Song evolution in Maluridae: influences of natural and sexual selection on acoustic structure

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Abstract. Many factors may influence the evolution of acoustic signals, including sexual selection, morphological constraints and environmental variation. These factors can play simultaneous and interacting roles in determining signal phenotypes. Here, we assess the evolution of song features in the Maluridae, a passerine family with significant variation among taxa in levels of sperm competition, morphological features and breeding habitats ranging from arid grasslands in Australia to tropical rainforests in New Guinea. We used phylogenetic comparative methods and a robust molecular phylogeny to compare song characteristics with a variety of other measures, including testes mass, body-size and latitude. Several aspects of the temporal and frequency structure of song were associated with relative testes mass, suggesting that sexual selection may influence some song characteristics in this family. The lowest frequencies of song were strongly predicted by body-size, indicating that morphological constraints have also likely influenced acoustic phenotypes. Song versatility, reflecting the diversity of note types in a song, was positively correlated with latitude, suggesting that complexity may increase in association with more temperate or variable environments. Variation in song structure across the family appears to reflect a complex interaction between natural and sexual selection.

Additional keywords: body-size, latitude, relative testes size.

Received 7 September 2012, accepted 10 April 2013, published online 15 August 2013

Introduction

Acoustic signals are subject to a wide variety of selective pressures. In birds, many factors have been shown to influence the structure and rate of evolutionary change of vocalisations, including sexual selection (Irwin 2000; Badyaev et al. 2002; Price and Lanyon 2004), body-size (Wallschläger 1980; Ryan and Brenowitz 1985; Price et al. 2006), habitat (Wiley 1991; Slabbekoorn and Smith 2002; Patten et al. 2004), breeding latitude (Weir and Wheatcroft 2011) and seasonal variability (Botero et al. 2009; Medina and Francis 2012). These patterns may be evident both within and between species, and they illustrate the wide variety of selective pressures that may influence acoustic phenotypes.

Given the range of selection pressures that can influence songs, making specific predictions concerning the evolution of acoustic signals is difficult. For example, sexual selection may favour the evolution of more complex or, alternatively, more simple vocalisations, depending upon what features are most attractive to females or most successful in male–male competitive interactions (Searcy and Andersson 1986; Cardoso and Hu 2011). Body-size may constrain the production of low frequencies (Ryan and Brenowitz 1985; Price et al. 2006), so evolutionary changes in body-size for social or ecological reasons may allow corresponding changes in the frequency composition of song. Alternatively, selection for low frequencies may cause a corresponding change in morphology (Fitch 1999). Acoustic adaptation to closed habitats may favour songs with slow rates of note repetition and lower sound frequencies, which transmit more effectively through dense vegetation than rapidly repeated or higher-frequency notes (discussed in Handford and Lougheed 1991; Wiley 1991; Patten et al. 2004; Seddon 2005). Finally, evidence suggests that birds living at higher latitudes may have songs that are longer, more complex or more variable, presumably due to the relatively variable environments at these latitudes (Botero et al. 2009; Weir and Wheatcroft 2011; Medina and Francis 2012). Ultimately, all of these selective pressures may interact and lead to different signal phenotypes in different taxa.

In examining potential factors influencing song evolution, it is critical to consider such interactions within a phylogenetic framework, because behavioural traits such as song may show significant phylogenetic dependence (Price and Lanyon 2002; Packert et al. 2003) and therefore trait values from related taxa may not be statistically independent (Felsenstein 1985; Freckleton et al. 2002). Accounting for phylogenetic relationships permits a statistically appropriate evaluation of potential evolutionary correlations, as well as an assessment of historical changes in traits. Thus,
a study group in which there is a well resolved phylogeny is essential for such an analysis, and such a phylogeny is available for the oscine family Maluridae. Additionally, songs in Maluridae range from fairly simple note repetitions to extremely complex assortments of rapidly repeated notes (Rowley and Russell 1997). Songs are used during territorial disputes and during dawn-chorus displays (Rowley and Russell 1997; Dalziell and Cockburn 2008; Greig and Pruett-Jones 2008, 2009, 2010; Hall and Peters 2008; Dowling and Webster 2013), and song playbacks are reported to elicit territorial responses in most studied species (Rowley and Russell 1997). Thus, song is likely important in mate attraction and territory defence in Maluridae, and other social functions of song such as mate guarding or within-group communication may be important as well (Payne et al. 1988, 1991; Hall and Peters 2008; Dowling and Webster 2013).

Here, we investigate associations of acoustic structure with levels of sperm competition, body-size and latitude in a comparative framework, using the Maluridae. This family is ideal for such an investigation for at least four reasons. First, species exhibit extreme variation in potential sexual selection pressures; the occurrence of extra-pair paternity ranges from as low as 6% of broods in some species to as high as 95% (Mulder et al. 1994; Kingma et al. 2009; Rowe and Pruett-Jones 2013). Although extra-pair paternity rates have not been measured for all species, relative testes mass (testes mass controlling for variation in body-weight) is known for most species and can be used as a proxy for levels of sperm competition (Rowe and Pruett-Jones 2006, 2011). Second, the species vary in body-size, from small fairy-wrens and emu-wrens (e.g. Stipiturus malachurus: 7.3 g) to large grasswrens (e.g. Amytornis striatus: 19.2 g) (Rowley and Russell 1997). Third, for a reasonably small family, species in Maluridae occupy a remarkable variety of habitats, from primary rainforest in New Guinea to the driest desert grasslands in Australia. And, lastly, all of the species in Maluridae are non-migratory, permanent residents of the habitats and territories on which they live, so vocal communication is presumably little affected by seasonal changes in habitat or by species interactions during migration. Overall, therefore, this family allows us to examine the influences of, and complex interactions between, natural and sexual selection in the evolution of bird song.

### Materials and methods

#### Study taxa

The Maluridae consists of five genera, the grasswrens (Amytornis) and emu-wrens (Stipiturus) that inhabit arid habitats throughout much of Australia, and the fairy-wrens (Malurus, Clytomyias and Sipodotus), which inhabit a variety of tropical and temperate habitats throughout Australia and New Guinea (Rowley and Russell 1997). Malurids are small, primarily insectivorous passerines and are part of the large Australasian radiation of the Meliphagoidae (Gardner et al. 2010). All species of the Maluridae are known, or believed, to exhibit cooperative breeding, although the extent of cooperation varies across species and across populations within a given species (Rowley and Russell 1997).

