BIOLOGY LETTERS

rsbl.royalsocietypublishing.org

Research

Cite this article: Davies S, Sewall KB. 2016 Agonistic urban birds: elevated territorial aggression of urban song sparrows is individually consistent within a breeding period. *Biol. Lett.* **12**: 20160315. http://dx.doi.org/10.1098/rsbl.2016.0315

Received: 14 April 2016
Accepted: 24 May 2016

Subject Areas:
behaviour, ecology

Keywords:
urbanization, territorial aggression, repeatability, testosterone

Author for correspondence:
Scott Davies
e-mail: sdavies1@vt.edu

Electronic supplementary material is available at http://dx.doi.org/10.1098/rsbl.2016.0315 or via http://rsbl.royalsocietypublishing.org.

THE ROYAL SOCIETY PUBLISHING

Animal behaviour

Agonistic urban birds: elevated territorial aggression of urban song sparrows is individually consistent within a breeding period

Scott Davies and Kendra B. Sewall

Department of Biological Sciences, Virginia Tech, Blacksburg, VA 24061, USA

SD, 0000-0003-3890-7372

Urban birds often more vigorously defend their territories during simulated intrusions than do their rural counterparts, but the factors responsible remain unclear. To address this issue, we investigated whether the disparity in territorial aggression of urban and rural male song sparrows, *Melospiza melodia*, is individually consistent within a breeding period. Additionally, to better understand the physiological and ecological factors underlying this behavioural difference, we examined whether territoriality was associated with plasma testosterone, a hormone that contributes to elevated aggression in vertebrates, and/or conspecific density, a factor often positively related to aggression. The urbanization-related difference in territoriality was individually consistent within a breeding period. However, the elevated territorial aggression of urban birds was not associated with plasma testosterone and, counter to our predictions, conspecific density was lower in urban compared with rural areas. We suggest that other aspects of testosterone signalling and features of the socio-ecological environment, such as the availability of breeding sites, may underlie increased territorial aggression in urban birds.

1. Introduction

Urban expansion presents a major challenge for organisms, because to survive they must adjust to novel environmental conditions. One such adjustment is an elevation in the intensity of conspecific territorial aggression of male urban birds [1–3], including song sparrows, *Melospiza melodia*, in Blacksburg and Radford, Virginia, USA [4], where this study was conducted. Territorial aggression is a crucial behaviour that is found throughout the animal kingdom and is used to defend limited resources, such as breeding territories, mates or food [5,6]. Although aggression has been the focus of considerable research, few studies have investigated the factors underlying interpopulation variation in territorial aggression [7]. In this study, we tested whether urbanization-related differences in territorial aggression were individually consistent within a breeding period and whether the hormone testosterone and/or conspecific density, two factors associated with territoriality, differed across habitat types. Examining the mechanisms and consistency of behavioural variation in response to habitat change is a first step toward understanding the impact of such changes on wild populations.

A crucial step in understanding the factors responsible for differences in behaviour is to establish whether the behaviour is individually consistent [8]. It is unclear, however, whether the elevated territoriality of urban birds is a consistent habitat-related behavioural difference. Territorial aggression is individually repeatable in rural male song sparrows [9,10], but it is unknown whether this behaviour is individually repeatable when simultaneously considering both urban and rural birds. To address this, we tested whether the territorial

aggression of urban and rural song sparrows is individually consistent and repeatable within a breeding period.

Testosterone contributes to the elevation of aggression in vertebrates, particularly in reproductive contexts [11]. Indeed, territorial aggression in many vertebrates coincides with peak circulating testosterone during the breeding period [12]. Experiments on wild birds have shown that territoriality during a simulated territorial intrusion, consisting of conspecific song playback and/or a caged live decoy bird, decreases following removal of the testes, and that this decrease is reversed by exogenous testosterone [13]. Testosterone is, therefore, a candidate to contribute to the elevated territorial aggression of urban birds. Only a few studies have examined whether plasma testosterone accounts for the elevated territoriality of urban birds [1,3]. Higher territorial aggression in urban birds was not mirrored by corresponding differences in plasma testosterone in two Sonoran Desert bird species [1], while testosterone was actually lower in urban dark-eyed juncos, *Junco hyemalis* [3]. Here, we investigated whether plasma testosterone parallels habitat-related differences in the territorial aggression of urban and rural song sparrows in Blacksburg and Radford, Virginia, USA.

