Author for correspondence:
Karan J. Odom
e-mail: kjo43@cornell.edu

THE ROYAL SOCIETY
PUBLISHING

Breeding season length predicts duet coordination and consistency in Neotropical wrens (Troglodytidae)

Emily L. Keenan[1], Karan J. Odom[2,3], Marcelo Araya-Salas[2,3,4], Kyle G. Horton[2,5], Matthew Strimas-Mackey[2], Megan A. Meatte[6], Nigel I. Mann[7], Peter J. B. Slater[8], J. Jordan Price[9] and Christopher N. Templeton[6]

[1]College of Arts and Sciences, [2]Laboratory of Ornithology, and [3]Department of Neurobiology and Behavior, Cornell University, Ithaca, NY, USA
[4]Recinto de Golfito, Universidad de Costa Rica, Golfito 60701, Costa Rica
[5]Department of Fish, Wildlife, and Conservation Biology, Colorado State University, Fort Collins, CO, USA
[6]Department of Biology, Pacific University, Forest Grove, OR, USA
[7]Biology Department, The Glasgow Academy, Glasgow G12 8HE, UK
[8]School of Biology, University of St Andrews, Fife KY16 9TH, UK
[9]Department of Biology, St Mary's College of Maryland, St Mary's City, MD, USA

KJO, 0000-0002-9074-135X; MA-S, 0000-0003-3594-619X; JJP, 0000-0002-0771-6269; CNT, 0000-0001-8701-3186

Many animals produce coordinated signals, but few are more striking than the elaborate male–female vocal duets produced by some tropical songbirds. Yet, little is known about the factors driving the extreme levels of vocal coordination between mated pairs in these taxa. We examined evolutionary patterns of duet coordination and their potential evolutionary drivers in Neotropical wrens (Troglodytidae), a songbird family well known for highly coordinated duets. Across 23 wren species, we show that the degree of coordination and precision with which pairs combine their songs into duets varies by species. This includes some species that alternate their song phrases with exceptional coordination to produce rapidly alternating duets that are highly consistent across renditions. These highly coordinated, consistent duets evolved independently in multiple wren species. Duet coordination and consistency are greatest in species with especially long breeding seasons, but neither duet coordination nor consistency are correlated with clutch size, conspecific abundance or vegetation density. These results suggest that tightly coordinated duets play an important role in mediating breeding behaviour, possibly by signalling commitment or coalition of the pair to mates and other conspecifics.

1. Background

Animals often use complex, coordinated displays to defend territories and attract mates [1,2]. Such signals range from coordinated courtship displays [3,4] to vocal duets [5–7]. In some tropical antiphonally duetting bird species, males and females combine their songs with such precision that they sound like a single bird singing [8–10]. While a number of studies have investigated the drivers responsible for duet evolution generally, few studies have investigated how and why especially coordinated duets evolved [5,7,11–14]. The remarkable level of duet coordination in some species suggests that precisely timed, highly coordinated duets have evolved for reasons beyond those that have been proposed to explain duet evolution in general [15–18].

Leading hypotheses for the evolution of duets include year-round territory defence, maintaining contact between mates and maintenance of long-term pair bonds [17–21]. The territory defence hypothesis posits that duets signal the ability
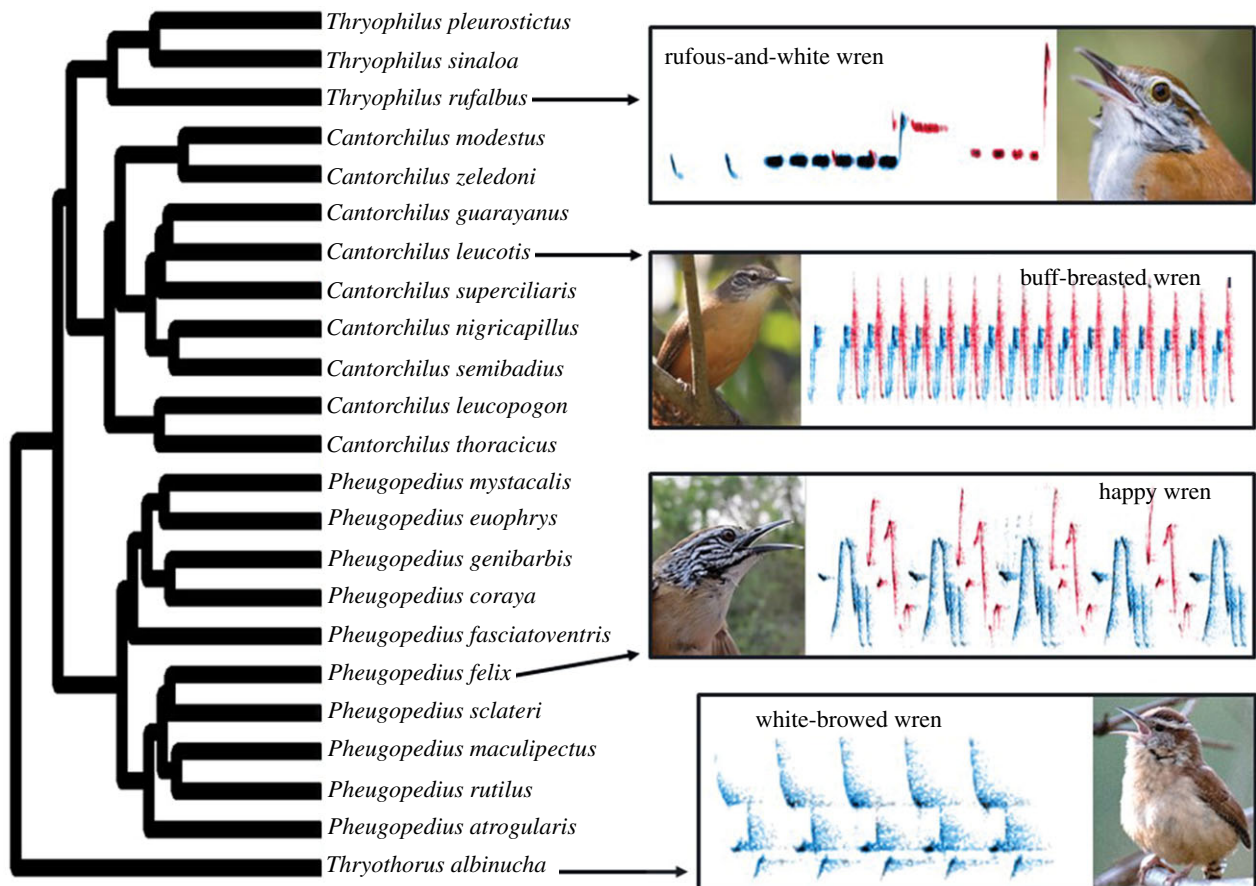
Figure 1. Phylogenetic relationships and spectrograms of duets for four wren species, showing variation in coordination across the four wren genera in the study. Male songs are highlighted in blue and female songs are highlighted in red. Photos by Daniel Mennill (rufous-and-white wren), Hector Bottai (buff-breasted wren), Christopher Templeton (happy wren) and T.C. Davis (Carolina wren, conspecific to white-browed wren in some taxonomies). Phylogeny based on [38]. (Online version in colour.)

