Project closing report

Project ID: PD 120998 (post-doctoral research grant, 2016. Oct. 1 – 2019. Sept. 30)
Principal investigator: Gábor Seress
– 2019. 18th of Oct. –

INTRODUCTION

During this research project my aim was to investigate the hypothesis that limited arthropod food availability could be responsible for the reduced breeding success and body size of a typical urban-dwelling bird species, the Great tit *Parus major*. To achieve this aim I conducted my research at multiple, complementing levels: at the level of the arthropod fauna in birds' environment, at the level of nestlings' diet and finally, at the level of birds' reproductive output. The research was carried out in accordance with the proposed research plan and, within the framework of this project, two other studies (with topics closely related to the aims of the current proposal) had also been carried out and published (see 1.2 and 3.2 below). Below I will summarize all the progress and results of the research project, and also list the related publications.

RESULTS

1. Survey on caterpillar abundance and birds' breeding success and nestling development *(Objectives 2.1 & 2.4. of the proposal)*

1.1. Caterpillar abundance and birds' breeding success. We analysed tree phenology, caterpillar biomass and Great tits' breeding success collected throughout four years and four habitats (two forest and two urban, 2013-2016). In the urban sites we found a trend of earlier leaf emergence in urban sites and also earlier seasonal onset of birds' breeding, however, this was not associated with consistently earlier peak in food availability. Despite this the seasonal dynamics of caterpillar biomass exhibited striking differences between habitat types with a clear peak in forests, and several much smaller peaks in urban sites. Caterpillar biomass was ca. 9-24 times higher in forests than urban sites during birds' chick rearing period (Fig.1 a). Additionally, urban Great tits laid smaller clutches, experienced more frequent nestling mortality from starvation, and reared fewer offspring (of lower body mass) to fledging age (Fig.1 b-c).

These results are published in *Ecological Applications* and in a Hungarian journal, and were also presented on an international and on two Hungarian conferences. A BsC thesis has also been prepared from the data collected within the framework of this project.

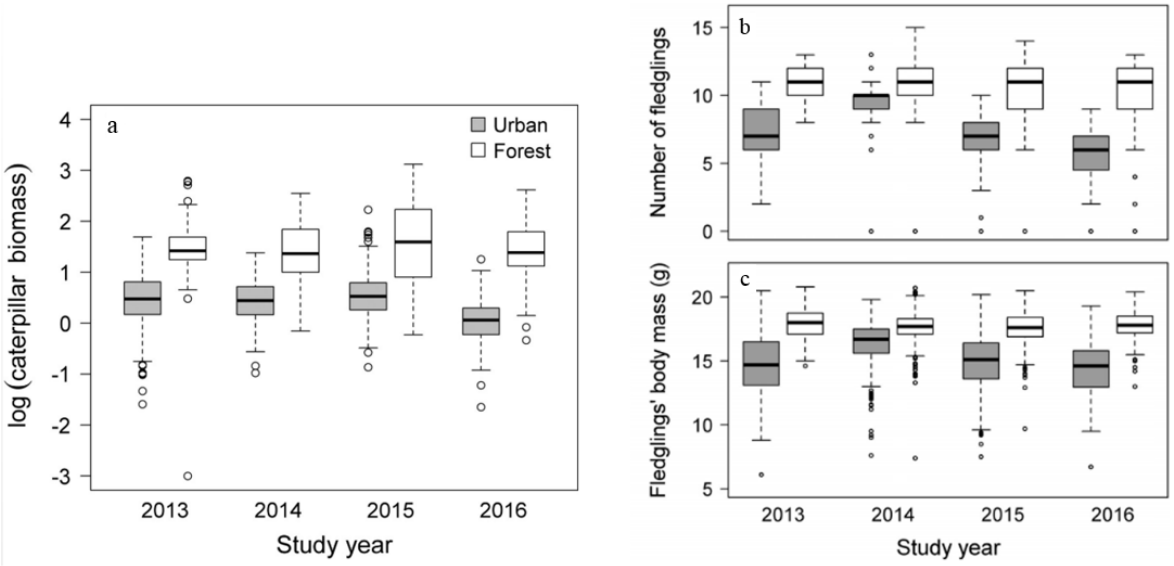

Fig.1. Differences between urban (gray) and forest (white) habitats, in the four years in **(a)** caterpillar biomass (mg/h) during the during the brood-rearing period, and Great tits' breeding success: **(b)** number of fledglings, and **(c)** body mass of fledglings (both measured at 14–16 days of chick age).

