EXTRA-PAIR PATERNITY IN THE BLACK-CAPPED CHICKADEE[1]

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Studies of the mating strategies of birds are increasingly shifting their focus to the role that females take in controlling copulation (Hunter et al. 1993, Petrie 1992, Birkhead and Møller 1993). In some avian species, females appear able to control whether copulations occur by accepting or rejecting copulation attempts (Björklund et al. 1992, Lifjeld and Robertson 1992, Westneat 1992). Similarly, females of some species actively solicit copulations by seeking the extra-pair male in his own territory (Smith 1988, Kempenaers et al. 1992, Venier et al. 1993). In species where females do control the outcome of copulation attempts, there is a need to, first, determine the extent that extra-pair copulations (EPCs) lead to fertilizations and, second, establish potential benefits that a female may obtain by engaging in EPCs.

Smith (1988) reported that female Black-capped Chickadees, *Parus atricapillus,* seek EPCs from males on territories adjacent to the territory defended by the female's mate. Interestingly, Smith observed that females preferentially sought males for EPCs that held higher dominance rank than the female's mate in the preceding winter's flock. Thus, female Black-capped Chickadees may be engaging in a mixed reproductive strategy of social monogamy while increasing reproductive success by extra-pair copulations with males of higher genetic quality than their own mate (Trivers 1972, Smith 1988, Hamilton 1990). Females may also benefit from engaging in EPCs with dominant males if, by doing so, this facilitates rapid pair bonding with the dominant male in the event that his mate dies. There is a need for data on whether EPCs are resulting in extra-pair paternity of the nestlings in order to imply a fitness benefit to the female's behavior.

In this study we document rates of extra-pair paternity in an Ontario population of Black-capped Chickadees, using DNA fingerprinting, and provide support for Smith's hypothesis by showing that extra-pair males were of higher dominance rank than the female's mate. We also suggest a paternity-related explanation for occasional observations of polyandry in chickadees (Waterman et al. 1989, Howitz 1991).

METHODS

We conducted parentage analyses on eight families of Black-capped Chickadees that bred in the 1992 season at the Queen's University Biological Station, Lake Op-

inicon, Ontario, Canada (44°3'N, 76°20'W). This population has been color banded since 1987. We caught adult birds with Potter traps baited with seed, or by netting adults at the nest cavity with mist netting fashioned into a loop on the end of extendible poles. We collected blood from adults and young by brachial puncture; 75 μL from adults and 50 μL from nestlings at 8–12 days posthatch, as nestlings will fledge prematurely if disturbed after 12 days (Smith 1991). Blood was stored in either 1× TNE_2 blood preservative or Queen's Lysis buffer (QLB) (Seutin et al. 1991). Samples were stored frozen (TNE_2) or at 4°C (QLB), until DNA was extracted.

Hae III cut DNA extracted from the blood was electrophoresed for up to 48 hr in 1.8% agarose gels and then Southern blotted onto Immobilon-N® transfer membranes. Membranes were hybridized with the minisatellite probe (per Shin et al. 1985) to create DNA fingerprints used to assess paternity of the nestlings. Nestlings were arranged on DNA fingerprints with the behavioral father and mother (feeding male and female who held the territory in which the nest occurred) as well as males from adjacent territories who may have engaged in EPCs with the female. We calculated band sharing between nestlings and adults (D statistic—see Wetton et al. 1987) and plotted the proportion of shared bands with behavioral parents against the number of unique bands that occurred in the nestling but in neither behavioral parent (Fig. 1a). As minisatellite loci follow a Mendelian pattern of inheritance (Jeffreys et al. 1985), a large number of unique bands in a nestling may indicate a mismatch with one or the other of the behavioral parents (Lifjeld and Robertson 1992). Similarly, a low band sharing coefficient suggests that two individuals are not related (Wetton et al. 1987, Lifjeld and Robertson 1992). Background band sharing between adults in our population was 0.10 ± 0.02 (SE), and no two adults had a band sharing above 0.25 (based on band sharing between mated pairs and between neighboring males and the mated pair at each of the eight nests—n = 18 comparisons). Based on plots of band sharing against unique bands (Fig. 1a), we developed a criterion of parental assignment similar to that used by Lifjeld and Robertson (1992). Behavioral parents were assigned parentage if nestling/parent band sharing was >0.30 and a nestling had <3 unique bands (bands not shared with one of the two behavioral parents). Unique bands were usually bands that were shared with neighboring males, with an average of only 1.9 ± 0.3 bands (53 nestlings) per fingerprint that could not be attributed to one adult. In cases where the band sharing and unique band values were close to critical values we scanned autoradiographs for distinctive bands