of a mated pair to jointly defend a territory [11,12], whereas the maintaining contact hypothesis posits that duets allow mated pairs to stay in contact in dense habitats where visual cues would be obscured [11,21]. By contrast, the pair bond maintenance hypothesis suggests that duets help to maintain long-term partnerships and signal commitment, which may aid in synchronizing reproduction and coordinating reproductive behaviours [22,23]. The pair bond maintenance hypothesis has some overlap with the other two hypotheses, as pairs may communicate the strength of their pair bond to conspecifics during territory defence and maintaining pair bonds may be aided by maintaining contact [15,22,24]. Indeed, duets may serve a variety of functions [14]. In Neotropical songbirds, evidence suggests that duets function in territory defence [25–30], contact maintenance [21] and long-term pair bond maintenance [31–33].

Duet evolution is often tied to tropical natural history [23,34]. The seasonal stability and high adult survival in tropical environments enables species to be sedentary, long-term monogamous and territorial year-round [23,35]. Competition with long-term neighbouring conspecifics in turn promotes the evolution of behaviours such as duets that help pairs defend year-round resources and territories [16,34,35]. In addition, many tropical species can breed over extended periods of the year, which may favour the evolution of behaviours that facilitate asynchronous reproduction by enabling pairs to coordinate reproductive activities, including onset of reproduction, parental care and 'bet hedging' strategies for when to reproduce based on environmental and partner

condition [23,36,37]. The importance of maintaining a social partner, coordinating breeding activities and territory defence over longer periods of the year may lead to the evolution of particularly elaborate signals that allow pair members to signal their commitment to each other or the strength of their pair bond to others [12,15,32,33].

Neotropical wrens (Troglodytidae) offer an exceptional system to study the evolution of duet coordination. Species in this family exhibit diverse singing patterns ranging from long, complex solos to highly coordinated male–female duets and choruses ([7], figure 1). Neotropical wren duets are particularly notable for their temporal coordination, which varies from antiphonal to polyphonal [7–9]. The extent to which polyphonal duet phrases overlap varies among both species and conspecific pairs within species [7,39].

We examined several possible factors responsible for the evolution of highly coordinated duets in the Neotropical wrens [12,13]: (i) breeding season length, (ii) clutch size, (iii) conspecific abundance, and (iv) vegetation density. We hypothesized that longer breeding seasons may select for more coordinated duets because synchronizing breeding for a large part of the year may be benefited by coordinated communication between mated pairs, including partners' ability to signal their commitment to one another or to signal coalition (e.g. strength of the pair bond) to conspecifics [15]. We hypothesized that larger clutch sizes would favour more coordinated duets, assuming that clutch sizes may lead to greater fecundity in long-lived, year-round territorial species. We hypothesized that conspecific abundance would select for increased duet

coordination, assuming that more abundant species experience higher levels of conspecific competition for resources and territories. Species with high conspecific abundance or that use duets in intrasexual competition might also develop highly coordinated duets to avoid masking of mates during territory defence [40]. Lastly, following the acoustic-contact hypothesis, we hypothesized that species in particularly dense forest habitats would have higher levels of duet coordination.

We assessed duet coordination and consistency across renditions of duets in 23 species of Neotropical wrens within a phylogenetic framework. We then compared duet coordination and consistency to the above natural history and environmental parameters. Our goal was to evaluate selective pressures that might be responsible for the evolution of highly coordinated duets.

2. Methods

(a) Field recordings

We measured and compared songs and duets for 23 species of wren, representing four closely related genera (figure 1). The recordings were gathered during previous fieldwork by N.I.M. and colleagues to quantify variation in wren duet structure, as described by Mann *et al*. [7]. To summarize, Mann *et al*. visited a variety of sites throughout Central and South America, banding and recording 1–6 pairs for each of the 23 wren species between January 2001 and May 2003. Each individual was marked with a unique combination of coloured leg bands and a blood sample was collected to later genetically confirm sex. For each pair, Mann *et al*. took four separate 90 min recordings on different days, typically in the morning, and noted each song that was sung by each sex. Each recording was paired with notes and spectrograms, which we used to establish when each sex sang within a duet. Mann *et al*.'s recording sessions sometimes resulted in hundreds of recorded songs per pair. We measured a subset of 10 randomly chosen duets per pair for each pair that was recorded. For the few species without 10 duets available, we measured all instances of duets. There were also a few species for which there were no instances of duets in the original data, so we selected solo vocalizations for our dataset. This resulted in a final dataset of 162 solos and 457 duets measured for the 23 species in the current study (see the electronic supplementary material, table S1). We defined a duet as any instance in which a pair member performed its song immediately following singing by its mate, either in an overlapping or alternating pattern [5,12,13].

(b) Sound analysis

Neotropical wren songs and duets typically consist of phrases, which are stereotyped groupings of 2–15 notes that each singer repeats throughout its song [1]. We used Raven Sound Analysis Software v. 1.5 [41] to select individual notes that comprised each phrase, as well as every phrase for each song within a duet. From these selections, we measured the beginning/end times for each note and phrase, the lengths of notes and phrases, the overlap between notes and between phrases, and the lengths of gaps between phrases (inter-phrase intervals). We extracted these values for males and females separately, so that we could compare timing of male and female songs within a duet.

(c) Coordination score

To statistically evaluate duet coordination, we used the Monte Carlo randomization test implemented in the coor_test function from the R package warbleR [42,43]. The function used the start and end times of vocalizations within a vocal bout (duet) to differentiate among song components and the intervals in between the songs. It then compared the observed duration of overlap between notes (and/or phrases) within a duet (i.e. the sum of the durations from all overlaps in a duet) to a null distribution of overlap durations calculated by repeatedly shuffling both signals and gaps (the silent space between notes or phrases). Null distributions were created using 10 000 iterations. A *p*-value was calculated as the proportion of random overlap durations that were equal to or more extreme than the observed value. We also calculated a duet coordination score, defined as the proportional difference between the observed overlap durations and those expected by chance according to our randomization procedure ((obs − exp)/exp). A coordination score of zero represented no coordination while larger integers (positive or negative) represented increasing coordination. Negative values corresponded to coordinated–alternating while positive values corresponded to coordinated–overlapping duets [44]. We calculated a duet coordination score for each duet and then averaged coordination scores by species. We measured duet consistency as the 95% confidence interval surrounding the mean coordination score across each species. Species without duets (soloing species) were assigned a score of zero, indicating that they had uncoordinated singing. We calculated coordination scores for both phrases and notes within duets, with similar results. Here, we present the results for male–female note placement within duets. These species-level mean note coordination and consistency scores were used for subsequent phylogenetic comparative analyses (electronic supplementary material, table S2).

