7-14-2009

Genetic structuring at a fine scale in the russet-crowned motmot (*Momotus mexicanus*) in a tropical dry forest in central Mexico

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The isolation-by-distance model of genetic structuring of populations predicts that genetic similarity between populations will decrease with increased geographic separation as the homogenizing influence of gene flow diminishes (Wright 1943). Dispersal is central to the demographic and evolutionary processes that shape genetic variation of bird populations (Sonsthagen et al. 2004). Because of their potential for long-distance flight, birds are generally believed to exhibit high levels of gene flow and low genetic structuring (Crochet 2000, McDonald 2003). Consequently, high genetic differentiation and isolation by distance may be expected to occur only at relatively large geographic scales (e.g., populations separated by at least 1000 km; Abbott et al. 2002). Nevertheless, site fidelity, which is common in birds (Greenwood 1980), may be expected to reduce genetic mixing at local scales and increase genetic structuring (Matthiopoulos et al. 2005).

In many tropical terrestrial birds, breeding sites are used also for foraging, roosting, and other maintenance activities (Stutchbury and Morton 2001). We envision a situation in which factors like site quality, previous breeding performance, or a knowledge of food sources, shelters, and interactions with predators could favor the development of high site fidelity through their effects on fitness (Greenwood 1980, Sedgwick 2004). Russet-crowned Motmots are year-round resident birds of the American tropics that use the same territories to breed and forage (Paniagua 2005). Because motmots make tunnel-nests in earthen banks (Howell and Webb 1995) vulnerable to floods, reproductive success depends critically on the selection of good-quality breeding sites (Paniagua 2005), which would favor the return to traditional nesting sites over successive years. We would expect species with strong nest site fidelity, like motmots, to show more-pronounced genetic structuring of their populations than species with weak nest site fidelity. In this context, we estimated the genetic structure of 3 populations of the territorial Russet-crowned Motmot (Momotus mexicanus) at a local scale in a tropical dry forest of central Mexico.

 METHODS

From April to August 2001, a total of 49 adult motmots were captured with mist nets at 3 breeding sites (Cruz Pintada, El Limón, and Tilzapotla) in the dry forest of the Reserva de la Biodiversidad de la Sierra de Huautla (RBSH) in central Mexico. The breeding sites were separated by a mean straight-line distance of 25 km (Fig. 1). We sampled 20 birds in Cruz Pintada, 14 in El Limón, and 15 in Tilzapotla. Blood samples were obtained from brachial
venipuncture using a disposable, sterile 1-mL hypodermic needle. Approximately 100 μL of blood was mixed with 1 mL of cell lysis buffer [700 μL TN2E, 60 μL SDS 10%, 10 μL proteinase K (10 mg · mL⁻¹)]. Samples were kept on ice until they could be stored at –76 °C.

DNA was obtained using a standard phenol-chloroform extraction (Gibbs et al. 1994) followed by ethanol precipitation and resuspension in 50 μL of Tris-EDTA buffer. DNA was quantified fluorimetrically (Gene Quant Pro, GE Healthcare, CT, USA). RAPD assays were performed in 25 μL total volume containing 1 ng · μL⁻¹ DNA template, Buffer 1X (Acetamide 50%, 300 mM Tricine, 500 mM KCL), 20 mM MgCl₂, 100 μM of each dNTP, 0.2 u Taq polymerase (GibcoRBL, MD, USA), 1 μM of primer (Operon Technologies, CA, USA). The reaction mixture was amplified with a PTC-100 thermocycler (MJ-Research, MA, USA) programmed as follows: 1 cycle of 1 minute at 94 °C, 44 cycles of 1 minute at 94°C, 2 minutes at 36 °C, and 2 minutes at 72°C, and 7 minutes at 72 °C. PCR products were analyzed by electrophoresis using 1.5% agarose gel for 1 hour at 100 V. Gels were stained with ethidium bromide and photographed under UV light. Twenty-five primers were tested. In order to reduce biases in scoring patterns, all reactions were amplified and scored twice to

Fig. 1. Study area in central Mexico. Dashed lines indicate geographic distance (km) between studied populations.
substantiate the existence of polymorphism and the reproducibility of bands. Bands were independently scored by 2 persons. Negative controls that contained no DNA were included in each PCR to assess for contamination. Nineteen primers revealed scarce polymorphisms or produced nonreproducible banding patterns and were dropped from further analyses. Primers OPA-01, OPA-07, OPB-08, OPB-11, OPB-15, and OPD-02 were selected for their ability to amplify clear, consistent, and polymorphic products.