Paper:

Seress, G., Hammer, T., Bókony, V., Vincze, E., Preiszner, B., Pipoly, I., Sinkovics, Cs., Evans, K. L. & Liker, A. (2018). *Impact of urbanization on abundance and phenology of caterpillars and consequences for breeding in an insectivorous bird.* Ecological Applications. https://doi.org/10.1002/eap.1730

Ágh, N. & **Seress, G**. (2019). *[In Hungarian] Az urbanizáció és a széncinegék – fiókanevelés az erdőben és nagyvárosban / Urbanization and birds – Raising young in forest and urban environments.* Természetbúvár, 2019 / 5.

Conference:

Seress, G., Hammer, T., Bókony, V., Vincze, E., Preiszner, B., Pipoly, I., Sinkovics, Cs., Evans, K.L., Liker, A. *Impact of urbanization on abundance and phenology of caterpillars and consequences for breeding in an insectivorous bird*. Oral presentation: Eighth International Hole-Nesting Birds Conference, Trondheim, Norway. 2017.10.31-11.02.

Seress, G., Hammer, T., Bókony, V., Vincze, E., Preiszner, B., Pipoly, I., Sinkovics, Cs., Evans, K. L., Liker, A. 2018. *[In Hungarian] A lombfakadás, a hernyók szezonalitása és tömegessége, és a széncinegék szaporodási sikere közötti összefüggések és eltérések városi és természetes élőhelyeken. / Differences and relationships between tree phenology, caterpillar abundance and avian breeding success in urban and forest environments.* Oral presentation: I. Urbanizációs Ökológia Konferencia, Veszprém, Hungary. 2018. 10.19-20.

Other:

Bubla, P., 2018: *Erdei és városi fán élő hernyópopulációk összehasonlítása [in Hungarian] / Comparison of the populations of arboreal caterpillars in urban and forest habitats.* BsC Thesis, University of Veterinary Medicine, Budapest. **Supervisors:** A. Liker & **G. Seress**.

Award

Seress, G. *[in Hungarian] Az élőhely urbanizáció hatásai a madarak táplálékbázisára és szaporodási sikerére / The effects of habitat urbanization on birds' food availability and breeding success*. Plenary talk on the forum of the *"Excellent Young Researcher of the Year"*, **a prize awarded to G.S.** by The Regional Committee in Veszprém of the Hungarian Academy of Sciences, Veszprém, Hungary. 2017.12.6

1.2. The effect of light pollution on caterpillar biomass in urban areas. To further explore the possible causes responsible for the drastically reduced caterpillar biomass in urban environments (see 1.1 above) we conducted a study closely related to the proposal's objectives. We studied the effect of artificial light at night (ALAN) on the biomass of arboreal caterpillar populations in four consecutive years (2014-2017) and two cities (Veszprém and Balatonfüred) with different characteristics. We predicted that increasing ALAN intensity is associated with reduced caterpillar biomass, because ALAN may increase predation risk for both caterpillars and adult lepidopterans (i.e. moths), and can also hinder the moths' reproductive success. We estimated caterpillar biomass from frass samples (n= 3061) collected from 36 focal trees (18 per city), following the same sampling design as in 1.1 (see above).

