Light suppresses acoustically triggered predator-avoidance behaviours in moths

Theresa Hügel[1]*, Holger R. Goerlitz[1*]

[1] Acoustic and Functional Ecology Group, Max Planck Institute for Ornithology, Seewiesen, Germany

***corresponding authors:**

Acoustic and Functional Ecology Group

Max Planck Institute for Ornithology

Eberhard-Gwinner-Str. 11

82319 Seewiesen, Germany

Email: thuegel@orn.mpg.de, hgoerlitz@orn.mpg.de

ORCID:

Theresa Hügel: 0000-0003-0500-1988

Holger Goerlitz: 0000-0002-9677-8073

ABSTRACT

Urbanization exposes wild animals to increased light. Nocturnal animals in particular are affected by artificial light. Under its influence, the balance of predator-prey interactions may shift. In the interaction between echolocating bats and eared moths, bats often benefit from the effects of light because moths exposed to light resort less often to last-ditch manoeuvres in response to attacking nearby bats. To what extent increased light affects moths' ability to engage in negative phonotaxis, a response to distant bats, is unknown. To quantify the overall effect of light on both last-ditch manoeuvres and negative phonotaxis, we counted the moths caught by two light traps over several nights. Traps were alternately equipped with speakers playing back ultrasonic simulations of hunting bats. The playback did not affect moth counts, indicating that light not only reduces the last-ditch manoeuvres of moths but also influences whether moths rely on negative phonotaxis. Increasing light pollution, therefore, interferes with different aspects of anti-predator behaviour in moths and, possibly, other eared insects. Our study demonstrates the negative effect of light on insects, clarifies the behavioural changes that result from light exposure and highlights the need for future research to uncover the mechanism(s) underlying these behavioural changes.

KEY WORDS

Artificial light at night, predator-prey, bats, Noctuidae, light traps, playback experiment

BACKGROUND

One-fifth of the world's human population can no longer see the Milky Way (1), because of the substantial increase of artificial light at night over the last decades (2,3). Plants and animals are also known to be adversely affected by light pollution (4–6). Light pollution not only impacts a single species' orientation, reproduction or communication (4), it also affects whole communities, for example, by shifting the balance of trophic interactions such as predator-prey interactions (7–12).

Echolocating bats preying upon eared moths represent a global predator-prey system with important ecological impacts (13–16). Bats and moths are primarily nocturnal and rely strongly on their sense of hearing to communicate, orientate, hunt or defend themselves. Bats echolocate to search for food (17,18). Eared moths are able to hear those echolocation calls and to respond with a two-staged anti-predator flight behaviour (19,20). During stage one, moths respond with negative phonotaxis, i.e. they fly away from the calling bat. Negative phonotaxis occurs when the bat is still far away and helps the moth avoid detection. During stage two, moths engage in last-ditch evasive behaviours such as attempting erratic manoeuvres or diving toward the ground. Last-ditch manoeuvres, which occur when the bat is nearby, enable the moth to avoid being captured by the bat. Whether negative phonotaxis or last-ditch manoeuvres are triggered depends on the acoustic properties of the bat's echolocation call as received at the moth's ear (19,21). The probably most important of these properties is received sound pressure level (SPL) (21–23), as received SPL of distant bats is fainter than that of nearby bats due to spherical spreading and atmospheric attenuation (24). In addition, call length and call repetition rate are also important (22,25). All of these properties provide information about the bat's distance and behaviour (17).

Despite their predominantly acoustic interaction, both bats and moths are also strongly affected by artificially introduced light sources. While some bats may profit from artificial light because they are able to exploit the resulting accumulation of prey animals (26,27), other species are negatively affected because they are unable to commute without interruption to new habitats (28–30). Generally all moths are negatively affected by light. Light strongly attracts moths, reducing the time they spend feeding (31,32) and interrupting their commuting behavior (33). Among the ecological consequences is reduced pollination (34) and population decline (31,35). Light also increases moths' predation risk in two ways. First, because the accumulation of moths around lights attracts bats, the predation pressure on moths increases (26,27). Second, light impedes moths' anti-predator flight behaviour. Several studies compared the sound-triggered anti-predator flights of moths under lit and unlit conditions, and reported that light reduces anti-predator flight behaviours. Wakefield et al. (36), who studied last-ditch manoeuvres, showed that only 24% of moths performed power-dives under LED illumination compared to 60% of moths in the dark; that is, LED-illumination inhibited last-ditch manoeuvres in 60% of the moths that reacted in darkness. Similarly, Svensson & Rydell (37) reported a reduction of ~60% in mostly last-ditch evasive behaviours in moths attempting to escape from bats within a radius of 1 meter around the light source compared to in moths in the dark (where all moths reacted). Minnaar et al. (38) reported the most extreme effect: while bat diet (as proxy for moth capture rate) in unlit conditions was best explained by a model that included anti-predator behaviour, bat diet in lit conditions was best explained by a model that included 100% reduction in anti-predator flight behaviour.

In another set of studies (39–41), light exposure was kept constant while the sound received by the moth was manipulated. Those results showed that moths still exhibited

some degree of anti-predator flight under illumination. Acharya & Fenton (39) compared last-ditch manoeuvres in eared and deafened moths under illumination, showing that 48% of eared moths exhibited last-ditch manoeuvres when preyed on by bats, whereas deafened moths did not. Agee & Webb (40) and Treat (41) compared the number of caught moths at light traps with and without ultrasonic stimuli. In both studies, capture rates at light traps with ultrasound playback were lower than capture rates at silent light traps; differences depended on stimulus design and moth family. Across different stimulus types, capture rates at light traps with ultrasound were reduced by 51-86 percentage points compared to capture rates at silent traps in a Noctuid species (*Heliothis zea*, (40)), and by 8-49 percentage points for different eared moth families (with minimum sample sizes of 37 individuals in total (41)).

