



Variable mating strategies and incest avoidance in cooperatively breeding grey-crowned babblers

CAROLINE J. BLACKMORE & ROBERT HEINSOHN

The Fenner School of Environment and Society, Australian National University

(Received 7 December 2006; initial acceptance 2 February 2007;
final acceptance 12 April 2007; published online ■ ■ ■; MS. number: 9199)

Understanding cooperative breeding requires an appreciation of the direct and indirect reproductive benefits available to group members, alongside the costs of group living such as increased potential for inbreeding. We use molecular techniques to determine sex, kinship and parentage to explore the benefits of helping and the mechanisms for incest avoidance in cooperatively breeding grey-crowned babblers, *Pomatostomus temporalis*. Generally, helpers gained only indirect fitness benefits through helping to raise kin. Breeding vacancies were usually filled by the oldest unrelated helper or by an immigrant if all helpers were related to the surviving breeder. A disproportionate number of helpers were unrelated to the dominant of the opposite sex. This suggests that related helpers disperse when their same-sex dominant dies rather than inherit the breeding position or help an unrelated immigrant to breed. Although monogamy was typical, extrapair paternity, joint-nesting and intraspecific brood parasitism were also revealed. Extrapair paternity was predicted by the relatedness of the dominant pair and appears to be an inbreeding avoidance mechanism, although some cases also suggest within-group reproductive conflict. This study is one of few to show a direct link between the rate of extrapair copulation and incest avoidance between the dominant breeding pair in a cooperatively breeding bird.

© 2007 The Association for the Study of Animal Behaviour. Published by Elsevier Ltd. All rights reserved.

Keywords: cooperative breeding; extrapair fertilization; grey-crowned babbler; incest avoidance; *Pomatostomus temporalis*

Cooperative avian societies, in which some individuals help the nesting attempts of others, show a plethora of mating systems (Cockburn 2004). These range from true monogamy where all helpers are nonbreeding, philopatric offspring (e.g. laughing kookaburra, *Dacelo novaeguineae*, Legge & Cockburn 2000) to polygynandrous systems where both sexes help and share reproduction (e.g. acorn woodpecker, *Melanerpes formicivorus*, Haydock et al. 2001). Even within a single species, alternative mating strategies may occur at varying levels. A female may raise her own young or parasitize the broods of other social groups (e.g. Australian magpie, *Gymnorhina tibicen*, Hughes et al. 2003). She may be either monogamous or polyandrous depending on how many helpers she has (e.g. superb fairy-wren, *Malurus cyaneus*, Mulder et al. 1994) or her relationship to them (e.g. stripe-backed wren, *Campylorhynchus nuchalis*, Piper & Slater 1993; white-browed scrubwren, *Sericornis frontalis*, Whittingham et al. 1997).

Correspondence: C. J. Blackmore, The Fenner School of Environment and Society, Australian National University, Canberra ACT 0200, Australia (email: caroline.blackmore@anu.edu.au).

Depending on the dynamics of group formation, females in cooperative species may be constrained in their choice of social partner. In particular, incestuous pairings may occur when philopatric young inherit breeding vacancies caused by the death of a parent. Females may move to avoid pairing with sons (e.g. long-tailed tit, *Aegithalos caudatus*, Hatchwell et al. 2000; superb fairy-wren, Cockburn et al. 2003), or young avoid mating with the surviving breeder (e.g. Florida scrub jay, *Aphelocoma coerulescens*, Woolfenden & Fitzpatrick 1990), although parent–offspring pairs may produce young in some species (e.g. acorn woodpecker, Haydock et al. 2001; moorhens, *Gallinula chloropus*, McRae 1996). As genetic benefits may be gained through alternative matings (Jennions & Petrie 2000; Cockburn 2004), females that find themselves paired to a relative could avoid inbreeding by seeking extrapair fertilizations (EPFs; Bensch et al. 1994; Blomqvist et al. 2002), although among cooperatively breeding birds this has only been recorded in the splendid fairy-wren, *Malurus splendens* (Tarvin et al. 2005) and the Mexican jay, *Aphelocoma ultramarina* (Eimes et al. 2005).