Our results did not support the negative effect of ALAN on urban caterpillar populations, because ALAN intensity was not related to caterpillar biomass (Fig.2), and this lack of effect was consistent between study sites and tree species. We suggest that the effect of ALAN on urban caterpillar biomass is either weak and thus can be masked by other, local environmental factors, or light pollution may have antagonistic effects acting during different stages of the lepidopteran life cycle. Another explanation could be that even the lower levels of our sites' public lighting are strong enough to cause serious detrimental effects for caterpillars, resulting in their uniformly low biomass.

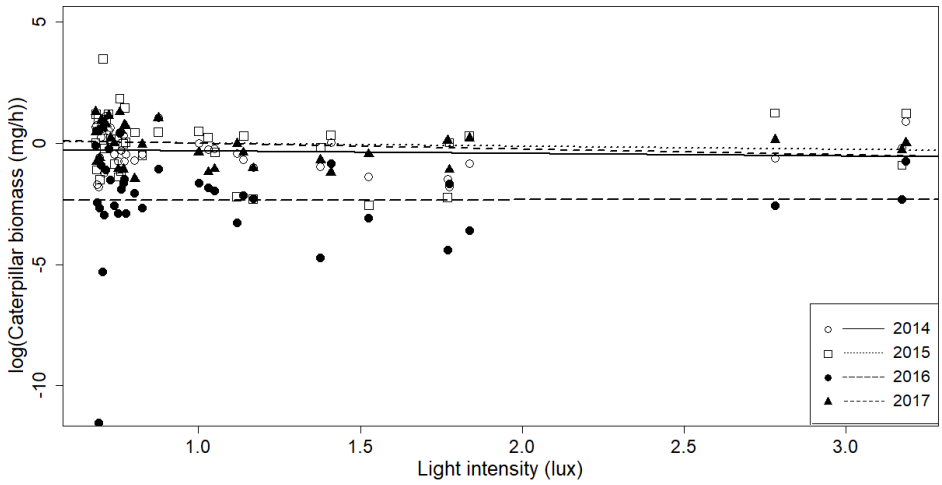

Fig.2. Scatterplot showing the relationship between light intensity and caterpillar biomass (log [mg/h/0.25m^2]), points showing up as mean values of focal trees per year, with regression lines illustrating the direction of the trends separately for the four years.

We published these results on two conferences, and prepared a manuscript which has been accepted for publication with major revision (currently under revision) in *Urban Ecosystems*. **Please note that the topic of**

this study is closely to the general aims of the proposal and though it was not planned in the original proposal but was carried out during the projects' duration.

Paper:

Péter, Á.*, **Seress, G**.*, Sándor, K., Klucsik, K.P., Vincze, E. & Liker, A. (2019) *The effect of artificial light at night on the biomass of caterpillars feeding in urban tree canopies*. Urban Ecosystems (under revision).

* ***Péter, Á. and Seress, G. are joint first authors of this study.***

Conference:

Péter, Á., Sándor, K., **Seress, G**., Vincze, E., Klucsik, K. P., Liker, A. 2018. *[In Hungarian] Az éjszakai mesterséges fény hatása a lombkoronában élő lepkehernyók biomasszájára. / The effect of artificial light at night on the biomass of caterpillars feeding in urban tree canopies.* Oral presentation: A Magyar Etológiai Társaság XX. Kongresszusa, Cluj Napoca, Romania. 2018.11.23-25.

Péter, Á., Sándor, K., **Seress, G**., Vincze, E., Klucsik, K. P., Liker, A. 2018. *[In Hungarian] Az éjszakai mesterséges fény hatása a lombkoronában élő lepkehernyók biomasszájára. / The effect of artificial light at night on the biomass of caterpillars feeding in urban tree canopies.* Oral presentation: I. Urbanizációs Ökológia Konferencia, Veszprém, Hungary. 2018. 10.19-20.