We obtained song recordings and collected published data on body-weight (g), habitat and mean latitude for 16 of the 21 malurid taxa that were included in a published molecular phylogeny for the Maluridae (Table 1; phylogeny based on Driskell et al. 2011). We also obtained data on combined testes mass (g) (Rowe and Pruett-Jones 2011) for 14 of these 16 taxa (all except Malurus cyanocephalus and Sipodotus wallacii). We were unable to obtain measurements for four of the taxa included by Driskell et al. (2011): Malurus splendidus musgravii, M. grayi, M. campbelli and Clytomyias insignis. The subspecies identity of song recordings for M. alboscapulatus was uncertain, so we collapsed the two subspecies present in the analysis of Driskell et al. (2011) into one taxon representing the M. alboscapulatus subspecies complex. Note that there are now three published phylogenies for the Maluridae (Gardner et al. 2010; Driskell et al. 2011; Lee et al. 2012). These three phylogenies generally agree in the overall arrangement of species and their topology except that in the phylogeny of Lee et al. (2012) M. amabilis is placed outside of the M. lamberti clade. Also, the position of

### Table 1. Habitat, mean latitude of breeding range, body-weight and combined testes mass for all taxa included in phylogenetic analyses

<table>
<thead>
<tr>
<th>Taxon</th>
<th>Habitat</th>
<th>Mean latitude (°S)</th>
<th>Body-weight (g)</th>
<th>Combined testes mass (g)</th>
<th>Body and testes mass source</th>
</tr>
</thead>
<tbody>
<tr>
<td>M. alboscapulatus</td>
<td>Grass and shrubland</td>
<td>6.0</td>
<td>11.2</td>
<td>0.25</td>
<td>Rowe and Pruett-Jones (2006)</td>
</tr>
<tr>
<td>M. amabilis</td>
<td>Closed forest</td>
<td>15.0</td>
<td>8.38</td>
<td>0.113</td>
<td>Rowe and Pruett-Jones (2013)</td>
</tr>
<tr>
<td>M. coronatus coronatus</td>
<td>Riparian and open forest</td>
<td>16.5</td>
<td>11.4</td>
<td>0.05</td>
<td>Rowe and Russell (1997) (body), Rowe and Pruett-Jones (2006) (testes)</td>
</tr>
<tr>
<td>M. cyaneus cyaneus</td>
<td>Open forest</td>
<td>42.5</td>
<td>9.4</td>
<td>0.48</td>
<td>Rowe and Pruett-Jones (2006)</td>
</tr>
<tr>
<td>M. cyaneus cyanochlamys</td>
<td>Open forest</td>
<td>30.5</td>
<td>8.93</td>
<td>0.29</td>
<td>Rowe and Pruett-Jones (2013)</td>
</tr>
<tr>
<td>M. cyaneus</td>
<td>Open forest</td>
<td>6.5</td>
<td>13.4</td>
<td></td>
<td>Rowley and Russell (1997)</td>
</tr>
<tr>
<td>M. elegans</td>
<td>Closed forest</td>
<td>33.0</td>
<td>9.8</td>
<td>0.203</td>
<td>Rowe and Pruett-Jones (2013)</td>
</tr>
<tr>
<td>M. lamberti assimilis</td>
<td>Mallee</td>
<td>26.5</td>
<td>8.29</td>
<td>0.18</td>
<td>Rowe and Pruett-Jones (2013)</td>
</tr>
<tr>
<td>M. lamberti lamberti</td>
<td>Open forest</td>
<td>29.0</td>
<td>8.2</td>
<td>0.25</td>
<td>Rowe and Pruett-Jones (2006)</td>
</tr>
<tr>
<td>M. leucopterus leuconotus</td>
<td>Grass and shrubland</td>
<td>27.0</td>
<td>7.7</td>
<td>0.326</td>
<td>Rowe and Pruett-Jones (2013)</td>
</tr>
<tr>
<td>M. melanochrophus melanochrophus</td>
<td>Open forest</td>
<td>22.0</td>
<td>7.57</td>
<td>0.213</td>
<td>Rowe and Pruett-Jones (2013)</td>
</tr>
<tr>
<td>M. pulcherrimus</td>
<td>Open forest</td>
<td>31.0</td>
<td>9.27</td>
<td>0.13</td>
<td>Rowe and Pruett-Jones (2013)</td>
</tr>
<tr>
<td>M. splendidus melanotus</td>
<td>Mallee</td>
<td>30.5</td>
<td>9.45</td>
<td>0.298</td>
<td>Rowe and Pruett-Jones (2013)</td>
</tr>
<tr>
<td>A. striatus striatus</td>
<td>Grass and shrubland</td>
<td>33.0</td>
<td>19.23</td>
<td>0.34</td>
<td>Rowe and Pruett-Jones (2011)</td>
</tr>
<tr>
<td>S. malachurus</td>
<td>Grass and shrubland</td>
<td>32.5</td>
<td>7.32</td>
<td>0.045</td>
<td>Rowe and Pruett-Jones (2011)</td>
</tr>
<tr>
<td>Sipodotus wallacii</td>
<td>Closed forest</td>
<td>5.0</td>
<td>7.9</td>
<td></td>
<td>Rowley and Russell (1997)</td>
</tr>
</tbody>
</table>
M. coronatus varies across all three phylogenies (see also Joseph et al. 2013). We chose the phylogeny of Driskell et al. (2011) in this study because that phylogeny includes the largest number of species of Malurus, the genus for which we had the most recordings. We do not test our results with the other phylogenetic hypotheses, but the similarity of the phylogenies overall led us to suspect that our results would be similar.

Measurements of testes mass were obtained from studies by Rowe and Pruett-Jones (2006, 2011, 2013), and body-weights were obtained from Rowley and Russell (1997). We calculated the mean breeding latitude of each taxon as the approximate midpoint between the highest and lowest latitudes of distribution maps in Rowley and Russell (1997). We classified habitat into five major types based on descriptions provided in Rowley and Russell (1997) (Table 1).

Song recordings

During the breeding seasons (October–December) of 2006–2010 we recorded songs during male dawn chorus displays from Lovely Fairy-wrens (M. amabilis), Red-winged Fairy-wrens (M. elegans), Variegated Fairy-wrens (M. assimilis), White-winged Fairy-wrens (M. leucopterus), Red-backed Fairy-wrens (M. melanochalas), Blue-breasted Fairy-wrens (M. pulcherrimus) and Splendid Fairy-wrens (M. splendens) (details of recording locations in Supplementary material, see http://www.publish.csiro.au/?act=view_file&file_id=MU12078_AC.pdf). For these recordings we used a Marantz PDM 661 solid-state digital recorder at 96-kHz sampling rate, 24-bit depth, or a Marantz PDM 670 at 48-kHz sampling rate, 16-bit depth (D&M Professional, Itasca, IL), combined with ME66 shotgun microphone capsules and K6 power modules (Sennheiser Electronic Corporation, Old Lyme, CT; frequency response 0.04–20.0 kHz).

We obtained additional song recordings from the Macaulay Library of Natural Sounds (Cornell University, Ithaca, NY), the xeno-canto online database (www.xeno-canto.org, accessed 6 March 2013), a commercially available audio CD (Plowright 2007) and the personal recording collection of Dr Michelle Hall (University of Melbourne) (details of recordings in Supplementary material). All recordings were uncompressed WAV files except for the three recordings from xeno-canto, which were compressed MP3 files. We included the latter three recordings in this analysis because those taxa were so poorly sampled; however, we acknowledge that the recording quality of those samples was lower than the quality of the other recordings in our database.

In many species of Maluridae both sexes are known to sing, especially members of the genus Malurus, in which at least one species (M. coronatus) performs coordinated male–female duets (Rowley and Russell 1997). Song in females is thought to function in territory defence (Cooney and Cockburn 1995; Hall and Peters 2008). Singing is thought to be primarily by males in emu-wrens and grasswrens, although this has been little studied (Rowley and Russell 1997). We were confident that the sampled individuals were males in all recordings collected by the senior author (E. I. Greig), by Michelle Hall and in most recordings from other sources; however, the sex was uncertain for some recordings (detailed in Supplementary material). In some species (e.g. M. splendens and M. cyaneus), males produce simpler trilled songs (Type II songs) in addition to their standard display vocalisations (Type I songs) (Langmore and Mulder 1992, Zelano et al. 2001). We avoided including Type II songs in our comparisons (see below).