Equally sized band fragments amplified with the same primer were interpreted as homologous products. Analyses of molecular variance (AMOVA; Excoffier et al. 1992) were performed on the RAPD phenotypes; this method partitions the total sum of squares into components representing variation among individuals within populations and among populations. The measure of population subdivision estimated by AMOVA is analogous to Wright’s $F_{ST}$. A series of 1000 permutations of individuals’ assignment to different populations was performed to estimate the level of significance of the fixation index. AMOVA was performed on a matrix of Euclidean distances previously obtained from the binary (0, 1) matrix with the software ARLEQUIN version 2.000 (Schneider et al. 2000). AMOVA also provides a measure of pairwise genetic differentiation. Indirect $F_{ST}$-derived estimates of gene flow were corrected for the number of subpopulations (Crow and Aoki 1984).

RESULTS

Analyses were performed on a subset of 83 loci of the whole data, as 17 loci were highly differentiated according to the criteria of Lynch and Milligan (1994). AMOVA revealed significant genetic differentiation among populations (Table 1). Differences among the 3 sites accounted for as much as 12.9% of the total genetic variation. Pairwise $F_{ST}$-based genetic differentiation was significant for all comparisons, $P < 0.0001$ (0.15 for El Limón vs. Cruz Pintada, 0.18 for El Limón vs. Tilzapotla, and 0.12 for Cruz Pintada vs. Tilzapotla). The $F_{ST}$ overall derived estimate of gene flow was 0.89 migrants per generation, with the pairwise estimates ranging from 0.56 to 0.91 migrants per generation.

DISCUSSION

The low genetic substructuring of populations commonly observed in birds reflects a demographic history characterized by extensive gene flow associated with high dispersal capacity (Crochet 2000, McDonald 2003). In stark contrast, Russet-crowned Motmots in central Mexico exhibited high genetic structuring, with almost 13% of the total RAPD genetic variance in our study area due to differences among breeding sites. This amount of genetic differentiation is exceedingly high considering the short distance (approximate average 25 km) between populations sampled.

Variation in life history can produce marked differences in dispersal capacity and, in turn, the genetic structuring of populations. For example, sedentary populations of the House Wren experience much-reduced gene flow compared to their migratory conspecifics (Arguedas and Parker 2000). Based on this result, we hypothesized that the evolution of high nest site fidelity in motmots could explain the high genetic differentiation observed in our study area.

Within their breeding range, Russet-crowned Motmots use a range of substrates to construct their nests, and nesting success is highly dependent on the physical properties of that substrate (Paniagua 2005). Suitable nest sites are limited, as tunnel-nests are vulnerable to floods. Nests that do not collapse tend to be located in scarce loam-textured soils (Paniagua 2009).
Thus, individuals might greatly benefit from discriminating among breeding areas and remaining at good-quality sites once these sites have been secured. These benefits would favor high site fidelity. In a recent study, 60% of previously marked birds nested in the same earthen bank at the El Limón breeding site (Osorio-Beristain unpublished observation). Also a bird banded 10 years ago was still in one of the breeding sites used in our study (T. Murphy, personal communication).

The Russet-crowned Motmot is broadly distributed along tropical deciduous forests from northwestern Mexico to Central America (Howell and Webb 1995). Thus, site fidelity is surely not the only factor affecting genetic structure at that geographic scale. This study is a first attempt at untangling factors that affect gene flow in this neotropical bird species. According to McDonald (2003), studies about neotropical birds are relatively scarce, particularly studies done at fine geographic scales. Our results do not support the hypothesis that genetic differentiation of bird populations will be apparent only at large geographic scales (see Abbott et al. 2002). Through future detailed multiyear studies of nesting success and return rates, we hope to support our interpretation of site fidelity as a strong determinant of the genetic structure of populations of Russet-crowned Motmots.

ACKNOWLEDGMENTS

This work was supported by El Consejo Nacional de Ciencia y Tecnología (project #32848 to Marcela Osorio-Beristain) and FOMES 2000. We thank E. Leyva for field and laboratory assistance, and E. Arellano and the group of the cuerpo académico Ecología Evolutiva at Universidad Autónoma del Estado de Morelos for their suggestions on this manuscript. We also thank F. Gonzalez, L. Kiere, D.B. McDonald, J. Hudon and 2 anonymous reviewers for their valuable comments that helped us to substantially improve the manuscript. This work complies with current Mexican laws.

LITERATURE CITED


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Received 20 February 2008
Accepted 8 October 2008