2. Manipulation of nestlings' diet *(objective 2.3 of the proposal)*

2.1. Pilot study. First, during a pilot study we captured urban and woodland Great tits, housed them in indoor aviaries (n=2 flocks, 8 individuals in total), and tested their food choice preferences by offering them several artificial food types. We prepared artificial food items from two types of basic material (a protein-rich mix used for hand-rearing nestlings (Orlux Handmix) and an insect-rich mix for feeding insectivorous birds (Orlux Insect Patee)), in different size and colour combinations in order to try to mimic the appearances of naturally occurring caterpillars and larvae. Although the birds tasted several food types during the experiment, only one of the offered food type and only by some of the birds was readily accepted (Orlux Handmix base, without any additional colouring, regardless of its shape).

Then, as the next step, in an outdoor pilot study we used this artificial food type at urban and woodland great tit nests (n=40), and video recorded parents' behaviour for 60 min long periods during the nestling rearing period. Although parent birds accepted the artificial food type in almost all of the involved nests they typically consumed the food themselves rather than feeding it to their young. Taken together this two-phased pilot study has shown us that wild living birds can readily accept and consume novel food items and that our method is suitable for this kind of data collection. However, we should change the applied food type as it was mainly consumed by adult birds and because both its warranty period and its handling were problematic in hot environmental conditions.

A BsC thesis work was prepared from the results of the indoor and outdoor pilot study.

Other:

Völgyi, N., 2017: [In Hungarian] Mesterséges kiegészítő táplálék kifejlesztése széncinegék számára, és tesztelése laboratóriumi és szabadon fészkelő madarakkal. / Developing an artificial supplemental food source for Great tits, and its testing in captive and wild-living urban and forest birds. BsC Thesis, University of Pannonia, Veszprém. **Supervisor: G. Seress**.

2.2. Food supplementation study. Between March and May (2017) we conducted an outdoor food manipulation experiment on active Great tit nests in an urban and a woodland study site. Supplemented (treatment) broods received extra food (nutritionally enhanced mealworms, *Tenebrio molitor* larvae) on a daily basis when nestlings were 3-15 days old. The food was provided in non-transparent plastic boxes attached right under the nest box entrance. The amount of mealworms were adjusted according to brood size and nestling age so that the extra food is estimated to cover 40-50% of chicks' daily food requirements. Control broods did not receive supplementary food but their nests were also checked daily for a similar amount of time to ensure that control and treatment broods received the same level of human disturbance. At 15-days of age (i.e., just prior to fledging) we ringed and weighed nestlings (± 0.1 g) and also measured the length of the left tarsus (± 0.1 mm) and right wing (± 1 mm). Final sample sizes were: urban control 10 broods, urban treatment 14 broods; forest control 12 broods and forest treatment 11 broods (n=47 broods in total).

We also used small concealed video cameras to monitor use of the feeder and parental provisioning behaviour n=42 broods). From the videos we extracted nestling provisioning rates and, for treatment broods only, we also categorized food items (mealworm, non-supplementary food (mainly caterpillars and other arthropods), or unidentified food item; Fig. 3 a). We recorded if birds (of any species) other than the focal brood's parents took supplementary food from the feeder and we also recorded if parents consumed supplementary food themselves.

We predicted that (1) control broods in the urban habitat would have reduced provisioning rates, nestling body size and survival compared to control broods in forest, indicating that food availability during the nestling

phase limits breeding success in urban but not in forested areas, and thus (2) control and supplementary fed broods in forested locations would exhibit negligible differences in provisioning rate, body size and nestling survival, whilst (3) urban treatment broods would have significantly increased provisioning rates, nestling body size and survival compared to urban control broods. Finally, if food limitation is indeed a major factor limiting breeding success and nestling development in cities, we expect to find that (4) extra insect food would eliminate or considerably mitigate the differences in reproductive success between urban treatment and forest control groups.