We obtained recordings from five additional taxa that were not included in the Driskell et al. (2011) phylogeny or for which we did not have morphological data (M. coronatus macgillivrayi, M. splendens splendens, M. s. emmottorum, A. merrootsyi and Stipiturus mallee: included in Table 2). We included these songs in analyses of note types (described below) and in assessing correlations between acoustic measurements, but we did not use these taxa in any phylogenetic analyses.

Acoustic analysis

We chose at least one example song from each male that was of high recording quality based on spectograms digitzed in Raven Pro 1.4 (Bioacoustics Research Program 2004; 16-bit sample format; discrete Fourier transform; DFT = 512 samples; frequency resolution = 124 Hz; time resolution = 11.6 ms; frame overlap = 50%). We made an effort to use only one song from any individual; however, for eight poorly sampled taxa we used multiple songs from the same individual when calculating species means, for a total of 184 songs from 161 individuals (mean ± s.e. = 8.6 ± 9.1 songs per taxon) (Table 2). For M. splendens and M. cyaneus, we excluded Type II songs (trills) from our analysis. We did include display songs that contained trill components, because these were often the primary songs given by some individuals during dawn chorus displays. For each song we manually measured individual note characteristics in Luscinia (Lachlan 2007); we defined a note as a continuous tracing on a spectrogram, and we measured only the fundamental frequency for each note and avoided measuring harmonics or sidebands. We summarised nine acoustic characteristics (listed in Table 2) for each song. We chose these acoustic characteristics because they captured aspects of variation related to song frequency (peak, high and low frequency, song bandwidth and note bandwidth), tempo (song length, note length, number of notes and note rate) and complexity (note variety and song versatility). Our sampling method did not permit an estimate of song repertoire size, but such an analysis would be a valuable line of future inquiry.

We categorised all notes in the dataset (n = 9995 notes) into note types using Ward’s agglomerative hierarchical clustering (Ward 1963) based on the peak, minimum and maximum note frequency, note bandwidth, note duration and the change in bandwidth over time (bandwidth divided by duration). From this cluster analysis we identified an optimal total number of clusters (i.e. note types) using a method in which the distances between adjoining clusters were plotted against the rank of those distances (described by Taft 2011). Clusters with a low rank have greater distances between them and their division explains a large proportion of variance in the data, whereas clusters with a high rank have very short distances between them and represent items that could be considered members of the same cluster. The result of this plot is an L-shaped distribution of points, with the elbow of this distribution representing a transition between numbers of clusters that explain a large proportion of variance in the data versus numbers of clusters that explain a small proportion of variance in the data (Taft 2011). To identify this elbow, we followed Taft (2011) and calculated linear fits for the distribution
Table 2. Means and standard deviations of all song variables for all taxa

Sample sizes of songs and individuals are given. Asterisks indicate taxa that were excluded from phylogenetic analyses, but which were used for classification of note types and assessment of correlations between acoustic variables. Taxa: *M*. *Malurus*; *A*. *Amytornis*; *S*. *Stipiturus*

