Spoelstra, K *et al.* 2015 Experimental illumination of natural habitat – an experimental set-up to assess the direct and indirect ecological consequences of artificial light of different spectral composition. *Phil. Trans. R. Soc. B* **370** doi: 10.1098/rstb.2014.0129

Supplementary material

Supplementary information on the setup of the research sites

site number	site name	light on since	coordinates	
1	Lebret's Hoeve	5/10/2012	52 5'53''	5 55'59''
2	Voorstonden	4/26/2013	52 7'21''	6 7'7''
3	Radio Kootwijk	3/12/2012	52 11'20''	5 48'17''
4	ASK Doornspijk 1	5/10/2012	52 22'36''	5 53'51''
5	ASK Doornspijk 2	5/10/2012	52 22'6''	5 53'11''
6	Klaterweg 1	4/21/2012	52 21'52''	5 51'25''
7	Klaterweg 2	4/21/2012	52 22'2''	5 51'13''
8	Hijkerveld	4/19/2012	52 54'44''	6 28'40''

Table S1. The number, name and location of the study sites, and the dates the lights went on.

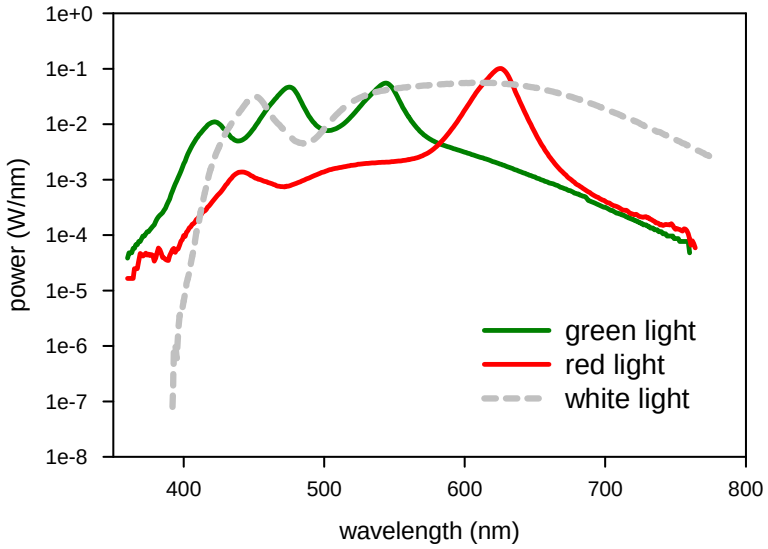

Figure S1. Spectral composition of the green (Philips Fortimo Clearsky; green line), red (Philips Fortimo ClearField; red line) and white light (Philips Fortimo White; yellow line).