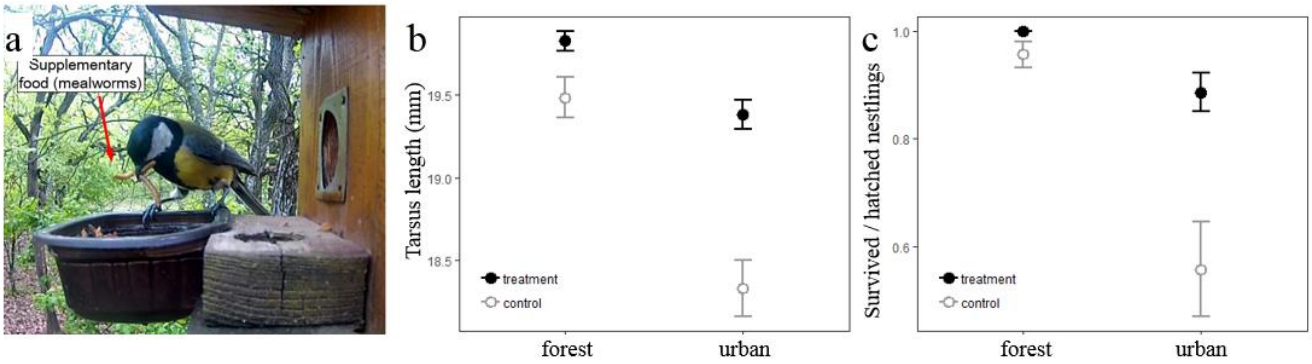

Fig. 3. (a) A forest Great tit parent takes mealworms from the plastic feeder box attached below the entrance hole, and differences (means ± SE) in 15-days old great tit nestlings' **(b)** tarsus length and **(c)** nestling survival in the groups of different habitat × treatment combinations

Our results show that provisioning rates were similar across habitats and control and treatment broods, but treatment (and not control) broods consumed large quantities of supplementary food. As predicted by the food limitation hypothesis we found that nestlings in (1) urban control broods had smaller body size and nestling survival rates than those in forest control broods, (2) forest treatment and control broods had similar body size and survival rates, (3) urban treatment nestlings had larger body size and survival rates than those in urban control broods, and crucially (4) urban treatment broods had similar body size and survival rates to nestlings in forest control broods Fig. 3 b-c).

We published these results on an international and on a Hungarian conference, and prepared a manuscript which has been accepted for publication with major revision (currently under revision) in *Journal of Animal Ecology*.

Paper:

Seress, G., Sándor, K., Evans, K.L. & Liker, A. (2019). *Food availability limits avian reproduction in the city: an experimental study on great tits (Parus major).* Journal of Animal Ecology (under revision).

Conference:

Seress, G., Sándor, K., Evans, K.L. & Liker, A. *Food availability limits avian reproduction in the city: an experimental study on great tits (Parus major).* Poster presentation: Conference of the European Ornithologists' Union, Cluj Napoca, Romania. 2019. 08. 20-26.

Seress, G., Sándor, K., Liker, A. 2017. *[In Hungarian] A szaporodási siker táplálék általi limitáltságának kísérletes tesztelése városi és erdei széncinegéken (Parus major). / An experimental test of food limitation on urban and forest breeding Great tits' breeding success.* Poster presentation: A Magyar Etológiai Társaság XIX. Kongresszusa, Dobogókő, Hungary. 2017.12.1-3. ***The poster has been awarded with the „Best poster presentation of the XIX. MET conference" prize.***

3. Components and biomass of nestlings' diet *(objective 2.2 of the proposal)*

To meet this objective we recorded urban and forest breeding Great tit parents' provisioning behavior on 60-min long video samples by using small, concealed video cameras attached to the nest boxes. From these video recordings we quantified the quality and quantity of the food items provisioned to nestlings (see 3.3 below), and also conducted two methodological study to evaluate the both this methods' accuracy (see 3.1 below) and its potential impacts on birds nestling provisioning behavior (see 3.2 below).