Here we provide the first genetic analysis of the mating system of grey-crowned babblers, *Pomatostomus temporalis*, a species that featured in some of the early research into cooperative breeding. Brown and colleagues showed that breeding grey-crowned babblers benefited from helpers through energy savings (Brown et al. 1978; Brown & Brown 1981), and pioneered the experimental removal of helpers to show that they both improve reproductive success and contribute to their own indirect fitness by doing so (Brown et al. 1982). However, these early studies relied on behavioural and morphometric cues to sex birds and infer parentage and kinship, and the mating system, demography and relatedness of group members remain unknown. Pairs are believed to be monogamous and social groups to form mostly through philopatry (King 1980), yet joint-nesting and polygyny have also been reported (e.g. Templeton 1955; Counsilman 1979). We use molecular genotyping techniques to determine sex, kinship and parentage in social groups of grey-crowned babblers. We examine the direct and indirect benefits available to helpers and explore extrapair mating as an incest avoidance mechanism.

METHODS

Study Species

The grey-crowned babbler is a group-living bird weighing about 80 g that mostly inhabits open forests and woodlands in northern and eastern Australia and southern New Guinea. It is multibrooded, generally commences breeding in July, and fledges the last broods before March, although breeding may occur outside of this period (Blackmore & Heinsohn in press). Groups usually consist of a socially monogamous, dominant pair and subordinate helpers of either sex, but unassisted pairs may breed successfully (Blackmore & Heinsohn in press). Helpers appear to be mostly offspring from previous broods although some are immigrants (King 1980). Gonadal development continues until 2 or 3 years of age, but males at least may produce gametes by age one (Counsilman 1979). Social rank corresponds to age (King 1980).

Field Methods

All research was carried out under authorization from the NSW Department of Environment and Conservation (DEC) and the Australian National University Animal Experimentation Ethics Committee. Between June 2003 and October 2005, 254 grey-crowned babblers (*P. t. temporalis*) in 47 social groups were captured in mistnets or as nestlings and colour-banded. Blood samples (20–70 µl) were collected from the brachial vein of 252 individuals and stored in 70% ethanol. Thirty-three groups (180 individuals) lived on contiguous territories in the east Pilliga Nature Reserve (PNR; 149°30'E, 30°53'S), and 14 groups (74 individuals) lived elsewhere in the PNR and on nearby private land near Coonabarabran, New South Wales, Australia.

Offspring from 2003 and 2004 were banded and sampled as 9- to 14-day-old nestlings where possible, or as

fledglings. All were assigned to first, second, third year and 3+ age cohorts by iris colour (King 1980). Birds first encountered as juveniles were allocated to broods according to relative physical appearance. Three groups contained more than three juveniles of indistinguishable age. As single females never laid clutches larger than three eggs, and all were later assigned as offspring of the dominant pair, they were allocated into two broods per group.

All colour-banded groups in the main study population and two in the surrounding district were monitored at least fortnightly in 2003 and 2004 between 1 July and the end of the following March, and censused again between July and October 2005. As group presence and composition changed within and between seasons, group size and structure were evaluated the first time each group was monitored in each season. First-year birds do not assist others (Brown et al. 1978), thus subordinate birds were counted as 'helpers' in seasons subsequent to that in which they fledged.

Molecular Methods

Extraction and genotyping techniques are described in Blackmore et al. (2006). Sex was determined with the molecular test developed by Griffiths et al. (1998). Males were genotyped at eight and females at seven polymorphic, unlinked microsatellite loci (Blackmore et al. 2006). The combined exclusion power of these loci was 0.99 with one parent known.

Parentage Assignment

Offspring born in the 2003 and 2004 breeding seasons were assigned to parents using a likelihood-based analysis of genotypes by the program Cervus (Marshall et al. 1998). Natural logarithm of the likelihood ratio (LOD) scores were calculated for offspring and candidate parents and compared with simulated critical LOD scores (the simulated critical difference between LOD scores for the first and second most likely candidate parent assigned with >95% and >80% confidence). Unsampled birds were included in simulations as candidate parents. Where possible they were sexed by breeding behaviour or sex-specific calls, or assigned as male or female so that the sex ratio among helpers in their group matched known levels (63% male; Blackmore 2006). To counteract the presence of close relatives among candidate parents, we adapted the assignment strategy of Richardson et al. (2001). Simulations were used to calculate critical LOD values for (1) maternity to a group female, (2) paternity to a group male, (3) maternity to an extragroup female, and (4) paternity to an extragroup male. The number of candidate parents for within-group assignment simulations was set at the maximum number of group members of the appropriate sex plus one, to simulate the chance that an extragroup bird was the true parent (Richardson et al. 2001). There were four female and five male within-group candidate parents for 2003 offspring simulations, with the proportion sampled set at 0.89 of females and 0.92 of males based on actual sampling rates. For

2004 offspring, there were five female and six male within-group candidate parents (0.94 and 0.91 sampled, respectively). For extragroup assignment, the number of candidate parents included all individuals of the appropriate sex and at least 1 year of age, including those of uncertain fate that had disappeared from their groups: 70 females and 84 males for 2003, and 90 females and 113 males for 2004. In both years, the proportions of extra-group females and males sampled were 0.88 and 0.92, respectively.