(bands of high intensity) common to the nestling and adult in question to assess whether parentage was misassigned (see for example Gibbs et al. 1990).

The minisatellite probe per detected an average of 20.26 ± 0.43 bands in each individual's fingerprints. In cases where parentage was in question, we reprobed the entire nest with a second minisatellite probe, Jeffreys' 33.15 (Jeffreys et al. 1985), which gave a higher number of bands (37.0 ± 0.73) for confirmation of the results. Results with either probe always agreed.

In addition to parentage analysis, we analyzed dominance interactions between males during the breeding season of 1992 and in the winter flocks of 1993. Observations in the winter of 1993 were conducted at three separate feeding trays at which 73 color banded birds fed (25 at site 1, 40 at site 2, and 8 at site 3). Similar to Ficken et al. (1990), we used behavior, such as directed aggression (chases and displays), supplanting (one bird displacing another from a perch) and waiting (waiting for another bird to leave a feeding site before approaching) as criteria for assessing relative dominance of birds. We observed 986 interactions at the three sites.

RESULTS

Three of the eight nests (37.5%) analyzed contained at least one nestling genetically mismatched with their behavioral father, whereas all nestlings had DNA profiles that could be attributed to their behavioral mother (Fig. 1b). Of a total of 53 nestlings sampled, there were nine (17%) extra-pair young. Within nests where extra-pair young occurred, nine of the 19 (47%) nestlings were genetically mismatched with their behavioral father. There were three different extra-pair males, one for each of the three nests where extra-pair young occurred. We were able to assign paternity of all these extra-pair young to a single extra-pair male for each nest. In Nest U, the extra-pair male fathered five of the seven nestlings, in Nest I the extra-pair male fathered three of the five nestlings, and in Nest JJ the extra-pair male fathered one of the seven nestlings.

In all cases of extra-pair paternity, the extra-pair male was dominant to the behavioral father of the nestlings. In the first nest (Nest I) the extra-pair male held the second highest rank out of 25 birds feeding at site 1 in winter 1993, whereas the behavioral father held the 13th rank. The extra-pair male dominated the cuckolded male in 4/4 interactions observed. At Nest JJ, the extra-pair male (highest ranked of 25 birds at site 1) was dominant to the behavioral father of Nest JJ (3rd highest ranked bird) in 7/7 interactions in winter 1993. During the 1992 breeding season, both the extra-pair male and the behavioral father of Nest U fed the female at the nest (see below) and the extra-pair male was seen chasing the behavioral father away from the nest tree twice. We did not observe these two males interacting together at feeders during the winter observations in 1993.