(d) Natural history data collection
(i) Breeding season length and clutch size

We gathered information on breeding season lengths and clutch sizes from *Handbook of the birds of the world* [45]. Breeding season length was counted as the total number of months listed for which each species has been observed breeding, and clutch size was counted as the number of eggs per clutch reported for each species. When more than one breeding season length or clutch size was provided for different parts of a species range, we took the median value for breeding season length and the average value for clutch size from the values listed.

(ii) Conspecific abundance

We calculated species prevalence as a proxy for conspecific abundance throughout each species' range using eBird data. Prevalence was calculated as the relative frequency of occurrence on eBird checklists within forested areas. To calculate this, we extracted all complete eBird checklists within each species range from 2010 to 2019 that were less than 5 km in length and 5 h in duration. We used BirdLife International's range polygons to define each species' range. To reduce the impact of spatial and temporal bias in the eBird data, we overlaid an equal area hexagonal grid with 5 km spacing between cell centres and assigned each checklist to a cell. Within each hexagonal cell for each year, we calculated the frequency of detection of the species on eBird checklists, then calculated the mean frequency across all hexagons to get a single prevalence value for each species. We restricted our estimates to hexagons that contained at least 500 m² of forest within a 2 × 2 km neighbourhood around an eBird observation. Forested habitat was determined based on land cover data from the NASA MODIS satellite (product names: MCD12Q1 v. 6).

(iii) Vegetation density

Vegetation density was calculated from Enhanced Vegetation Index (EVI) data extracted from the NASA MODIS satellite (product: MOD12A3 v6). We used EVI over Normalized Difference

Vegetation Index (NDVI), as EVI has higher resolution for habitats with dense vegetation (e.g. rainforest). We calculated both mean EVI and the standard deviation of EVI for each species based on annual 2 km resolution MODIS data. Using the same set of eBird checklists assigned to hexagonal cells as described for calculating species prevalence, we extracted EVI values for the location of each checklist using the MODIS data from the year of the checklist. The annual MODIS EVI data for 2019 were unavailable at the time of analysis, so 2018 EVI data were used for 2019 eBird checklists. To reduce spatial and temporal bias, for each hexagonal cell and each year, we calculated the mean EVI for checklist locations in which the species occurred. Finally, we calculated the mean and standard deviation of EVI across all hexagonal cells and years. We used the current best practices for both prevalence and vegetation density estimates [46].

(e) Phylogenetic reconstruction

We examined evolutionary patterns of duet coordination and consistency using the phytools and geiger packages in R [43,47,48]. We evaluated the robustness of ancestral state reconstructions of duet coordination and consistency by comparing several reconstruction methods, including squared-change parsimony, maximum likelihood (ML) and Bayesian reconstructions (using the *ace* and *anc.bayes* functions in phytools) [47]. We also assessed the fit of alternative underlying evolutionary models to evolutionary patterns of duet coordination and consistency, including Brownian motion (BM), Ornstein–Uhlenbeck, early burst models and Pagel's lambda, kappa and delta models with the fitContinuous function in geiger [48]. This verified that our data met assumptions of most reconstruction methods of an underlying distribution similar to BM (electronic supplementary material, table S3). Reconstructed ancestral states were visualized and compared among methods using both the *phenogram* function in phytools to focus on the state of the root node and the *contMap* heat map function in phytools to compare how states changed over time [47]. All reconstruction and comparative analyses were conducted on a phylogenetic tree for the wrens with good coverage for species in this study [38].

(f) Phylogenetic correlative analyses

We compared evolutionary patterns of duet coordination and consistency to natural history predictors (breeding season length, clutch size, conspecific abundance and vegetation density). Comparative correlational analyses were conducted using phylogenetically controlled Bayesian models with the R package MCMCglmm [49]. For all models, we used a non-informative, parameter-extended prior to improve mixing with R-structure $V = 1$, nu = 0.002 and G-structure $V = 1$, nu = 1, alpha.mu = 0, and alpha.$V = 25^2$. We also ran all comparative analyses as phylogenetic least squares (PGLS), including selection and evaluation of the best underlying model of evolution (electronic supplementary material). Duet coordination, consistency and all predictor variables were compared visually on the phylogeny with the *contMap* function using the *ML* reconstruction in phytools [47]. The results were qualitatively equivalent for both sets of correlative analyses. Phylogenetic signal was calculated using both Blomberg's K [50] and Pagel's Lambda [51] with the phylosig function in phytools [47].

3. Results

Most Neotropical wren species produced duets classified as coordinated–alternating, meaning that males and females alternated their song phrases and overlapped each other significantly less than expected by chance (figure 2; electronic

supplementary material, table S2). However, there was considerable variation among species in the extent to which song phrases overlapped within duets. Interestingly, species with the most coordinated duets were not each other's closest relatives (figure 2). The highest levels of coordination in alternating duets (i.e. the most negative coordination scores) were observed in *Cantorchilus modestus*, *Cantorchilus leucotis*, *Cantorchilus nigricapillus* and *Pheugopedius euophrys*. The lowest levels of coordination (i.e. scores closest to zero) were observed in *Thryothorus albinucha*, *Cantorchilus leucopogon*, *Thryophilus pleurostictus*, *Thryophilus sinaloa* and *Thryophilus rufalbus*, all of which are solo singers except *T. rufalbus*. Solos were all scored as uncoordinated, but *T. rufalbus* duets were just as uncoordinated on average as the songs of soloist species (figure 2; electronic supplementary material, table S2).

Most Neotropical wrens also showed high levels of within-species consistency in duet coordination, indicated by low confidence intervals for the coordination scores (figure 2; electronic supplementary material, figure S1 and table S2). As with the coordination scores, consistency varied across species, with *Cantorchilus guarayanus* displaying notably inconsistent duets.

Ancestral state reconstruction indicated that the common ancestor of these Neotropical wrens probably produced duets that were slightly coordinated and alternating (figure 2; electronic supplementary material, figure S2). However, the strength of this result varied with reconstruction method: ML and Bayesian approaches reconstructed the ancestor as having an intermediately coordinated duetting ancestor, whereas parsimony suggested an uncoordinated or non-duetting ancestor (figure 2; electronic supplementary material, figure S2 and table S3). As ML and Bayesian approaches take branch lengths into account and maximize parameters to reflect the distribution of the data, we have greater confidence in these approaches.