In summary, while the first set of studies shows that light suppresses the sound-triggered anti-predator flight of moths (ranging from 60-100 percentage points suppression), the second set of studies shows that sound can still trigger anti-predator flight even in the light (ranging from 8-86 percentage points). Therefore, how and to what extent light suppresses anti-predator flight in moths is unclear. Importantly, however, in most of the studies, only last-ditch manoeuvres are reported (36,37). Even in the studies of Treat (41) and Agee & Webb (40), where the stimuli may have elicited both stages of evasive flight, last-ditch manoeuvres likely dominated the outcome due to the study. As both studies used directional light- and sound sources, it is likely that the moths that were attracted to the light were always exposed to such high sound pressure levels that last-ditch manoeuvres were elicited. Only Minnaar et al. (38) investigated both stages of evasive flight, albeit indirectly. They observed the diet of naturally foraging bats under lit and dark conditions, suggesting that light under more naturalistic conditions may completely suppress anti-

110 predator flight throughout all moth populations. Yet, no direct quantification of the overall
111 effect of light on both stages of the anti-predator flight in moths has been made. Here, we
112 designed an experiment to investigate the effects of light on both stages of evasive flight,
113 negative phonotaxis and last-ditch manoeuvres. Like Agee & Webb and Treat (40,41), we
114 compared moth captures at two light traps, and then simulated bat predation via alternating
115 ultrasonic playback at one of the traps. We combined an omnidirectional light field that
116 would attract moths equally with a directional sound field. Therefore, sound intensities
117 around the light trap varied and triggered either last-ditch manoeuvres (high on-axis SPL) or
118 negative phonotaxis (low off-axis SPL) in proximity to the trap.

119 We predicted three different outcomes, depending on the effect of light on the two
120 stages of moths' anti-predator flight: (1) No moth captures at the ultrasonic light trap,
121 indicating that both stages of sound-elicited anti-predator flight behaviour are functional
122 and neither stage is suppressed by light. (2) Reduced moth capture rates at the ultrasonic
123 light trap, indicating that light partly suppresses sound-elicited anti-predator flight
124 behaviour. (3) Equal captures at both light traps, indicating that light completely suppresses
125 both stages of sound-elicited antipredator flight.

126

127

128 **METHODS**

129 **Setup, study site, moth capture and measurement**

130 We compared the number of caught moths between two light traps, one of which had been
131 additionally equipped with a loudspeaker, to examine the effect of light on the evasive flight
132 behaviours of eared moths. We set up two equal-intensity light traps (Sylvania, blacklight,
133 F15W/350BL-T8, USA, Fig. 1A) at 30 m distance to each other; one was situated at a path in a

134 forest (trap A) and one was in the forest itself (trap B), close to the Max Planck Institute for
135 Ornithology, Seewiesen, Germany. Both traps hung freely at ~1.7 m above ground, radiating
136 light at 360° in the horizontal plane (Fig. 1A). In 2018, we collected data over 15 rainless
137 nights between July 19 and August 16. The number of nights for data collection was based
138 on the number of nights sampled in Agee & Webb and Treat (6 nights and 2-12 nights,
139 respectively (40,41)). Each test night, we equipped one or the other of the two traps with
140 two loudspeakers (Ultrasonic Dynamic Speaker ScanSpeak, Avisoft Bioacoustics, Glienicke,
141 Germany); both broadcasting an ultrasonic stimulus (see below) to simulate echolocating
142 bats. Both speakers were fixed back to back facing in opposite directions and were mounted
143 above one of the light traps at ~2 m above ground. Thus, each trap was associated for 7-8
144 nights with the sound of predating bats. In the evening (between 20:30h and 23:40h), we
145 turned on the light of the traps and the acoustic playback, and the next morning (between
146 7:00h and 9:40h), we turned off the light and the playback. Every morning we emptied the
147 traps and counted all moths from the three ear-possessing families Noctuidae, Geometridae
148 and Erebidae. We measured individuals' body lengths along the middorsal line (from the
149 head to the end of the abdomen), to correct for the fact that larger moths tend to be more
150 sensitive to sound than smaller moths (42,43). For statistical analysis, we assigned
151 individuals to one of six categories, covering sizes from 1 to 3.9 cm in bins of 0.5 cm (for
152 details, see R-script in Supplemental Information, SI). For those individuals whose body
153 length we could not measure (e.g., due to a missing abdomen, N = 137, 15.5%), we used
154 either the mean value of the same species or, if this was not possible (N = 1), the mean value
155 of the family.
156
157

Ultrasonic playback stimulus design and evaluation

We simulated the predation pressure that echolocating bats represent by playing an ultrasonic stimulus consisting of repeated pure tones. The stimulus was designed based on the acoustic properties of bat echolocation calls (call frequency, duration, and interval) and the hearing system of moths. Pulse frequency was 35 kHz to match most moths' best hearing threshold between 20-50 kHz (Noctuidae: (43,44); Erebidae: (45); Pyralidae: (25); Geometridae: (46,47); Sphingidae/Drepanidae: (44)). Pulse duration was 10 ms including 2 ms linear rise and fall time, which corresponds to the longer calls of European open space bats (48,49) and according to which we hoped to optimize detection by moths and information transfer about bat distance (22). The interpulse interval was 100 ms, matching the call interval of bats' echolocation calls in their search phase (60 - 200 ms, (48,50)). On-axis sound pressure level (SPL) was 98 dB SPL re. 20 µPa RMS at 1 m distance (see below for a detailed description of the sound field). This stimulus was presented continuously in a loop throughout the night via the loudspeakers using Avisoft-RECORDER software (Avisoft Bioacoustics), a sound card (Avisoft UltraSoundGate 116, Avisoft Bioacoustics) and a laptop computer.

