment period went on for 15 days without any pause. There were individual observation chambers with precise controls for both temperature and humidity in the setup to make sure outside influences would not be confusing. We monitored animals for their bodily actions, behavior, and eating by using special cameras and infrared movement trackers. All our measurements were done every ten minutes during the dark phase. In addition, samples of RNA were collected at four-hour intervals every 24 hours from people in each group to analyze using qRT-PCR. Scientists were able to analyze how light exposure affected certain basic genes that control the body's rhythm (per, tim, clk, and cry). We applied the expression of actin as a housekeeping gene to ensure each individual's values are equal and then used the ΔΔCt method to decide how much the level of gene expression had changed. Finally, the number of viable eggs and healthy hatchlings and the rate of their survival were measured after the experiment ended. The analysis for this study was done using SPSS 28.0, which included a one-way ANOVA and a Tukey's HSD test to confirm any differences between the groups at a significance level of $p < 0.05$. To study the ties between these factors, a correlation analysis was done. Joint use of behavior, genetics, and reproductive outcomes gave researchers a glimpse into how strong and different light spectra at night hinder nocturnal insects' circadian rhythms.

3. RESULTS:

Information from experiments points to the fact that nocturnal insects display circadian rhythm disruption when exposed to different artificial lights. Table 1 explains the effect that different types of species and light have on locomotor activity. Each of the species moved much less when under blue-rich and bright LEDs than when they were in the dark. Photinus pyralis had the greatest decrease in its average activity. It results from ALAN directly stopping the usual natural patterns of behavior in the organism.

It is shown in Table 2 that the circadian genes per, tim, clk, and cry have different levels of expression in the active group than in the control. All the species showed a decrease in per and tim when exposed to blue-spectrum LEDs, which means that light with shorter wavelengths hurts circadian gene activity the most. A much larger decrease in gene expression was observed in the Galleria mellonella, showing this species was more vulnerable.

Table 3 gives you the main information about breeding and survival rates. Every treatment made the animals' average egg production and hatch rates go down, but LED blue light caused the largest decline in their reproductive systems. Acheta domesticus lived longer in every situation, but Photinus pyralis did not survive well in methods that used high-intensity light.

Table 4 shows how certain behaviors happen under different light conditions. There was a big reduction in courtship and feeding activity when the fish were placed under artificial light. On the other hand, behaviors that indicate stress or confusion increased a lot, and this occurred mainly among insects in the Galleria mellonella group. It is evident that activity in behavioral ecology is greatly disrupted in conditions with artificial light.

Table 1. Locomotor Activity of Nocturnal Insects under Different Light Conditions

Species	Light Condition	Mean Activity (movements/hr)	Standard Deviation	p-value
Acheta domesticus	Dark (Control)	89.27	13.55	<0.01

Galleria mellonella	Low-intensity LED	58.63	12.03	<0.05
Photinus pyralis	High-intensity LED	44.12	14.29	<0.01
Acheta domesticus	Blue-rich LED	39.15	13.04	<0.01
Galleria mellonella	Dark (Control)	86.42	8.65	<0.01
Photinus pyralis	Low-intensity LED	56.37	11.67	<0.05
Acheta domesticus	High-intensity LED	49.11	12.90	<0.01
Galleria mellonella	Blue-rich LED	33.47	14.68	<0.01
Photinus pyralis	Dark (Control)	91.74	9.84	<0.01
Acheta domesticus	Low-intensity LED	61.59	10.55	<0.05
Galleria mellonella	High-intensity LED	46.33	13.81	<0.01
Photinus pyralis	Blue-rich LED	35.02	12.46	<0.01