Candidate parents often included full siblings of the offspring. Where one parent is unsampled, even half-siblings may emerge as the 'most likely' parent (Marshall et al. 1998). With the mother known, a proportion of full siblings emerged as the 'most likely' father. The dominant male was assigned as the true father if he also had a high LOD score and no mismatches; otherwise parentage was assigned to the most senior group candidate with a positive LOD score and no mismatches. Unsampling dominants were assumed to be true parents once offspring were successfully assigned to the opposite-sex dominant. Where more than one within-group female returned positive LOD scores and no mismatches, each was run as a 'known parent' for within-group paternal assignment. The pair with the highest summed LOD scores and no mismatches were considered true parents, although the confidence of assignment to an individual other than the 'most likely' parent could not be estimated. While this strategy increased the risk of falsely excluding true helper or extragroup parents, it ensured the most conservative interpretation of the mating system.

In describing the mating system, the term 'extrapair' refers to matings with an individual other than the social partner, including within-group helpers. 'Extragroup' describes matings obtained outside the social group.

Relatedness and Assignment to Kinship Groups

Pairwise relatedness was estimated with symmetrical Lynch & Ritland (1999) relatedness estimators calculated in GenAIEx 6 (Peakall & Smouse 2006). To determine whether helpers of unknown lineage were first-order relatives of other group members, we used the program Kinship 1.3.1 (Goodnight & Queller 1999) to calculate the likelihood that helper–dominant and helper–offspring dyads fell into dichotomous categories of 'nonrelatives' or 'relatives'. As most helpers are likely to be philopatric young, the null hypothesis was set at R_m (relatedness with mother) = 0.5 and R_f (relatedness with father) = 0.5. The primary hypothesis was that helpers were nonrelatives ($R_m = 0$, $R_f = 0$). If pairwise log likelihood ratios were greater than the 95% critical log likelihood ratio limit generated from 10 000 simulations (0.5087), the null hypothesis was rejected and those dyads were classified as nonrelatives ($P < 0.05$). Structuring the hypotheses in this way resulted in a large type II error rate ($P = 0.25$) of failing to detect nonrelatives, but increased the reliability of detecting unrelated helpers.

Statistical Analyses

All data were analysed in the statistical modelling program Genstat (Release 8.2; Lawes Agricultural Trust, Harpenden, U.K.). Relationships between EPF and relatedness were calculated with Generalized Linear Mixed Models, with the proportion of extrapair young in each brood included as the binomial response variable. In all models, 'group' was included as the random term to avoid pseudoreplication because of repeated sampling of social groups. Residuals and fitted values were plotted to confirm data were normally distributed.

RESULTS

Parentage Assignment

Parentage was assigned for 118 offspring in 60 broods: 55 offspring from 32 broods in 2003 and 63 offspring from 28 broods in 2004. Fifty-eight per cent of maternal and 64% of paternal assignments were made with 95% confidence, 12% and 14%, respectively, were made with 80% confidence, and 11% and 6%, respectively, were not assigned to the 'most likely' candidate. Fourteen per cent of offspring were assigned to an unsampled dominant female and 4% to an unsampled dominant male. Where all group members were sampled, 6% of young could not be assigned to a sampled female, either within- or extra-group, and were attributed to unsampled extragroup mothers. Similarly, 12% were assigned to an unsampled extragroup father. Eight helpers born in 2002 were assigned to the within-group dominant pair, and are included in analyses of relatedness and kinship but not in mating system analyses.

Group Structure and Kin Relationships

Over three seasons, group sizes ranged from two to 13, with a median size of four (mean \pm SE = 4.28 ± 0.22 ; $N = 85$ group-years). Unassisted pairs comprised 16% of groups. Most dominant females (89%; $N = 76$ group-years) and males (86%; $N = 80$) were at least 3 years of age (Fig. 1). Helpers censused over three seasons were mostly 1 year olds (51%; $N = 146$ helper-years), but could be aged up to 3+ (Fig. 1). Male (mean \pm SE = 1.72 ± 0.07 years) and female helpers (mean \pm SE = 1.69 ± 0.12 years) did not differ in age ($t_{144} = 0.31$, $P = 0.76$; Fig. 1).

Pairwise relatedness estimates for first-order relatives identified through parentage assignment averaged 0.253 (± 0.01 SE; $N = 382$ dyads). Helpers of unknown lineage were on average related to their dominants by 0.118, similar to estimations from half-siblings (Fig. 2). Socially paired dominants averaged lower relatedness estimates than half-siblings and were the least closely related dyad in a group ($F_{5,508} = 33.46$, $P < 0.001$; Fig. 2).

