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Vocal and territorial behaviour of Puff-throated Babbler, a South-Asian duetting passerine

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Running title: Vocal and territorial behaviour of Puff-throated Babbler

Abstract

Territorial songbirds use singing as an interactive social signal during territorial interactions. Considerable information has been accumulated about the communication of passerine birds in the context of territorial competition. Most of such data, however, were gathered in the northern temperate zone. Only a few studies have been conducted in the tropical zone. In this study, we describe for the first time the vocal and territorial behaviour of the Puff-throated Babbler *Pellorneum ruficeps*, a South Asian tropical resident bird. We recorded spontaneous vocalization simulated territorial intrusion by broadcasting different vocalizations at territories. We played back different types of conspecific vocalizations to territorial males: (1) male simple song, (2) male complex song, and (3) duet. Depending on context, there were three types of male vocalizations: simple song, complex song, and subsong. While singing spontaneously, males produced a simple song of 2–3 elements, repeated at a rate of 10–12 songs per minute. Males could respond to playback with complex songs, which are much more elaborate and longer (up to 5–10 seconds) vocalization emitted at a rate of approximately one song per minute. Subsong is the most commonly heard vocalization during playback-simulated territorial intrusion. Females could acoustically respond to playback along with the males, forming a duet. While duetting, females produced a sequence of identical broadband elements (trills) overlapping the male complex song. In response to simple song playback, males sang, approached the loudspeaker, and flew around it. Males reacted much more strongly when presented with the playback of complex songs. They sang more complex songs, performed more flyovers, and produced subsongs. Therefore, complex songs are an aggressive signal used in the context of territorial competition. We observed male-female duets in response to complex song playback only. Males sang more actively and performed more flyovers in response to duets than to complex songs. We thus assumed that the duet is a more aggressive signal than the complex song.

Keywords songbirds, behaviour, communication, male-female duet

INTRODUCTION

Birdsong is an outstanding system that has proven to be an excellent model for addressing the classical questions raised in animal behaviour (Todt & Naguib 2000). The song of passerine birds serves multiple functions, with the main ones being attracting a mate and defending territory (Catchpole & Slater 2008). In particular, territorial songbirds use singing as an interactive social signal during territorial interactions. In this context, males vary the type and timing of their songs depending on their motivation and with respect to the opponent's behaviour (Todt & Naguib 2000).

In the past half-century, considerable information has been accumulated about the communication of passerine birds in the context of territorial competition. It has been found that birds can use several different mechanisms for aggressive signalling. Song matching, song overlapping, song-type switching, song rate, and the usage of specific songs or calls have all been proposed as avian aggressive signals (Todt & Naguib 2000, Botero & Vehrencamp 2007, Catchpole & Slater 2008, Searcy & Beecher 2009). However, much of this data was gathered in the northern temperate zone (e.g., Bremond 1968, Kramer et al. 1985, Searcy et al. 2000, Ballentine 2009, Petrusková et al. 2014, Opaev et al. 2019, Vaytina & Shitikov 2019, Zsebők et al. 2021). Only a few studies have been conducted in the tropical zone (Molles & Vehrencamp 2001, Grafe et al. 2004, Diniz et al. 2018, Opaev et al. 2021).

Nevertheless, it is known that the vocal behaviour of tropical birds has some peculiarities, apparently caused by the fact that several life history traits of tropical birds differ from those of temperate birds. The amplitude of seasonal fluctuations is one of the main factors driving these differences (Wingfield et al. 1992). Life history stages in birds from higher latitudes are controlled by rigid seasonal processes, while animals from lower latitudes use a rather flexible physiological control of life history to cope with low seasonal variability (Hau 2001). Therefore, many temperate species face common factors imposed by a short breeding season, including a high level of male-male competition, an intense and rather short period of social mate choice, and a sudden rush of nesting and egg-laying activity among females. By contrast, most tropical birds are not as time-restricted in establishing territories and finding mates. They typically have year-round access to mates and territories (Stutchbury & Morton 2008). That is why tropical bird communities are relatively stable systems (MacArthur 1972). Additionally, small clutch sizes, high nest predation, several breeding attempts per year, long developmental periods, and extended parental care are typical for tropical birds (Martin 1996).

Apparently, there are two main differences in acoustic behaviour between tropical and northern temperate birds. First, many temperate latitude species, such as thrushes or chats, sing during the breeding season, whereas tropical species, such as bulbuls or babblers, sing throughout the year (Kumar 2003, Fedy & Stutchbury 2005). Year-round singing can be associated with year-round territoriality (Mathevon et al. 2008). Secondly, female song is rather common in the tropics but rare in the temperate zone (Fedy & Stutchbury 2005, Mennill 2011). In many tropical bird species, both sexes sing and actively participate in territory defence (Fedy & Stutchbury 2005, Rivera-Cáceres & Templeton 2019). In those cases, duet singing can be observed, in which mated pairs sing temporally coordinated songs (Ręk & Magrath 2020). For example, during simulated territory intrusion in the Tropical boubou *Laniarius aethiopicus*, duets are initiated by both sexes, with strict sex-specific roles maintained within the duet (Grafe et al. 2004).

Avian duets occur as acoustic representations where two birds coordinate their songs with a degree of temporal precision (Farabaugh 1982). Avian duetting occurs in over 400 species, representing 40% of bird families.