<table>
<thead>
<tr>
<th>Taxon</th>
<th>Sample size</th>
<th>Song length (s)</th>
<th>Peak frequency (kHz)</th>
<th>High frequency (kHz)</th>
<th>Low frequency (kHz)</th>
<th>Song bandwidth (kHz)</th>
<th>Note bandwidth (kHz)</th>
<th>Note length (ms)</th>
<th>Number of notes</th>
<th>Note rate (notes/s)</th>
<th>Note variety</th>
<th>Song versatility</th>
</tr>
</thead>
<tbody>
<tr>
<td>M. alboscapulatus</td>
<td>2 (2)</td>
<td>4.2 ± 0.7</td>
<td>5.4 ± 1.0</td>
<td>9.3 ± 1.9</td>
<td>2.5 ± 0.8</td>
<td>6.9 ± 1.0</td>
<td>0.37 ± 0.09</td>
<td>40.6 ± 5.9</td>
<td>59 ± 21.2</td>
<td>13.9 ± 2.8</td>
<td>10.0 ± 1.4</td>
<td>0.18 ± 0.04</td>
</tr>
<tr>
<td>M. amabilis</td>
<td>9 (9)</td>
<td>2.1 ± 0.2</td>
<td>6.5 ± 0.2</td>
<td>9.5 ± 0.3</td>
<td>3.5 ± 0.9</td>
<td>5.9 ± 1.0</td>
<td>0.71 ± 0.12</td>
<td>46.4 ± 11.1</td>
<td>31 ± 6.0</td>
<td>14.6 ± 2.8</td>
<td>7.7 ± 1.7</td>
<td>0.25 ± 0.06</td>
</tr>
<tr>
<td>M. coronatus coronatus</td>
<td>9 (9)</td>
<td>4.6 ± 1.9</td>
<td>4.7 ± 0.5</td>
<td>8.3 ± 0.7</td>
<td>1.4 ± 0.2</td>
<td>6.9 ± 0.7</td>
<td>1.27 ± 0.25</td>
<td>61.7 ± 8.0</td>
<td>43 ± 16.8</td>
<td>9.4 ± 1.2</td>
<td>6.1 ± 2.0</td>
<td>0.15 ± 0.05</td>
</tr>
<tr>
<td>M. coronatus macgillivrayi</td>
<td>3 (2)*</td>
<td>3.9 ± 1.0</td>
<td>4.8 ± 0.2</td>
<td>8.0 ± 0.5</td>
<td>1.3 ± 0.2</td>
<td>6.7 ± 0.3</td>
<td>1.12 ± 0.12</td>
<td>59.6 ± 19.7</td>
<td>37 ± 9.6</td>
<td>9.5 ± 3.2</td>
<td>5.5 ± 2.4</td>
<td>0.16 ± 0.06</td>
</tr>
<tr>
<td>M. cyaneus cyaneus</td>
<td>2 (1)</td>
<td>3.5 ± 0.2</td>
<td>5.6 ± 0.7</td>
<td>9.7 ± 1.1</td>
<td>2.3 ± 0.2</td>
<td>7.4 ± 1.3</td>
<td>0.96 ± 0.47</td>
<td>68.3 ± 15.4</td>
<td>35 ± 0.7</td>
<td>9.8 ± 0.8</td>
<td>11.5 ± 2.1</td>
<td>0.33 ± 0.05</td>
</tr>
<tr>
<td>M. cyaneus cyanochlamys</td>
<td>8 (6)</td>
<td>3.1 ± 0.9</td>
<td>5.8 ± 0.5</td>
<td>9.6 ± 0.7</td>
<td>2.9 ± 0.5</td>
<td>6.7 ± 0.8</td>
<td>0.97 ± 0.34</td>
<td>44.3 ± 6.9</td>
<td>45 ± 17.2</td>
<td>14.2 ± 1.7</td>
<td>11.1 ± 2.6</td>
<td>0.27 ± 0.08</td>
</tr>
<tr>
<td>M. cyanocephalus</td>
<td>5 (1)</td>
<td>4.7 ± 1.4</td>
<td>2.4 ± 0.0</td>
<td>3.8 ± 0.3</td>
<td>1.3 ± 0.3</td>
<td>2.5 ± 0.4</td>
<td>0.38 ± 0.03</td>
<td>106.5 ± 8.5</td>
<td>25 ± 7.2</td>
<td>5.4 ± 0.4</td>
<td>20.0 ± 0.0</td>
<td>0.09 ± 0.03</td>
</tr>
<tr>
<td>M. elegans</td>
<td>10 (10)</td>
<td>3.3 ± 0.4</td>
<td>5.3 ± 0.3</td>
<td>8.8 ± 0.6</td>
<td>2.3 ± 0.2</td>
<td>6.5 ± 0.6</td>
<td>0.76 ± 0.15</td>
<td>62.1 ± 15.8</td>
<td>29 ± 7.6</td>
<td>8.8 ± 2.3</td>
<td>9.3 ± 1.6</td>
<td>0.33 ± 0.07</td>
</tr>
<tr>
<td>M. lamberti assimilis</td>
<td>23 (23)</td>
<td>2.9 ± 0.4</td>
<td>5.5 ± 0.4</td>
<td>8.9 ± 0.8</td>
<td>2.8 ± 0.5</td>
<td>6.1 ± 0.8</td>
<td>1.21 ± 0.36</td>
<td>39.4 ± 6.8</td>
<td>53 ± 8.9</td>
<td>18.0 ± 2.4</td>
<td>11.6 ± 1.6</td>
<td>0.22 ± 0.05</td>
</tr>
<tr>
<td>M. lamberti lamberti</td>
<td>1 (1)</td>
<td>3.63</td>
<td>9.3</td>
<td>3.5</td>
<td>5.8</td>
<td>1.03</td>
<td>37.7</td>
<td>55</td>
<td>18.5</td>
<td>13</td>
<td>0.24</td>
<td></td>
</tr>
<tr>
<td>M. leucopterus leuconotus</td>
<td>22 (22)</td>
<td>4.8 ± 0.8</td>
<td>4.4 ± 0.2</td>
<td>7.9 ± 1.0</td>
<td>1.9 ± 0.2</td>
<td>6.1 ± 1.0</td>
<td>0.66 ± 0.17</td>
<td>21.4 ± 1.9</td>
<td>119 ± 21.9</td>
<td>24.8 ± 2.0</td>
<td>10.0 ± 1.6</td>
<td>0.09 ± 0.02</td>
</tr>
<tr>
<td>M. melanochephalus melanochephalus</td>
<td>39 (39)</td>
<td>3.8 ± 0.6</td>
<td>6.3 ± 0.3</td>
<td>10.5 ± 0.4</td>
<td>3.0 ± 0.5</td>
<td>7.5 ± 0.6</td>
<td>0.86 ± 0.15</td>
<td>37.7 ± 6.3</td>
<td>56 ± 10.7</td>
<td>14.7 ± 2.1</td>
<td>14.4 ± 2.4</td>
<td>0.26 ± 0.05</td>
</tr>
<tr>
<td>M. pulcherrimus</td>
<td>6 (6)</td>
<td>2.9 ± 0.3</td>
<td>5.8 ± 0.3</td>
<td>9.2 ± 0.4</td>
<td>2.6 ± 0.5</td>
<td>6.6 ± 0.6</td>
<td>0.80 ± 0.17</td>
<td>36.4 ± 6.4</td>
<td>39 ± 10.6</td>
<td>13.2 ± 2.6</td>
<td>10.0 ± 2.5</td>
<td>0.27 ± 0.08</td>
</tr>
<tr>
<td>M. splendens splendens</td>
<td>4 (4)*</td>
<td>3.3 ± 0.8</td>
<td>5.6 ± 0.3</td>
<td>7.9 ± 0.7</td>
<td>2.3 ± 0.6</td>
<td>5.5 ± 1.0</td>
<td>0.89 ± 0.18</td>
<td>52.8 ± 11.8</td>
<td>42 ± 10.9</td>
<td>12.6 ± 1.9</td>
<td>9.8 ± 2.2</td>
<td>0.24 ± 0.05</td>
</tr>
<tr>
<td>M. splendens emmottorun</td>
<td>5 (5)*</td>
<td>4.0 ± 0.9</td>
<td>4.8 ± 0.4</td>
<td>8.1 ± 0.7</td>
<td>2.1 ± 0.2</td>
<td>6.0 ± 0.7</td>
<td>0.48 ± 0.13</td>
<td>47.8 ± 5.5</td>
<td>53 ± 11.4</td>
<td>13.3 ± 1.2</td>
<td>9.2 ± 1.3</td>
<td>0.18 ± 0.02</td>
</tr>
<tr>
<td>M. splendens melanotus</td>
<td>12 (12)</td>
<td>3.0 ± 0.4</td>
<td>4.9 ± 0.4</td>
<td>8.0 ± 1.0</td>
<td>2.4 ± 0.5</td>
<td>5.5 ± 0.7</td>
<td>0.67 ± 0.39</td>
<td>45.2 ± 4.1</td>
<td>45 ± 8.9</td>
<td>14.9 ± 1.5</td>
<td>9.3 ± 1.9</td>
<td>0.21 ± 0.04</td>
</tr>
<tr>
<td>A. striatus striatus</td>
<td>5 (1)</td>
<td>3.0 ± 1.7</td>
<td>6.7 ± 1.2</td>
<td>11.6 ± 0.3</td>
<td>1.3 ± 0.2</td>
<td>10.3 ± 0.5</td>
<td>1.00 ± 0.34</td>
<td>65.6 ± 7.3</td>
<td>20 ± 10.3</td>
<td>6.7 ± 0.8</td>
<td>6.2 ± 0.8</td>
<td>0.39 ± 0.24</td>
</tr>
<tr>
<td>A. merrotsyi</td>
<td>9 (1)*</td>
<td>2.7 ± 1.2</td>
<td>7.5 ± 0.3</td>
<td>11.7 ± 0.4</td>
<td>2.3 ± 0.4</td>
<td>9.4 ± 0.4</td>
<td>1.34 ± 0.36</td>
<td>79.3 ± 12.5</td>
<td>18 ± 6.9</td>
<td>6.8 ± 1.0</td>
<td>5.9 ± 1.6</td>
<td>0.36 ± 0.13</td>
</tr>
<tr>
<td>S. malachurus</td>
<td>3 (1)</td>
<td>3.0 ± 0.4</td>
<td>8.0 ± 0.1</td>
<td>11.2 ± 0.0</td>
<td>4.7 ± 0.3</td>
<td>6.4 ± 0.4</td>
<td>0.64 ± 0.07</td>
<td>58.7 ± 2.0</td>
<td>35 ± 2.5</td>
<td>11.8 ± 0.8</td>
<td>8.0 ± 1.0</td>
<td>0.23 ± 0.02</td>
</tr>
<tr>
<td>S. mallee</td>
<td>3 (2)*</td>
<td>4.0 ± 1.1</td>
<td>7.2 ± 0.3</td>
<td>10.5 ± 1.3</td>
<td>2.5 ± 0.2</td>
<td>8.0 ± 1.5</td>
<td>1.13 ± 0.37</td>
<td>89.4 ± 9.8</td>
<td>26 ± 5.9</td>
<td>6.5 ± 0.5</td>
<td>7.3 ± 0.6</td>
<td>0.30 ± 0.10</td>
</tr>
<tr>
<td>Spodotus wallacii</td>
<td>2 (2)</td>
<td>1.6 ± 0.6</td>
<td>8.4 ± 0.0</td>
<td>10.1 ± 0.9</td>
<td>5.2 ± 0.4</td>
<td>4.9 ± 0.5</td>
<td>0.62 ± 0.03</td>
<td>71.8 ± 11.4</td>
<td>16 ± 8.5</td>
<td>9.8 ± 1.9</td>
<td>3.0 ± 0.0</td>
<td>0.22 ± 0.12</td>
</tr>
</tbody>
</table>
statistical dependence of traits (Pagel 1999; Freckleton et al. 2002). We ran two
iterations of this analysis, one with all 16 taxa for which we had
body-weight and latitude data (using those two factors as pre-
dictor variables), and one with the subset of 14 taxa for which we had
body-weight, body-weight and latitude data (with all three
factors as predictor variables).