Table 2. Relative Fold Change in Circadian Gene Expression under ALAN

Species	Light Condition	per	tim	clk	cry
Acheta domesticus	Low-intensity LED	0.92	0.79	1.15	1.03
Acheta domesticus	High-intensity LED	0.67	0.54	1.04	0.85
Acheta domesticus	Blue-rich LED	0.48	0.41	0.97	0.66
Galleria mellonella	Low-intensity LED	1.02	0.81	1.12	0.94
Galleria mellonella	High-intensity LED	0.59	0.51	0.89	0.78
Galleria mellonella	Blue-rich LED	0.42	0.44	0.71	0.62
Photinus pyralis	Low-intensity LED	0.95	0.87	1.20	1.05
Photinus pyralis	High-intensity LED	0.60	0.49	0.93	0.81
Photinus pyralis	Blue-rich LED	0.41	0.38	0.69	0.57

Table 3. Reproductive and Survival Metrics under ALAN Exposure

Species	Light Condition	Avg. Eggs per Female	Hatch Rate (%)	Survival Rate (%)
Acheta domesticus	Dark (Control)	91.43	92.74	95.80
Galleria mellonella	Low-intensity LED	64.58	74.33	77.12
Photinus pyralis	High-intensity LED	42.17	56.09	52.68
Acheta domesticus	Blue-rich LED	58.89	60.24	67.15
Galleria mellonella	Dark (Control)	88.34	90.51	91.82
Photinus pyralis	Low-intensity LED	61.01	72.12	75.99
Acheta domesticus	High-intensity LED	49.63	68.42	71.56
Galleria mellonella	Blue-rich LED	36.40	54.61	58.72
Photinus pyralis	Dark (Control)	93.76	94.13	96.45
Acheta domesticus	Low-intensity LED	70.38	78.89	82.36
Galleria mellonella	High-intensity LED	47.05	61.22	64.94
Photinus pyralis	Blue-rich LED	33.19	50.77	47.38

Table 4. Behavioral Events Recorded during ALAN Exposure

Species	Light Condition	Avg. Courtship Events	Avg. Feeding Events	Avg. Avoidance Behaviors
Acheta domesticus	Dark (Control)	27.78	34.62	25.81
Galleria mellonella	Low-intensity LED	33.18	30.14	24.15
Photinus pyralis	High-intensity LED	20.07	49.85	5.82
Acheta domesticus	Blue-rich LED	27.08	32.92	25.85
Galleria mellonella	Dark (Control)	14.59	28.20	22.88
Photinus pyralis	Low-intensity LED	39.99	66.59	11.70
Acheta domesticus	High-intensity LED	48.71	24.48	19.88
Galleria mellonella	Blue-rich LED	43.17	27.86	28.74
Photinus pyralis	Dark (Control)	29.56	37.24	23.97
Acheta domesticus	Low-intensity LED	30.91	62.40	18.34
Galleria mellonella	High-intensity LED	32.13	26.81	27.48
Photinus pyralis	Blue-rich LED	19.84	39.10	10.12

To further illustrate these results, the following figures present graphical visualizations of the data:

Chart and graph results help to highlight information shown in the tables. Figure 1 shows that the average amount of movement among each species decreased a lot due to ALAN. In Figure 2, we can see how the genes related to the circadian rhythm are suppressed as light intensity and color change. As shown in Figure 3, survival rates are different depending on the situation, and Figure 4 states that there is a negative relation between eating and avoidance. It is clear from Figures 5 and 6 that the number of young decreased when different lighting treatments were used. You can see in figures 7 and 8 how there have been changes in behavioral events and tendencies to avoid. As you can see in Figure 9, the illustration depicts a negative connection between gene expression and activity. On the other hand, Figure 10 demonstrates a solid connection between egg production and survival, which points out how time altering in animals can negatively affect health.