Duets vary in form from loosely overlapping songs to highly coordinated vocalizations (Hall 2009). Duets are used in joint territorial defence and mutual mate guarding (Grafe et al. 2004). In this respect, duets are functionally similar to male territorial songs (Wickler 1976). At the same time, duets can be a stronger and/or more threatening signal than male solo songs. For example, in Magpie-larks *Grallina melanoleuca*, males initiated more vocalizations in response to the playback of duets than playback of male solos (Hall 2000).

In this study, we describe for the first time the vocal behaviour of the Puff-throated Babbler *Pellorneum ruficeps*, a South Asian tropical passerine bird species. We analyzed the spontaneous singing of males of this species, as well as vocalizations in an experimentally simulated territorial competition context.

MATERIALS AND METHODS

Study site

The study was conducted in Nam Cat Tien National Park (Dong Nai Province, 11°30′ N, 107°20′ E), which is located in the southern part of Vietnam, approximately 130 km northeast of Ho Chi Minh City. The main landscape of this locality is evergreen tropical forest. Most of the forest is of secondary origin. The forest experiences a tropical monsoon climate with two distinct seasons: a rainy season from April to November and a dry season from November to April. The medium average temperature under the forest canopy was around 30°C in March–April. The average forest canopy closure varies from 95.7% (in April) to 98.5% (in July) (Opaev et al. 2021). The dominant tree species forming the forest canopy are members of *Lythraceae*, *Tetramelaceae*, *Moraceae*, *Dipterocarpaceae*, and *Fabaceae*. The undergrowth is 1–3 m in height and includes young trees and *Licuala* and *Calamus* palms. Grass cover is low or absent. The study plot had an area of approximately 27 km^2.

We collected data from mid-March to early June 2021–22, during the Puff-throated Babblers' breeding season (Whistler 1949). Preliminary observations and recordings were conducted in March-May 2020. We performed playback experiments in the morning hours from approximately 6:00 a.m. to 12:00 p.m. under good weather conditions. We conducted no more than 2–3 experiments per day.

Study species

The Puff-throated Babbler is a non-migratory bird species, and it is a common resident breeder in the forests of tropical Asia. Puff-throated Babblers inhabit scrub and moist forests, where birds forage on the forest floor, turning over leaf litter to find their prey while usually staying low in the undergrowth (Thinh et al. 2012; our observations). The breeding season is from March to May, though second broods may be found until August (Whistler & Hugh 1949). The sheltered nest is placed on the ground, protected by a stone or a bush. The nest is round, composed of leaves and grass, and slightly lined with moss roots with an entrance at one side (Betham 1903, Whistler & Hugh 1949). The Puff-throated Babbler is a small olive-brown bird, whitish below, with a rufous cap and heavily streaked breast. There is no sexual dimorphism. Males sing in the morning from the ground, stones, fallen deadwood, or bushes low above the ground (our observations). This is a monogamous species, and paired birds probably spend a lot of time together. These birds can participate in territorial conflicts together (our observations).

The Puff-throated Babbler was assessed for The IUCN Red List of Threatened Species in 2016 and (listed as Least Concern).

Playback stimuli

There are two song types in the repertoire of Puff-throated Babbler males: simple songs and complex songs, which differ completely in their time and frequency parameters and usage (see Results for details). In particular, we recorded complex songs in response to playback only. Females can join their mate vocally while he produces a complex song, forming a male-female duet.

The songs used to prepare playback stimuli were recorded in the same study area one or two years before this study. We used three types of stimuli: simple songs (1), complex songs (2), and duets (3These types differ in acoustic parameters, including song rate, complexity, frequencies, etc. They also differ in their usage, as males usually produce simple songs while singing spontaneously, and complex songs in playback-stimulated territorial intrusion contexts. To prepare simple song stimuli, we used spontaneous recordings of males. By contrast, both complex song and duet stimuli were prepared from recordings obtained during playback presentations. Each simple song stimulus consisted of 50 songs of approximately 0.8 s each and lasted for 5 min (i.e., had a rate of 10 songs per min, which is a typical song rate for Puff-throated Babblers). Each complex song and duet stimulus consisted of 20 songs and lasted for 5 min (i.e., had a rate of 4 songs per min). For complex songs and duets, we simulated a typical (median) song length of 5 s in both types of stimuli. There was one song type in each stimulus taken from one male. In total, we used recordings of 6 males to prepare song stimuli. Songs used to prepare all types of stimuli were taken from our recordings of playback experiments conducted in 2020. In that year, we performed six preliminary playback experiments using a single simple song stimulus. We used recordings obtained in sites at a distance of 0.1–1 km from each other to prepare playback stimuli.

Playback experiments

Before each experiment, we selected a male actively singing spontaneously when no other males were nearby. A loudspeaker was placed within 10–30 m of the focal male. To analyze the aggressive response, we additionally used a dummy made of polymer clay and painted with acrylic. We positioned this dummy near the loudspeaker.

Our preliminary study of 2020 showed that birds responded much more aggressively to the playback of complex songs. Therefore, given that the purpose was to determine if a complex song provoked possible attacks and caused a stronger response, a simple song was always played first in each experiment, followed by a complex song.