To validate the effect of the stimulus in the dark, without the potentially suppressing influence of light, we exposed free-flying moths in a dark flight room (5.3 m x 3.5 x 3 m^3) to the stimulus. We caught moths at trap A over the course of four nights and tested them within 30 hours after capture. We placed moths on the ground of the flight room and recorded the flight paths of upward-flying moths with an IR-sensitive camera (Sony HDR-CX560, Sony, Tokio, Japan) under IR illumination (850 nm, Mini IR Illuminator TV6700, ABUS Security-Center, Wetter, Germany). Using the same audio equipment as described above, we presented the stimulus by manually triggering it when a moth flew in front of the speaker.

182 The moth's distance to the speaker at stimulus onset was about 1-2 m. We categorized the
183 recorded flight paths as "reaction" when the flight direction, level of erraticness or both
184 changed with the stimulus onset (for examples, see video in SI); as "no reaction" when we
185 did not observe those changes; or as "ambiguous" when we could not assign the flight path
186 to one of the other categories. In total, of the 32 flight paths we evaluated, ten moths (31.2
187 %) showed a reaction, twelve moths (37.5%) did not show a reaction, and eleven moths
188 (34.3%) were categorized as "ambiguous".
189
190 **Overlap of sound and light field**
191 The range and geometry of the presented sound and light fields differed. While the light was
192 emitted omnidirectionally in the horizontal plane, the sound field was directional (Fig. 1). We
193 estimated the biologically relevant range for attracting moths by light and the biologically
194 relevant sound fields for triggering moths' evasive flight based on literature values and our
195 own measurements.
196 Light traps can attract released moths from up to 80 m, yet recapture dramatically
197 decrease beyond 15 m, and additionally depended on family (51,52). Family-specific models
198 estimated 5%-recapture rate to be 10 ± 6 m (mean ± SEM) for Noctuidae and 23 ± 12 m for
199 Geometridae (51). Note, however, that these data were obtained with a different light
200 source than ours (6W actinic vs. 15W blacklight in our case), and that the radius of attraction
201 to light is not necessarily equivalent to the radius over which light interferes with anti-
202 predator behaviour. Yet, we used these family-specific values as first estimates of the
203 biologically relevant distances beyond which moths would not be attracted to our light trap
204 (Fig. 1B, dashed lines).

To estimate the effect of the playback, we measured the playback level of the loudspeaker from 0° on-axis to 90° off-axis in steps of 5° (for details about calculations, see SI). The on-axis source level was 97 dB SPL re. 20 µPa RMS @ 1 m distance, i.e., within the lower range of natural bat call levels in the field (100-120 dB peSPL @ 1 m (50), with corresponding RMS-SPL values being ~3-7 dB lower than the peSPL values (53,54)). With increasing off-axis angle, playback source level was reduced by up to ~30 dB at 45°, resulting in a minimum playback level of 70 dB SPL RMS @ 1m. We then calculated the distance to the loudspeaker at which the playback would reach biologically relevant levels of 60 and 80 dB SPL RMS as a function of angle around the loudspeaker. We chose 60 and 80 dB SPL as approximate thresholds for eliciting negative phonotaxis and last-ditch manoeuvres, respectively, based on several lines of evidence. The triggering of negative phonotaxis and last-ditch manoeuvres in moths is thought to be correlated with the activity of moths' auditory receptor neurons, A1 and A2 (22,55,56). The neuronal thresholds of these neurons vary with frequency and species (22,43,55,57–59), with A1 being generally about 20 dB more sensitive than A2 (43,55,60). The best thresholds of A1 range from ~35-55 dB SPL at the position of the moth, and the best thresholds of A2 range from ~52-72 dB SPL (20,22,43,55,57,58). Behavioural thresholds in moths are often unknown, but those that are known tend to be ~10 dB higher than neuronal thresholds (reviewed in (54)). Indeed, our own data indicate that sound pulses of ~80 dB SPL elicit last-ditch manoeuvres (61), which is 8-28 dB above known A2 thresholds (20,22,43,55,57,58). We thus defined 80 dB SPL as threshold which securely elicits last-ditch maneouvres. As no behavioural data exist for the threshold of negative phonotaxis, we chose 60 dB SPL, which is 20 dB below our threshold for last-ditch manoeuvres and matches the difference between the A1 and A2 thresholds.

We calculated the distance-isolines for 60 and 80 dB SPLs; these varied with the angle around the loudspeaker, ranging from 3.6 to14.6 m and 0 to 5.6 m, respectively (Fig. 1B).

Statistical analysis

To test for an effect of light on the moths' negative phonotaxis, we fitted linear mixed-effects models to the moth count data as a function of the fixed effects *playback*, *trap*, *moth family*, and *moth body length*, the random factor *date* (to account for repeated measures), and the interactions of *playback* and *moth family*, and *playback* and *moth body length*. We modelled error distribution using a negative binomial distribution and tested for overdispersion using the "DHARMa" package (62). We defined the moth family Noctuidae as intercept, as it had the largest sample size and thus was the most reliable reference. To test which factors significantly contributed to the model fit, we conducted backwards model reduction (63). Hence, the full model was successively reduced, by stepwise removing covariates, starting wit the covariate with the highest p-value of t-statistics provided by the model summary. We compared nested models with likelihood ratio tests (type I) using a chi-squared statistic to find the minimal adequate model. All statistical analyses were conducted in R version 3.3.2 (R Foundation for Statistical Computing, Vienna, Austria) using the packages "lme4"(64), "lmerTest"(65), "MASS" (66), and "blmeco" (67). For further details, see R-script included in SI.