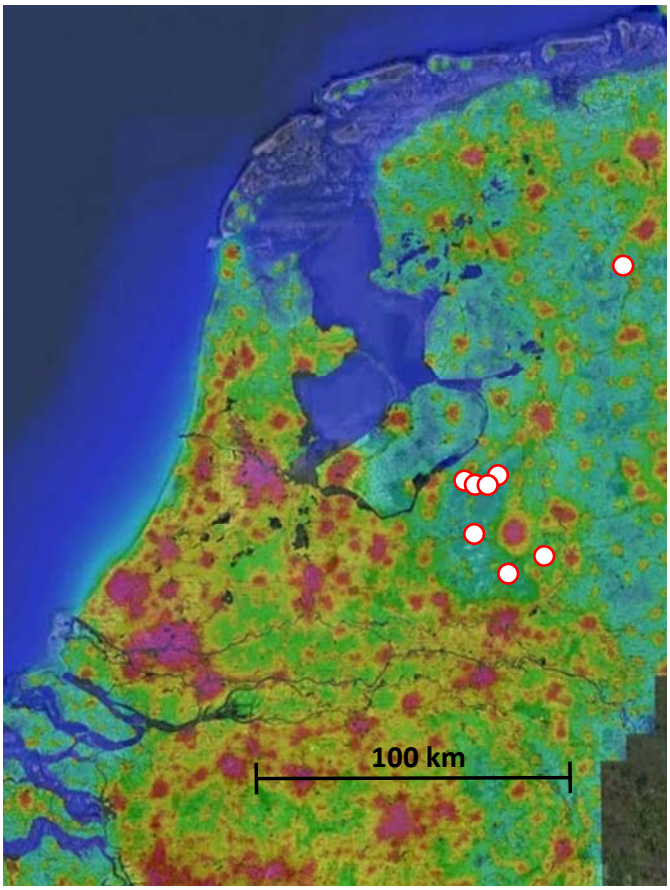

Figure S2. Location of research sites plotted on a nightly light emission map of the Netherlands. Blue indicates low, red indicates high exposure to artificial light at night. The research sites are located in the darker areas (map image courtesy of Avex's files, http://www.avex-asso.org).

Supplementary information on species assessment

Mice and other (non-flying) mammals.

All non-flying mammals were recorded with camera traps (Reconyx HC500, Reconyx, Holmen, WI, US). The most relevant settings used: motion sensor = on; sensitivity = high; pictures per trigger = 1; quiet period = no; AM period = off, PM period = off, resolution = 3.1 MP; night mode = high quality. The traps were set without delay as differences in vegetation height around the concrete tiles may disturb unbiased recordings: in low vegetation, an approaching mouse may trigger the trap sensor before the mouse arrives on the tile. Hence a mouse may not be photographed on the tile if it leaves before the delay time expires.

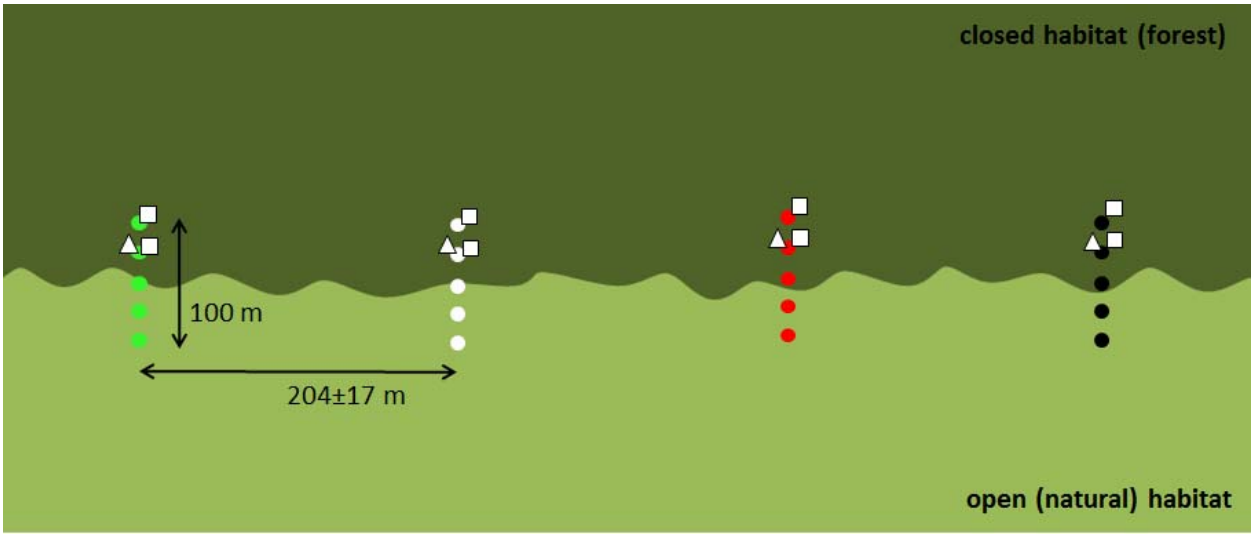

Figure S3. Placement of camera traps: squares indicate the location of a camera trap aimed at a fish oil baited tile; these were placed near the middle and last light post in the forest. Camera traps were attached to trees such that tiles were directly exposed to the light of the nearest light post. Distances between light posts and tiles were standardized between the four transects within one site, such that the distance between tile and middle light post at all four transects was equal, and the distance between the tile and the last light post at all four transects was equal. Triangles indicate the position of a third camera trap placed at 1.5 meter height on a tree facing horizontally into the forest, for the assessment of presence of large mammals (data not presented in this paper).