Most helpers (58%; $N = 59$) were first-order relatives of all offspring they helped to raise. Thirty-two per cent were first-order relatives of only some offspring, with all but one of these related to at least half of all offspring

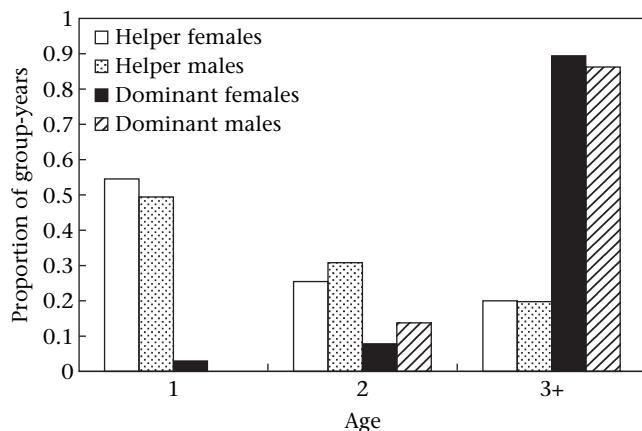


Figure 1. The age distribution of female and male helpers ($N = 55$ and 91 group-years, respectively) and dominant birds ($N = 76$ and 80 group-years, respectively).

they raised across broods. Ten per cent of helpers were nonrelatives of all offspring they helped to rear.

Where helpers and both dominants were sampled, 74% of helpers ($N = 95$) were first-order relatives of both dominants, suggesting that they were caught in their natal group, including 32 helpers assigned as offspring of the dominant pair. Twelve per cent were nonrelatives of the dominant male, 9% were nonrelatives of the dominant female, and 5% of helpers were not relatives of either dominant. Helpers that were only related to one dominant were more likely to be unrelated to the dominant of the opposite sex ($\chi^2_1 = 5.30$, $P = 0.02$; Fig. 3).

Eleven male and two female breeding vacancies arose in groups with helpers of the appropriate sex to fill the position. Female vacancies were filled by the oldest helper,

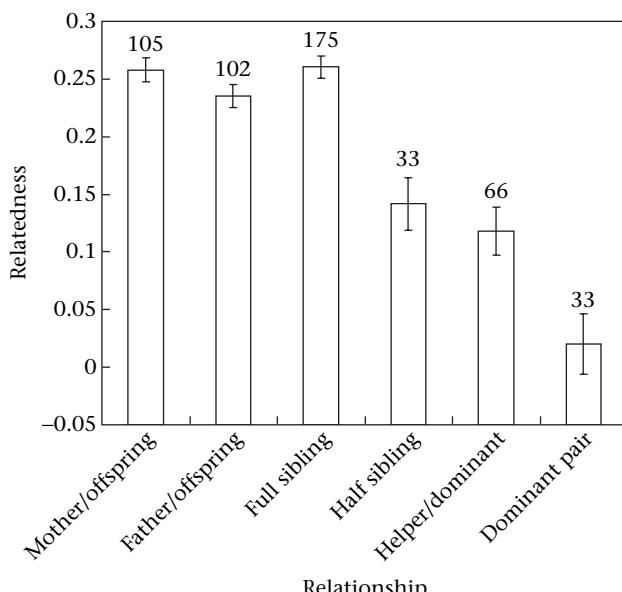


Figure 2. Mean (\pm SE) pairwise relatedness estimates ($N = 514$ dyads) between known first-order relatives ($N = 382$), known half-siblings ($N = 33$), helpers and their dominants ($N = 66$), and dominant pairs ($N = 33$).

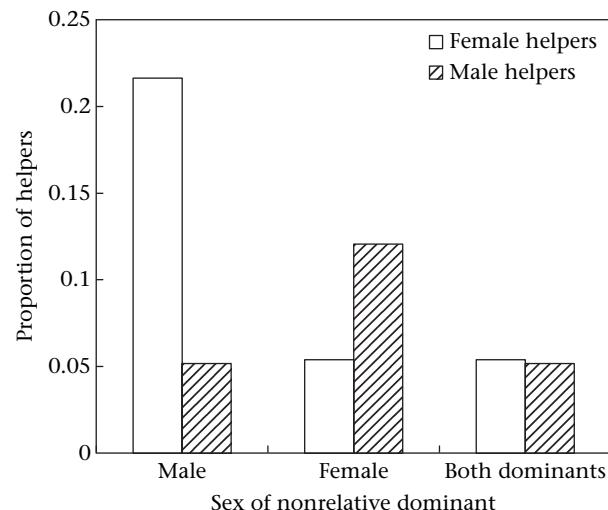


Figure 3. The proportion of female ($N = 37$) and male ($N = 58$) helpers that were nonrelatives of the male but relatives of the female dominant; nonrelatives of the female but relatives of the male dominant; or nonrelatives of both dominants.