To complement our estimations of $\lambda$, we also calculated the
descriptive statistic $K$ for all song traits ($n = 16$ taxa) to compare
the relative amount of convergence or divergence among traits
(Blomberg et al. 2003). Values of $K$ less than 1 indicate that
closely related taxa are more different than would be expected by
chance under a Brownian motion model of evolution, and values
of $K$ greater than 1 indicate that related taxa are more similar than
would be expected under a Brownian motion model (Blomberg
et al. 2003). We used the software package Picante (Kembl et al.
2010) implemented in R to calculate $K$ for all traits, and we used
the randomisation procedure implemented with the function
‘phylosignal’ (using 10,000 repetitions) to determine whether
traits exhibited significantly more similarity between related
individuals than would be expected by chance.

We reconstructed evolutionary changes in song on the phy-
logeny using MacClade 4.08 (Maddison and Maddison 2005).
For this analysis, we converted our measurements of continuous
song features into discrete, ordered characters by plotting means
and standard errors for taxa and then dividing these measures into
states where error bars did not overlap (see Price and Lanyon
2002, 2004 for more detailed explanations of this technique).
Only taxa for which a minimum of three song samples were
available (12 of the 16 taxa included in the phylogenetic
analysis: Table 2) were used in the defining of character states
to ensure a minimal degree of accuracy in the placement of error
bars, and divisions between states were positioned equidistant
from nearest error bars. We then scored all taxa based on mean
measurements of song features, including those with fewer than
three representative songs, and mapped these discrete character
states onto the tree using simple parsimony. This method
allowed us to identify statistically discontinuous evolutionary
changes in song patterns while controlling for within-taxon
variability. All mean measurements based on fewer than three
songs fell within the character states calculated using other taxa,
so these measurements would not have generated additional
character states.

As will be evident below, the results of our analyses examining
factors related to song variation varied depending on whether we
included two variables (body-weight and latitude) across 16 taxa
or three variables (body-weight, testes mass and latitude) across
14 taxa, as well as whether we consider the results of the
regression analysis directly or adjusted the table-wide signifi-
cance values with a Bonferroni correction. In the results
section below, we present the results based on the regression analysis,
but note when these results are questioned due to the Bonferroni
correction.

Our habitat classifications for each taxon were very general
and do not include variation in microhabitats or intraspecific
variation in habitat use. Thus, we did not conduct a thorough
analysis of habitat in relation to acoustic characteristics. How-
ever, as a preliminary assessment of song variation in relation to
habitat type, we used one-way ANOVAs to compare mean song
trait values across habitat types.

Results
Song variables differed considerably among the 21 taxa for
which we had recordings (ANOVA: $n = 184$ songs, $P < 0.001$
for all 11 variables in Table 2). Naturally, several song variables
were correlated with one another (Table 3). For example, the
number of notes, mean note rate and mean note length in a song
were highly correlated with each other. Thus, songs tended to be
composed of either many short, quickly repeated notes or fewer
long, widely spaced notes. Song bandwidth was strongly and
positively correlated with the highest frequencies but not the
lowest frequencies of songs, suggesting that variation in song
bandwidth among species was largely explained by the presence
or absence of high-frequency notes.

Most song traits were significantly more similar between
closely related taxa than would be expected by chance (i.e. more
similar than if trait values were distributed randomly on the tree),
but all had values of $K$ that were less than 1 (Table 4), indicating
that closely related taxa tended to be more divergent in these
characteristics than would be expected under a Brownian motion
model of evolution. Our maximum-likelihood estimates of $\lambda$
for all song traits except note bandwidth were significantly different
from 1 but not significantly different from 0 (Table 5), indicating
some degree of phylogenetic independence of these traits. It
should be noted, however, that estimates of $K$ and $\lambda$ were not
always consistent; in some cases traits exhibited significant
phylogenetic signal based on analysis with ‘phylosignal’, but
not in our estimates of $\lambda$ in multiple regression models (Tables 4, 5, 6).

In both regression models, body-weight was strongly and negatively correlated with lowest song frequency ($P=0.002$ for the model including 16 taxa, $P=0.026$ for the model including 14 taxa: Tables 5, 6, Fig. 1 top), indicating that in larger species, males tended to sing lower-frequency songs. This relationship remained significant in the analysis with 16 taxa after sequential Bonferroni correction for 22 comparisons (Holm 1979), but not in the analysis with 14 taxa and 33 comparisons (Tables 5, 6). Body-weight was not a significant predictor of any other song variable in the analysis with 16 taxa, and although body-weight showed significant relationships with some song traits in the analysis with 14 taxa, these relationships were not significant after sequential Bonferroni correction. Overall, therefore, body-weight was associated with the low frequency of songs but not strongly associated with other song variables.

Mean latitude was a significant positive predictor of song versatility in both models ($P=0.004$ for the model including 16 taxa, $P=0.003$ for the model including 14 taxa: Tables 5, 6, Fig. 1 bottom). Thus, more complex songs tended to occur at higher latitudes (i.e. farther south), although these relationships did not remain significant after sequential Bonferroni correction for multiple comparisons. Mean latitude was also significantly associated with note bandwidth; however, this was only in the model with 16 taxa ($P=0.001$ for the model including 16 taxa, which remained significant after sequential Bonferroni correction, $P=0.141$ for the model including 14 taxa: Tables 5, 6). Mean latitude was associated with song length, note length and number of notes (i.e. tempo-related song traits) in the model including 14 taxa, but none of these relationships remained significant after sequential Bonferroni correction.

Testes mass was a significant predictor of song structure for several song traits, including the length, number, rate and variety of notes in a song (Table 6). Taxa with larger testes therefore tended to produce songs with shorter, more rapidly repeated notes that included more note types. Additionally, these song traits had values of $K<1$ and did not exhibit significant similarities between related individuals (i.e. traits appeared to be distributed randomly on the tree relative to phylogenetic relationships). This departure from a Brownian model of evolution could potentially indicate the presence of selection on these song traits. Larger testes mass was also significantly correlated with lower peak frequencies ($P=0.016$), and a trend existed between testes mass and the highest frequency ($P=0.058$) and lowest frequency ($P=0.059$) components of songs. All relationships with testes mass were, however, not significant after sequential Bonferroni correction.