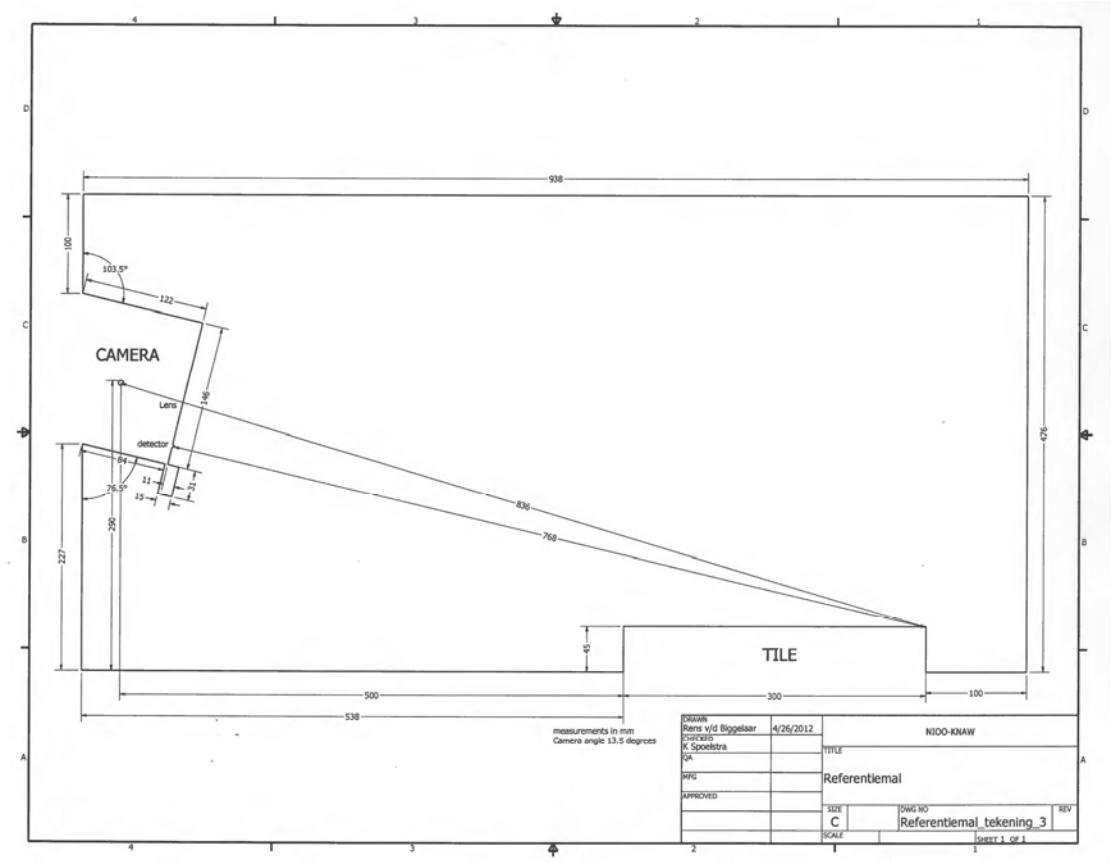

Figure S4. Position of camera trap relative to a 30x30 cm concrete tile (courtesy Rens van den Biggelaar, NIOO-KNAW). The camera trap was always placed in a custom made clamp permanently fixed to the tree; the clamps were made such that the camera trap angle was adjustable. When placing the camera traps, wooden moulds were always used to ensure the camera trap had exactly the right angle and distance to the tile.