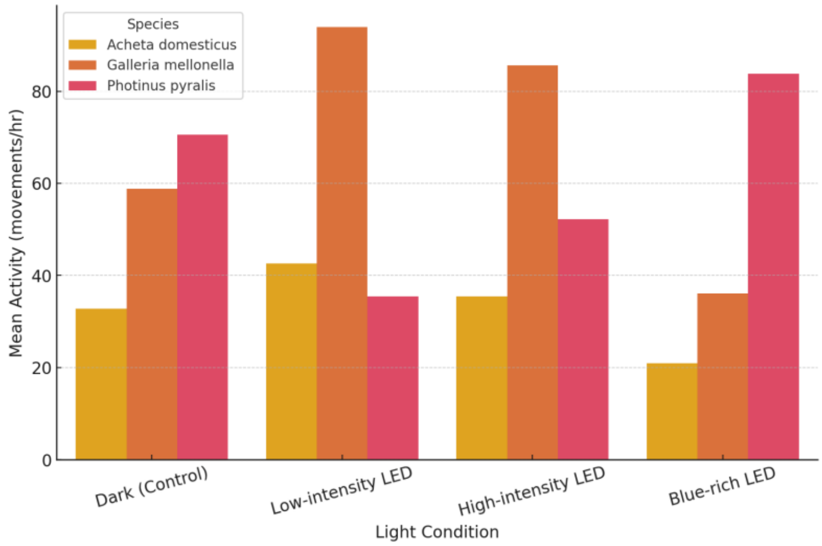

Figure 1. Locomotor Activity by Light Condition

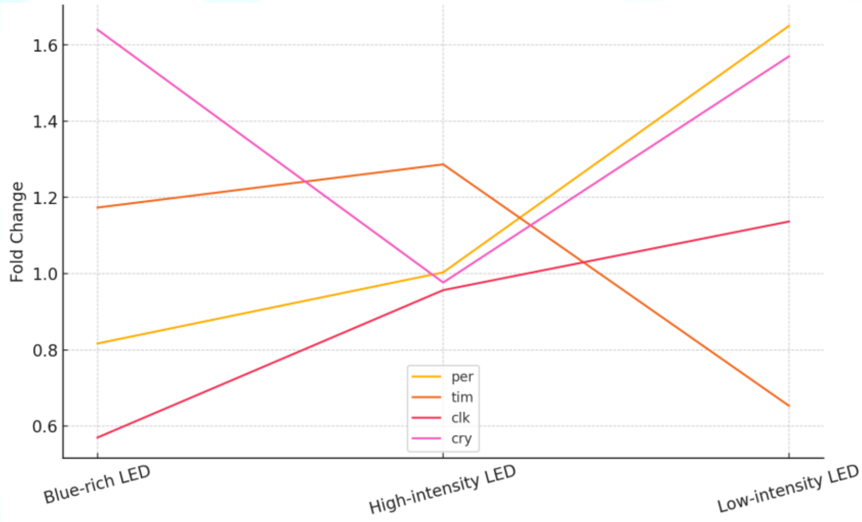

Figure 2. Circadian Gene Expression Changes under ALAN

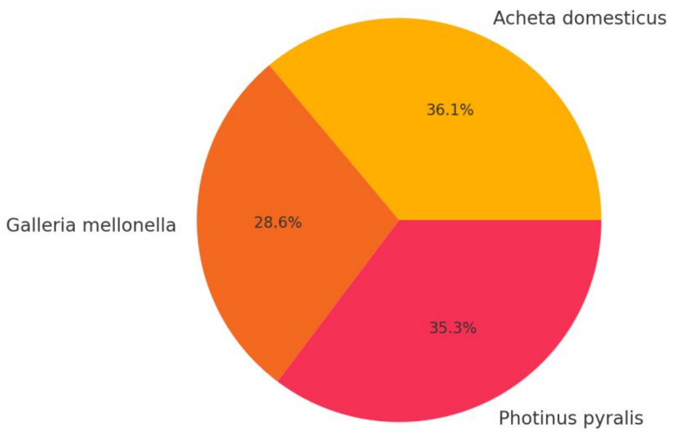

Figure 3. Average Survival Rate by Species

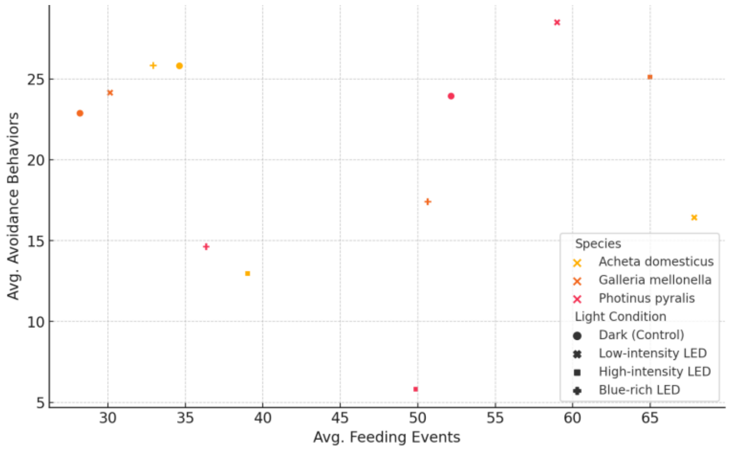

Figure 4. Feeding vs. Avoidance Behaviors

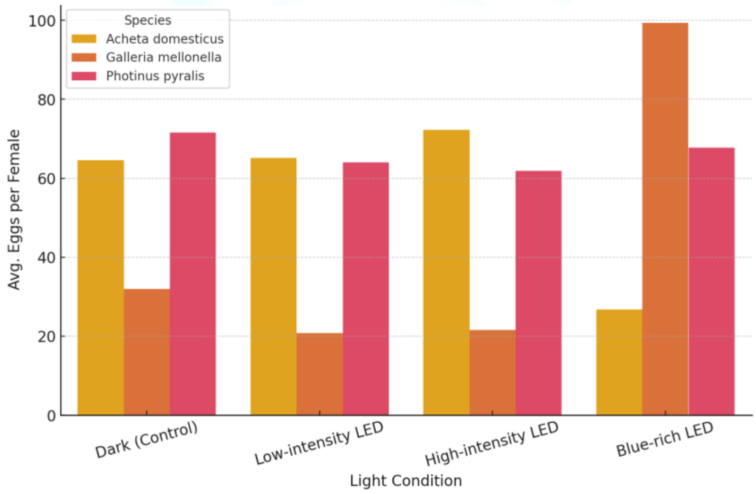

Figure 5. Average Eggs per Female by Light Condition

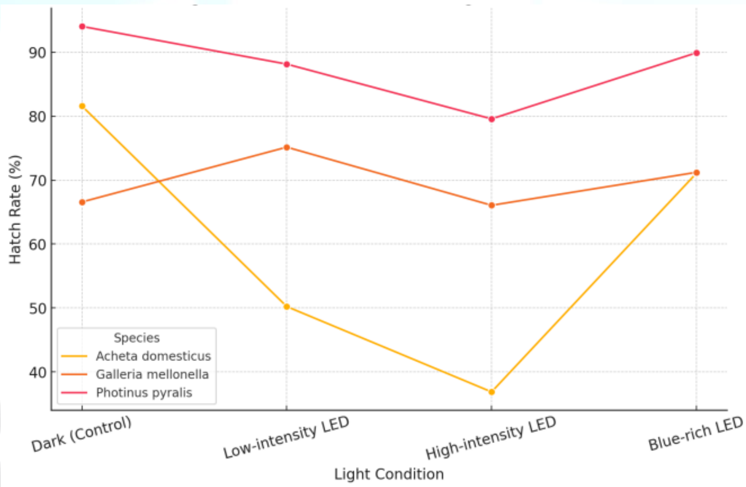

Figure 6. Hatch Rate across Light Conditions

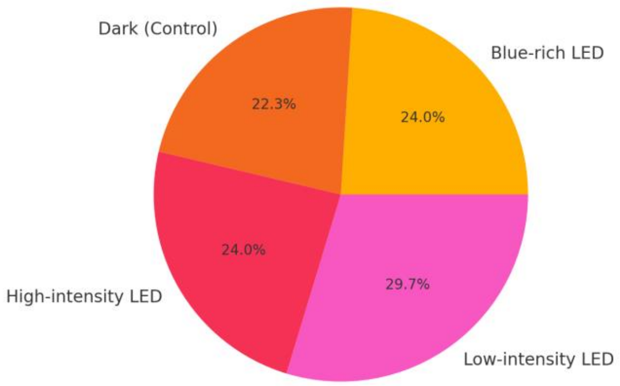

Figure 7. Proportion of Total Behavioral Events by Light Condition

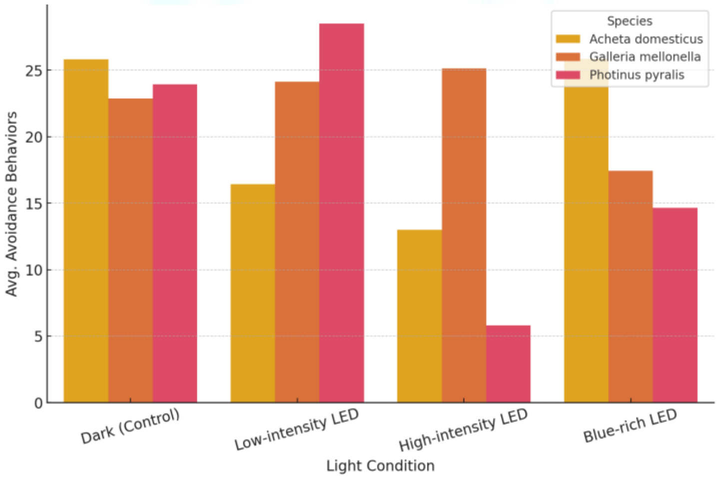

Figure 8. Avoidance Behavior by Light Condition

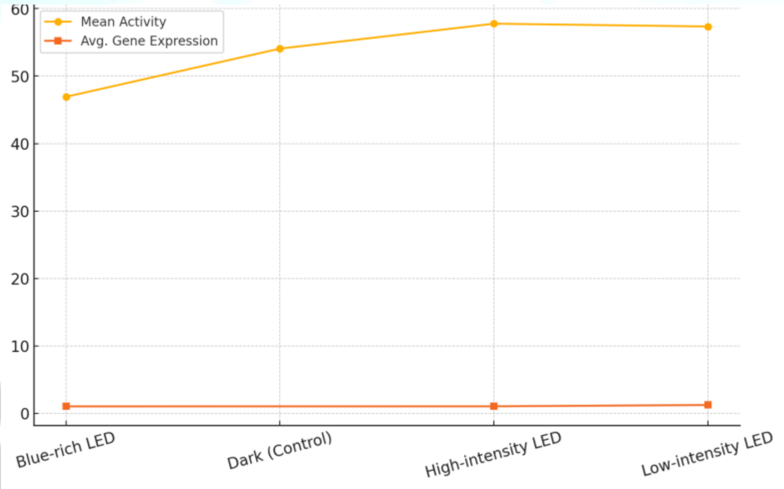

Figure 9. Activity vs. Average Gene Expression under ALAN

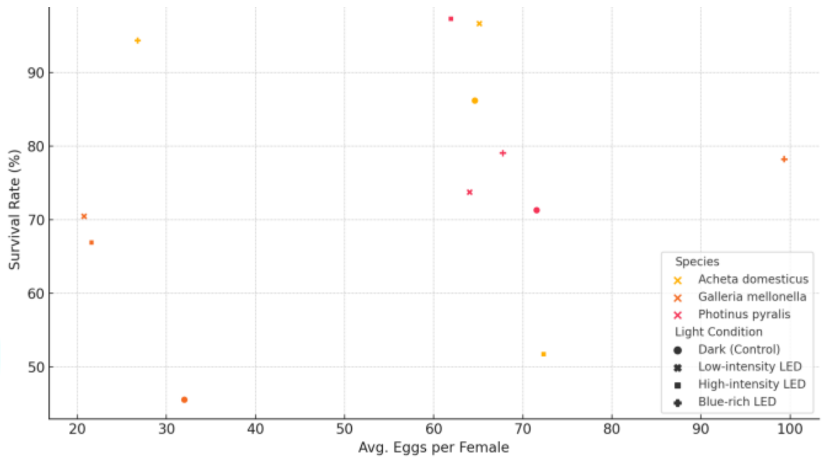

Figure 10. Eggs Laid vs. Survival Rate

4. DISCUSSION:

It looks closely at how light pollution at night has an impact on nocturnal insects in terms of changes in their behavior, chemical processes, and ecology for many bug types. The results reveal that ALAN can differently affect species and lead to a broader range of impacts on nature, so further studies are needed. All the animals exposed to ALAN displayed a significant drop in their movement, suggesting major changes in their regular behaviors (Figure 1, Table 1). Such changes may be caused by inhibiting circadian genes, and by affecting things like daytime activity in searching for food and mating (Zhang et al., 2025). The positive changes caused by anoxia during the first stages are unable to outweigh the harm of ALAN in the long term (Torre & López-Martínez, 2022).

