Title: Light Pollution Increases West Nile Virus Competence in a Ubiquitous Passerine Reservoir Species

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One sentence summary: Light pollution increases host infectiousness.

Abstract: Light pollution is a growing problem, but its impacts on infectious disease risk have not been considered. Previous research has revealed that dim light at night (dLAN) dysregulates various immune functions and biorhythms, which hints that dLAN could change the risk of disease epidemics. Here, we demonstrate that dLAN enhances infectiousness of the house sparrow (*Passer domesticus*), an urban-dwelling avian host of West Nile virus (WNV). Sparrows exposed to dLAN maintained viral titers above the transmission threshold to a biting vector (10^5 plaque-forming units) for two days longer than controls but did not die at higher

rates. A mathematical model revealed that such effects could increase WNV outbreak potential

by ~41%. dLAN likely affects other host and vector traits relevant to transmission, so additional

research is needed to advise management of zoonotic diseases in light polluted areas.

35   **Main Text:** Among the many anthropogenic changes that impact humans and wildlife, one of

the most pervasive but least understood is light pollution (*1*). Light pollution takes many forms

(i.e. sky glow, light clutter, glare; *2*), but dim light at night (dLAN) is exceptionally common and

important in both urban centers and non-urban areas including farms, airports, warehouses, and

wherever lighting is necessary for human activities at night (Fig. 1). Indeed, greenspaces near

40   roadways experience extensive dLAN exposure from billboards, street lamps, residential

lighting, and headlights emanating from passing vehicles (*3*).

Although dLAN is widespread, its effects on human and wildlife health have been under-studied.

Early research on human health found that individuals working throughout the night routinely

45   suffer higher rates of Type II diabetes, heart conditions and other non-infectious maladies

compared to day-working staff (*4*). The recent switch to energy efficient night-lighting, which

has converted incandescent bulbs to short wavelength cool white LEDs, may increase health risk

further. In domesticated rodents, such short wavelength light has been linked to metabolic

dysregulation, immunosuppression, and the development of some cancers (*4*). Levels of blue

50   light as low as 0.2 lux can suppress melatonin secretion in humans (*5, 6*), and in wildlife,

comparable forms of dLAN alter many behavioral, life history, and physiological traits (*7, 8*).

Despite the diverse and strong effects of dLAN, no study has yet investigated whether and to

what degree it might affect infectious disease risk. This is surprising given the pervasiveness of

55   light pollution, especially because hosts and vectors have evolved to use light cues to coordinate

multiple daily and seasonal rhythms (*9, 10*). Light is among the most reliable environmental

cues, and light regimes induce variation in the timing of animal activities as well as temporal

fluctuations in immune defenses and other factors that influence risk of infection (*11*). Our goal

was to discern whether light pollution, and dLAN specifically, could alter zoonotic disease risk

60   for humans and wildlife by changing the ability of a reservoir host to amplify virus for

subsequent uptake by a vector. Heterogeneity in such an ability, which we term host competence

(*12- 14*), is mediated by physiological pathways sensitive to light signals (*11*) and those

coordinating organismal responses to stressors, namely the glucocorticoids (*15*). Melatonin and

glucocorticoids have profound effects on both host behaviors that affect parasite exposure risk

65   and anti-vector defenses (*16-17*).

In the present study, we investigated dLAN effects on WNV infections in house sparrows

(*Passer domesticus*) because they are among the most common birds in light-polluted areas (*18*).

They are also among the more competent species for WNV (*19*), and a close commensal of

70   humans. We chose WNV for two reasons: i) >46,000 cases of human disease have been reported

across the US since its introduction to New York in 1999 (*20*), and ii) it has decimated avian

populations, particularly corvids and other passerine species that commonly occupy light-

polluted habitats (*21*). In previous work with WNV, we found that corticosterone, the

aforementioned avian stress hormone, enhances WNV competence by elevating attractiveness to

75   *Culex* mosquito vectors for blood-feeding (*22*) and increasing WNV viremia above the

transmission-to-vector threshold (10^5 log plaque-forming units (PFUs) (supp. material)). We

thus designed our study to determine whether any effects of dLAN on WNV competence were

mediated by corticosterone dysregulation.