Phylogenetically controlled comparative analyses revealed that duet coordination and duet consistency were both positively correlated with breeding season length ($p = 0.019$ and $p = 0.032$, respectively; table 1), with more coordinated and consistent duets occurring in species with longer breeding seasons (figure 3). However, neither coordination nor consistency were correlated with other natural history traits, including clutch size ($p = 0.328$ and 0.269), species abundance ($p = 0.575$ and 0.826), vegetation density ($p = 0.886$ and 0.730) or variation in vegetation density ($p = 0.639$ and 0.333; table 1; electronic supplementary material, figure S3). These results were also supported by phylogenetic least squares using a model selection approach (electronic supplementary material, tables S5 and S6) and were robust to removal of species without duets (electronic supplementary material, table S7).

Phylogenetic signal for duet coordination was high ($K = 1.23$, lambda = 1.28), whereas phylogenetic signal for duet consistency was relatively low ($K = 0.77$, lambda < 0.001). This is consistent with findings by Mann *et al.* [7] that there are similarities in duet coordination and overlap within genera.

4. Discussion

While most Neotropical wrens we examined produced coordinated and alternating duets, the degrees of coordination, consistency and amount of overlap of song phrases varied considerably among species. Some species produced duets
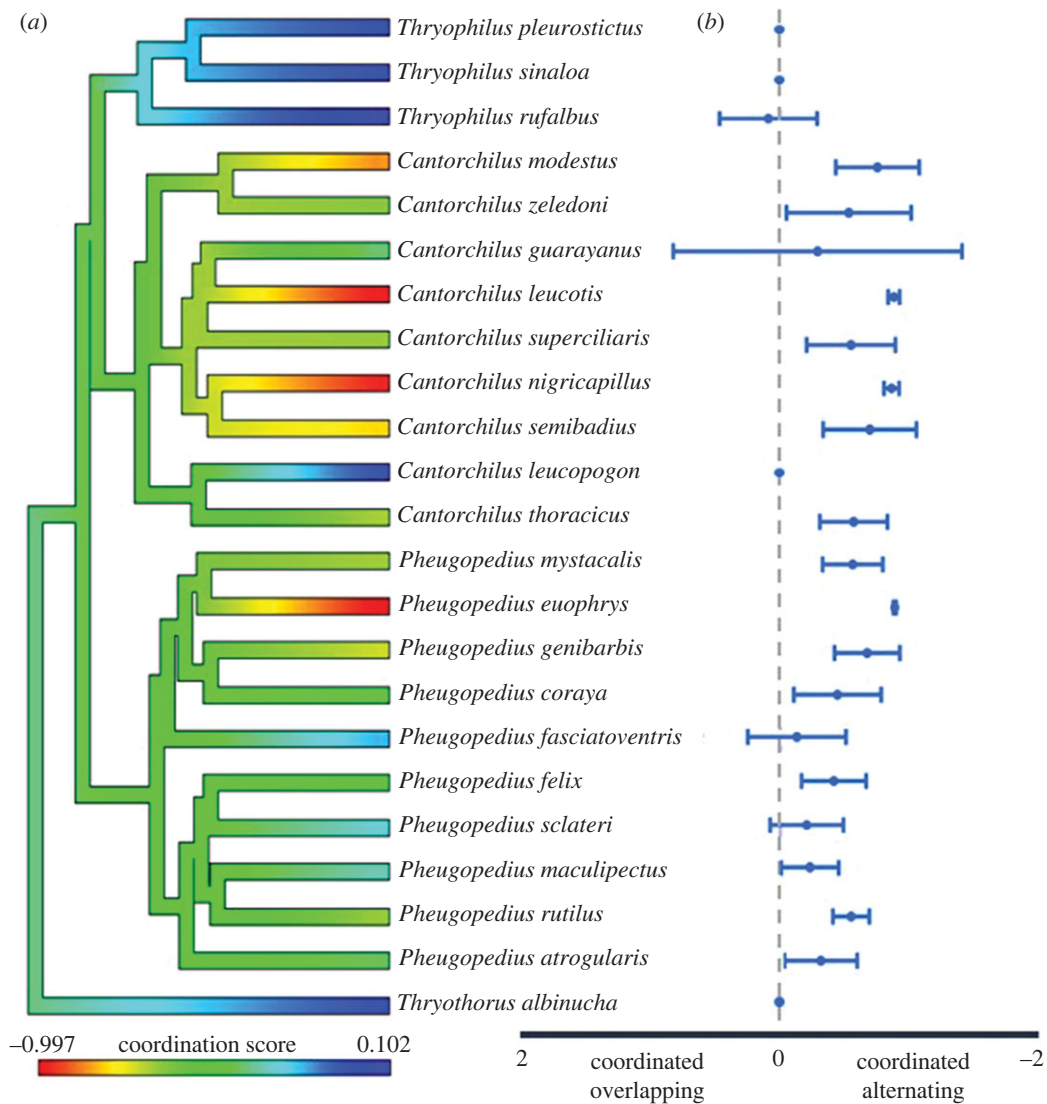
Figure 2. (*a*) Maximum-likelihood ancestral state reconstruction of duet coordination and (*b*) means and standard deviations for duet coordination scores by species. Coordination scores closer to −1 (red) indicate coordinated duets in which partners alternate their vocalizations and values closer to 0 (blue) indicate uncoordinated duets. *Thryophilus pleurostictus, Thryophilus sinaloa, Cantorchilus leucopogon* and *Thryothorus albinucha* are primarily soloists and, therefore, were assigned coordination scores of zero. Phylogeny based on [38]. (Online version in colour.)

with exceptionally high precision and consistent coordination, and the species with the most coordinated duets (e.g. *C. leucotis, C. nigricapillus,* and *P. euophrys*) were not each other's closest relatives. In fact, we demonstrated that exceptionally coordinated duets have independently evolved multiple times within Neotropical wrens and appear to be a derived state exhibited by only some species. Of all the natural history features we examined, only breeding season length explained a species' degree of duet coordination and consistency. This result suggests that highly coordinated duets might benefit species that breed over longer parts of the year, perhaps by helping them coordinate breeding activities or communicate strength of the pair bond.

(a) Duet coordination and breeding season length

At least two hypotheses have been proposed for how duet coordination could reflect pair bond strength: signalling commitment to a partner and signalling pair coalition to conspecifics [12–14]. The rationale for both hypotheses is that temporal coordination of duets is challenging to coordinate, requiring individuals to continually listen to their partner's

song and modify their own output [32,52,53]. The addition of male and female-specific song phrases [7], as well as pair-specific answering rules (duet codes) [54], both commonly found in Neotropical wrens, create further challenges for coordinating duet songs [55]. Recent evidence also indicates that duet coordination is learned over time in Neotropical wrens [33,55–57]. By taking the time and effort to learn to coordinate songs, both partners are signalling their commitment or willingness to invest in territory defence, reproductive efforts and parental care [12,22,32,55].