The intensity of conspecific territorial aggression in a population is also shaped by conspecific density [5,6]; environments with high conspecific density are thought to favour elevated territoriality [7]. The elevated territorial aggression in urban male song sparrows leads us to predict that the urban areas considered here would have a higher density of birds than the surrounding rural areas, which we tested by comparing point counts of song sparrows in the two habitat types.

2. Material and methods

(a) Territorial aggressive behaviour

We measured territorial aggression in 35 urban and 38 rural male song sparrows at three rural and three urban sites (see the electronic supplementary material for details and estimates of urbanization for each site) using standardized conspecific song playback [9]. Briefly, between 8 April and 19 May 2015, we positioned a speaker (Micro II; JBL, Northridge, CA, USA) in the estimated centre of the focal male's territory and randomly selected one of 16 playback stimuli (broadcasted for 6 min at one song per 10 s). Each playback stimulus consisted of two song types recorded from a single male from a population in Durham, NC, USA, and approximated natural song (79–85 dB at 1 m). During each playback, we recorded the average distance the focal male was from the speaker, as well as the number of songs, soft songs (lower amplitude songs produced during aggressive contests) and wing waves (a postural aggressive display) [14]. We combined these behaviours into a single territorial aggression score using principal components analysis (PCA; electronic supplementary material, table S2), which we then analysed using an ANOVA. Greater values indicate that a bird had a shorter average distance from the speaker and performed more songs, soft songs and wing waves.

We measured territoriality of 25 rural and 28 urban birds a second time during the 2015 breeding period, between 8 and 11 June 2015 (average of 26 ($\pm$0.5) days after the first measure). We identified males for repeated assays of territorial aggression using unique colour band combinations. To test whether there is a consistent habitat-related difference in territorial aggression score, we used a repeated measures ANOVA. Additionally, we used the intraclass correlation to test whether territorial aggression score is individually repeatable.

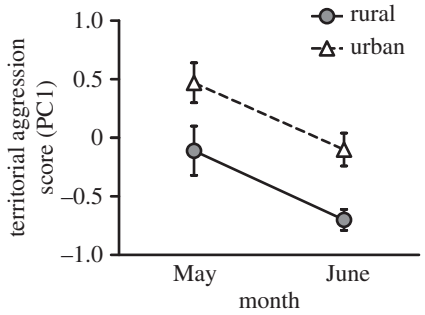

Figure 1. The elevated territorial aggression towards conspecific song playback of urban ($n = 28$) compared with rural ($n = 25$) male song sparrows was individually consistent within a breeding season. Points depict mean ($\pm$s.e.m.) principal component scores of territorial behaviours.

(b) Plasma testosterone

Immediately after measuring behaviour of the birds sampled in April and May, we set up a mistnet, caught the focal bird using song playback, and then collected a blood sample (total of 47 birds, 16 urban and 31 rural) to quantify plasma testosterone using enzyme-linked immunoassays (ADI-900-065, Enzo Life Sciences), which we validated for use in the song sparrow. For details of blood collection and the hormone assay, see the electronic supplementary material. We analysed these data using a generalized linear model (GLM) with a Gamma distribution and log link. For all GLMs, assessment of the residual deviance showed that the selected link was the most appropriate for our data.

(c) Conspecific density

To estimate the density of song sparrows in urban and rural areas, we conducted 10 min point counts at 40 randomly selected rural ($n = 20$) and urban ($n = 20$) sampling locations across all six field sites between 4 and 7 May 2015. We counted the number of song sparrows seen or heard singing within 50 m of the observer. Counts were conducted by the same observer on mornings of clear weather from 30 min before to 3 h after sunrise. We analysed these data using a GLM with a Poisson distribution and log link.