Historical reconstructions of song evolution based on unambiguous discrete changes complement these analyses (Fig. 2) and indicate the importance of latitude in song evolution. Although changes in song structure have occurred throughout the evolution of the clade (Fig. 2), most changes appear to be associated with historical movements between tropical and temperate environments. For example, the songs of *M. anabilibis* and *M. coronatus* (both tropical species) differ in a variety of ways from songs of closely related taxa with ranges farther south (*M. lamberti*, *M. pulcherrimus* and *M. elegans*). Similarly, *M. alboscapulatus* (tropical) and *M. melanopechus* (subtropical) differ from their sister taxon *M. leucopterus* (a temperate species), and *M. cyanopechus* (distributed in New Guinea) has songs that are strikingly different from any Australian species (see spectrograms in Fig. 2). Songs from *S. wallacii*, another species distributed in New Guinea, appeared visibly different from other taxa in spectrograms (Fig. 2), but our quantitative analyses did not reveal any large historical changes in this lineage explaining these differences. Song versatility (SV), a trait shown

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**Table 3. Correlation coefficients ($r$) between all song variables across all taxa (n = 184 songs)**

Strong correlations ($r > 0.500$) are highlighted in bold.

<table>
<thead>
<tr>
<th>Song variable</th>
<th>Peak frequency</th>
<th>High frequency</th>
<th>Low frequency</th>
<th>Song bandwidth</th>
<th>Note bandwidth</th>
<th>Note length</th>
<th>Number of notes</th>
<th>Note rate</th>
<th>Note variety</th>
</tr>
</thead>
<tbody>
<tr>
<td>Peak frequency</td>
<td>–0.351</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>High frequency</td>
<td>–0.307</td>
<td>0.938</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>Low frequency</td>
<td>–0.291</td>
<td>0.909</td>
<td>0.745</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>Song bandwidth</td>
<td>–0.007</td>
<td>0.575</td>
<td>0.706</td>
<td>0.405</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>Note bandwidth</td>
<td>–0.148</td>
<td>0.302</td>
<td>0.433</td>
<td>0.037</td>
<td>0.381</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>Note length</td>
<td>–0.165</td>
<td>0.064</td>
<td>0.223</td>
<td>–0.120</td>
<td>0.041</td>
<td>0.173</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>Number of notes</td>
<td>0.623</td>
<td>–0.364</td>
<td>–0.446</td>
<td>–0.188</td>
<td>–0.119</td>
<td>–0.192</td>
<td>–0.708</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>Note rate</td>
<td>0.191</td>
<td>–0.278</td>
<td>–0.432</td>
<td>–0.074</td>
<td>–0.219</td>
<td>–0.153</td>
<td>–0.852</td>
<td>0.853</td>
<td>–</td>
</tr>
<tr>
<td>Note variety</td>
<td>0.174</td>
<td>0.188</td>
<td>0.075</td>
<td>0.271</td>
<td>0.246</td>
<td>0.069</td>
<td>–0.586</td>
<td>0.376</td>
<td>0.436</td>
</tr>
<tr>
<td>Song versatility</td>
<td>–0.561</td>
<td>0.544</td>
<td>0.589</td>
<td>0.420</td>
<td>0.452</td>
<td>0.330</td>
<td>0.335</td>
<td>–0.615</td>
<td>–0.497</td>
</tr>
</tbody>
</table>

---

**Table 4. Values of Blomberg’s $K$ for all song variables and results of randomisation test assessing whether traits were more similar in related individuals than would be expected by chance (i.e. if traits were distributed randomly on the tree)**

Significant $P$-values ($P < 0.05$) are highlighted in bold. Values of $K < 1$ indicate that relatives are less similar than would be expected under a Brownian motion model of evolution.

<table>
<thead>
<tr>
<th>Song variable</th>
<th>$K$</th>
<th>$Z$</th>
<th>$P$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Song length</td>
<td>0.75</td>
<td>–1.57</td>
<td>0.012</td>
</tr>
<tr>
<td>Peak frequency</td>
<td>0.73</td>
<td>–1.49</td>
<td>0.007</td>
</tr>
<tr>
<td>High frequency</td>
<td>0.86</td>
<td>–1.24</td>
<td>0.024</td>
</tr>
<tr>
<td>Low frequency</td>
<td>0.64</td>
<td>–1.58</td>
<td>0.008</td>
</tr>
<tr>
<td>Song bandwidth</td>
<td>0.84</td>
<td>–1.41</td>
<td>0.005</td>
</tr>
<tr>
<td>Note bandwidth</td>
<td>0.52</td>
<td>–0.54</td>
<td>0.339</td>
</tr>
<tr>
<td>Note length</td>
<td>0.61</td>
<td>–1.02</td>
<td>0.104</td>
</tr>
<tr>
<td>Number of notes</td>
<td>0.74</td>
<td>–0.82</td>
<td>0.219</td>
</tr>
<tr>
<td>Note rate</td>
<td>0.58</td>
<td>–0.99</td>
<td>0.130</td>
</tr>
<tr>
<td>Note variety</td>
<td>0.64</td>
<td>–1.33</td>
<td>0.045</td>
</tr>
<tr>
<td>Song versatility</td>
<td>0.68</td>
<td>–1.42</td>
<td>0.026</td>
</tr>
</tbody>
</table>
to correlate significantly with latitude, decreased in at least three separate lineages (M. alboscapulatus, M. coronatus and M. cyanocephalus), and note bandwidth (NB) also decreased in two of these taxa. Each of these changes clearly corresponded with movements from temperate to tropical breeding latitudes. Despite the relationships between song traits and latitude, we found little evidence for an association of song traits and habitat. No song trait showed significant variation among habitats (P > 0.10 for all 11 traits).

Song changes have also been associated with changes in morphology. For example, lowest frequencies (LF) have decreased in the songs of the Maluridae (Fig. 2), and both of these species have large body-sizes in comparison to congeners (Table 1). Changes in song characters also complement correlations between song traits described in Table 3; for example, all four changes in note length (NL) were associated with changes in note rate (NR), and these variables always changed in opposite directions, with increases in note lengths occurring with decreases in note rates and vice versa.

### Discussion

Signal evolution in any family of birds is obviously a complex interaction of selection pressures and responses to environmental variability, and interspecific variation in the songs of the Maluridae illustrates this complex interaction. Species in the Maluridae experience variation along several gradients that should relate to signal evolution. First, with respect to geographical distribution, malurid species occur from near the equator in New Guinea south to a latitude of ~42°S. Second, with respect to ecology, species occur in diverse habitats ranging from primary rainforests to open and desolate deserts in Australia. Third, with respect to mating behaviour, although all of the species are known to be socially monogamous, the extent of reproductive promiscuity in this one family varies as much as it does across all other birds of the world combined, from almost non-existent to more than 90% of broods. Natural selection due to environmental factors (latitude and habitat) should be expected to influence interspecific variation in song structure (e.g. Wiley 1991; Botero et al. 2009; Weir and Wheatcroft 2011); however, the effects of extra-pair paternity on song evolution are less well understood (Garamszegi and Hu 2011; Soma and Garamszegi 2011). Nevertheless, if song characteristics are important in mate choice and reflect rates of extra-pair paternity within species, interspecific variation in promiscuity might also be reflected in song variation across species.