Longer breeding seasons could also promote commitment to a pair bond if reproductive success increases with pair bond length and throughout the season. In some monogamous species, reproductive success does increase with subsequent broods and length of the pair bond [58]. While many Neotropical wren species form long-term pair bonds with especially low levels of extra-pair mating [59], pair bond length varies across pairs [29,31,57,60]. In buff-breasted wrens (*C. leucotis*), a species with especially coordinated duets and long breeding seasons, pairs that have bred together show high mate and territory fidelity, whereas individuals newly entering the population are much more likely to divorce and often do so within the
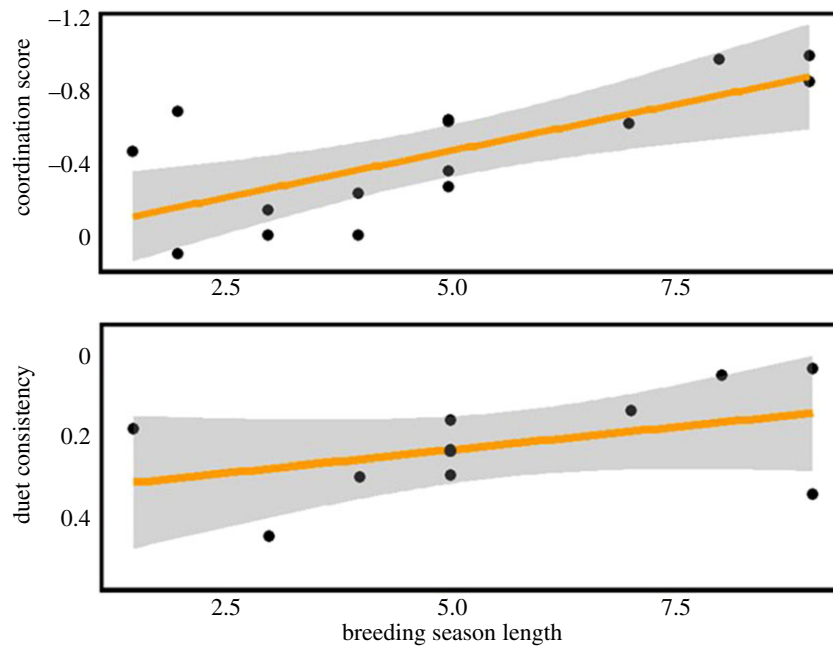
6

royalsocietypublishing.org/journal/rspb   Proc. R. Soc. B **287**: 20202482

Figure 3. Duet coordination and consistency are both greater in species with longer breeding seasons. More negative coordination scores represent species with more strongly coordinated, alternating duets. Species with duet consistency values closer to zero have more consistently coordinated duets (lower 95% confidence intervals). Note that the y-axes of both graphs are flipped to more readily show that coordination and consistency increase with breeding season length. Black points in each plot represent species with coordination and consistency score data. (Online version in colour.)

Table 1. Results from phylogenetically controlled Bayesian models used to evaluate correlated evolution of duet coordination and consistency versus predictor variables. (Values in italics are statistically significant according to a Bayesian p-value < 0.05.)

explanatory variable	predictor variable	posterior mean	lower 95% CI	upper 95% CI	effective sample size	Bayesian p-value
coordination	breeding season length	−0.21	−0.39	−0.04	49 177	*0.021*
	clutch size	0.08	−0.09	0.26	48 988	0.326
	species abundance	−0.04	−0.18	0.11	49 984	0.571
	EVI mean	0.01	−0.13	0.15	49 984	0.900
	EVI s.d.	−0.03	−0.17	0.10	49 984	0.637
consistency	breeding season length	−0.12	−0.23	−0.01	49 984	*0.031*
	clutch size	−0.16	−0.44	0.13	43 541	0.263
	species abundance	0.03	−0.22	0.28	41 342	0.829
	EVI mean	−0.03	−0.24	0.17	49 984	0.724
	EVI s.d.	−0.10	−0.30	0.11	47 518	0.333

first several months of a partnership [61]. There may be true reproductive benefits for an individual to accurately assess its partner's commitment to the pair bond.

Alternatively, the fact that duets are learned and become more coordinated over time could signal to other conspecifics the length or strength of the pair bond and, therefore, the ability of a pair to jointly defend a territory [15,18]. In at least some Neotropical wren species, more coordinated duets are not perceived as more threatening signals [39]; however, it seems plausible that conspecifics could obtain additional information about the quality or strength of a pair from the fine structure of the duets [25–30]. Furthermore, signalling commitment and coalition are not necessarily mutually exclusive functions. Duets are known to be multifunctional, and it seems that duet coordination could

simultaneously benefit pairs and individuals as a signal both within and between pairs [14].

Duet coordination could also be indicative of a pair's ability or willingness to coordinate reproductive activities, including reproductive synchrony [12,14]. Yet, some species duet well outside of the breeding season, suggesting synchronizing reproductive behaviour is not the sole function of duets in those species [62,63]. Nevertheless, both male and female songs are known to stimulate gonadal growth and bring birds into breeding condition in some species [64,65], and female song can be used to coordinate breeding activities such as nest relief and care for young [66–68].

Another alternative is that high levels of duet coordination arise in species with longer breeding seasons because of 'masking avoidance'. Under this scenario, the efforts of each pair

member to ensure that its own song is heard leads to duets with increasing coordination [40,69,70]. This phenomenon could be particularly pronounced in species with longer breeding seasons because these individuals may duet over longer periods of the year and, therefore, achieve greater coordination through enhanced masking avoidance. In addition, species with longer breeding seasons may face more intraspecific and intrasexual competition for resources, creating a heightened need for each pair member to most effectively compete with same or opposite sex intruders. However, we did not find evidence that duet coordination varies with intraspecific competition (e.g. high population densities) and other metrics of intrasexual conflict (high extra-pair paternity or divorce rates [37,40,57]), which seem to be low for these wrens [59,71]. Nevertheless, territory defence appears to be a primary function of duets in general [11,12,69] and our coarse metric for assessing population density could have missed the importance of intraspecific competition on duet structure. Indeed, we think masking avoidance is a compelling and parsimonious hypothesis that could influence duet coordination in these and other duetting species and we encourage more studies that directly test the contributions of masking avoidance and signalling commitment to duet coordination in a range of species [40].

Similarly, Neotropical wren species with longer breeding seasons may have more coordinated duets simply because they have practised their duets over a longer period of the year. Wrens learn to coordinate both duet codes and response timing [33,57]. However, in the few species studied, wrens achieve a high level of duet coordination within the first few days of a new partnership [55,56]. Therefore, it seems unlikely that the additional practise during longer breeding seasons is a primary factor leading to more coordinated duets in species with long breeding seasons. In addition, duets are not tied exclusively to the breeding season for many duetting species, so most wren species would have opportunities to practise duet coordination throughout the year. Lastly, if practising was the only factor influencing duet coordination, we would expect more species-level variation in general, as we presumably sampled individuals of each species that had been together for different lengths of time.