3. Results

(a) Territorial aggressive behaviour

The territorial aggression score (PC1) was greater in urban than rural sparrows sampled in April and May (rural: -0.01 ± 0.17; urban: 0.64 ± 0.18; $F_{1,53} = 5.47$, $p = 0.023$), and was not affected by date ($F_{1,53} = 0.012$, $p = 0.91$). This difference in territorial aggression persisted in May and June; territorial aggression score remained greater in urban compared with rural song sparrows later in the breeding period ($F_{1,51} = 14.4$, $p < 0.0001$; figure 1). The intensity of territorial aggression in both habitats declined between May and June ($F_{1,51} = 13.35$, $p = 0.001$). However, there was no interaction between habitat and month ($F_{1,51} = 0.002$, $p = 0.96$). Furthermore, territorial aggression score was individually repeatable (intraclass correlation = 0.32, $F_{52,52} = 2.20$, $p = 0.003$).

(b) Plasma testosterone

Plasma testosterone decreased over the breeding season ($\chi^2_1 = 4.63$, $p = 0.031$), but did not differ between urban and rural sparrows ($\chi^2_1 = 1.68$, $p = 0.20$; figure 2). There was also no effect of playback duration ($\chi^2_1 = 0.32$, $p = 0.57$) or the

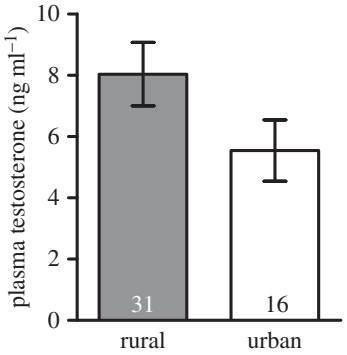

Figure 2. Plasma testosterone of urban and rural male song sparrows was not significantly different (GLM: $\chi^2_1 = 1.68$, $p = 0.20$) when duration of playback and date were included in the model. Columns depict mean ($\pm$s.e.m.).

two-way interactions between habitat and either date ($\chi^2_1 = 2.60, p = 0.11$) or playback duration ($\chi^2_1 = 1.36, p = 0.24$).

(c) Conspecific density

Fewer song sparrows were detected in urban compared with rural areas (urban: 0.85 ± 0.15; rural: 2.1 ± 0.25; $\chi^2_1 = 9.90$, $p = 0.002$). Likewise, fewer song sparrows were heard singing in urban than rural areas (urban: 0.5 ± 0.11; rural: 1.3 ± 0.13; $\chi^2_1 = 6.59$, $p = 0.010$).

4. Discussion

We found that the greater intensity of conspecific territorial aggression of urban male song sparrows in Blacksburg and Radford, Virginia, USA, is individually consistent within a breeding period [9]. Previous studies in songbirds, including song sparrows [1–4], have found that urban birds are more territorially aggressive than their rural counterparts. However, our study is the first to demonstrate that this urbanization-related difference in territoriality is individually consistent within a breeding period. Our findings also extend the individual repeatability of territorial aggression in male song sparrows [9,10] to both urban and rural birds.

Although testosterone is frequently positively related to the intensity of aggression in vertebrates [11], we found that the elevated territoriality of urban male song sparrows was not associated with levels of plasma testosterone. Our finding is consistent with other studies that have examined whether plasma testosterone accounts for urbanization-related differences in territoriality [1]. However, we cannot exclude the possibility that testosterone contributes to the habitat difference in territoriality through other mechanisms. For example, testosterone is synthesized *de novo* in the avian brain [15], which may exert local effects on the neural networks that modulate territoriality [16] independent of circulating testosterone. Furthermore, the hormone dehydroepiandrosterone, which can be metabolized into androgens and oestrogens in target tissues, promotes territorial behaviours in the song sparrow [16] and, therefore, may contribute to the urbanization-associated difference in territoriality.

Similarly, conspecific density is a socio-ecological factor thought to modulate territorial aggression, with higher conspecific density promoting more intense territoriality [5,6]. Given that urban song sparrows more intensely defended their territories, our finding that urban areas had lower conspecific density is counterintuitive. Our finding, together with the finding of Foltz *et al.* [4] that the proximity of neighbouring territorial song sparrows did not explain territoriality, suggests that the positive relationship between conspecific density and territorial aggression may not apply in urban areas.

Although our results do not support plasma testosterone or high conspecific density driving elevated territoriality in urban birds, we suggest that other relatively stable socio-ecological factors, such as differences in resource abundance across rural and urban areas [4], could be responsible for differences in territorial aggression. For example, urban areas could provide a more predictable or better quality food source [4,17], giving urban sparrows more time or motivation to be vigilant for conspecific intruders. Continued investigation of the proximate mechanisms and socio-ecological drivers of elevated territoriality in urban birds will improve our fundamental understanding of this social behaviour and also of the effects of urbanization on wild populations.

