CFAES

Department of Entomology

Artificial light at night (ALAN) changes seasonal responses in mosquitoes

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Figure 1. Night sky in rural and urban areas, demonstrating effect of ALAN on "sky glow". *Photo credit: J. Stanley*

INTRODUCTION

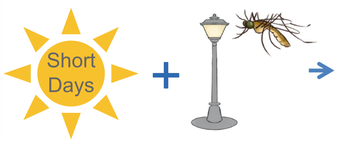
- Light pollution caused by artificial light at night (ALAN) used to illuminate human structures causes overall brightening of night sky
- Over half of United States experiences light-polluted skies, with brightest pockets occurring over cities [1]
- ALAN alters normal light cycles, and thus impacts insect physiology, behavior, and ecology [2]
- The Northern house mosquito, *Culex pipiens*, is the primary vector of West Nile Virus (WNV) in the U.S. among other diseases [3]
- Female *Cx. pipiens* enter a programmed dormancy known as diapause to survive harsh winter conditions
  - In response to short days, females cease reproductive development, halt blood-feeding, and accumulate fat, and cannot transmit disease [4]

Figure 2. During summer, female mosquitoes take a blood-meal (left). During winter, they will instead seek out shelters to survive the winter (right). *Photo Credit: left: A. Farajollahi, right: J. Layton*

QUESTION: Does ALAN interfere with seasonal responses in female *Cx. pipiens* mosquitoes?

HYPOTHESIS: Dim ALAN will inhibit diapause initiation in female mosquitoes

PREDICTIONS: ALAN-exposed mosquitoes will exhibit...



1) **Larger egg follicles**
2) **Less fat content**
3) **Higher proclivity to blood-feed**

METHODS

Insect Rearing

- ALAN created using individual LED diodes to mimic dim light at night (~4 lux)
- Lab colony mosquitoes reared from larvae to adulthood in diapause-inducing conditions (11.5 Light:12.5 Dark, 20 °C) with ALAN or no light at night (Control)
- 3 cages per treatment across 3 cohorts (n = 9 cages per treatment)
- All subsequent measurements performed on seven-day-old females

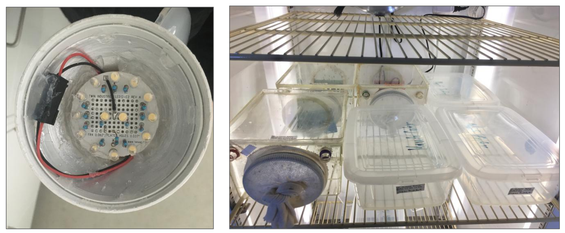
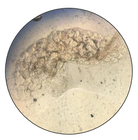


Figure 3. ALAN-treatment mosquitoes were reared under dim ALAN constructed from LEDs (left, lens removed) in clear, plastic containers (right)

Prediction 1) Ovarian Development

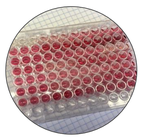
- Ovaries dissected in 0.9% saline solution
- 10 egg follicles per female measured under inverted microscope
- 20 females per cage ($n_{control}$ = 160, n_{ALAN} = 179)
- Data analyzed in R using LMM (nlme package)



Dissected ovary under microscope

Prediction 2) Fat Content

- Fat content measured using vanillin-phosphoric acid assay
- 4-8 females per cage ($n_{Control}$ = 62, n_{ALAN} = 68)
- Data analyzed in R using LMM (nlme package)



Vanillin-phosphoric acid assay samples for plate-reader

Prediction 3) Blood-Feeding Proclivity

- Females starved from sugar solution for 24 hours prior to blood-feeding
- Offered chicken blood via artificial membrane feeding system for one hour at dusk
- Number of females with blood in gut counted after feeding
- Percent fed per cage calculated ($n_{Control}$ = 6, n_{ALAN} = 5)
- Data analyzed using Chi-Square Goodness of Fit test
- Last two blood-fed cages offered oviposition water; number of egg rafts laid counted and monitored for viable larvae

Cages set-up to offer blood-meal using feeding system

RESULTS

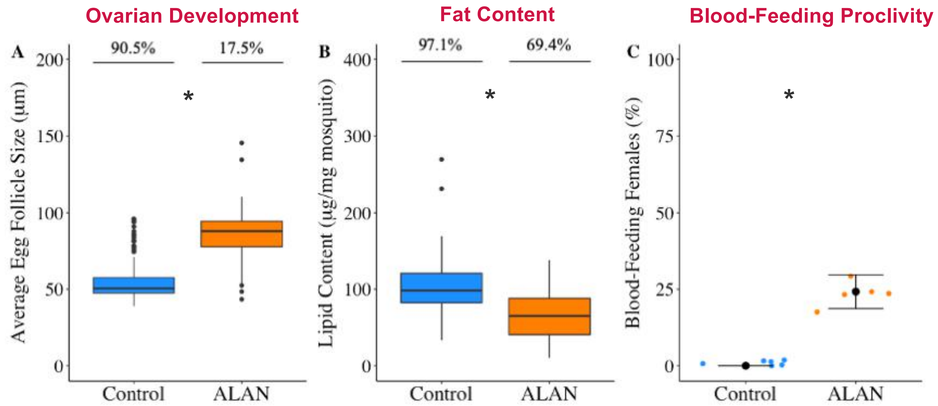


Figure 4. Exposure to dim ALAN prevents female mosquitoes from entering diapause.

Outliers are indicated by black dots in panels A and B. Mean is shown by black dot in panel C. Percentages indicate number of mosquitoes considered in diapause (average egg follicle <75 µm and lipid content >50 µg/mg mosquito). Asterisks indicate significant differences (p < 0.0001).

- ALAN-exposed females had significantly larger egg follicles than controls
- ALAN mosquitoes accumulated less fat than controls. However 70% were higher than non-diapause threshold
- 25% of ALAN-exposed females took a blood meal, while no control females blood-fed
- When offered oviposition water, 67% of blood-fed females laid egg rafts. All rafts hatched viable larvae

Figure 5. ALAN-exposed females show a combination of seasonal phenotypes. *Egg follicles (left) and fat content (right). Short-day mosquitoes entered diapause, while long-day averted diapause.*

CONCLUSIONS

- ALAN-exposed females showed evidence of reproductive development (increased egg follicle size, higher blood-feeding, producing viable larvae), indicating they averted diapause
- Exposure to ALAN similarly decreased fat accumulation, but they were still fatter than long-day, non-diapausing mosquitoes

SIGNIFICANCE

- ALAN-exposed mosquitoes are reproductively active longer, but also accumulate fat so may be able to survive food scarcity during winter
- Mosquito populations higher in cities [5], and particularly in low-income areas [6]
- Results indicate are actively biting for longer periods of time, thus increasing disease risk
- Increasing mosquito control and surveillance in cities may mitigate the impact of altered seasonal responses in mosquitoes

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