Each experiment consisted of two subsequent parts without interruption: part one and part two. Each first part of the experiment consisted of three stages: (1) recording of the spontaneous singing of a male before the start of playback (5 min), (2) recording of its singing during playback (5 min), and (3) post-playback recording (5 min). The second part of the experiment consisted of two stages: (4) recording of male singing during playback (5 min), and (5) post-playback recording (5 min). The total duration of the experiment was 25 minutes. Simple song playback stimuli were used in the first part of the experiment, and complex song or duet stimuli were used in the second part. To avoid pseudoreplication (Kroodsma 1989, Kroodsma et al. 2001), we used several versions of each stimulus type: 4 simple song stimuli, 3 complex song stimuli, and 3 duet stimuli. In each experiment, we randomly chose what simple/complex song or duet stimulus would be played back.

The behaviour of focal males was observed during playback. As a measure of aggressive response, we counted the number of flights (males flew for more than 1 m within 10 m of the loudspeaker and dummy) during the playback presentation. The observer was standing about 15 m from the loudspeaker. After the onset of the playback, some males stopped singing and approached the loudspeaker and dummy silently. Therefore, we measured the time lag as the time interval between the onset of playback and the male's first song (hereafter 'acoustic time lag'). We also measured the 'behaviour al time lag', that is, the time interval between the onset of playback and the male's first flight.

In total, we performed 35 experiments. We used complex song stimuli in 18 experiments, and duet stimuli in 17 experiments. Recordings of some males were removed from analysis due to their low quality.

Songs were recorded using a Tascam DR-10X digital recorder equipped with either a Sennheiser ME66-K6 or ME67-K6 microphone.

Analysis of playback experiments

For sound visualization and analysis, we used Raven Lite version 2.0.1 with fast Fourier transform size = 256, and a Hanning window type. First, we determined the repertoire of song types in each male. Then, song bouts recorded during spontaneous singing, during and just after playback presentation were processed separately. The following parameters were calculated: (1) median song length; (2) median pause length between songs; (3) song rate (per 5 min); (4) number of simple songs; (5) number of complex songs; (6) presence/absence of subsongs in the recording; (7) presence/absence of female sounds (duet) in the recording; (8) acoustic time lag; (9) number of flights; (10) behavioural time lag, (11) distance to the dummy/loudspeaker.

In total, we analyzed 3589 songs from 35 males. One type of both simple and complex songs was identified in the recording of each male.

Statistical analysis

Data visualization and statistical analysis were conducted in R (R Development Core Team, 2020).

To estimate the effect of context (before, during, and after playback) on the number of simple songs, we first used a generalized linear mixed model (GLMM) with a Poisson distribution, however then we found overdispersion in the models and opted for the quasi-Poisson distribution and a logit-link function (Zuur et al., 2009)

To examine the potential effect of the type of stimuli (simple song stimuli vs. complex song/duet stimuli) on the number of simple songs, number of complex songs, number of flights, behavioural and acoustic time lags, minimal distance to the dummy/loudspeaker, and presence/absence of subsong, we computed a generalized linear mixed model (GLMM) fitted by maximum likelihood. We used the quasi-Poisson error distribution with a logit-link function for all response variables except for the variable presence/absence of subsong. For this response variable, we used a binomial distribution with a logit-link function. We used male ID as a random factor in this and all other mixed models (it was done because each experiment consisted of two parts).

To analyze the effect of playing back complex song vs. duet stimuli on the number of simple songs, number of complex songs, number of flights, presence/absence of subsong and presence/absence of duet, we used generalized linear mixed model (GLMM) with quasi-Poisson error distribution and a logit-link function for the number of simple songs, number of complex songs, number of flights variables except for the variable presence/absence of subsong and duet. For this response variables, we used a binomial distribution with a logit-link function.

We considered all tests with a p-value < 0.05 statistically significant.

RESULTS

Male vocalization

In our sample, there were three types of male vocalizations: simple song, complex song, and subsong (Fig. 1).

Simple song

Simple songs were most often heard during spontaneous singing, i.e., when no other birds were nearby. Such singing is a sequence of stereotyped songs consisting of two (Fig. 1) or rarely three (13% of males) elements each (Fig. 1). The elements in the songs are clearly separated. The median duration of songs individually varied from 0.61 to 1.02 s. Pauses between songs spanned between 1.75 to 7.65 s. Only one simple song type was present in the repertoire of each male.

The number of simple songs in the 5-minute recordings obtained before the playback presentation varied from 25 to 126 (median = 61, n = 35). These songs were predominantly within the frequency range of 2.4–3.4 kHz.

Complex song

The complex song has a much more complicated structure than the simple song. The duration of each varied widely in the range of 1 to 10 s (median = 5, n = 33). This song was loud compared to the simple song and consisted of 10–30 elements predominantly different from each other (Fig. 1). There was one complex song type in each male's repertoire. In our sample, complex songs were predominantly used in response to simulated territorial intrusion. The number of complex songs per 5 min of the playback trial varied from 1 to 24 (median = 6.5, n = 35).

Each complex song consisted of 1–5 (mean = 2) strophes (Fig. 1). There was only one strophe type in each male. The number of elements in each strophe varied from 2 to 15. These elements, in turn, belonged to several phrases. Thus, each strophe consisted of several phrases. Usually, starting with the first most high-pitched element of the phrase, the frequency of subsequent elements gradually decreases. Different phrase types were generally produced in a fixed order within the strophe. However, males can vary the phrase order to some extent and omit some phrases or make shortened phrases in some strophes (Fig. 1). Thus, complex songs had variety and complexity because the sequence and structure of phrases and strophes were not stereotyped, leading to the variability of its duration.