Bats

Bats were recorded with Pettersson D500x detectors (Pettersson Elektronik, Uppsala, Sweden). Detectors were placed in Explorer Cases (www.explorercases.com) permanently mounted in the nearest tree to the light post in the forest edge. The detectors were placed such that the microphone of the detectors was behind a 40 mm hole in the Explorer Case, facing the open area. The D500x detectors were programmed to Profile 3 (sampling frequency = 500 kHz; pre-trigger = off; recording length = 5 seconds; high-pass filter = enabled (cuts off sound below 15 kHz); autorecording = enabled; trigger sensitivity = 1). The recording settings were set as follows: input gain = 40; trigger level = 40; interval = 0. The detectors were programmed to be active starting 10 minutes before sunset, and stop recording 30

minutes before sunrise. All detectors were connected to external 6V Panasonic lead-acid batteries (model LC-R0612P, Panasonic Corporation, Japan).

Moths

Moths were recorded using Heath traps (Vermandel Hulst, The Netherlands). Moths are attracted to a 6 Watt actinic fluorescent tube (Philips Lighting Amsterdam) placed in between three Perspex vanes (Fig. S5). Moths then drop via a funnel into the trap, and land on egg boxes where moths can rest until collected. The actinic lamps are powered by small 12 Volt lead acid batteries. Moths are trapped twice a month from May till September on evenings with little wind (below 4 BFT), no rain, and temperature above 8 ^{0}C. The traps are placed underneath the middle light post of the four treatments, in the forest edge. The trap lights are switched on before sunset and left in the field overnight. The following morning the traps are closed and collected. Moths are removed from the trap under a mosquito net to prevent escaping and identified. All individuals are identified to species level except for species that require preparation of the genitalia.

Figure S5. Heath trap used for the collection of moths.

species group	method	sampling routine	number of sites sampled				analysed and presented in this paper	sample size	dimension
			2011	2012	2013	2014			
mice	camera traps	two weeks in sept/oct, and two weeks in nov/dec	0	7	8	8	sept/oct 2012	523	number of mice photographed on baited tiles per night
bats	ultrasound detectors	one week in jun/jul and one week in aug/sep	0	7	8	8	jun/jul 2013	136	five second call sequences
birds	standardized observations	eight visits from early march to early july	5	7	8	8	interval 2011-2012 and 2012-2013	1144	difference per species per transect, interval 2011-2012 and 2012-2013
birds	netting	six times from early march to early july	3	7	8	8	-	-	-
moths	Heath traps	eight nights from early june to half September	2	5	5	5	2012 and 2013	338	number of individuals per family per transect
plants	permanent quadrats	2011, repeat 2014	8	0	0	8	-	-	-

Table S2. The sampling method, yearly sampling routine per species group, the number of sites sampled per year, and the data presented in this paper.

Supplementary results

Birds

Species	**common species name**	**relative increase in observations (average between sites)**	**S.E.**	**n sites**
Turdus merula	common blackbird	0.63	0.23	5
Aegithalos caudatus	long-tailed tit	0.53	0.23	5
Regulus regulus	goldcrest	0.49	0.24	5
Phylloscopus trochilus	Willow warbler	0.31	0.31	7
Parus major	great tit	0.29	0.37	8
Turdus philomelos	song thrush	-0.09	0.16	6
Phoenicurus phoenicurus	common redstart	-0.09	0.11	6
Periparus ater	coal tit	-0.12	0.12	6
Lophophanes christatus	European crested tit	-0.21	0.24	7
Certhia brachydactyla	short-toed treecreeper	-0.23	0.20	6

Table S3. Five common species which showed the largest relative increase (upper five) and decrease (lower five) in observations between 2012 and 2013 at illuminated transects (colours combined per site) compared to dark control. For each species we calculated the impact of light by averaging over all sites the species occurred (at least 5 sites) the difference between the change in absolute numbers in the light (white, red and green combined) and in the dark transects, dividing by the absolute number of birds in 2012 on the entire site (summed for all four treatments). A value of 0.2 thus means that (averaged over the sites) on the white, red and green transects combined the species increased with 20% more than in the dark transect. N sites indicates the number of sites the species was found.