one a relative (replacing a nonrelative) and the other a nonrelative of the dominant male. Six male vacancies occurred where male helpers were nonrelatives of the dominant female. Four were filled by the oldest male helper. In two the only helper disappeared with the dominant male, and both were replaced by two immigrant males, the elder of which bred with the surviving female. Five male vacancies occurred where male helpers were relatives of the dominant female: two were filled by immigrants; one group disbanded after the female did not re-pair; and two were filled by related helpers, both replacing dominant males that had also been relatives of the breeding female. Thus, in this sample, the most senior nonrelative helper always inherited the male breeding vacancy, and when all helpers were relatives, vacancies were usually filled by immigrants. However, recruitment to incestuous pairings occurred in groups where the breeding female was previously mated to a relative.

Mating System

In most broods (65%; $N = 60$) all offspring were assigned to the dominant pair, but 20% of broods contained only young not assigned to one or both of the dominant pair, and 15% contained both. Most young (76%; $N = 117$) were the 'legitimate' offspring of the dominant pair (Fig. 4). This includes the only two individuals not matched to a group female but sired by the dominant male. Both were caught as fledglings when groups of three were first caught, and the most likely explanation is that the true mother had been recently replaced by the captured female. Five young (4%; 4/38 broods from groups with male helpers) were sired by a helper male, and 16 (14%; 11/60 broods) by an extragroup male (Fig. 4). Five offspring (4%; 4/60 broods) in three completely sampled groups could not be assigned to any individual, despite repeated genotyping to exclude laboratory error. We

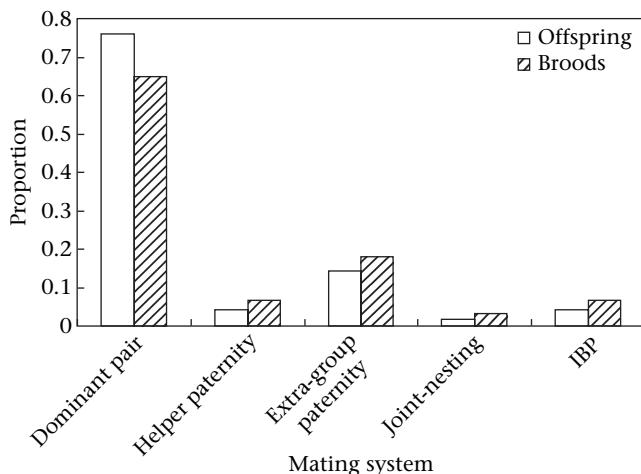


Figure 4. The relative frequency of monogamy, extrapair paternity, joint-nesting and intraspecific brood parasitism among offspring ($N = 117$) and broods ($N = 60$).

conclude these eggs were dumped by unsampled extra-group females (intraspecific brood parasitism or IBP). Two IBP nestlings found in the same brood had a relatedness estimate of 0.5, suggesting that only one extra-group female had contributed to the clutch. In one of the groups where IBP was detected we also found one nestling in each of two successive broods (2% of offspring; 2/22 broods hatched in groups with female helpers) laid by a female helper mated with an extra-group male (joint-nesting; Fig. 4). At 4 years of age, the helper that joint-nested was the oldest female helper in the study and one of only two that were unrelated to the dominant female.

Genotypes were available for both putative parents and all offspring in 24 groups. Extrapair young were produced by 10 females (15/42 broods). All females in pairs with relatedness estimates greater than 0.2 (indicative of close relatives; Fig. 2) produced extrapair young ($N = 4$). The proportion of extrapair young in a brood increased with the relatedness of the dominant pair ($\chi^2 = 4.83$, $P = 0.028$; Fig. 5). The random term 'group' indicated high within-term correlation ($r = 0.995$). Only six

comparisons were possible between the relatedness estimates of females and extra-pair sires, and females and their dominant partners. There was a trend towards females being less related to extra-pair sires (mean \pm SE = 0.01 ± 0.06) than their social partners (mean \pm SE = 0.18 ± 0.08), but this was not significant (paired t test: $t_5 = 1.80$, $P = 0.066$).