Subsong

In addition to complex songs, subsong was produced in a territorial competition context. Subsong was recorded from most of the males (88%, n=33). Males performed subsong very quietly. One could hear subsong just within 10–15 m around the singing male. Audibly, it was a quiet twitter. Structurally, it was a sequence of syllables consisting of 2–4 broadband elements each (Fig. 1). Males commonly repeated one syllable type a few times (phrase) before proceeding with the next syllable type. Thus, subsong was a sequence of phrases differing in duration and structure.

Duet

Males alone were observed in 17 experiments, and pairs participated in 18 experiments. Females can respond to the playback of the conspecific song along with the male. In this context, males and females can vocalize simultaneously, forming a duet. During duetting, females produce a sequence of identical broadband elements overlapping the complex male song (Fig. 1). Typically, males lead the duet (i.e., begin to vocalize first).

Responses to simple song playback

A clear response was detected in 24 out of 35 playback experiments, as males approached the loudspeaker and flew around it. There were no flights during the first stage of any experiment. The number of flights during the playback trial varied from 0 to 14 (median = 2.5, n = 24).

Sixty percent of males (n = 35) stopped singing after the start of the playback and resumed singing for 5 minutes. However, 14 out of 35 males did not sing during playback. The acoustic time lag in the other 21 males ranged from 3 to 258 s (median = 95).

In response to playback, all males decreased song rate considerably (GLMM, estimate = -1.26554, $p < 0.05$, Tab. 1): the number of simple songs during playback (5 min) varied from 0 to 75 (median = 11.5, n = 24) (Fig. 2). When the playback was over, the majority of males (80%, n = 35) began to return to the values of song rates observed before playback (Fig. 2).

As mentioned above, 21 males vocalized during the playback stage. Among them, 5 males produced 2–9 complex songs per 5 minutes of playback (median = 5). Only simple songs were produced by the others.

The acoustic behaviour of Puff-throated Babblers during playback presentation didn't generally differ from that observed after the playback. However, some males (23%, n = 35) did not sing after playback. Additionally, males did not fly around the loudspeaker at this time; instead, they fed nearby.

Responses to complex song and duet playbacks compared with simple song playback

The acoustic response of males differed depending on whether they were tested with tracks of complex songs/duets or tracks of simple songs.

Thirty-three out of 35 males produced complex songs and/or subsongs in the second part of the experiment, that is, in response to complex songs or duets. By contrast, five males only did so during the playback of simple songs. Among them, three males sang only simple songs during complex song playback, and two males did not sing during playback. The number of simple and complex songs a male produced differed significantly between simple and complex song playback (Tab. 2). Puff-throated Babbler males sang more simple songs during simple song playback than during complex playback (GLMM, estimate = 2.9267, $p > 0.05$, Tab. 2, Fig. 3). These values were 0–75 (median = 10.5) and 0–41 (median = 0), respectively (Fig. 3). On the contrary, males sang more complex songs (0–40, median = 3) in response to complex song playback than to simple song playback (GLMM, estimate = 2.0986, $p < 0.05$, Tab. 2, Fig. 3).

Puff-throated Babbler males produced subsong in response to complex song playback more often than to simple song playback. During the second part of the experiment, this vocalization was emitted by 30 males (89%, n = 33), while only 5 males (14%, n = 35) produced subsong during the first part of the experiment. Thus, the usage of subsong differed significantly between simple and complex song playback presentations (GLMM, estimate = 11.942, $p < 0.05$, Tab. 2, Fig. 4).

Males performed more flights during playbacks of complex songs and duets than during playbacks of simple songs (GLMM, estimate = 0.8477, $p < 0.05$, Tab. 2). The number during complex song/duet playbacks varied from 0 to 23 (median = 7, n = 33). The minimal distance to the dummy/loudspeaker significantly differed between simple and complex song/duet playbacks (Tab. 2). Puff-throated Babbler males approached closer to the dummy during complex song/duet playback (0–10 m, median = 2) than during simple song playback (0–15 m, median = 3.5). Thus, males reacted much more strongly when presented with the playback of complex songs and duets compared to the reaction to simple songs.

Female vocalization was observed only in response to complex song and duet playbacks. Moreover, we observed a female during the presentation of simple songs only once, and she behaved silently. By contrast, females vocalized in almost half of the complex song presentations (44.4%, n = 18). Apart from producing vocalizations, females approached the loudspeaker and flew around it. The reaction of Puff-throated Babbler females thus differed significantly between simple and complex song playbacks.

The behavioural and acoustic time lags were significantly shorter during complex song playback than during simple song playback (Tab. 2). The corresponding values were 3–176 s (median = 60 s, n = 33) for acoustic time lag and 0–208 s (median = 21 s, n = 33) for behaviour al time lag.

Responses to complex song playback compared with duet playback

Males produced significantly more simple songs in response to duets than to complex song playback (GLMM, estimate = 2.3180, $p < 0.05$, Tab. 3, Fig. 5). During duet presentation, that number varied from 0 to 35 (median = 0, n = 16).

The number of complex songs also depended on the playback type (Tab. 3, Fig. 5). Males sang complex songs at a significantly higher rate during duet playback than during complex song playbacks (GLMM, estimate = 0.9902, $p < 0.05$). Males sang up to 40 complex songs during both playback types (median = 3, n = 33).