252 **RESULTS**

253 We caught a total of 878 moths over 15 nights (Fig. 2). The minimal adequate model
254 included *playback*, *family* and *size* as fixed factors and *date* as a random factor. Capture
255 success varied among moth families (likelihood ratio test for factor *family*, $\chi^2 = 59.43$, d.f. =
256 2, $p < 0.001$). Most of the counted moths (80.6 %) belonged to the family Noctuidae; the
257 remaining 19.4 % was composed of Geometridae (11.3%) and Erebidae (8.1%). We caught
258 more small than large moths (likelihood ratio test for factor *size*, $\chi^2 = 89.76$, d.f. = 5,
259 $p < 0.001$). Although total capture rates seemed to differ between traps (total capture of 506
260 (trap A) vs. 365 (trap B) moths), *trap* was not a significant factor of the minimal adequate
261 model (likelihood ratio test, $\chi^2 = 2.46$, d.f. = 1, $p = 0.116$). That is, the capture rates per night
262 did not significantly differ between the traps (Fig. 2A).

263 The number of caught moths was similar on nights with and without playback for
264 both traps (Fig. 2A), that is, our playback treatment had no detectable effect on the number
265 of caught moths (likelihood ratio test of factor *playback*, $\chi^2 = 2.44$, d.f. = 1, $p = 0.118$). The
266 number of caught moths fluctuated strongly between nights (7-116 moths per trap and
267 night), which might be one reason for the low power of our dataset (~38%, see R-script in SI
268 for more information).

269 We did not detect an interaction between *moth size* and *playback* and between *moth*
270 *family* and *playback* (likelihood ratio tests, $\chi^2 = 4.54$, d.f. = 3, $p = 0.208$ and $\chi^2 = 0.38$, d.f. = 2,
271 $p = 0.826$, respectively), that is, moths of different sizes and families did not react differently
272 to the playback. This missing interaction is likely due to *playback* not being a significant
273 factor of the minimal adequate model.

DISCUSSION

Echolocating bats and eared insects offer a textbook example of predator-prey interactions, which is based on acoustic information (20). Increasing light pollution (2,3), however, severely impacts both bats and moths (e.g. (27,28,32), and has potentially cascading effects on their predator-prey-interactions, population dynamics and ecosystems (11,38). Light reduces the acoustically triggered last-ditch manoeuvres in eared moths, which in turn makes them more vulnerable to nearby predating bats. It is less clear, however, whether light also reduces the ability of eared moths to perform negative phonotaxis, a behaviour that serves as the moths' first line of defence against distant echolocating bats. To better understand how artificial light at night affects the eared moths' overall ability to avoid predators, we compared moth captures at two light traps. One trap was silent, while the other trap broadcast bat-like ultrasonic stimuli that triggered last-ditch manoeuvres as well as negative phonotaxis under dark conditions. The number of caught moths did not significantly differ between traps. Thus, the presented playback did not repel the moths, suggesting that the light suppressed both anti-predator behaviours, negative phonotaxis and last-ditch manoeuvres.

In contrast to our results, both Treat (41) and Agee & Webb (40) observed reduced moth capture rates at the ultrasonic trap, showing that sound repelled eared moths. Differences in stimulus design and habituation effects of the moths are in our opinion insufficient to explain the variation in capture rates. Treat (41) and Agee & Webb (40) broadcast a variety of different stimuli varying in pulse rate, frequency, pulse duration, and SPLs (ranges: pulse rate: 0.7-155 pulses/s; frequency: 12.5-200 kHz; pulse duration: 2-10 ms; SPL: ~60-100 dB SPL @ 1 m distance), all of which elicited varying degrees of evasive flight in eared moths. The acoustic properties of our stimulus was within this range and did elicit

evasive flight under dark control conditions, but did not elicit evasive flight under lit conditions. Habituation to the ultrasound could have affected individuals that flew close to the light traps for extended time periods. It is more likely, however, that most caught individuals randomly encountered and approached the traps over larger distances and thus did not perceive our presented stimulus continuously. Likewise, Agee & Webb (40) also presented their ultrasonic stimuli over the course of a whole night and found that ultrasonic playbacks repelled the moths.

We suggest that major differences in the geometry and overlap of light- and sound-fields explain these contrasting results. In the setups of Treat (41) and Agee & Webb (40), sound and light fields almost overlapped. Before entering the light traps in these setups, approaching moths had to pass through high levels of sound pressure; such levels likely have been high enough to elicit last-ditch manoeuvres. In both studies, fewer moths were caught in the ultrasonic traps compared to the silent traps, suggesting that the playback elicited some anti-predator flight (likely last-ditch manoeuvres) under these light and sound conditions. Specifically, for stimuli similar to ours, noctuid moths were captured at a rate of 35% for a 37.5 kHz pulsed tone stimulus vs. 65% at the silent trap (41), while in Agee & Webb (40), the moths were captured at a rate of ~15% for a 30 kHz pulsed tone stimulus vs. 85% at the silent trap for two species of the families Noctuidae and Pyralidae. In contrast, our setup combined omnidirectional light fields with directional sound fields, a design which exposed the moths to different direction-dependent sound pressure levels while approaching the light. Moths approaching the trap on-axis were exposed first to low SPLs and then to increasingly high SPLs. This spatial pattern of increasing SPL was intended to first elicit negative phonotaxis and later last-ditch manoeuvress. In contrast, moths approaching the traps off-axis were exposed to low SPLs that can elicit only negative phonotaxis. As our