(b) Duet coordination and tropical life history

Prevailing theory and empirical data support duet evolution being tied to natural history traits that are particularly common in the tropics, including long-term monogamy and year-round territoriality [17–20,34,35]; however, to our knowledge, all species in the current study are year-round territorial. Therefore, we suggest that there are selection pressures that select for duet evolution generally (e.g. a sedentary lifestyle) and additional ecological factors that favour particular duet structures or levels of coordination [13,14,16,17].

We found no relationship between duet coordination or consistency versus conspecific abundance or vegetation density. Associations between year-round territoriality and duets have been supported by other studies [17–19], although a global analysis of duet evolution did not find an association with open versus closed habitat [18]. However, it is important to point out that both of these variables were measured relatively coarsely in our analysis compared to the fine scale at which we measured duets. Therefore, we might not have been able to detect biologically relevant patterns owing to the scale at which both of these metrics were calculated.

Thus, it would be worth revisiting these hypotheses in a comparative context with improved resolution for these variables or through field studies that directly test these hypotheses in individual species.

To further tease apart existing hypotheses for the evolution of highly coordinated duets, more natural history knowledge is needed for these wrens and other tropical species, such as territory sizes, conspecific densities, pair bond lengths, number of nesting attempts per year, and resource availability. Breeding season length data could come from one or a few locations for each species [45], and, therefore, could be more variable than estimated here. Moreover, our coordination scores are only a snapshot of Neotropical wren behaviour gathered during a narrow window of time and in specific locations. There may be more variability to these behaviours among all these species; for instance, duets may be more or less coordinated during certain times of year, and this warrants future study incorporating seasonal and geographical variation in duet coordination and structure, particularly for species for which we had small sample sizes (e.g. fawn-breasted wren, *C. guarayanus*). Studies like these will benefit greatly from additional data as we continue to compile natural history data for tropical species from various locations.

(c) Duet structure in Neotropical wrens

Our findings extend the comprehensive descriptions of wren duet form by Mann *et al.* [7] by quantifying levels of coordination in these same species. Mann *et al.* [7] demonstrated that members within each genus—*Cantorchilus*, *Thryophilus*, *Pheugopedius* or *Thryothorus*—tend to sing in a similar style. We found similar patterns in duet coordination levels within genera, with the exception of a few species. For example, three of the four most highly coordinated duets occur within the *Cantorchilus* genus, while *Thryophilus*, *Pheugopedius* and *Thryothorus* were generally less coordinated, with the exception of *P. euophrys*. As in Mann *et al.*'s [7] description of singing styles, the phylogenetic signal we observed for duet coordination was high. Duets by species within the same genus tend to be similarly structured—with *Pheugopedius* notable for their overlapping duets and *Cantorchilus* their introductory phrases and alternating duets—which may also be reflected by the high phylogenetic signal. Along these lines, coordination scores were lower in many *Pheugopedius* species compared to *Cantorchilus*, which is probably owing to the overlap often seen between the beginnings and ends of phrases within *Pheugopedius*. Nevertheless, while phylogenetic signal is clearly important, the species with the most highly coordinated duets were not closely related to each other, indicating that precisely coordinated duets evolved multiple times independently within the Neotropical wrens. Ultimately, our study demonstrates that other ecological and natural history factors—especially breeding season length and the large degree of within-pair commitment that must be necessary in these species—are important factors in the evolution of highly coordinated duets.

Authors' contributions. C.N.T., J.J.P. and K.J.O. conceived of the project. N.I.M. collected and organized the field recordings with help from P.J.B.S. E.L.K. and M.A.M. extracted the acoustic measurements. M.S.-M. and K.G.H. computed the prevalence and vegetation density

References

1. Catchpole CK, Slater PJB. 2008 *Bird song: biological themes and variations*, 2nd edn. Cambridge, UK: Cambridge University Press.

2. Bradbury JW, Vehrencamp SL. 2011 *Principles of animal communication*, 2nd edn. Oxford, UK: Oxford University Press.

3. Tinbergen N. 1951 *The study of instinct*. Oxford, UK: Clarendon Press.

4. Lindsay WR, Houck JT, Giuliano CE, Day LB. 2015 Acrobatic courtship display coevolves with brain size in manakins (Pipridae). *Brain Behav. Evol.* **85**, 29–36. (doi:10.1159/000369244)

5. Farabaugh SM. 1982 The ecological and social significance of duetting. In *Acoustic communication in birds*, vol. 2 (eds DE Kroodsma, EH Miller, H Ouellet), pp. 85–123. New York, NY: Academic Press.

6. Wachtmeister C-A. 2001 Display in monogamous pairs: a review of empirical data and evolutionary explanations. *Anim. Behav.* **61**, 861–868. (doi:10.1006/anbe.2001.1684)

7. Mann NI, Dingess KA, Barker FK, Graves JA, Slater PJB. 2009 A comparative study of song form and duetting in neotropical *Thryothorus* wrens. *Behaviour* **146**, 1–43. (doi:10.1163/156853908X390913)

8. Mann NI, Marshall-Ball L, Slater PJB. 2003 The complex song duet of the plain wren. *Condor* **105**, 672–682. (doi:10.1093/condor/105.4.672)

9. Mann NI, Dingess KA, Slater PJB. 2006 Antiphonal four-part synchronized chorusing in a neotropical wren. *Biol. Lett.* **2**, 1–4. (doi:10.1098/rsbl.2005.0373)

10. Cuthbert JL, Mennill DJ. 2007 The duetting behavior of Pacific coast plain wrens. *Condor* **109**, 686–692. (doi:10.1093/condor/109.3.686)

11. Thorpe WH. 1972 *Duetting and antiphonal song in birds: its extent and significance. Brill Archive* **18**.

12. Hall ML. 2004 A review of hypotheses for the functions of avian duetting. *Behav. Ecol. Sociobiol.* **55**, 415–430. (doi:10.1007/s00265-003-0741-x)

13. Hall ML. 2009 Chapter 3: a review of vocal duetting in birds. In *Advances in the study of behavior* (eds M Naguib, K Zuberbuumlhler, NS Clayton, VM Janik), pp. 67–121. New York, NY: Academic Press.