The behavior of the two males at Nest U is consistent with polyandry; the extra-pair male and the behavioral father of the nest both fed the behavioral mother and nestlings. Polyandry is uncommon in chickadees, with only two reported cases to our knowledge (Waterman et al. 1989, Howitz 1991). This appears to be a true case of polyandry where the female has managed to enlist the help of two males, rather than divorcing her first mate with the arrival of the extra-pair male. The extra-pair male and his mate occupied a territory adjacent to the behavioral parents of Nest U. On 19 May, 1992 (approximately the fifth day of laying for the female at Nest U, as estimated by backdating 12–13 days incubation from the date of hatch (Smith 1991)) the extra-pair male and mate were seen on their own territory where they had excavated a nest; this was the last day that we saw the extra-pair male's mate. On 24 May, the behavioral father was found feeding the behavioral mother at Nest U (approximately day four of incubation) within their own territory. On 25 May, the extra-pair male entered the territory of the pair from Nest U, landed in the tree above Nest U and chased the behavioral father from the tree. The extra-pair male's nest appeared deserted and the extra-pair male was seen in the vicinity of Nest U over the next several days. The extra-pair male began feeding the behavioral female of Nest U on 29 May (day 9 of incubation), and continued to feed her even after the behavioral parents were seen feeding nestlings (8 June, approximately five days post-hatch). By 10 June, the extra-pair male was feeding the nestlings at a similar rate to their behavioral father (over a 2-hr period the two males fed the nestlings seven and eight times, respectively). As five of the seven nestlings were fathered by the extra-pair male, and the extra-pair male was seen on his own territory with his mate on 19 May, two days prior to the laying of the last two eggs, this suggests that the extra-pair male must have engaged in one or more EPCs with the behavioral mother of Nest U rather than pairing with her only after his own mate had died.

DISCUSSION

Based on our sample of eight families, it appears that the extent of extra-pair paternity in Black-capped Chickadees (37.5%) is comparable to that in another parid, the Blue Tit (31%) (Kempenaers et al. 1992). A total of 17% of chickadee nestlings from the eight families were extra-pair young (Blue Tits—11%) and 47% of chickadee nestlings in nests with mixed parentage were extra-pair young (Blue Tits—37%). All extra-pair young in our study were sired by a single extra-pair male per nest, although in two of these nests there was more than one male with a territory adjacent to the focal pair. Smith's (1988) and our observations (unpubl. data) that female chickadees actively solicit copulations, usually by seeking the male in his own territory, suggest that females may be preferentially selecting some neighboring males over others. In all three nests with extra-pair paternity, the extra-pair male appeared to be dominant to the behavioral father, as Smith (1988) predicted. Ficken et al. (1990) suggest that dominance may be correlated with access to resources, therefore dominance in winter flocks may be an accurate measure of quality that females may use to assess males.