322 capture rates did not differ among the light traps -- indicating that the playback had no
323 repelling effect on moth flight -- we suggest that the light strongly reduced the negative
324 phonotaxis of eared moths. Even if the moths still exhibited some degree of last-ditch
325 manoeuvre close to the trap (as shown by Treat and Agee & Webb, (40,41), these
326 manoeuvres might have brought the moth into a position off-axis to the loudspeaker, either
327 to the sides or below, where the SPL is lower. From there, no further last-ditch manoeuvres
328 was elicted due to the lower SPL, and the light kept attracting the moth into the trap. Our
329 results are in line with those of Minnaar et al. (38), who found that a model of escape
330 behaviour in moths assuming 0% efficiency best explained bats' diet under lit conditions. We
331 therefore suggest that light suppresses not only last-ditch manoeuvres, as previously shown,
332 but also negative phonotaxis.

333 In summary, our results underline the strong effect of light on eared moths, providing
334 an additional explanation for shifts in predator-prey interactions between bats and moths
335 (11,12,38). Because negative phonotaxis, along with last-ditch manoeuvres, is suppressed by
336 light, moths are not only unable to escape nearby predators, but they are also unable to
337 avoid them by flying away. Similarly, fish are attracted to lit areas, where they are "trapped"
338 and preyed upon by seals (9) and other fish (68). Our results emphasize the need to
339 understand the mechanism(s) underlying animals' attraction to light and to quantify the
340 effects of light on animals' behaviour, such as predator avoidance. Light pollution is
341 increasing and insect populations are declining. Only by understanding the mechanism(s)
342 underlying these issues can we apply efficient conservation measures.

343 **Ethics**

344 The experiments in this study did not require an approval by an ethical committee.

345

346 **Data accessibility**

347 All data, including R-scripts and supplementary video, is attached via electronic

348 supplemental material.

349

350 **Author contributions**

351 TH and HRG conceived of and designed the study. TH collected and analysed the data. TH

352 and HRG wrote the manuscript. All authors gave final approval for publication and agree to

353 be held accountable for the work performed therein.

354

355 **Competing interests**

356 We have no competing interests.

357

358 **Funding**

359 This study was supported by funds of the Deutsche Forschungsgemeinschaft

360 (Emmy Noether research grants GO2029/2-1 and GO2029/2-2 to H.R.G.) and the

361 International Max Planck Research School for Organismal Biology (student grant to T.H.)

362

363 **Acknowledgements**

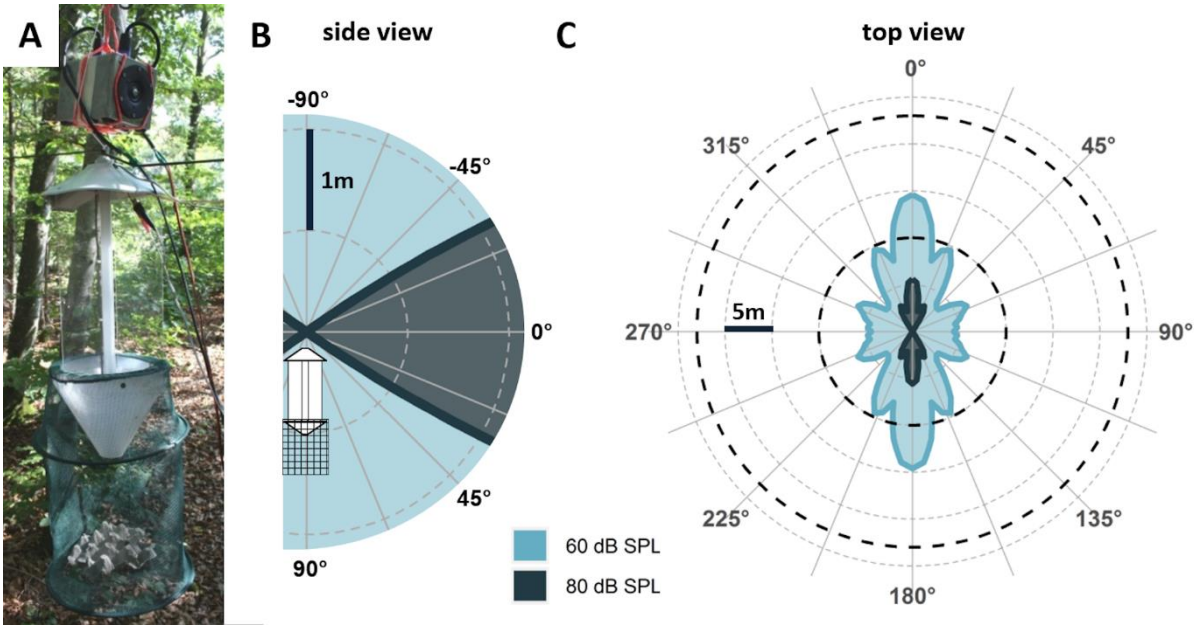
364 We thank the IMPRS for support and especially the participants of the course for “Scientific

365 Writing” 2019 for comments on previous versions of the manuscript. We further thank the

Max Planck Institute for Ornithology Seewiesen for providing excellent infrastructure and support.