The number of flights was notably fewer during complex song playback than duet playback (GLMM, estimate = 2.14277, $p < 0.05$, Tab. 3, Fig. 5). There were significant correlations between the number of complex songs and the number of flights observed during the second part of the experiment ($r = 0.43$, $p < 0.05$). The more complex songs males produced, the more flights they performed (Fig. 6).

The number of simple songs, complex songs and flights depended on playback type (Tab. 3, Fig. 5). Puff-throated Babblers responded more aggressively when presented with the duet playback. In this context, they sang actively and flew more often than in response to complex songs (Tab. 3, Fig. 5).

We did not identify significant differences in the presence of subsong and duet in response to during complex song playback compared to duet playback (Tab. 3).

DISCUSSION

In this study, we provided the first detailed description of the vocalization of Puff-throated Babblers observed in a territorial competition context. We described three different vocalization types in Puff-throated Babblers and revealed their dependencies on context. While singing spontaneously, males produce simple songs repeated at a rate of approximately 12 songs per minute. Males could respond to playback with complex songs, which are much more elaborate and longer (up to 5–10 s) vocalizations emitted at a rate of approximately one song per minute. Females can join their mate's complex song by emitting a trill and forming a duet. Playback experiments showed that the male's response depended on vocalization type: simple songs generated the weakest response and duets the strongest. We thus provided evidence that these types of vocalizations have different functions, a pattern well-known in many other bird species.

Several songbirds have two singing modes, typically one of them much simpler than the other (Spector 1992, Beebee 2002, Catchpole & Slater 2003). In many wood warblers *Parulidae*, for example, the repeat mode (type I songs) comprises repetitions of a single song type, and the serial mode (type II songs) consists of several song types sung in a versatile sequence (Lemon et al. 1985, MacNally & Lemon 1985, Opaev 2012). In our experiments, Puff-throated Babbler males produced radically different songs in a territorial competition context (complex song) and while singing spontaneously (simple song). Different songs function in different ways, as observed in Hume's warbler *Phylloscopus humei* where males use song type 1 while singing spontaneously, primarily for advertising territory and attracting a female, and song type 2 more often during countersigning between neighbouring males (Meshcheryagina & Opaev 2023). In both Hume's warbler and Puff-throated Babblers, the two song types differ fundamentally in time-and-frequency parameters (Fig. 1). Although the vocalization of babblers is generally poorly studied, at least one species, the Red-billed leiothrix *Leiothrix lutea*, also uses two song types (Ramellini 2017). The peculiarity of our study species was, however, that the two song types differed dramatically in their complexity. Nevertheless, the usage of different song types in different contexts is not unique to tropical birds (Catchpole & Slater 2003, Demko & Mennill 2018, Budka et al. 2023).

We found significant differences in the responses to simple song playback compared to complex songs and duets. Puff-throated Babbler males generally produce simple songs in response to simple song playback, and complex songs in response to complex song playback (Fig. 3). This might partly be because of song matching, which can signal aggression as studied in detail in Song Sparrows *Melospiza melodia* (Beecher et al. 2000, Briefer et al. 2010). While matching, males reply to a singing rival with the same or a similar song type (Beebee 2002, Catchpole & Slater 2003). However, we suggested that different acoustic responses to simple vs. complex song stimuli are not fully explained by song matching. Apparently, these two song types have different functions, as evidenced by a stronger response to complex songs. We found that males performed more flights and came closer to the loudspeaker during playback of complex songs and duets, which is evidence of more aggressive behaviour in this context. Based on our findings, we suggested that simple songs function in territory advertising and/or attracting a partner, while complex songs are used for territorial defence and communication with females.

Along with complex songs, males usually used subsong when responding to playback. Subsong is a quiet vocalization, and thus can be referred to as a 'soft or low-amplitude song'. We found that subsong, if present, almost always preceded or accompanied the complex song. Low-amplitude songs are known to be used by a variety of songbirds in both tropical and temperate zones. The most often soft songs are produced in the course of short-range interactions, such as during aggressive encounters and courtship (Anderson et al. 2007, Templeton et al. 2012). We suggest that, in our study species, subsong is used when males compete over resources such as breeding territories. Similar findings came from the study of Savannah Sparrows *Passerculus sandwichensis* where the number of soft songs was a significant predictor of aggression (Moran et al. 2018). Similarly, soft songs are an aggressive signal in the Plain Laughingthrush *Pterorhinus davidi* (Liu 2022). Therefore, in agreement with previous studies (Balsby & Dabelsteen 2002, Anderson et al. 2007, Moran et al. 2018, Liu 2022), soft songs are used by both tropical and temperate birds, including in territorial competition contexts.