We found that fairy-wrens, grasswrens and emu-wrens varied significantly in the acoustic structure of their songs and exhibited a high degree of divergence in most song traits. Estimates of K as well as reconstructions of evolutionary changes on the phylogeny suggest that many traits have departed from Brownian motion and may have been influenced by selection (Blomberg et al. 2003). This was complemented by our estimates of λ, which for most acoustic traits were not significantly different from 0 but were significantly different from 1, indicating phylogenetic independence for most traits.

Our result that low frequencies were influenced by body-size (Fig. 1 top, Tables 5, 6) complements other work comparing body-size and sound frequencies in bird song, which suggest that

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**Table 5. Correlations of song variables with body-weight and mean breeding latitudes in multiple regression models (n = 16 taxa)**

<table>
<thead>
<tr>
<th>Song variable</th>
<th>R²</th>
<th>F</th>
<th>λ</th>
<th>P (λ = 0)</th>
<th>P (λ = 1)</th>
<th>Predictor</th>
<th>Slope</th>
<th>t</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Song length</td>
<td>0.38</td>
<td>3.92</td>
<td>0.00</td>
<td>1.000</td>
<td>0.001</td>
<td>Body</td>
<td>1.32</td>
<td>1.30</td>
<td>0.215</td>
</tr>
<tr>
<td>Peak frequency</td>
<td>0.18</td>
<td>1.43</td>
<td>0.00</td>
<td>1.000</td>
<td>0.001</td>
<td>Body</td>
<td>−2.50</td>
<td>−1.57</td>
<td>0.141</td>
</tr>
<tr>
<td>High frequency</td>
<td>0.02</td>
<td>0.16</td>
<td>0.00</td>
<td>1.000</td>
<td>0.005</td>
<td>Body</td>
<td>−1.09</td>
<td>−0.50</td>
<td>0.626</td>
</tr>
<tr>
<td>Low frequency</td>
<td>0.52</td>
<td>7.17</td>
<td>0.00</td>
<td>1.000</td>
<td>0.000</td>
<td>Body</td>
<td>−3.40</td>
<td>−3.78</td>
<td>0.002*</td>
</tr>
<tr>
<td>Song bandwidth</td>
<td>0.11</td>
<td>0.84</td>
<td>0.00</td>
<td>1.000</td>
<td>0.021</td>
<td>Body</td>
<td>2.31</td>
<td>1.28</td>
<td>0.223</td>
</tr>
<tr>
<td>Note bandwidth</td>
<td>0.64</td>
<td>7.15</td>
<td>0.98</td>
<td>0.014</td>
<td>0.859</td>
<td>Body</td>
<td>−0.04</td>
<td>−0.20</td>
<td>0.845</td>
</tr>
<tr>
<td>Note length</td>
<td>0.22</td>
<td>1.87</td>
<td>0.51</td>
<td>0.177</td>
<td>0.010</td>
<td>Body</td>
<td>40.73</td>
<td>1.74</td>
<td>0.106</td>
</tr>
<tr>
<td>Number of notes</td>
<td>0.13</td>
<td>0.98</td>
<td>0.00</td>
<td>1.000</td>
<td>0.000</td>
<td>Body</td>
<td>−25.80</td>
<td>−9.91</td>
<td>0.381</td>
</tr>
<tr>
<td>Note rate</td>
<td>0.26</td>
<td>2.34</td>
<td>0.00</td>
<td>1.000</td>
<td>0.000</td>
<td>Body</td>
<td>−10.55</td>
<td>−2.15</td>
<td>0.051</td>
</tr>
<tr>
<td>Note variety</td>
<td>0.16</td>
<td>1.22</td>
<td>0.22</td>
<td>0.824</td>
<td>0.037</td>
<td>Body</td>
<td>−3.63</td>
<td>−0.95</td>
<td>0.359</td>
</tr>
<tr>
<td>Song versatility</td>
<td>0.49</td>
<td>6.15</td>
<td>0.00</td>
<td>1.000</td>
<td>0.000</td>
<td>Body</td>
<td>0.08</td>
<td>1.02</td>
<td>0.324</td>
</tr>
</tbody>
</table>

*Significant P-values (P < 0.05) are highlighted in bold. Asterisks indicate relationships that remained significant after sequential Bonferroni correction for 22 comparisons. Maximum-likelihood estimates of λ are shown and compared to models in which λ is set to 0 or 1.
Song length $0.45$ | $2.74$ | $1.00$ | $0.242$ | $1.000$ | Body $-0.76$ | $-1.25$ | $0.240$  
| Testes $0.33$ | $1.22$ | $0.250$  
| Latitude $-0.04$ | $-2.24$ | $0.049$  

Peak frequency $0.60$ | $0.02$ | $0.00$ | $1.000$ | $0.016$ | Body $0.33$ | $0.28$ | $0.788$  
| Testes $-1.05$ | $-2.90$ | $0.016$  
| Latitude $0.02$ | $1.12$ | $0.290$  

High frequency $0.35$ | $1.80$ | $0.00$ | $1.000$ | $0.020$ | Body $1.82$ | $1.25$ | $0.240$  
| Testes $-0.94$ | $-2.14$ | $0.058$  
| Latitude $0.02$ | $0.70$ | $0.497$  

Low frequency $0.71$ | $8.18$ | $0.71$ | $0.302$ | $0.145$ | Body $-1.77$ | $-2.61$ | $0.026$  
| Testes $-0.54$ | $-2.13$ | $0.059$  
| Latitude $0.01$ | $0.87$ | $0.403$  

Song bandwidth $0.61$ | $5.29$ | $0.00$ | $1.000$ | $0.006$ | Body $3.88$ | $3.83$ | $0.003$  
| Testes $-0.27$ | $-0.88$ | $0.398$  
| Latitude $0.02$ | $1.37$ | $0.200$  

Note bandwidth $0.27$ | $1.23$ | $0.93$ | $0.011$ | $0.465$ | Body $0.17$ | $0.85$ | $0.415$  
| Testes $-0.06$ | $-0.65$ | $0.528$  
| Latitude $0.01$ | $1.60$ | $0.141$  

Note length $0.69$ | $7.47$ | $0.00$ | $1.000$ | $0.026$ | Body $35.20$ | $3.23$ | $0.009$  
| Testes $-10.65$ | $-3.24$ | $0.009$  
| Latitude $0.51$ | $3.14$ | $0.011$  

Number of notes $0.53$ | $3.74$ | $0.00$ | $1.000$ | $0.047$ | Body $-52.08$ | $-2.23$ | $0.050$  
| Testes $15.82$ | $2.24$ | $0.049$  
| Latitude $-0.79$ | $-2.27$ | $0.046$  

Note rate $0.64$ | $5.81$ | $0.00$ | $1.000$ | $0.041$ | Body $-14.39$ | $-3.90$ | $0.003$  
| Testes $3.33$ | $2.99$ | $0.013$  
| Latitude $-0.09$ | $-1.65$ | $0.130$  