368 **FIGURES**

369



370

371

372 **Figure 1: (A) Experimental setup.** The photo shows one of the light traps with two
373 loudspeakers attached above the trap, both of which broadcast the acoustic stimulus in
374 opposite directions. **(B) Side view of the biologically relevant sound field around the light**
375 **trap with attached loudspeakers.** Coloured areas indicate areas with minimum sound
376 pressure levels of 60 and 80 dB SPL re. 20 µPa, which are biologically relevant acoustic
377 thresholds for eliciting negative phonotaxis and last-ditch manoeuvres, respectively, in eared
378 moths. Schematic indicates position and dimension of the light trap. **(C) Top view of the**
379 **biologically relevant sound field (coloured areas) and light field (dashed lines)**. Dashed lines
380 indicate the distance over which 5% of released noctuid (10 m) and geometrid (23 m) moths
381 are recaptured at the light trap, respectively (51), as an indicator of the maximum range of
382 light attraction.

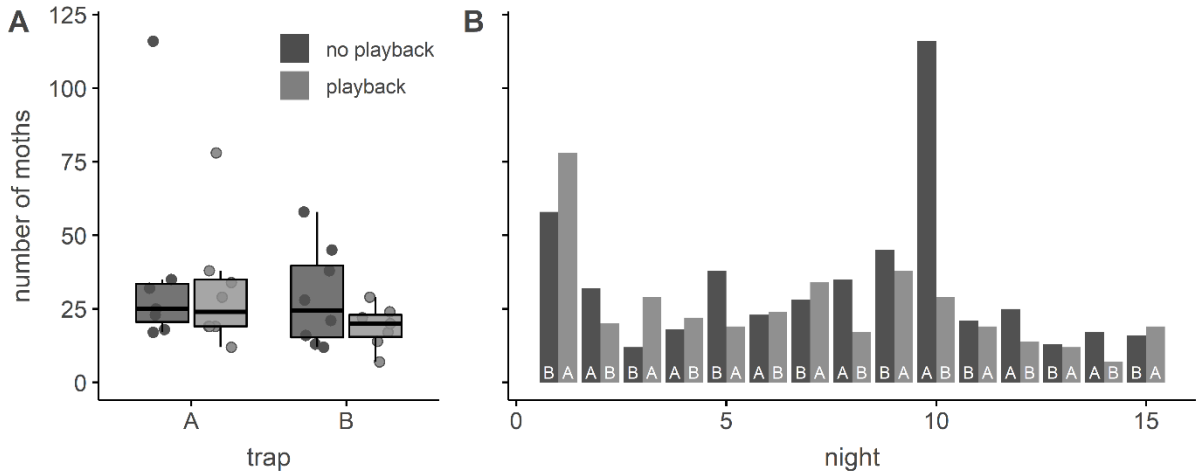


Figure 2: Number of caught moths per night and per playback treatment. **(A) Number of caught moths at both light traps, with and without playback.** Boxplots show median, quartiles and whiskers (up to 1.5 x inter-quartile range) of the daily counts (dots). **(B) Daily counts of caught moths per night and per trap for all 15 nights.** White letters at the base of the bars indicate the trap.

REFERENCES

1. Falchi F, Cinzano P, Duriscoe D, Kyba CCM, Elvidge CD, Baugh K, et al. The new world atlas of artificial night sky brightness. Sci Adv. 2016;2(6):e1600377.

2. Hölker F, Moss T, Griefahn B, Kloas W, Voigt C, Henckel D, et al. The dark side of light: a transdisciplinary research agenda for light pollution policy. Ecol Soc. 2010;15(4).

3. Fouquet R. Seven centuries of energy services: the price and use of light in the United Kingdom (1300-2000). Energy Journal. 2006;27:139–77.

4. Longcore T, Rich C. Ecological light pollution. Front Ecol Environ. 2004;2(4):191–8.

5. Davies TW, Smyth T. Why artificial light at night should be a focus for global change research in the 21st century. Glob Chang Biol. 2018;24(3):872–82.

6. Knop E, Zoller L, Ryser R, Gerpe C, Hörler M, Fontaine C. Artificial light at night as a new threat to pollination. Nature. 2017;548(7666):206–9.

7. Davies TW, Bennie J, Inger R, Ibarra NH, Gaston KJ. Artificial light pollution: are shifting spectral signatures changing the balance of species interactions? Glob Chang Biol. 2013;19(5):1417–23.

8. Davies TW, Duffy JP, Bennie J, Gaston KJ. The nature, extent, and ecological implications of marine light pollution. Front Ecol Environ. 2014;12(6):347–55.

9. Yurk H, Trites AW. Experimental attempts to reduce predation by harbor seals on out-migrating juvenile salmonids. Trans Am Fish Soc. 2000;129(6):1360–6.

10. Miller Colleen R., Barton Brandon T., Zhu Likai, Radeloff Volker C., Oliver Kerry M., Harmon Jason P., et al. Combined effects of night warming and light pollution on predator–prey interactions. P Roy Soc P B Biol Sci. 2017;284(1864):20171195.

11. Russo D, Cosentino F, Festa F, De Benedetta F, Pejic B, Cerretti P, et al. Artificial illumination near rivers may alter bat-insect trophic interactions. Environ Pollut. 2019;252:1671–7.

12. Bailey LA, Brigham RM, Bohn SJ, Boyles JG, Smit B. An experimental test of the allotonic frequency hypothesis to isolate the effects of light pollution on bat prey selection. Oecologia. 2019;190(2):367–74.

13. Van Toor ML, O'Mara MT, Abedi-Lartey M, Wikelski M, Fahr J, Dechmann DKN. Linking colony size with quantitative estimates of ecosystem services of African fruit bats. Curr Biol. 2019;29(7):R237–8.