Seven females produced offspring sired by seven extra-group males, three of which were identified and were from adjacent territories. Only one social partner cuckolded by an extra-group male gained a share of paternity. The three dominant males cuckolded by a helper also sired offspring in each brood. Two of the three dominants were close relatives of the breeding female. All four helpers that gained paternity were relatives of the dominant male they parasitized, and two were relatives of the breeding female. Overall, helper paternity occurred in two (40%) of five groups with unrelated male helpers but only in one (7%) of 14 groups that only had males related to the breeding female.

DISCUSSION

Our study reveals both a complex mating system and important incest avoidance behaviour in a cooperatively breeding bird. While most groups of grey-crowned babblers consisted of a monogamous pair assisted by non-breeding helpers, we also detected low levels of extra-pair paternity, joint-nesting and IBP. Importantly, their social system also entails some incestuous pairing of the dominant birds, and the results provide rare evidence that extra-pair mating has evolved as a mechanism to avoid potential inbreeding when this occurs.

Grey-crowned babblers are one of few species in which helpers have been shown experimentally to increase group productivity (Brown et al. 1982); our genetic analysis of their mating system enables further evaluation of the costs and benefits of group living and helping behaviour. Most (84%) breeding pairs had helpers even though the benefits of helping may be outweighed by the direct costs of not breeding (Brown & Brown 1981). Only a small proportion of helpers acquired direct benefits through extra-pair paternity or joint-nesting, or stood to inherit the breeding vacancy. That helpers were usually younger and breeders older suggests that young birds are constrained from both independent and within-group breeding. Most helpers, however, stood to improve their indirect fitness by helping to raise kin.

Most young were offspring of the dominant pair, and most helpers were offspring from previous seasons. However, the rates of monogamy reported here may have been overestimated: for example, IBP may have been more frequent because maternity was assigned first to within-group females (Richardson et al. 2001). None the less, even the conservative use of biological logic to resolve parentage under high levels of philopatry (Double et al. 1997) led to estimates of one in four offspring resulting from EPF, IBP or joint nesting.

Female grey-crowned babblers produced extra-pair young when paired to close relatives. Incest avoidance appears to be the most likely incentive for such extra-pair

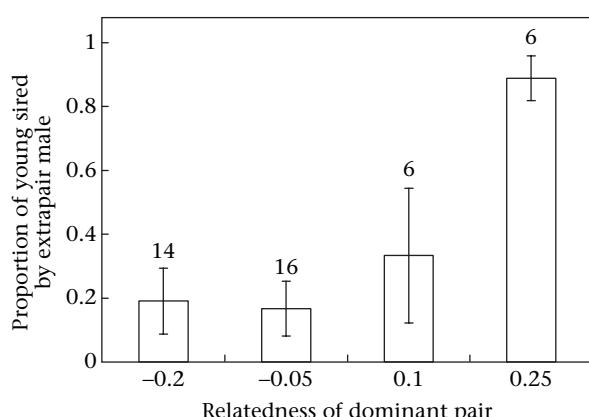


Figure 5. Pairwise relatedness estimates of dominant pairs and the average proportion (\pm SE) of extra-pair young in broods ($N = 42$). Value labels indicate the number of broods.

mating. An alternative explanation could be that female babblers routinely engage in extrapair copulations, perhaps selecting extrapair mates that are phenotypically superior to their social partner (e.g. Kempenaers et al. 1992; Hasselquist et al. 1996), but that fertilization is more likely to result when they are socially paired to a relative. This could occur if differential success in sperm competition (Birkhead & Møller 1992) favoured fertilization by unrelated males, or the offspring of incestuous fertilization were more likely to die at the embryonic stage. We did not find evidence that incestuous offspring were more likely to die as embryos, as sampled broods were no smaller when they resulted from incestuous matings ($\text{mean} \pm \text{SE} = 2.67 \pm 0.33$ nestlings; $N = 3$, for incestuous, and $\text{mean} \pm \text{SE} = 2.73 \pm 0.20$ nestlings; $N = 11$, for nonincestuous matings). Without knowledge of female copulatory behaviour we cannot determine whether the pattern of EPF resulted from sperm competition or female-controlled copulations. The relationship between EPF and male quality also warrants further investigation.

However, several factors indicate that female babblers do not routinely gain EPF. The overall incidence of EPF was low (18% of offspring); extrapair young were generally found only in groups where the dominant pair was related (Fig. 5); and all known extrapair mates were from adjacent territories, suggesting that proximity may be important to mate choice. A relationship between EPF and genetic similarity of pairs has been found in only two other cooperative breeders, the splendid fairy-wren (Tarvin et al. 2005) and Mexican jay (Eimes et al., 2005). In some other avian cooperative breeders, high rates of extragroup or EPF may have the effect of reducing inbreeding even if that is not the primary function of such behaviour (Koenig & Haydock 2004).