Note variety $0.66$ | $6.33$ | $0.00$ | $1.000$ | $0.005$ | Body $-6.71$ | $-3.51$ | $0.006$  
| Testes $2.21$ | $3.82$ | $0.003$  
| Latitude $-0.03$ | $-1.22$ | $0.250$  

Song versatility $0.60$ | $5.09$ | $0.00$ | $1.000$ | $0.037$ | Body $0.15$ | $2.05$ | $0.067$  
| Testes $-0.01$ | $-0.30$ | $0.774$  
| Latitude $0.00$ | $3.85$ | $0.003$  

Table 6. Correlations of song variables with body-weight, mean breeding latitudes and combined testes mass in multiple regression models ($n=14$ taxa)

Significant $P$-values ($P<0.05$) are highlighted in bold. No relationships remained significant after sequential Bonferroni correction for 33 comparisons. Maximum-likelihood estimates of $\lambda$ are shown and are compared to models in which $\lambda$ is set at 0 or 1.
M. melanocephalus, M. leucopterus and M. splendens, which have large testes mass (Table 1) and reasonably note-rich songs. Note length, number of notes and note rate have K values that are <1 and the traits do not exhibit significant phylogenetic signal (i.e. related taxa are not more similar than if traits were distributed randomly on the tree), suggesting the possible presence of selection on these traits. Additionally, maximum-likelihood estimates of λ for these traits indicated phylogenetic independence (Tables 5, 6). Although note rate, number and length are not necessarily aspects of song performance per se (e.g. Podos 1997; Ballentine et al. 2004; Cardoso et al. 2009), acoustic features such as these may be associated with varying levels of song difficulty or stimulatory potential (e.g. Vallet et al. 1998) and are reasonable candidates for sexually selected song traits in the Maluridae.

The lack of a clear relationship between song versatility and relative testes mass suggests that sexual selection plays little role in this aspect of song. In one sense, this is surprising because vocalisations and vocal complexity appear to be so critical to the integrity of social groups of fairy-wrens and, by extension, to the process of mate choice and male–male competition. Nevertheless, the importance of song versatility in social behaviour may not necessarily translate to variation among taxa that have different rates of extra-pair paternity, especially if this song characteristic has multiple functions. Other factors, such as note number or rate, might play more direct roles in reproductive success, leading to interspecific differences. Furthermore, previous phylogenetic studies have shown that, although sexual selection can cause dramatic evolutionary changes in song, no single aspect of song is clearly associated with variation in mating systems (Price and Lanyon 2004). Complexity may instead change stochastically, particularly in learned bird songs where copying errors and improvisation occur (e.g. Payne 1996; Kroodsma et al. 1999; Podos et al. 1999; Johnson 2006). Thus, our findings suggest that there may be many solutions to the problem of effectively advertising to potential mates and competing with rivals.

One important aspect of interspecific variation in songs in fairy-wrens that we did not analyse here is the use of predator-elicited Type II songs by males. In Superb Fairy-wrens (M. cyaneus) (Langmore and Mulder 1992), Splendid Fairy-wrens (M. splendens melanotus) (Zelano et al. 2001, Greig and Pruett-Jones 2009, 2010) and Variegated Fairy-wrens (M. lamberti assimilis) (Greig et al. 2010), males give Type II songs in response to hearing predators call. Greig and Pruett-Jones (2009, 2010) showed that in Splendid Fairy-wrens this vocalisation is not an alarm call but rather a display vocalisation that is given when conspecifics are particularly alert (i.e. in the presence of predators). In a broad, interspecific comparison Greig and Webster (unpublished data) have shown that males of many other species of Malurus in Australia respond to hearing predators by giving a vocalisation similar to those described above. The species that occur in New Guinea have not been surveyed with respect to this vocalisation, and thus it is not clear whether it is a trait common to many members of the subfamily Malurinae or whether it is unique to the Australian taxa. Overall, predator-elicited trills may be an important sexual or social signal and a thorough study of the evolution of this song type would be valuable.

Although our focus here was on interspecific variation, it is becoming increasingly clear that the study of song dynamics within species is yielding remarkable insights into communication in fairy-wrens (Hall and Peters 2008; Cockburn et al. 2009, Greig et al. 2012, Dowling and Webster 2013, Kleindorfer et al. 2013). Song in males, besides having an obvious territorial function, is implicated in aspects of mate choice (Cockburn et al. 2009; Greig 2010), although a specific correlation between song and mating success, either within-pair or extra-pair, has yet to be determined. Similarly, although female song has been studied in two species (Cooney and Cockburn 1995; Hall and Peters 2008; Kleindorfer et al. 2013), the fact that female song is such a prominent feature of communication in fairy-wrens suggests that this would be an extremely fruitful area for future research. Finally, intraspecific geographic variation in songs and associated conspecific responses of Red-backed and Superb Fairy-wrens suggests that song may be an important isolating mechanism between divergent populations (Greig and Webster 2013; Kleindorfer et al. 2013).

In summary, songs in Maluridae are influenced by morphological constraints and show some clear patterns of evolution in response to environmental factors. The influences of sexual selection are less clear, but such influences are undoubtedly complex given the variety of social functions of song in this system. Our findings complement previous comparative and meta-analyses that suggest that patterns of directional song evolution in response to sexual selection are not straightforward (Garamszegi and Moller 2004; Price and Lanyon 2004; Cardoso and Hu 2011; Soma and Garamszegi 2011). The correlation we
observed between aspects of song complexity and latitude complements work in other avian systems that suggests that higher latitudes support more variable, versatile or complex songs (e.g. Weir and Wheatcroft 2011), for example by a general 'acoustic release' if higher latitudes have lower species abundance (Podos et al. 2004), or if acoustic versatility is advanta-
geous in climatic conditions that are more variable (Botero et al. 2009). Finally, song frequency characteristics in Maluridae appear to be constrained by body-size in a manner similar to other avian taxa. Overall, therefore, there appear to be a variety of both natural and social selective pressures that have influenced songs in Maluridae, and these factors interact in complex ways to create the acoustic phenotypes characteristic of this avian family.

Acknowledgements

For assistance in the field, logistical or financial support, and advice or discussion we thank Mike Webster, Melissa Rowe, Michelle Hall, Ben Taft, Dan Baldassarre, Becky Cramer, Jenélle Dowling, Sara Kaiser, Shalene Shah, Duncan Yandell, Joanne Heathcote, John and Lisa at Dryandra, Lyanne Brouwer, Marina and Simon, Tim Daniels, Matt Medler, Jan Lewis, Doug Barron, Karen and Angus Emmott, Graham Chapman, Leslie and Michael Brooker, Mornington Wildlife Sanctuary, Macaulay Library of Natural Sounds, Alex White, Nick Brandley, Bryce Masuda and Karon Odom. Financial support was provided by the National Science Foundation (MSW and SP-J), the Cornell Laboratory of Ornithology, the University of Chicago Hinds Fund, American Ornithologists’ Union, Animal Behaviour Society, Frank M. Chapman Memorial Fund and GAANN National Training Fellowship (EO). All work was conducted with approval from appropriate animal ethics and permitting agencies (University of Chicago Animal Care and Use Committee approval #71708, Cornell University Animal Care and Use Committee approval 2009-0105, James Cook University Ethics approval #A1340, Scientific Purposes Permit #WISP07773610, Regulation 17 licence #SF007698).

References