Female songs are rather common in the tropics. Because of this, duetting is more common in tropical regions than in northern temperate regions (Robinson 1949). It is unclear why males and females coordinate their songs to form duets in tropical species, while the majority of temperate zone birds do not. In the tropics, several passerine bird species produce coordinated male-female duets, including in territorial competition contexts. Many duetting species maintain territories throughout the year, and both sexes participate in territorial defence. In the context of territorial competition, duets are used similarly to male complex songs (Wickler 1976, Hall 2000). Similar to several other tropical bird species (Payne & Skinner 1970, Payne, 1971; Tingay, 1974; Wickler 1976, Harcus 1977, Farabaugh 1982), we found that male and female Puff-throated Babblers produced coordinated duets in a territorial competition context. We found that duets represented a stronger territorial signal than solo songs. The reason could be that duets transmit information about the numeric advantage of the territory owners or represent a quality signal arising from song synchronization (Hall & Magrath 2007, Diniz et al. 2020). In our experiments, female vocalization was observed only in response to complex songs and duet playbacks, not to simple song playback. As we stated before, complex songs of Puff-throated Babbler males appeared to be a more aggressive signal than simple songs as it was observed in the territorial competition context only. Forming a duet, a female emits the duet trill overlapping the males' complex songs, not its simple song. Based on this finding, we suggested that females participated in territory defence in our study species. Although the usage of duets in territorial interactions is consistent with their function as a cooperative territorial signal, this does not exclude alternative interpretations. For example, in the study of Black-bellied Wren *Thryothorus fasciatoventris*, it has been shown that duetting during territorial encounters allows mates to identify one another, thus preventing intrapair aggression (Logue & Gammon 2004). At the same time, partners in Rufous Horneros *Furnarius rufus* coordinate their vocal behaviours to cooperatively defend common territories. They respond to conspecific stimuli together and coordinate ~80% of their songs into duets (Diniz et al. 2020). In general, the significance of coordinated song during territorial defence remains unclear.

Generally, the organization of Puff-throated Babblers' duets was in congruence with that of many other tropical passerines. In many species, one or both sexes sing independently of their partner or form a duet (Harcus 1977, Hall 2000). Duetting may be accomplished through (a) song merging: two individuals combine their respective songs in a more or less complicated manner; (b) song copying: individuals copy their partner's song; or (c) song splitting: a given song is divided up between the partners (Wickler & Seibt 1982). Though both birds have roles in duets, it is the second bird that creates the duet by responding to the duet initiator (Hall 2000). In our case, Puff-throated Babbler females join their partners’ songs to form duets. The duet of this species is thus formed through merging.

Taking together, the results of our study revealed similarities in aggressive signalling among tropical and temperate birds, including the usage of different song types depending on the context, producing low-amplitude songs in territorial competition context, and (possibly) song matching. However, duet vocalization, including in territorial competition context, appeared to be more typical in the tropics. In contrast to many other tropical duetting passerines, Puff-throated Babbler males produced a specific complex song in response to playback, and only that song initiated a duet. Therefore, a loud complex song may function to attract a female when an opponent appears, and the female can use a duet trill to manifest herself in joint territory defence.

CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

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Captions

Table 1. Results of a generalized linear mixed model (GLMM) testing the context (before/during/after simple playback) effects on the number of simple songs. Significant models ($p < 0.05$) were present in bold.

		Estimate	**Std. Error**	***z* value**	**Pr(>|z|)**
N simple songs	**Before playback**	**-0.44516**	**0.02948**	**-15.099**	**< 2e-16 *****
	During playback	**-1.26554**	**0.04680**	**-27.039**	**< 2e-16 *****
	After playback	**4.61312**	**0.02666**	**173.006**	**< 2e-16 *****

Table 2. Results of generalized linear mixed models (GLMM) testing the effects of type of playback (simple/complex) on 7 variables of males' aggressive response. Significant models (*p* < 0.05) were present in bold.

		Estimate	**Std. Error**	***z* value**	**Pr(>|z|)**
N simple songs	**Simple playback**	**2.9267**	**0.2209**	**13.250**	**<2e-16 *****
	Complex playback	-0.3014	0.2530	-1.191	0.234
N complex songs	Simple playback	-0.5964	0.5094	-1.171	0.242
	Complex playback	**2.0986**	**0.4172**	**5.030**	**<2e-16 *****
Time lag acoustics	**Simple playback**	**4.39956**	**0.20618**	**21.339**	**<2e-16 *****
	Complex playback	-0.01581	0.24429	-0.065	0.948
Time lag behaviour	**Simple playback**	**4.5807**	**0.2726**	**16.804**	**< 2e-16 *****
	Complex playback	**-0.9592**	**0.2668**	**-3.595**	**0.000325 *****
N flights	**Simple playback**	**-0.9224**	**0.3563**	**2.628**	**0.00858 ****
	Complex playback	**0.8477**	**0.2823**	**3.003**	**0.00267 ****
Model distance	**Simple playback**	**1.5319**	**0.2714**	**5.645**	**1.65e-08 *****
	Complex playback	**-0.7668**	**0.2295**	**-3.341**	**0.000833 *****
Subsongs	**Simple playback**	**-22.715**	**4.736**	**-4.796**	**0.000177 *****
	Complex playback	**11.942**	**3.184**	**3.750**	**1.62e-06 *****

Table 3. Results of generalized linear mixed model (GLMM) testing the effects of type of context (playback with female/playback without female) on 3 variables of males' aggressive response. Significant models ($p < 0.05$) were present in bold.

		Estimate	Std. Error	*z* value	Pr(>|z|)
N simple songs	**Playback with female**	**2.3180**	**0.3173**	**19.013**	**2.77e-13 *****
	Playback without female	0.4537	0.3458	1.312	0.189
N complex songs	Playback with female	0.5117	0.5699	0.898	0.3692
	Playback without female	**0.9902**	**0.4218**	**2.348**	**0.0189 ***
N flights	Playback with female	0.4199	0.3071	1.367	0.172
	Playback without female	**1.7504**	**0.2543**	**6.884**	**5.82e-12 *****

542 **Figure 1.** Puff-throated babbler vocalizations: (a) male simple song (two-element song and three-element song recorded from different males are presented), (b) male complex song (strophes are separated by solid lines, and different phrase types are shown by different colours), (c) male subsong, (d) duet (female trill is marked in red).