14. Kunz TH, Braun de Torrez E, Bauer D, Lobova T, Flemming TH. Ecosystem services provided by bats. Ann NY Acad Sci. 2011;1223(1):1–38.

15. Kasso M, Balakrishnan M. Ecological and economic importance of bats (order Chiroptera). ISRN Ecol. 2013;Article ID 187415:9 pages.

16. Boyles JG, Cryan PM, McCracken GF, Kunz TH. Economic importance of bats in agriculture. Science. 2011;332(6025):41–2.

17. Schnitzler H-U, Moss CF, Denzinger A. From spatial orientation to food acquisition in echolocating bats. Trends Ecol Evol. 2003;18:386–94.

426 18. Fenton MB. Eavesdropping on the echolocation and social calls of bats. Mammal Rev.
427 2003;33:193–204.

428 19. Roeder KD. The behaviour of free flying moths in the presence of artificial ultrasonic pulses.
429 Anim Behav. 1962;10(3–4):300–4.

430 20. Ter Hofstede HM, Ratcliffe JM. Evolutionary escalation: the bat-moth arms race. J Exp Biol.
431 2016;219(Pt 11):1589–602.

432 21. Roeder KD. Aspects of the noctuid tympanic nerve response having significance in the
433 avoidance of bats. J Insect Physiol. 1964;10(4):529–46.

434 22. Gordon, Ter Hofstede HM. The influence of bat echolocation call duration and timing on
435 auditory encoding of predator distance in noctuoid moths. J Exp Biol. 2018;221(6):jeb171561.

436 23. Svensson M, Rydell J, Brown R. Bat predation and flight timing of winter moths, *Epirrita* and
437 *Operophtera* species (Lepidoptera, Geomentridae). Oikos. 1999;84(2):193–8.

438 24. Goerlitz HR. Weather conditions determine attenuation and speed of sound: Environmental
439 limitations for monitoring and analyzing bat echolocation. Ecol Evol. 2018;8(10):5090–100.

440 25. Skals N, Surlykke A. Hearing and evasive behaviour in the greater wax moth, *Galleria mellonella*
441 (Pyralidae). Physiol Entomol. 2000;25(4):354–62.

442 26. Rydell J. Exploitation of insects around streetlamps by bats in Sweden. Funct Ecol.
443 1992;6(6):744–50.

444 27. Cravens ZM, Brown VA, Divoll TJ, Boyles JG. Illuminating prey selection in an insectivorous bat
445 community exposed to artificial light at night. J Appl Ecol. 2018;55(2):705–13.

446 28. Stone EL, Jones G, Harris S. Street lighting disturbs commuting bats. Curr Biol.
447 2009;19(13):1123–7.

448 29. Mathews F, Roche N, Aughney T, Jones N, Day J, Baker J, et al. Barriers and benefits:
449 implications of artificial night-lighting for the distribution of common bats in Britain and Ireland.
450 Philos Trans R Soc Lond B Biol Sci. 2015;370(1667).

451 30. Straka TM, Lentini PE, Lumsden LF, Wintle BA, Ree R van der. Urban bat communities are
452 affected by wetland size, quality, and pollution levels. Ecol Evol. 2016;6(14):4761–74.

453 31. van Langevelde Frank, van Grunsven Roy H. A., Veenendaal Elmar M., Fijen Thijs P. M. Artificial
454 night lighting inhibits feeding in moths. Biol Lett. 2017;13(3):20160874.

455 32. Macgregor CJ, Pocock MJO, Fox R, Evans DM. Effects of street lighting technologies on the
456 success and quality of pollination in a nocturnally pollinated plant. Ecosphere.
457 2019;10(1):e02550.

458 33. Owens ACS, Lewis SM. The impact of artificial light at night on nocturnal insects: A review and
459 synthesis. Ecol Evol. 2018;1–22.

460 34. Macgregor CJ, Evans DM, Fox R, Pocock MJO. The dark side of street lighting: impacts on moths
461 and evidence for the disruption of nocturnal pollen transport. Glob Chang Biol. 2017;23(2):697–
462 707.

463 35. Wilson JF, Baker D, Cheney J, Cook M, Ellis M, Freestone R, et al. A role for artificial night-time
464 lighting in long-term changes in populations of 100 widespread macro-moths in UK and Ireland:
465 a citizen-science study. J Insect Conserv. 2018;22(2):189–96.

466 36. Wakefield A, Jones G, Stone EL, Harris S, Jones G, Harris S. Light-emitting diode street lights
467 reduce last-ditch evasive manoeuvres by moths to bat echolocation calls. R Soc open sci.
468 2015;2(8):150291.

469 37. Svensson AMA, Rydell J. Mercury vapour lamps interfere with the bat defence of tympanate
470 moths (Operophtera spp.; Geometridae). Animal Behav. 1998;55(1):223–6.

471 38. Minnaar C, Boyles JG, Minnaar IA, Sole CL, McKechnie AE. Stacking the odds: light pollution may
472 shift the balance in an ancient predator-prey arms race. J Appl Ecol. 2015;52(2):522–31.

473 39. Acharya L, Fenton MB. Bat attacks and moth defensive behaviour around street lights. Can J
474 Zool. 1999;77(1):27–33.

475 40. Agee HR, Webb JC. Effects of ultrasound on capture of *Heliothis zea* and *Ostrinia nubilalis*
476 moths in traps equipped with ultraviolet lamps. Ann Entomol Soc Am. 1969;62(6):1248–52.