3.1. Methodological study #1. In this methodological research our aim was to assess the accuracy and repeatability of estimates on the size of prey items extracted from parental provisioning video recordings (a method we used in 3.2, see below). We used three approaches to test the reliability of prey size measurements. First, by using artificially manufactured plasticine caterpillars (in a size range similar to that of real caterpillars), we compared their known sizes to their sizes obtained from measurements from video recordings. Second and

third, we examined within-and among-observer repeatability of measurements on the size and volume of actual prey items delivered to nestlings by adult Great tits. Overall, all of the three analyses yielded very high repeatability (rICC = 0.99, 0.98 and 0.93 for the three objectives, respectively), suggesting that the size of prey items delivered to nestlings can be accurately measured from video recordings, and these measurements, in turn, can be used to accurately determine the volume of insect prey items.

We published these results in *Journal of Field Ornithology*. **Please note that this methodological study was not planned in the original proposal but was carried out during the projects' duration**.

Paper:

Sinkovics, C., **Seress, G.**, Fábián, V., Sándor, K., & Liker, A. (2018). *Obtaining accurate measurements of the size and volume of insects fed to nestlings from video recordings.* Journal of Field Ornithology. https://doi.org/10.1111/jofo.12248

3.2. Methodological study #2. In addition, during the reporting period we prepared and published a paper which is methodologically closely related to the objectives of 3.3. In order to collect data on parents' nestling provisioning behaviour we usually capture and individually mark breeding birds on their nest (during the nestling rearing period) and also mount small, concealed video cameras to the nest boxes – but these procedures may have undesired effects on the behaviour of birds that have rarely been quantified. Thus, in this research we studied the effects of both capturing, weighing and measuring, and the presence of video-cameras on the behaviour Great tits breeding in urban and forest habitats. We found no significant effects of the presence of the video camera on birds' behaviour, but found that males captured in their nest boxes were more vigilant and hesitated longer before entering nest boxes, and also had slightly lower provisioning rates than males that had not been captured. Captured females also tended to be more vigilant than non-captured females, but their provisioning rates were not affected. In males, capturing also influenced the behaviour of their non-captured mates. We found no habitat-related difference in these, and our treatments also had no effect on birds' breeding success. Our results suggest that while capturing can influence parents' behaviour even several days later, the presence of the small video camera on the nest does not introduce bias into the collected data, therefore can be a suitable method in ecological studies.

We published these results in *Journal of Field Ornithology*. **Please note that this methodological study was not planned in the original proposal but was published during the projects' duration**.

Paper:

Seress, G., Vincze, E., Pipoly, I., Hammer, T., Papp, S., Preiszner, B., Bókony, V. & Liker, A. (2017). *Effects of capture and video-recording on the behavior and breeding success of Great Tits in urban and forest habitats.* Journal of Field Ornithology. https://doi.org/10.1111/jofo.12205

3.3. Quantity and quality of nestling diet. To quantify the volume and composition of Great tit nestlings' we processed 60-min long video samples of n=153 broods (76 urban and 77 forest broods) recorded in 2014-2016. In all of these broods both parents were confirmed to be alive at the time of the video recording. From each video we extracted data on provisioning events and calculated the following variables: (1) number of feeding events per hour, (2) provisioning rate (i.e. nr. of feeding events per nestling per hour), (3) average prey volume per feeding event (delivered prey item), (4) average prey volume per nestling per hour, and (5) nestling food composition (food items categorized as 'caterpillar', 'other arthropod', or 'other food item', unidentified food items excluded; n=3274 feeding events in total (urban = 1650, forest = 1624)).

In order to calculate prey biomass, we applied the following method. For each feeding event when the prey item was clearly visible in the parent's beak and was hold right before the nest box entrance we took a screenshot. From these screenshots we measured prey length and width (legs and wings were not taken into account; Fig. 4), then calculated prey volume (in mm^3) using the following equation $V=\pi*h*(0.5s)^2$ (V is prey volume, and l and w are length and average width of a prey item; i.e. we treated prey items as cylinder-like objects). We used the diameter of the entrance hole as a size reference for the measurements, as it is uniformly 32 mm in all the involved nest boxes. Note that only a subset of the videos were suitable for such precise measurements (either due to inadequate light conditions, or due to that parents' movement was too fast to take sharp screenshots of the delivered food items), so in total we could involve n=92 broods (43 urban, 49 forest) to these measurements. All measurements were conducted in ImageJ software.