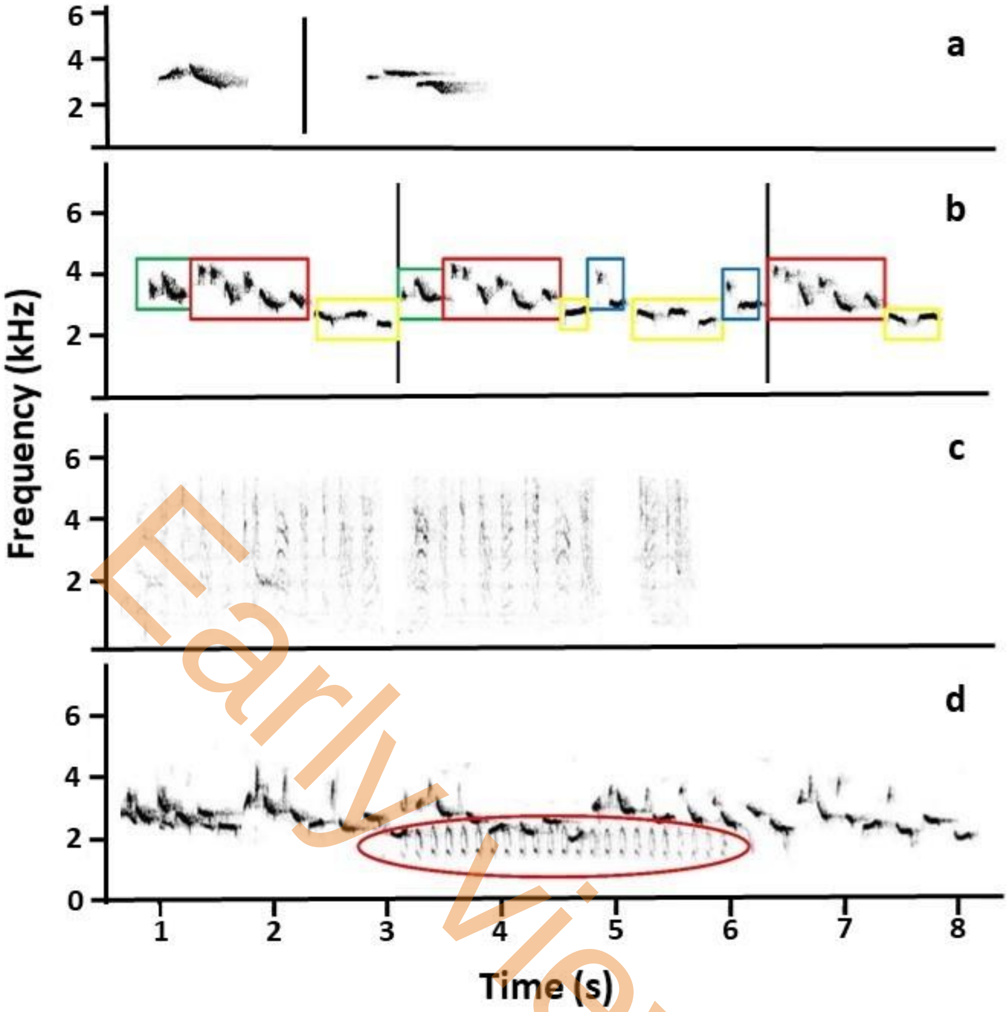

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Figure 2. Number of simple songs per 5 min across three experimental stages: before, during, and after simple song stimuli presentation.

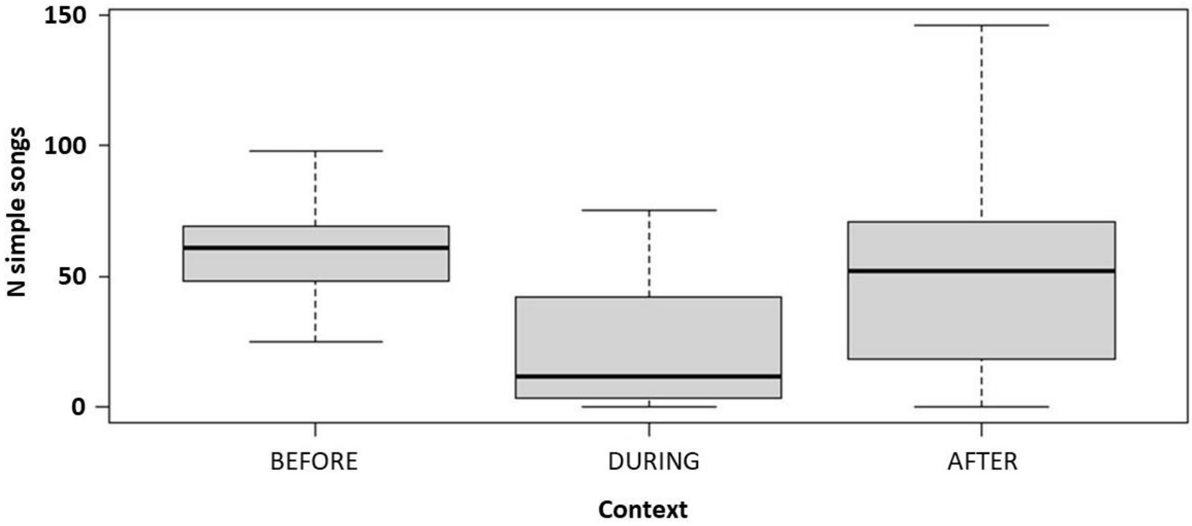

Figure 3. Number of simple and complex songs recorded in response to simple vs. complex song playbacks.