Ethics. Our methods were approved by the Virginia Tech institutional animal care and use committee (protocol: BIOL 13-074) and conducted under permits from the Virginia department of game and inland fisheries (permit no. 48639), the US fish and wildlife service (permit: MB08005B-0) and the USGS bird banding laboratory (permit no. 23818).

Data accessibility. The full dataset used in this study is available in the electronic supplementary material.

Authors' contributions. S.D. and K.B.S. conceived of and designed the study. S.D. collected and analysed the data, carried out the hormone assay and wrote the manuscript. K.B.S. assisted with data collection, contributed to the interpretation of results and provided editorial input on the manuscript. Both authors agree to be held accountable for the content herein and approve the final version of the manuscript.

Competing interests. We have no competing interests.

Funding. Funding was provided by the Department of Biological Sciences and the Fralin Institute at Virginia Tech.

Acknowledgements. We thank H. Butler for assistance and S. Foltz for information about field sites.

References

1. Fokidis HB, Orchinik M, Deviche P. 2011 Context-specific territorial behavior in urban birds: no evidence for involvement of testosterone or corticosterone. *Horm. Behav.* **59**, 133–143. (doi:10.1016/j.yhbeh.2010.11.002)
2. Scales J, Hyman J, Hughes M. 2011 Behavioral syndromes break down in urban song sparrow populations. *Ethology* **117**, 887–895. (doi:10.1111/j.1439-0310.2011.01943.x)
3. Atwell JW, Cardoso GC, Whittaker DJ, Price TD, Ketterson ED. 2014 Hormonal, behavioral, and life-history traits exhibit correlated shifts in relation to population establishment in a novel environment. *Am. Nat.* **184**, E147–E160. (doi:10.1086/678398)
4. Foltz SL, Ross AE, Laing BT, Rock RP, Battle KE, Moore IT. 2015 Get off my lawn: increased aggression in urban song sparrows is related to resource availability. *Behav. Ecol.* **26**, 1548–1557. (doi:10.1093/beheco/arv111)
5. Grant JWA. 1993 Whether or not to defend? The influence of resource distribution. *Mar. Behav. Physiol.* **23**, 137–153. (doi:10.1080/10236249309378862)
6. Brown JL. 1964 The evolution of diversity in avian territorial systems. *Wilson Bull.* **76**, 160–169.

7. Yoon J, Sillett TS, Morrison SA, Ghalambor CK. 2012 Breeding density, not life history, predicts interpopulation differences in territorial aggression in a passerine bird. *Anim. Behav.* **84**, 515–521. (doi:10.1016/j.anbehav.2012.05.024)
8. Debeffe L *et al.* 2015 Short- and long-term repeatability of docility in the roe deer: sex and age matter. *Anim. Behav.* **109**, 53–63. (doi:10.1016/j.anbehav.2015.08.003)
9. Nowicki S, Searcy WA, Krueger T, Hughes M. 2002 Individual variation in response to simulated territorial challenge among territory-holding song sparrows. *J. Avian Biol.* **33**, 253–259. (doi:10.1034/j.1600-048X.2002.330307.x)
10. Hyman J, Hughes M, Searcy WA, Nowicki S. 2004 Individual variation in the strength of territory defense in male song sparrows: correlates of age, territory tenure, and neighbor aggressiveness. *Behaviour* **141**, 15–27. (doi:10.1163/156853904772746574)
11. Wingfield JC, Moore IT, Goymann W, Wacker DW, Sperry T. 2006 Contexts and ethology of vertebrate aggression: implications for the evolution of hormone–behavior interactions. In *Biology of aggression* (ed. RJ Nelson), pp. 179–210. New York, NY: Oxford University Press.
12. Wingfield JC, Ball GF, Dufty AM, Hegner RE, Ramenofsky M. 1987 Testosterone and aggression in birds. *Am. Sci.* **75**, 602–608.
13. Wingfield JC. 1994 Regulation of territorial behavior in the sedentary song sparrow, *Melospiza melodia morphna*. *Horm. Behav.* **28**, 1–15. (doi:10.1006/hbeh.1994.1001)
14. Searcy WA, Anderson RC, Nowicki S. 2006 Bird song as a signal of aggressive intent. *Behav. Ecol. Sociobiol.* **60**, 234–241. (doi:10.1007/s00265-006-0161-9)
15. Soma KK, Scotti ML, Newman AEM, Charlier TD, Demas GE. 2008 Novel mechanisms for neuroendocrine regulation of aggression. *Front. Neuroendocrinol.* **29**, 476–489. (doi:10.1016/j.yfrne.2007.12.003)
16. Goodson JL, Wilson LC, Schrock SE. 2012 To flock or fight: neurochemical signatures of divergent life histories in sparrows. *Proc. Natl Acad. Sci. USA* **109**(suppl. 1), 10 685–10 692. (doi:10.1073/pnas.1203394109)
17. Deviche P, Davies S. 2014 Reproductive phenology of urban birds: environmental cues and mechanisms. In *Avian urban ecology: behavioral and physiological adaptations* (eds D Gil, H Brumm), pp. 98–115. Oxford, UK: Oxford University Press.