Our results, coupled with Smith's observations on EPC behavior, suggest that female chickadees might be engaging in a mixed reproductive strategy. Females form socially monogamous pairs with particular males,
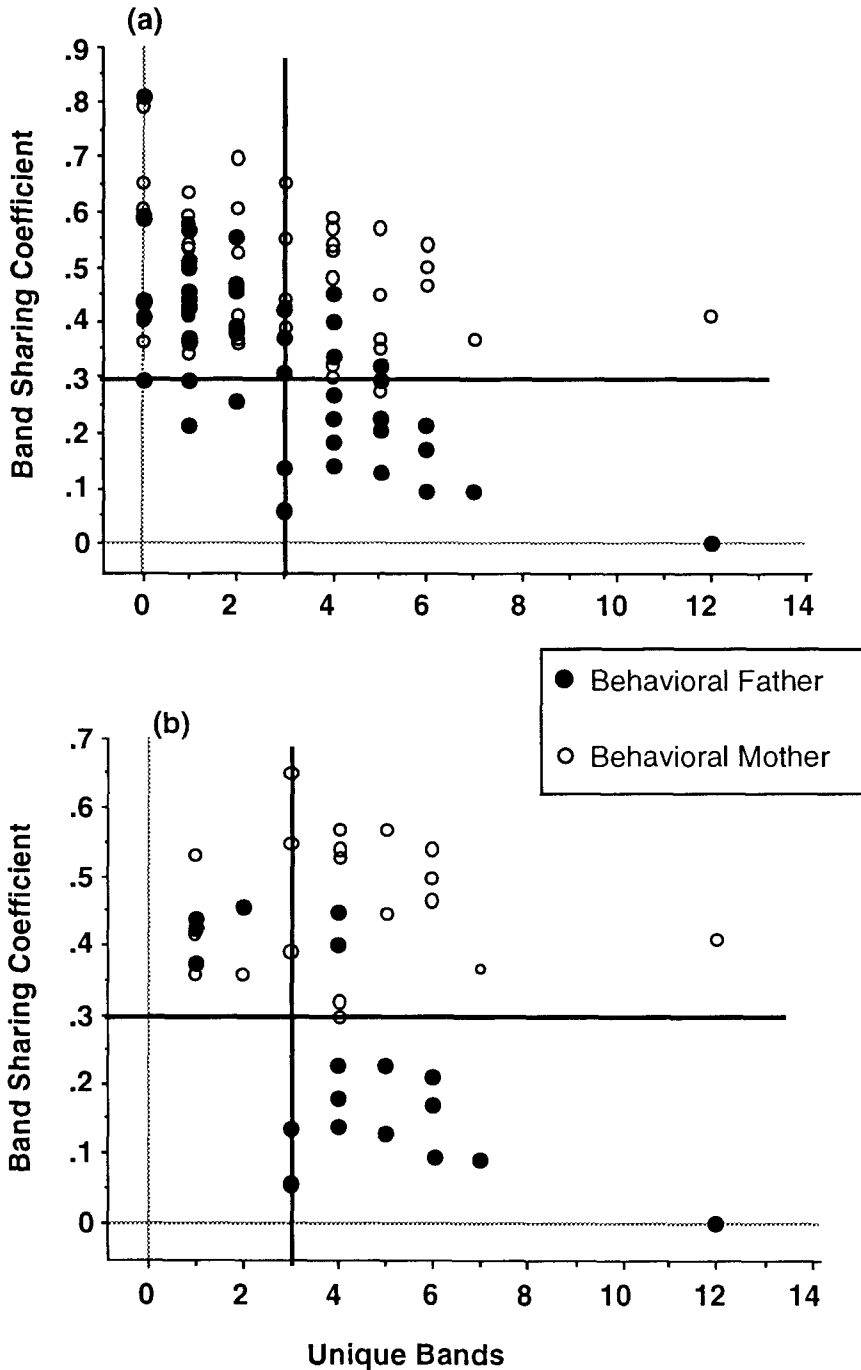
FIGURE 1.   Band sharing between behavioral parents and nestlings. Each point represents the proportion of bands in a nestling's fingerprint shared with one of the putative parents, plotted against the number of bands in the nestling's fingerprint that were not found in the fingerprints of either putative parent (Unique Bands). Paternity/maternity was assigned to adults if the band sharing coefficient with offspring was >0.3 and the number of Unique Bands was <3. In cases where band sharing coefficients were questionable, parentage was confirmed by distinctive bands on DNA fingerprints. Data from 53 nestlings from all eight nests is given in (a). Nineteen nestlings from three nests where at least one nestling was genetically mismatched from at least one parent are

but could conceivably increase genetic quality of their offspring by engaging in EPCs with males of higher dominance rank than their own mates (Trivers 1972). There would be ample opportunity for female chickadees to assess the relative rank of their own mate in relation to other mated males in the population based on social interactions within winter flocks (Smith 1988). In addition to attempting to increase the sample size of the present study, our future work will focus on more direct evidence as to whether females actively seek EPCs with males based on male quality, measured in part by male dominance.

Alternately, high ranking males may be better at forcing extra-pair copulations on females. We feel that this is less likely in the Black-capped Chickadee. We have never witnessed an attempted forced copulation in chickadees. Males do not appear able to gain even within-pair copulations without cooperation of the female (a short, distinctive chase usually occurs prior to copulation, copulation only occurs if the female stops and the pair begin to display. If no display is given by the female copulation does not occur—Smith 1991). Thus the observations of active female solicitation of EPCs (Smith 1988, pers. observ.) support the mixed strategy view of the extra-pair paternity that we found.

Finally, we found a possible explanation for the occasional observations of polyandry in Black-capped Chickadees. The two reported cases of polyandry in chickadees (Waterman et al. 1989, Howitz 1991) have one thing in common; the second male at the nest was a neighboring male whose own mate had disappeared and subsequently had been seen associating with the polyandrous female. In both previous accounts, authors either witnessed copulation of the female with both males (Howitz 1991) or suggested that it was likely (Waterman et al. 1989) although neither had information on whether the second male had any genetic relationship to the nestlings it was helping to feed. In our case of polyandry, the second male was the genetic father of five of the seven young in the nest. As the extra-pair male lost his original mate late in the breeding season, he may not have been able to secure a new mate in time to begin a new clutch. Thus, polyandry in chickadees may occur in cases where extra-pair males, having lost their own mates, attempt to salvage their annual reproductive effort by helping at a nest where they are likely to have fathered nestlings through EPCs.