Other predictions associated with EPF to avoid genetic similarity (Griffith et al. 2002) could not be tested. Most EPFs were obtained from extragroup males, and because the social partners of females that produced extragroup young did not usually share paternity, the relative heterozygosity of extrapair and within-pair young could not be compared. There was, however, a nonsignificant trend suggesting that extrapair males were less genetically similar to breeding females than social partners.

In established groups, the effect of incest avoidance on suppression of helper reproduction is difficult to disentangle from reproductive competition, but can be illuminated when breeding vacancies occur (Koenig et al. 1998). In grey-crowned babbler groups where all helpers were related to both dominants, breeding vacancies were usually filled from outside. However, helpers were sometimes related to only one of the dominants, presumably because of extrapair mating or because an immigrant replaced a deceased parent. In this situation, a disproportionate number of helpers were nonrelatives of the dominant of the opposite sex (Fig. 3). This could arise if helpers dispersed when their same-sex dominant died (Koenig et al. 1998), were perceived as a threat by an incoming same-sex step-parent and evicted (e.g. Goldstein et al. 1998), or the opposite sex of helper was more likely to delay dispersal because of the chance to mate with the incoming step-parent (Balcombe 1989). In grey-crowned babblers,

related helpers that do not disperse when their same-sex breeder dies have little chance of inheriting the breeding vacancy and would also suffer from reduced inclusive fitness. Sons, at least, have little chance of gaining reproduction if incest avoidance is the motivation behind extrapair mating in this species. As the immigrant dominant is likely to be a nonrelative, breeding females may not seek EPF. Helpers may also conflict with the new dominant over resources and reproduction (Koenig et al. 1998).

Females that obtained extragroup fertilizations did not usually obtain any fertilizations from their partner. However, females that mated with a helper always mated with the dominant too. This suggests that the observed pattern of mixed paternity in broods may sometimes result from reproductive sharing among group males. Female grey-crowned babblers probably also compete for reproduction within their social groups. Although parentage assignment confirmed only one case of joint-nesting, there were two other nests with four eggs or young we could not sample, and these were the only other two groups with older (3+) female helpers. Thus joint-nesting may occur more frequently than detected here.

Intraspecific brood parasitism (IBP) also occurs at low levels in grey-crowned babblers. IBP is thought to be rare among passerines because the high maternal investment required to raise altricial young selects for retaliation against egg dumping (Sorenson 1992; Geffen & Yom-Tov 2001). However, the presence of helpers in some cooperatively breeding species, including grey-crowned babblers, reduces the provisioning required of parents (Brown et al. 1978), and may reduce selection for retaliation. Some authors consider IBP to be closely linked to joint-nesting (McRae 1996; Cockburn 2004) and mutually beneficial as females that dump eggs may be related to the females they parasitize (Andersson & Ahlund 2000), although others have suggested it is 'bet-hedging' (Hughes et al. 2003) or an epiphenomenon arising from nest distribution (Reyer et al. 1997). With further study the grey-crowned babbler provides a rare opportunity to test these and other predictions about the evolution of IBP versus joint-nesting, which may be seen to represent a continuum of secondary female parental care (Zink 2000).

Simultaneous expression of multiple mating systems seems to be relatively prevalent among Australian cooperative breeders, many of which do not fit existing frameworks of the evolution of cooperative breeding (Koenig & Dickinson 2004). As much of the interspecific variation in alternative mating systems occurs at the taxonomic family level (Arnold & Owens 2002), future molecular analyses may reveal similar levels of mixed mating strategies, including joint-nesting and brood parasitism, in other Pomatostomidae species.

Acknowledgments

This research was funded by the Stuart Leslie Bird Research Award, the Australian Bird Environment Fund, Australian Geographic and the Centre for Resource and Environmental Studies. We thank Rod Peakall, Greg Adcock, Dan Ebert, Nadeena Beck, Mike Double and Rose Andrew for

assistance with molecular techniques and analyses; Scott Filmer, Chris Timewell, Lisa Minchin, Nora Leuschner, Molly Dickens, Christine Yong and Philippa Nelson for help in the field; the New South Wales DEC; and the Brookhouse, Graham, Lang, Drysdale, Jerry and Middleton families for access to their properties.