80   To assess the effects of dLAN on house sparrow competence for WNV, we exposed wild-caught

birds to dLAN (12h light: 12h 8 lux dim light) or natural light conditions (12h light: 12h dark)

for 1-3 weeks before and after exposure to 10^1 (PFUs) of WNV, NY 1999 strain (*13*; dLAN

exposure duration was not impactful in models, so it is not further addressed below). Following

WNV exposure, we sampled serum on days 2, 4, 6, and 10 to quantify WNV viremia in

85   circulation (*13*), and we measured body mass (to 0.1g prior to and on each blood sampling day)

to assess effects on individual health. We detected a significant effect of dLAN on WNV viremia

in house sparrows (dLAN x time: $F_{4,124} = 2.9$, $P = 0.023$; Figure 2A). Over the course of the first

four days post-exposure (dpe; all animals in both groups became infected), both dLAN and

control birds had comparable viral titers, but 6 days post-exposure (dpe), dLAN individuals had

90   significantly higher viral titers than control individuals ($\beta = 1.7$ +/-0.60, t = -2.7, P = 0.006).

Previous studies have found that the minimum circulating viral titer needed to transmit WNV to

vectors is ~10^5 PFUs (horizontal dashed line in Fig. 2A).

We then asked whether dLAN might modify the capacity of individuals to transmit WNV to

95   biting vectors. When we queried whether dLAN generally affected WNV tolerance (in the form

of defense of body mass sensu *23*; % change relative to pre-WNV exposure values), we found no

effect across the entire post-exposure period (dLAN x integrated WNV titer: $F_{1,34} = 1.3$, $P = 0.26$). However, birds with the highest WNV titers overall lost more body mass than birds with

lower cumulative titers (integrated WNV titer: $F_{1, 34} = 6.6$, $P = 0.02$). Subsequently, we assessed

100   directly whether body mass changed differently post-infection in dLAN and control birds. We

found that birds in the control group tended to gain mass post-infection, whereas body mass

reached a nadir in dLAN birds on d6 post-infection (Fig. 2B). When we analyzed how body mass

and WNV titer were related over the infectious period, we found that the interaction varied with

days post-exposure (dpe x dLAN x WNV titer: $F_{3, 83} = 3.11$, $P = 0.03$). This three-way

105   interaction was driven by WNV x dLAN effects on d6 ($\beta = 1.3$ +/-0.60, $t = 2.1$, $P = 0.04$; Fig.

2C): at this time, only dLAN-exposed birds (~1/2 of dLAN birds) maintained WNV titers above

the transmission threshold; no control birds were infectious on d6. Given the stronger effects of

WNV on body mass loss in the dLAN versus control group, one might expect greater mortality

in the former. However, we found no effect of dLAN on survival of WNV infection post-

110   exposure ($\chi^2_1 = 0.26$, $P = 0.61$; Fig. 2D). About 60% of birds in each group survived to d6.

The second goal of our study, to evaluate one putative mechanism whereby dLAN affected

WNV viremia, revealed that dLAN modestly affected CORT levels but this variation did not

predict any aspect of WNV competence (see supplemental material). To evaluate the

115   epidemiological implications of dLAN on WNV risk in a wild bird population (Fig. 3), we

calculated the relative change in the pathogen basic reproductive number, R_0, in the presence and

absence of dLAN effects based on a single host, single vector model of WNV transmission using

parameters derived from literature and these experiments (24). We found that dLAN-induced

changes in the avian host's infectious period increased R_0 by 41% (Supplementary Material).

120   Assuming that dLAN had no other effects on avian or vector competence and abundance, we

estimated that dLAN would increase R_0 from 8.95 to 12.66 in a WNV-naive songbird

community where House Sparrows are the dominant species (Supplementary Material).

Our results motivate further investigation of the ecological and molecular mechanisms whereby

125   dLAN alters zoonotic risk. In addition to effects observed here, light pollution might alter vector

and host diversity as well as the nature and timing of their interactions (over days and seasons; *25*). Most WNV vectors, for instance, take blood meals at dusk and dawn (*26*); with dLAN, the blood-meal feeding window might be extended, or vectors might arouse too early to find a blood meal before they are forced to forage on nectars or other substrates just to remain viable. The

130   pineal-derived hormone, melatonin, which coordinates circadian oscillators that reside in cells throughout the body, often regulates such behaviors (*27*). Dysregulation of melatonin synthesis and secretion, which has been observed in response to dLAN in the lab (*4*), could have complex effects on WNV dynamics, particularly as vectors too rely on melatonin for temporal coordination of behaviors. In urban areas, comparatively warmer and more stable temperatures

135   and numerous habitats for breeding could augment mosquito populations. Conversely, urban vectors might lay few eggs collectively because of difficulties in synchronizing blood-meal searching with periods of optimal host availability. Coupled with the ability of melatonin to modify host and vector immune functions (*28*), it is very difficult to predict now how dLAN will truly affect disease risk in natural settings.