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shown again in (b). Minisatellite probe used is per, but results were confirmed with a second probe, Jeffreys 33.15. In no cases were behavioral mothers mismatched with nestlings, thus genetic mismatching is attributed to extra-pair paternity. Unique Bands predominantly matched fingerprints of a neighboring male.

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SEX-RELATED DIFFERENCES IN DISTRACTION-DISPLAYS BY FLORIDA SANDHILL CRANES[1]

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Key words: Florida Sandhill Cranes; Grus canadensis pratensis; sexual differences; distraction displays.

In the wild, parent birds with young have been observed to distract predators with a wide range of behaviors. These range from active defense by attacking the predator (e.g., Audubon's Crested Caracara, *Polyborus plancus*; Yosef and Yosef 1992) and trying to prevent its approach of the area where the young are (e.g., Black-capped Chickadees, *Parus atricapillus*, Long 1982; Redshank, *Tringa totanus*, Warburg 1952), to distraction of predators by sneaking away from the location and then flying away in a conspicuous manner (e.g., Eastern Meadowlarks, *Sturnella magna*, pers. observ.), or even "*mock brooding*" in which the bird sits on the ground as if incubating and occasionally bends down as if to arrange imaginary eggs (e.g., Dotterel, *Eudromias morionellus*; Nethersole-Thompson and Nethersole-Thompson 1986).

One of the most well-known distraction displays in birds is the "*broken-wing act*" (Skutch 1976). It is also known as feigning injury, feigning a broken wing, broken wing ruse, parental ruse, lure display, disablement reaction, diversionary display. The parent bird feigns injury, and limps while holding a wing down as if it was broken. When the predator perceives the parent as potentially easy prey, it attempts to catch it. When sufficiently distanced from the young, the parent flies away. This behavior is well-developed in many species including McCowan's Longspur, (*Calcarius mccoenii*) that have been observed in "*cooperative injury simu-*

lation" (Skutch 1976, Nethersole-Thompson and Nethersole-Thompson 1986). This specific behavior has been extensively described in a wide range of species, mainly waders (see Nethersole-Thompson and Nethersole-Thompson 1986) and nighthawks (*Chordeiles chloris*; Skutch 1976). However, it is virtually unknown in larger species. This includes cranes (*Grus* spp.), except for the Common Cranes (*G. grus*; Cramp 1980).

I have been unsuccessful in finding similar observations in the existing literature on Sandhill Cranes (*G. canadensis*) (e.g., Bent 1926, Walkinshaw 1949, Voss 1977, Nesbitt and Archibald 1981, Johnsgard 1983). Here I report observations recorded at the MacArthur Agro-ecology Research Center (MAERC) of the Archbold Biological Station, Highlands County, southcentral Florida. MAERC is a 4,200-ha working cattle ranch that has extensive Bahia-grass (*Paspalum notatum*) pastures. Barbed wire fences bound the pastures. These observations were made during 1990–1993 in the post-hatching period (April–May).

I accidentally witnessed my first distraction display by a female Sandhill Crane in May 1990. While driving through pastures on a four-wheeled motorcycle (ATV), I observed two adult Florida Sandhill Cranes (*G. canadensis pratensis*) running from me while I was about 300 m away from them. I drove to about 75 m from the pair and observed them through binoculars. I noticed that they had two young running with them. The young were less than two weeks post-hatch and retained their reddish-brown down feathers. When I attempted to get closer, the male started to call loudly and attempted a "*directed walk-threat*" (Nesbitt and Archibald 1981) towards me. The young then disappeared from sight, and the male and female flew away and landed about 50 m away from their previous position. I walked into the general area and searched the ground. After an extensive search I found only one of