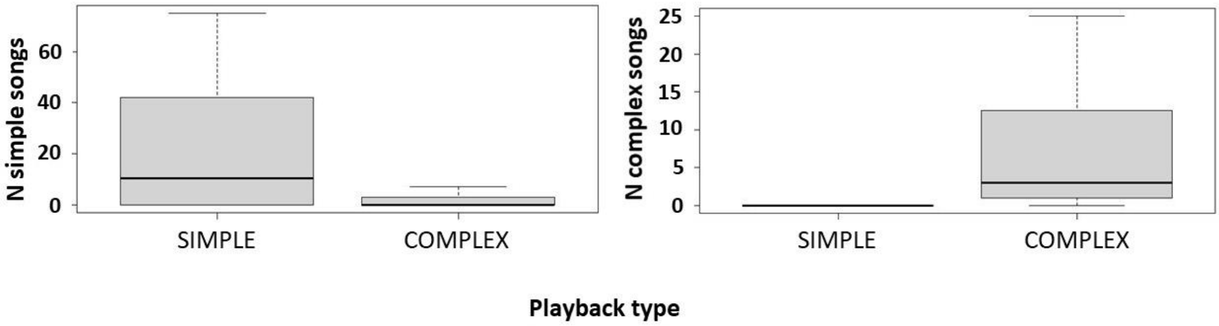

Figure 4. Presence/absence of subsongs in response to simple vs. complex song playback presentation (yes – subsong present, no – absent).

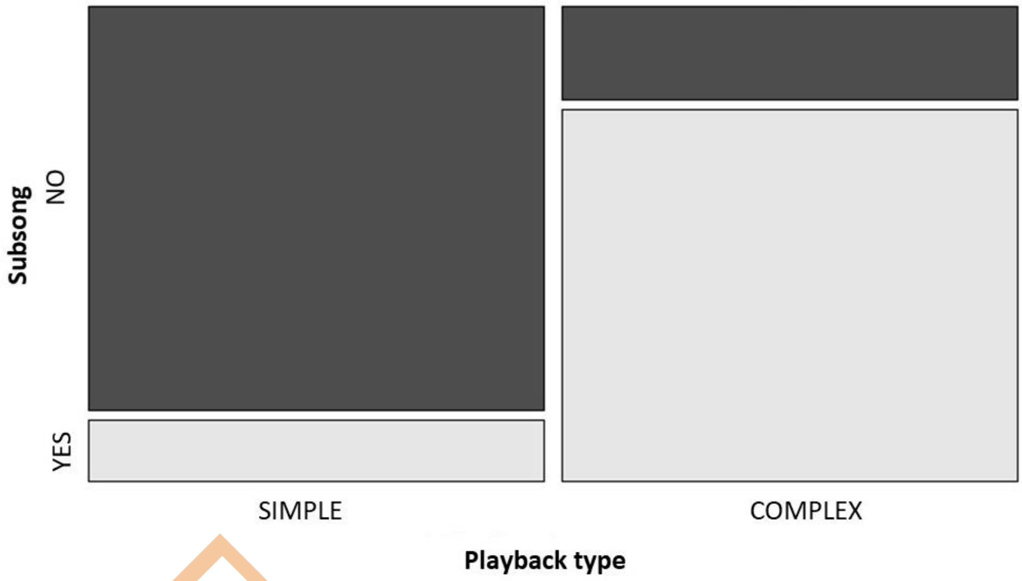

Figure 5. Number of simple songs, complex songs and flights of puff-throated babbler males observed during the playback of the second part of the experiment depending on playback type (complex song vs. duet).

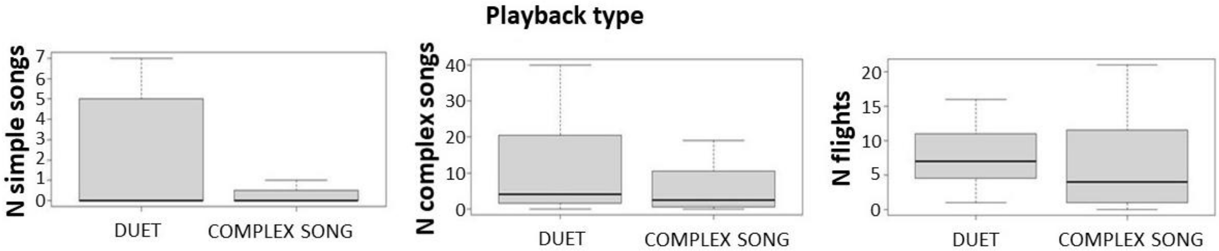

Figure 6. Bivariate plot showing the correlation between the number of complex songs and the number of flights observed during complex song/duet presentation.