477 41. Treat AE. Comparative moth catches by an ultrasonic and a silent light trap. Ann Entomol Soc
478 Am. 1962;55(6):716–20.

479 42. Surlykke A, Filskov M, Fullard JH, Forrest E. Auditory relationships to size in noctuid moths:
480 bigger ist better. Naturwissenschaften. 1999;86(5):238–41.

481 43. Ter Hofstede HM, Goerlitz HR, Ratcliffe JM, Holderied MW, Surlykke A. The simple ears of
482 noctuoid moths are tuned to the calls of their sympatric bat community. J Exp Biol.
483 2013;216(21):3954–62.

484 44. Nakano R, Takanashi T, Surlykke A. Moth hearing and sound communication. J Comp Physiol A.
485 2015;201(1):111–21.

486 45. ter Hofstede HM, Ratcliffe JM, Fullard JH. Nocturnal activity positively correlated with auditory
487 sensitivity in noctuoid moths. Biol Lett. 2008;4(3):262–5.

488 46. Rydell J, Skals N, Surlykke A, Svensson AM. Hearing and bat defence in geometrid winter moths.
489 Proc Biol Sci. 1997;264(1378):83–8.

490 47. Surlykke A, Filskov M, Sanderson CG, Lal D, Peters B, Handb, et al. Hearing in geometrid moths.
491 Naturwissenschaften. 1997;84:356–9.

492 48. Skiba R. Europäische Fledermäuse. 2. Edition. Magdeburg: VerlagsKG Wolf; 2014.

493 49. Obrist MK, Boesch R, Fluckiger PF. Variability in echolocation call design of 26 Swiss bat species:
494 consequences, limits and options for automated field identification with a synergetic pattern
495 recognition approach. Mammalia. 2004;68:307–22.

496 50. Holderied MW, von Helversen O. Echolocation range and wingbeat period match in aerial-
497 hawking bats. Proc Royal Soc B. 2003;270:2293–9.

498 51. Merckx T, Slade EM. Macro-moth families differ in their attraction to light: implications for
499 light-trap monitoring programmes. Insect Conserv Divers. 2014;7(5):453–61.

500 52. Truxa C, Fiedler K. Attraction to light - from how far do moths (Lepidoptera) return to weak
501 artificial sources of light? Eur J Entomol. 2012;109(1):77–84.

502 53. Seibert A-M, Koblitz JC, Denzinger A, Schnitzler H-U. Bidirectional echolocation in the bat
503 *Barbastella barbastellus*: different signals of low source level are emitted upward through the
504 nose and downward through the mouth. PLoS One. 2015;10(9):e0135590.

505 54. Lewanzik D, Goerlitz HR. Continued source level reduction during attack in the low-amplitude
506 bat *Barbastella barbastellus* prevents moth evasive flight. Funct Ecol. 2017;32(5):1251–61.

507 55. Madsen BM, Miller LA. Auditory input to motor neurons of the dorsal longitudinal flight
508 muscles in a noctuid moth (*Barathra brassicae L.*). J Comp Physiol. 1987 Jan 1;160(1):23–31.

509 56. Roeder KD. Responses of the less sensitive acoustic sense cells in the tympanic organs of some
510 noctuid and geometrid moths. J Insect Physiol. 1974;20(1):55–66.

511 57. Surlykke A. Hearing in hooktip moths (Drepanidae: Lepidoptera). J Exp Biol. 2003;206(15):2653–
512 63.

513 58. Waters DA, Jones G. The peripheral auditory characteristics of noctuid moths: responses to the
514 search-phase echolocation calls of bats. J Exp Biol. 1996;199(4):847–56.

515 59. Zha Y, Chen Q, Lei C. Ultrasonic hearing in moths. Ann Soc Entomol Fr. 2009;45(2):145–56.

516 60. Goerlitz HR, ter Hofstede HM, Zeale MRK, Gareth J, Holderied MW. An aerial-hawking bat uses
517 stealth echolocation to counter moth hearing. Curr Biol. 2010;20:1568–72.

518 61. Hügel T, Goerlitz HR. Species-specific strategies increase unpredictability of escape flight in
519 eared moths. Funct Ecol. 2019;0(0).

520 62. Hartig F. DHARMa: Residual Diagnostics for Hierarchical (Multi-Level / Mixed) Regression
521 Models [Internet]. 2019. Available from: https://CRAN.R-project.org/package=DHARMa

522 63. Lewis F, Butler A, Gilbert L. A unified approach to model selection using the likelihood ratio test.
523 Methods Ecol Evol. 2011;2(2):155–62.

524 64. Bates D, Maechler M, Bolker B, Walker S. Fitting linear mixed-effects models using lme4. J Stat
525 Softw. 2015;67(1):1–48.

526 65. Kuznetsova A, Brockhoff PB, Christensen RHB. lmerTest package: tests in linear mixed effects
527 models. J Stat Softw. 2017;82(1):1–26.

528 66. Venables WN, Ripley BD. Modern Applied Statistics with S. Fourth Edition. New York: Springer;
529 2002.

530 67. Korner-Nievergelt F, Rpth T, von Felten S, Guelat J, Almasi B, Korner-Nievergelt P. blmeco
531 [Internet]. [cited 2019 Mar 21]. (R package). Available from: https://rdrr.io/cran/blmeco/

532 68. Becker A, Whitfield AK, Cowley PD, Järnegren J, Næsje TF. Potential effects of artificial light
533 associated with anthropogenic infrastructure on the abundance and foraging behaviour of
534 estuary-associated fishes. J Appl Ecol. 2013;50(1):43–50.