BIOLOGY LETTERS

rsbl.royalsocietypublishing.org

Correction

Cite this article: Davies S, Sewall KB. 2016 Correction to 'Agonistic urban birds: elevated territorial aggression of urban song sparrows is individually consistent within a breeding period'. *Biol. Lett.* **12**: 20160900. http://dx.doi.org/10.1098/rsbl.2016.0900

Correction to 'Agonistic urban birds: elevated territorial aggression of urban song sparrows is individually consistent within a breeding period'

Scott Davies and Kendra B. Sewall

Biol. Lett. **12**, 20160315. (Published online 21 June 2016). (doi:10.1098/rsbl.2016.0315)

We have been made aware that the estimate of individual repeatability that we used in our study (i.e. the intraclass correlation coefficient) overestimates repeatability, and that calculating repeatability using the analysis of variance-based method, as described by Lessells & Boag [1]), is a more appropriate estimate. Consequently, we have recalculated repeatability using this more appropriate approach and describe these changes below.

When pooling the behaviour of urban and rural birds, there was a non-significant trend towards low repeatability of territorial aggression score ($r = 0.21$, $F_{52,105} = 1.54$, $p = 0.06$). However, when considering each population alone there was a trend toward repeatability in rural, but not urban birds (urban: $r = 0.02$, $F_{24,49} = 1.04$, $p = 0.46$; rural: $r = 0.28$, $F_{27,55} = 1.78$, $p = 0.07$). Overall, when considering both populations simultaneously or each alone, territorial aggression score was not significantly repeatable within individuals.

In our study, we considered 'repeatability' to be the proportion of variation that is reproducible among repeated measurements. By contrast, we use the term 'consistency' to refer to the consistent difference in population means when measuring the same individuals at multiple times. Although the territorial aggression of individual male song sparrows was not individually repeatable, urban male song sparrows were more territorially aggressive than their rural counterparts at both time points, making the difference in behaviour across habitat types consistent. Thus, our corrected results show consistency in population differences without individual repeatability of behavioural scores. Further, our title should read '. . .elevated territorial aggression of urban song sparrows is consistent within a breeding period' in reference to the consistent difference in population means, not the proportion of variation that is reproducible among repeated measurements.

Additional research is required to determine whether there is a habitat-related difference in the repeatability of territorial aggression in male song sparrows, which our re-analyses and prior work [2,3] suggest may be the case.

References

1. Lessells CM, Boag PT. 1987 Unrepeatable repeatabilities: a common mistake. *Auk* **104**, 116–121. (doi:10.2307/4087240)
2. Scales J, Hyman J, Hughes M. 2011 Behavioral syndromes break down in urban song sparrow populations. *Ethology* **117**, 887–895. (doi:10.1111/j.1439-0310.2011.01943.x)
3. Akçay Ç, Campbell SE, Beecher MD. 2014 Individual differences affect honest signaling in a songbird. *Proc. R. Soc. B* **281**, 20132496. (doi:10.1098/rspb.2013.2496)

THE ROYAL SOCIETY PUBLISHING