14. Dahlin CR, Benedict L. 2014 Angry birds need not apply: a perspective on the flexible form and multifunctionality of avian vocal duets. *Ethology* **120**, 1–10. (doi:10.1111/eth.12182)

15. Hall ML, Magrath RD. 2007 Temporal coordination signals coalition quality. *Curr. Biol.* **17**, R406–R407. (doi:10.1016/j.cub.2007.04.022)

16. Odom KJ, Omland KE, Price JJ. 2015 Differentiating the evolution of female song and male–female duets in the New World blackbirds: can tropical natural history traits explain duet evolution? *Evolution* **69**, 839–847. (doi:10.1111/evo.12588)

17. Mikula P, Tószögyová A, Hořák D, Petrusková T, Storch D, Albrecht T. 2019 Female solo song and duetting are associated with different territoriality in songbirds. *Behav. Ecol.* **31**, 322–329. (doi:10.1093/beheco/arz193)

18. Tobias JA, Sheard C, Seddon N, Meade A, Cotton AJ, Nakagawa S. 2016 Territoriality, social bonds, and the evolution of communal signaling in birds. *Front. Ecol. Evol.* **4**, 74. (doi:10.3389/fevo.2016.00074)

19. Logue DM, Hall ML. 2014 Migration and the evolution of duetting in songbirds. *Proc. R. Soc. B* **281**, 20140103. (doi:10.1098/rspb.2014.0103)

20. Benedict L. 2008 Occurrence and life history correlates of vocal duetting in North American passerines. *J. Avian Biol.* **39**, 57–65. (doi:10.1111/j.0908-8857.2008.04103.x)

21. Mennill DJ, Vehrencamp SL. 2008 Context-dependent functions of avian duets revealed by microphone-array recordings and multispeaker playback. *Curr. Biol.* **18**, 1314–1319. (doi:10.1016/j.cub.2008.07.073)

22. Wickler W. 1980 Vocal dueting and the pair bond. *Z. Tierpsychol.* **52**, 201–209. (doi:10.1111/j.1439-0310.1980.tb00711.x)

23. Stutchbury BJ, Morton ES. 2001 *Behavioral ecology of tropical birds*. New York, NY: Academic Press.

24. Benedict L. 2010 California towhee vocal duets are multi-functional signals for multiple receivers. *Behaviour* **147**, 953. (doi:10.1163/000579510X498633)

25. Mennill DJ. 2006 Aggressive responses of male and female rufous-and-white wrens to stereo duet playback. *Anim. Behav.* **71**, 219–226. (doi:10.1016/j.anbehav.2005.05.006)

26. Templeton CN, Rivera-Cáceres KD, Mann NI, Slater PJB. 2011 Song duets function primarily as cooperative displays in pairs of happy wrens. *Anim. Behav.* **82**, 1399–1407. (doi:10.1016/j.anbehav.2011.09.024)

27. Logue DM. 2007 Duetting in space: a radio-telemetry study of the black-bellied wren. *Proc. R. Soc. B* **274**, 3005–3010. (doi:10.1098/rspb.2007.1005)

28. Marshall-Ball L, Mann N, Slater PJB. 2006 Multiple functions to duet singing: hidden conflicts and apparent cooperation. *Anim. Behav.* **71**, 823–831. (doi:10.1016/j.anbehav.2005.05.021)

29. Levin RN. 1996 Song behaviour and reproductive strategies in a duetting wren, *Thryothorus nigricapillus*: I. Removal experiments. *Anim. Behav.* **52**, 1093–1106. (doi:10.1006/anbe.1996.0257)

30. Levin RN. 1996 Song behaviour and reproductive strategies in a duetting wren, *Thryothorus nigricapillus*: II. Playback experiments. *Anim. Behav.* **52**, 1107–1117. (doi:10.1006/anbe.1996.0258)

31. Gill SA. 2012 Testing hypotheses for the evolution of long-term monogamy in neotropical buff-breasted wrens (*Cantorchilus leucotis*). *J. Ornithol.* **153**, 125–134. (doi:10.1007/s10336-011-0804-0)

32. Rivera-Cáceres KD. 2015 Plain wrens *Cantorchilus modestus zeledoni* adjust their singing tempo based on self and partner's cues to perform precisely coordinated duets. *J. Avian Biol.* **46**, 361–368. (doi:10.1111/jav.00575)

33. Rivera-Cáceres KD, Quirós-Guerrero E, Araya-Salas M, Searcy WA. 2016 Neotropical wrens learn new duet rules as adults. *Proc. R. Soc. B* **283**, 20161819, (doi:10.1098/rspb.2016.1819)

34. Slater PJB, Mann NI. 2004 Why do the females of many bird species sing in the tropics? *J. Avian*

Biol. **35**, 289–294. (doi:10.1111/j.0908-8857.2004.03392.x)

35. Price JJ. 2009 Evolution and life-history correlates of female song in the New World blackbirds. *Behav. Ecol.* **20**, 967–977. (doi:10.1093/beheco/arp085)

36. Griebeler EM, Caprano T, Böhning-Gaese K. 2010 Evolution of avian clutch size along latitudinal gradients: do seasonality, nest predation or breeding season length matter? *J. Evol. Biol.* **23**, 888–901. (doi:10.1111/j.1420-9101.2010.01958.x)

37. Griffith SC. 2019 Cooperation and coordination in socially monogamous birds: moving away from a focus on sexual conflict. *Front. Ecol. Evol.* **7**, 455. (doi:10.3389/fevo)

38. Mann NI, Barker FK, Graves JA, Dingess-Mann KA, Slater PJB. 2006 Molecular data delineate four genera of 'Thryothorus' wrens. *Mol. Phylogenet. Evol.* **40**, 750–759. (doi:10.1016/j.ympev.2006.04.014)

39. Kovach KA, Hall ML, Vehrencamp SL, Mennill DJ. 2014 Timing isn't everything: responses of tropical wrens to coordinated duets, uncoordinated duets and alternating solos. *Anim. Behav.* **95**, 101–109. (doi:10.1016/j.anbehav.2014.06.012)

40. Tobias JA, Seddon N. 2009 Signal jamming mediates sexual conflict in a duetting bird. *Curr. Biol.* **19**, 577–582. (doi:10.1016/j.cub.2009.02.036)

41. Bioacoustics Research Program. 2017 *Raven Pro: interactive sound analysis software*, version 1.5. Ithaca, NY: Cornell Laboratory of Ornithology.

42. Araya-Salas M, Smith-Vidaurre G. 2017 warbleR: an r package to streamline analysis of animal acoustic signals. *Methods Ecol. Evol.* **8**, 184–191. (doi:10.1111/2041-210x.12624)

43. R Core Team. 2019 *R: a language and environment for statistical computing*. Vienna, Austria: R Foundation for Statistical Computing.

44. Araya-Salas M, Wojczulanis-Jakubas K, Phillips EM, Mennill DJ, Wright TF. 2017 To overlap or not to overlap: context-dependent coordinated singing in lekking long-billed hermits. *Anim. Behav.* **124**, 57–64. (doi:10.1016/j.anbehav.2016.12.003)

45. del Hoyo J, Elliott A, Sargatal J, Christie DA, de Juana E. 2010 *Handbook of the birds of the world alive*. Barcelona, Spain: Lynx Edicions.

