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Cooperative Breeding at a Nest of Slaty-backed Nightingale-Thrushes (*Catharus fuscater*)

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ABSTRACT.—Our observations on the reproductive habits of the Slaty-backed Nightingale-Thrush (*Catharus fuscater*) were made at a single nest between 2–12 November 2009 at an elevation of 2,050 m, in the vicinity of the Yanyacu Biological Station and Center of Creative Studies (00° 36′ S, 77° 53′ W), 5 km west of Cosanga (Napo Province, northeastern Ecuador). During the first 3 days following hatching, the only adult which provisioned nestlings was a color-banded female. Beginning with day 4, however, we observed five other individuals bringing food to the nest, including three color-banded males, one unmarked male, and one unmarked individual presumed to be female. The last two birds and one of the banded males were sexed using morphological differences, the remaining banded individuals were sexed molecularly. Most (72%) of provisioning visits to 4–9 day old nestlings were made by the color-banded female which also incubated the eggs. Our observations suggest the existence of a potentially

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complex cooperative breeding system in Slaty-backed Nightingale-Thrush. *Received 24 March 2014. Accepted 17 November 2014.*

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World-wide, more than 300 species of birds have been found to breed cooperatively, but among the ~140 species of thrushes (Turdidae), fewer than 5% have been reported to have helpers at the nest (Cockburn 2006). Among thrushes known to, at least occasionally, have helpers during reproduction, are two of the 12 species in the genus *Catharus* (Goetz et al. 2003, Halley and Heckscher 2012). The Slaty-backed Nightingale-Thrush (*Catharus fuscater*) is a non-migratory member of the genus which breeds from north-western Bolivia to Costa Rica, at elevations of 800–2600 m (Collar 2005). Despite its broad geographical range, its breeding biology is poorly known, with only one detailed study of nesting behavior (Beltrán and Kattan 2001, Collar 2005). Here, we provide the first documentation of more than two adults attending a nest of the Slaty-backed Nightingale-Thrush.

METHODS

We observed a nest of Slaty-backed Nightingale-Thrushes from 2–12 November 2009 at the Yanyacu Biological Station and Center of Creative Studies (00° 36′ S, 77° 53′ W, elevation 2,050 m). The station is located 5 km west of Cosanga, adjacent to the private reserve of Cabañas San Isidro, Napo Province, north-eastern Ecuador. AD made observations using 10x40 binoculars from a hide placed 10 m from the nest. The open-cup nest contained two eggs which hatched on 4 November. Observation periods ranged from 4–11 hrs daily, for a total of 70 hrs.

Sexes of Slaty-backed Nightingale-Thrush look alike but males have a brighter bill and a more orange eye-ring than females (Ridgely and Greenfield 2001, Collar 2005), which under field conditions can be difficult to assess. As part of an unrelated study, however, several of the birds observed in this study had been banded previously. To determine sex of the banded birds, we used brachial venipuncture to collect approximately 100–200 µl of whole blood from adult thrushes captured with mist nets. Blood was stored in 250–500 µl of Queen's lysis buffer (Seutin et al. 1991) depending on the amount of the blood sample. Blood was stored at ~20°C in the field and later at −20°C in the laboratory. We extracted whole genomic DNA from blood samples (200–300 µl and brought to a final volume of 500 µl with lysis buffer) collected in the field with standard phenol-chloroform-isoamyl based extractions. This was followed by ethanol precipitation and washes. After extraction, we stored genomic DNA at −20°C in 1x TE (pH 8.0). Genetic sexing was performed using a set of molecular primers (P2/P8, Griffiths et al. 1998). PCR reactions were performed in 10 µl of final volume using 5 µl 2X GoTaq Mastermix (Promega Corp., Madison, WI, USA), 1 µl 10 mM each of forward and reverse primer, 0.5 µl ultrapure H_20, and 2.5 µl genomic DNA.

The PCR reaction consisted of an initial denaturing step at 94 °C for 60 secs, followed by 30 cycles of denaturation (94 °C for 60 secs), annealing (48 °C for 60 secs) and extension (72 °C for 2 mins), and one final cycle of 72 °C for 10 mins. PCR products were separated in 3% agarose gels run in standard TAE buffer and visualized by ethidium bromide staining. Birds were sexed as females (heterogametic, WZ) when both fragments were amplified, and as males (homogametic, ZZ) when only a single band was visible.

RESULTS

We observed six individuals visit the nest. Four of these were color-banded individuals, and two were unmarked. We sexed three of the banded birds from blood taken at the time of banding to confirm that one (the individual observed incubating) was a female, and two were males. The fourth banded bird was sexed as male based on bill and eye-ring color. Of the two unmarked individuals, one showed the brighter eye-ring and bill of a male, while the second had a dark bill and dull-yellow eye-ring. The latter showed no signs of juvenal plumage and was in all ways similar to the individual confirmed as a female, but we cannot rule out the possibility that it was a juvenal male.

During 2 days of observation prior to the eggs hatching, we observed only the banded female incubating, but both of the molecularly-confirmed males perched on the rim of the nest for short periods, once each. During both male visits, the female was absent from the nest and the males peered briefly into the nest before flying away. During the first 3 days after the eggs hatched, we observed the nest for a total of 25.5 hrs, and the only adult we recorded provisioning the nestlings

was the same female that had been incubating. Beginning on day 4, however, we observed five other individuals bringing food to the nest at least once. Nestlings were fed mainly by the molecularly-sexed and banded female (150 provisioning visits, 72%, $n = 209$). The two molecularly-sexed males provisioned the nestlings 17 and 4 times, respectively. The two males that were sexed using morphological features fed the nestlings 10 and 8 times, respectively, and the un-sexed individual which had the morphological characteristics of a female fed the nestlings on 20 occasions.

DISCUSSION

Although the natural history of Slaty-backed Nightingale-Thrushes has been studied extensively in our area during the past 10 years (HFG, unpubl. data), color-banding of adults has just begun, and it is unknown how frequently nests are attended by multiple adults. Bicknell's Thrush (*Catharus bicknelli*) has a complex breeding system referred to as cooperative polyandry, whereby females occupy small, non-overlapping home ranges and mate with one or more males who assist in provisioning young (Goetz et al. 2003, Strong et al. 2004). Males, in contrast, occupy large home ranges that overlap extensively with those of both males and females, mate with multiple females, and feed multiple broods concurrently. The observations presented here suggest that the breeding system of the Slaty-backed Nightingale-Thrush may show a similar complexity, as is also suggested for a third congener, the Veery (*Catharus fuscescens*; Halley and Heckscher 2012). The breeding behavior of most tropical *Catharus* species is poorly studied, and cooperative breeding systems in the relatively well-studied northern species were discovered only recently (Goetz et al. 2003, Halley and Heckscher 2012), suggesting the possibility that cooperative breeding may be more wide-spread within this genus than previously recognized.

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