We found no statistically significant differences in (1) the number of feeding events per hour between habitats, but (2) provisioning rates were significantly higher in urban compared to forest broods in two (2015 and 2016) out of the three years (Fig. 5a). We also found that (3) the average prey volume per prey item was significantly higher in forest compared to urban habitats in in two (2015 and 2016) out of the three years. The higher feeding frequencies but smaller food loads per feeding in urban compared to forest broods resulted in that

we found no significant difference between habitats in (4) the average amount of food a nestling received (i.e. the average prey volume per nestling per hour) (Fig. 5b). In the composition of nestling diet we found that urban chicks received much less caterpillars than forest chicks (ratio of caterpillars of all identified food items, urban: c. 0.59-0.70, forest: c. 0.83-0.91; depending on year), a difference that was statistically significant in all three years.

Fig. 4. To determine prey volume (biomass) we measured the length (indicated by the white dotted line) and width (averaged from three measurements as indicated by the black, solid lines) of the delivered prey items from screenshots taken from nestling provisioning video recordings. The vertical diameter of the nest box entrance hole served as size reference for measurements. Screenshot taken by C.S.

To summarize these results we found that, compared to forest broods, in urban nests feeding rates are somewhat higher but received food loads (prey volume) are smaller, resulting in no habitat difference in the received amount of food per nestling per hour. However, urban chicks receive lower quality food due to the relative scarcity of caterpillars, their preferred and optimal prey items, in their diet, which probably result in their lower body mass and higher starvation induced mortality we found in the same years and study sites (see 1.1. above).

We already published these results on an international and on two Hungarian conferences, and a manuscript is currently in preparation (we plan to publish it in an international peer-reviewed journal). A BsC thesis has also been prepared from the data collected within the framework of this project.

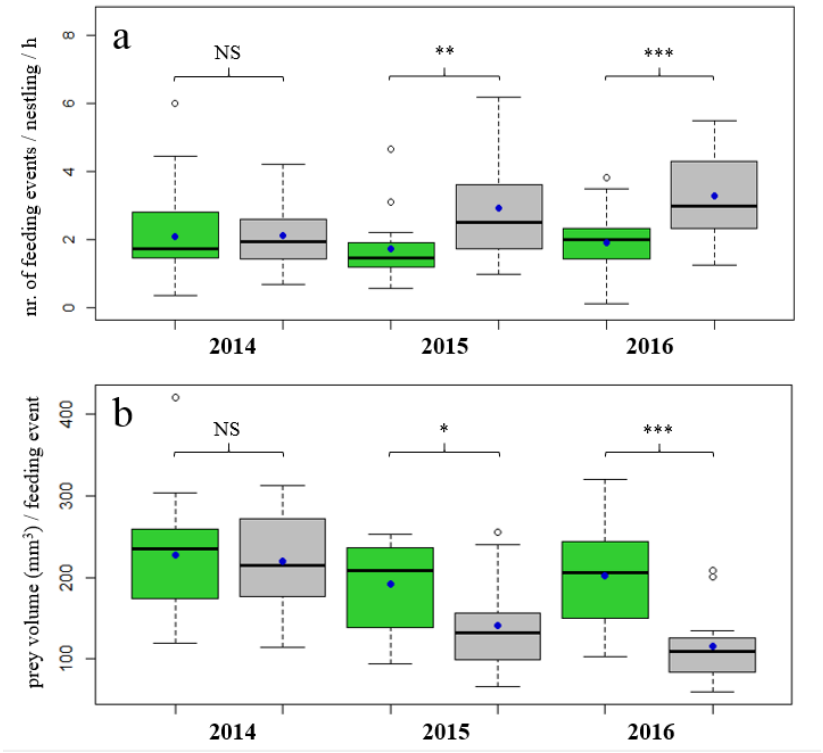

Fig. 5. Differences in **(a)** provision rates and **in (b)** the amount of food received by nestlings per provisioning event between urban (grey) and forest (green) habitats. The thick, middle lines represent medians while the blue dots show means.