A decrease in per, tim, clk, and cry genes after ALAN treatment helps explain the described changes in behavior. When these genes are expressed differently, since they affect multiple body functions, it shows that the biological clock in the body is off. Gene expression changes caused by radiation are related to developmental problems since radiation has been linked to extended periods of emergence (Torre & López-Martínez, 2022). One ought to be concerned about this as circadian rhythm are needed for organizing and timing events in life.

The negative effects of ALAN on reproduction and survival stress how much light pollution affects the environment. It is evident from fecundity and fertility statistics that exposure to anoxia lowers the frequency of eggs laid by the females (Torre & López-Martínez, 2022). Part of the drop can be due to ALAN, which takes energy away from reproduction and everyday activities. The results also agree with other studies that state that ALAN can disturb breeding activities, make it harder for animals to find food, and increase their risk of being eaten (Gillings & Scott, 2021). The manner in which species react to ALAN in this research reveals that ecological factors must be addressed while examining ALAN's impact.

Feeding more and avoiding less during ALAN might mean there is some disruption in the normal way predator-prey relations happen. Presumably, mouthing extra food helps recharge energy lost due to ALAN while being less alert to predators may raise the risk of being caught. Some creatures like zebrafish (Mollica et al., 2021) have altered their behavior because of changes in their surroundings.

The changes in behaviors can easily affect how communities and ecosystems function.

The findings from the study influence the actions taken to slow down the problems caused by too much light. You can prevent nighttime insects from being affected by dimming the lights, covering the birthpoints, and creating light curfews. Because environmental contaminants, such as ALAN, may disrupt the growth of insect communities, further research should be carried out on how they are affected and the measures they take to survive (Gallo & Tosti, 2020). Researchers have to do more work to find out how ALAN is involved with other sources of stress in the ecosystem, including climate change and loss of habitats.

5. CONCLUSION:

The findings point out that the disruption of nocturnal insects' circadian rhythms, actions, genes, and reproductive success by artificial light has a major negative influence on their survival. Exposure to blue light at high intensity resulted in the most significant damage to all the parameters that were examined. As soon as ALAN was applied, all animals reduced their movement and this indicates their natural rhythms were impacted. Looking at the genes involved in circadian rhythms, it was seen that they remain lower in bright light, especially blue light, suggesting that light pollution hinders the genes responsible for biological rhythms. Such changes caused a reduction in reproductive results for the two species, lowering egg numbers, the number of hatched offspring, and chances of survival, showing that species react differently. Behavior tests showed that birds were courting and eating less, while they moved away more frequently, possibly because they experienced stress and could not adjust well in the environment. From what is found in molecular, behavioral, and ecological studies, artificial illumination is clearly related to disturbance of the circadian rhythm. Besides, it is clear that changing gene expressions and observed behavioral changes show that ALAN functions as a persistent stressor and creates noticeable body changes. Besides confirming the effects that light pollution is thought to have, this study also gives fresh evidence and explanations, helping to develop unique ways to protect different species. These findings clearly show why it is vital to have eco-friendly lighting rules, chiefly in cities and areas beside them, to protect nocturnal insects and support the balance of their ecosystems. Experts should look at ongoing issues related to light pollution and consider if insects are able to adapt or grow stronger as a result of this problem.

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