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As we start to investigate dLAN effects on infectious disease risk, zoonotic and otherwise, it will be important to study how the lighting spectra we currently favor (and when and where we use lighting) can be adjusted to mitigate risk. Motion-activated or directionally-targeted light sources can be substituted for the constant illumination practices, and lighting overall could also be

145   reduced when it would have the greatest impacts on wildlife (i.e., migrations, breeding seasons).

A good example to emulate comes from the International Dark-Sky Association, which has led efforts to eliminate lighting in tall urban buildings during avian migrations; this practice reduces the extensive window strikes that occur during those periods (*2*). An equivalent example for vectored-disease in the southeastern US would entail a diminution in lighting of vulnerable areas during the height of arbovirus transmission season (e.g., late Fall; *29*). Additional mitigation opportunities likely reside in the advent of new technologies detectable by human, but less so wildlife, vision (e.g., high-wavelength (red) wavelengths versus the broad-spectrum options typically used; *30*).

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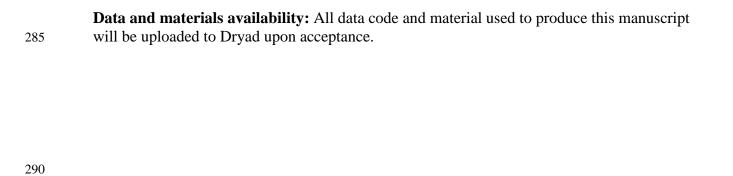
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305   **Fig. 1.**

Suomi NPP satellite derived VIIRS image of artificial nighttime illumination across the United States in 2012. Photo credit to NASA Earth Observatory. *Light Pollution*. Digital Image. *NASA Earth Observatory.* n.p, n.d. Web 15 Jan 2018
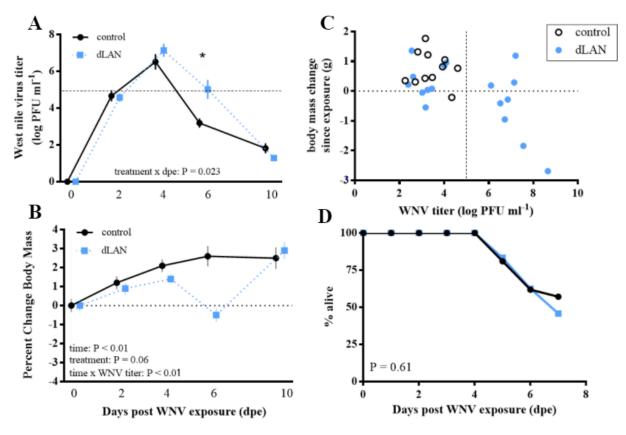<https://earthobservatory.nasa.gov/IOTD/view.php?id=84722>

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Fig. 2. Effects of experimental West Nile virus exposure and impacts on house sparrows (*Passer domesticus*) exposed to dim light at night (dLAN; 8 lux during night hours for 2-3 weeks prior to WNV exposure) versus controls (animals kept on 12h light:12 dark for duration of experiment). Blue points and lines signify dLAN-exposed individuals, and black points and solid lines signify
315 controls. (A) Individuals exposed to dLAN had significantly higher viral titers on d6 post exposure, indicated by the asterisk. The horizontal dashed light represents the transmission threshold, or the minimum amount of virus in circulation required to transmit WNV to a vector (i.e. 10^5 PFUs). (B) Effects of WNV and dLAN on change in body mass throughout the course of WNV infection. On d6, dLAN exposed individuals loss appreciable mass whereas controls
320 continued to gain body mass. C) Relationship between WNV titer and body mass change on d6 post WNV exposure. The vertical dashed line represents the WNV transmission threshold; individuals to the right of this dashed line are infectious to mosquitoes, and individuals to the left of this dashed line are not. Only dLAN exposed individuals were infectious on d6. D) No effect of dLAN on WNV-associated mortality. Although some dLAN exposed birds lost more mass
325 and maintained higher viral titers for longer periods of time than controls (in (B)), mortality rate did not differ between groups.

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