Conference:

Liker, A., Sinkovics, C., Pipoly, I., Vincze, E., Evans, K.L., **Seress, G**. *Effects of urbanization on nestling diet and reproductive success in great tits (Parus major).* Poster presentation: Conference of the European Ornithologists' Union, Cluj Napoca, Romania. 2019. 08. 20-26.

Sinkovics, C., **Seress, G.**, Pipoly, I., Vincze, E., Liker, A. 2018. *[In Hungarian] Urbanizáció hatása széncinegék (Parus major) fiókaetetési viselkedésére / The effect of urbanization on the nestling provisioning behaviour of Great tits (Parus major).* Oral presentation: I. Urbanizációs Ökológia Konferencia, Veszprém, Hungary. 2018. 10.19-20.

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Other:

Fábián, V., 2018: *[In Hungarian] Ivari különbségek a széncinegék (Parus major) utódgondozó viselkedésében természetes és urbanizált környezetben / Sexual differences in the nestling provisioning behaviour of Great tits (Parus major) in urban and forest habitats.* BsC thesis, University of Pannonia, Veszprém. Supervisors: **G. Seress** & C. Sinkovics.

PUBLICATIONS

Articles published in English:

Seress, G., Hammer, T., Bókony, V., Vincze, E., Preiszner, B., Pipoly, I., Sinkovics, Cs., Evans, K. L. & Liker, A. (2018). *Impact of urbanization on abundance and phenology of caterpillars and consequences for breeding in an insectivorous bird.* Ecological Applications. https://doi.org/10.1002/eap.1730

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Currently under revision:

Seress, G., Sándor, K., Evans, K.L. & Liker, A. (2019). *Food availability limits avian reproduction in the city: an experimental study on great tits (Parus major).* Journal of Animal Ecology (accepted with major revision, currently under revision). Pre-print uploaded to the REAL-MTA repository.

Péter, Á.*****, **Seress, G.***, Sándor, K., Klucsik, K.P., Vincze, E. & Liker, A. (2019) *The effect of artificial light at night on the biomass of caterpillars feeding in urban tree canopies*. Urban Ecosystems (accepted with major revision, currently under revision). Pre-print uploaded to the REAL-MTA repository.
**** Péter, Á. and Seress, G. are joint first authors of this study.***

Articles published in Hungarian:

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Conference materials (in English)

Seress, G., Hammer, T., Bókony, V., Vincze, E., Preiszner, B., Pipoly, I., Sinkovics, Cs., Evans, K.L., Liker, A. *Impact of urbanization on abundance and phenology of caterpillars and consequences for breeding in an insectivorous bird*. Oral presentation: Eighth International Hole-Nesting Birds Conference, Trondheim, Norway. 2017.10.31-11.02.

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Seress, G. *[in Hungarian] Az élőhely urbanizáció hatásai a madarak táplálékbázisára és szaporodási sikerére / The effects of habitat urbanization on birds' food availability and breeding success*. Plenary talk on the forum of the *"Excellent Young Researcher of the Year",* **a prize awarded to G.S.** by The Regional Committee in Veszprém of the Hungarian Academy of Sciences, Veszprém, Hungary. 2017.12.6

Péter, Á., Sándor, K., **Seress, G**., Vincze, E., Klucsik, K. P., Liker, A. 2018. *[In Hungarian] Az éjszakai mesterséges fény hatása a lombkoronában élő lepkehernyók biomasszájára. / The effect of artificial light at night on the biomass of caterpillars feeding in urban tree canopies*. Oral presentation: A Magyar Etológiai Társaság XX. Kongresszusa, Cluj Napoca, Romania. 2018.11.23-25.

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