46. Strimas-Mackey M, Hochachka WM, Ruiz-Gutierrez V, Robinson OJ, Miller ET, Auer T, Kelling S, Fink D, Johnston A. 2020 *Best practices for using eBird data version 1.0*. Ithaca, NY: Cornell Laboratory of Ornithology.

47. Revell LJ. 2012 Phytools: an R package for phylogenetic comparative biology (and other things). *Methods Ecol. Evol.* **3**, 217–223. (doi:10.1111/j.2041-210X.2011.00169.x)

48. Pennell MW, Eastman JM, Slater GJ, Brown JW, Uyeda JC, FitzJohn RG, Alfaro ME, Harmon LJ. 2014 geiger v2.0: an expanded suite of methods for fitting macroevolutionary models to phylogenetic trees. *Bioinformatics* **30**, 2216–2218. (doi:10.1093/bioinformatics/btu181)

49. Hadfield JD. 2010 MCMC methods for multi-response generalized linear mixed models: the MCMCglmm R package. *J. Stat. Softw.* **33**, 1–22. (doi:10.18637/jss.v033.i02)

50. Blomberg SP, Garland Jr T, Ives AR. 2003 Testing for phylogenetic signal in comparative data: behavioral traits are more labile. *Evolution* **57**, 717–745. (doi:10.1111/j.0014-3820.2003.tb00285.x).

51. Pagel M. 1999 Inferring the historical patterns of biological evolution. *Nature* **401**, 877–884. (doi:10.1038/44766)

52. Logue DM, Chalmers C, Gowland AH. 2008 The behavioural mechanisms underlying temporal coordination in black-bellied wren duets. *Anim. Behav.* **75**, 1803–1808. (doi:10.1016/j.anbehav.2007.10.036)

53. Templeton CN, Mann NI, Ríos-Chelén AA, Quiros-Guerrero E, Macías Garcia C, Slater PJB. 2013 An experimental study of duet integration in the happy wren, *Pheugopedius felix. Anim. Behav.* **86**, 821–827. (doi:10.1016/j.anbehav.2013.07.022)

54. Logue DM. 2006 The duet code of the female black-bellied wren. *Condor* **108**, 326–335. (doi:10.1093/condor/108.2.326)

55. Rivera-Cáceres KD, Templeton CN. 2019 A duetting perspective on avian song learning. *Behav. Processes* **163**, 71–80. (doi:10.1016/j.beproc.2017.12.007)

56. Rivera-Cáceres KD, Quirós-Guerrero E, Araya-Salas M, Templeton CN, Searcy WA. 2018 Early development of vocal interaction rules in a duetting songbird. *R. Soc. Open Sci.* **5**, 171791. (doi:10.1098/rsos.171791)

57. Quirós-Guerrero E, Janeiro MJ, Cresswell W, Templeton CN. 2018 Evidence of repertoire sharing and stability despite a high turnover rate in a duetting neotropical wren. *J. Avian Biol.* (doi:10.1111/jav.02382)

58. Poirier NE, Whittingham LA, Dunn PO. 2004 Males achieve greater reproductive success through multiple broods than through extrapair mating in house wrens. *Anim. Behav.* **67**, 1109–1116. (doi:10.1016/j.anbehav.2003.06.020)

59. Douglas SB, Heath DD, Mennill DJ. 2012 Low levels of extra-pair paternity in a neotropical duetting songbird, the rufous-and-white wren (*Thryothorus rufalbus*). *Condor* **114**, 393–400. (doi:10.1525/cond.2012.110028)

60. Hennin HL, Barker NKS, Bradley DW, Mennill DJ. 2009 Bachelor and paired male rufous-and-white wrens use different singing strategies. *Behav. Ecol.*

Sociobiol. **64**, 151–159. (doi:10.1007/s00265-009-0832-4)

61. Gill SA, Stutchbury BJM. 2006 Long-term mate and territory fidelity in neotropical buff-breasted wrens (*Thryothorus leucotis*). *Behav. Ecol. Sociobiol.* **61**, 245–253. (doi:10.1007/s00265-006-0255-4)

62. Topp SM, Mennill DJ. 2008 Seasonal variation in the duetting behaviour of rufous-and-white wrens (*Thryothorus rufalbus*). *Behav. Ecol. Sociobiol.* **62**, 1107–1117. (doi:10.1007/s00265-007-0538-4)

63. Odom KJ, Omland KE, McCaffrey DR, Monroe MK, Christhilf JL, Roberts NS, Logue DM. 2016 Typical males and unconventional females: songs and singing behaviors of a tropical, duetting oriole in the breeding and non-breeding season. *Front. Ecol. Evol.* **4**, 14. (doi:10.3389/fevo.2016.00014)

64. Kroodsma DE. 1976 Reproductive development in a female songbird: differential stimulation by quality of male song. *Science* **192**, 574–575. (doi:10.1126/science.192.4239.574)

65. Cheng M-F. 1992 For whom does the female dove coo? A case for the role of vocal self-stimulation. *Anim. Behav.* **43**, 1035–1044. (doi:10.1016/S0003-3472(06)80016-3)

66. Ritchison G. 1983 The function of singing in female black-headed grosbeaks (*Pheucticus melanocephalus*): family-group maintenance. *Auk* **100**, 105–116. (doi:10.1093/auk/100.1.105)

67. Halkin SL. 1997 Nest-vicinity song exchanges may coordinate biparental care of northern cardinals. *Anim. Behav.* **54**, 189–198. (doi:10.1006/anbe.1996.0415)

68. Rose EM, Coss DA, Haines CD, Danquah SA, Studds CE, Omland KE. 2019 Why do females sing?—pair communication and other song functions in eastern bluebirds. *Behav. Ecol.* **30**, 1653–1661. (doi:10.1093/beheco/arz130)

69. Lohr B, Wright TF, Dooling RJ. 2003 Detection and discrimination of natural calls in masking noise by birds: estimating the active space signal. *Anim. Behav.* **65**, 763–777. (doi:10.1006/anbe.2003.2093)

70. Brumm H, Slater PJB. 2007 Animal communication: timing counts. *Curr. Biol.* **17**, R521–R523. (doi:10.1016/j.cub.2007.04.053)

71. Cramer ER, Hall ML, Kort SRD, Lovette IJ, Vehrencamp SL. 2011 Infrequent extra-pair paternity in the banded wren, a synchronously breeding tropical passerine. *Condor* **113**, 637–645. (doi:10.1525/cond.2011.100233)

72. Keenan EL *et al.* 2020 Data from: Breeding season length predicts duet coordination and consistency in Neotropical wrens (Troglodytidae). Dryad Digital Repository. (https://doi.org/10.5061/dryad.pk0p2ngm6)