References

- Andersson, M. & Ahlund, M.** 2000. Host-parasite relatedness shown by protein fingerprinting in a brood parasitic bird. *Proceedings of the National Academy of Sciences of the United States of America*, **97**, 13188–13193.
- Arnold, K. E. & Owens, I. P. F.** 2002. Extra-pair paternity and egg dumping in birds: life history, parental care and the risk of retaliation. *Proceedings of the Royal Society of London, Series B*, **269**, 1263–1269.
- Balcombe, J. P.** 1989. Non-breeder asymmetry in Florida scrub jays. *Evolutionary Ecology*, **3**, 77–79.
- Bensch, S., Hasselquist, D. & Vonschantz, T.** 1994. Genetic similarity between parents predicts hatching failure – nonincestuous inbreeding in the great reed warbler. *Evolution*, **48**, 317–326.
- Birkhead, T. R. & Möller, A. P.** 1992. *Sperm Competition in Birds: Evolutionary Causes and Consequences*. San Diego: Academic Press.
- Blackmore, C.J.** 2006. Breeding biology and behaviour of the grey-crowned babbler. Ph.D. thesis, Australian National University.
- Blackmore, C.J. & Heinsohn, R.** In press. Reproductive success and helper effects in the cooperatively breeding grey-crowned babbler *Pomatostomus temporalis*. *Journal of Zoology*.
- Blackmore, C. J., Adcock, G. J., Ebert, D. & Heinsohn, R.** 2006. Microsatellite loci for population and behavioural studies of grey-crowned babblers (*Pomatostomus temporalis*: Aves). *Molecular Ecology Notes*, **6**, 412–414.
- Blomqvist, D., Andersson, M., Kupper, C., Cuthill, I. C., Kis, J., Lanctot, R. B., Sandercock, B. K., Szekely, T., Wallander, J. & Kempenaers, B.** 2002. Genetic similarity between mates and extra-pair parentage in three species of shorebirds. *Nature*, **419**, 613–615.
- Brown, J. L. & Brown, E. R.** 1981. Kin selection and individual selection in babblers. In: *Natural Selection and Social Behavior: Recent Research and New Theory* (Ed. by R. D. Alexander & D. W. Tinkle), pp. 244–256. New York: Chiron Press.
- Brown, J. L., Dow, D. D., Brown, E. R. & Brown, S. D.** 1978. Effects of helpers on feeding of nestlings in the grey-crowned babbler (*Pomatostomus temporalis*). *Behavioral Ecology and Sociobiology*, **4**, 43–59.
- Brown, J. L., Brown, E. R., Brown, S. D. & Dow, D. D.** 1982. Helpers: effects of experimental removal on reproductive success. *Science*, **215**, 421–422.
- Cockburn, A.** 2004. Mating systems and sexual conflict. In: *Ecology and Evolution of Cooperative Breeding in Birds* (Ed. by W. D. Koenig & J. Dickinson), pp. 81–101. Cambridge: Cambridge University Press.
- Cockburn, A., Osmond, H. L., Mulder, R. A., Green, D. J. & Double, M. C.** 2003. Divorce, dispersal and incest avoidance in the cooperatively breeding superb fairy-wren *Malurus cyaneus*. *Journal of Animal Ecology*, **72**, 189–202.
- Counsilman, J. J.** 1979. Notes on the breeding biology of the grey-crowned babbler. *Bird Behaviour*, **1**, 114–124.
- Double, M. C., Cockburn, A., Barry, S. C. & Smouse, P. E.** 1997. Exclusion probabilities for single-locus paternity analysis when related males compete for matings. *Molecular Ecology*, **6**, 1155–1166.
- Eimes, J. A., Parker, P. G., Brown, J. L. & Brown, E. R.** 2005. Extrapaternal fertilization and genetic similarity of social mates in the Mexican jay. *Behavioral Ecology*, **16**, 456–460.
- Geffen, E. & Yom-Tov, Y.** 2001. Factors affecting the rates of intra-specific nest parasitism among Anseriformes and Galliformes. *Animal Behaviour*, **62**, 1027–1038.
- Goldstein, J. M., Wolfenden, G. E. & Hailman, J. P.** 1998. A same-sex stepparent shortens a prebreeder's duration on the natal territory: tests of two hypotheses in Florida scrub-jays. *Behavioral Ecology and Sociobiology*, **44**, 15–22.
- Goodnight, K. F. & Queller, D. C.** 1999. Computer software for performing likelihood tests of pedigree relationship using genetic markers. *Molecular Ecology*, **8**, 1231–1234.
- Griffith, S. C., Owens, I. P. F. & Thuman, K. A.** 2002. Extra pair paternity in birds: a review of interspecific variation and adaptive function. *Molecular Ecology*, **11**, 2195–2212.
- Griffiths, R., Double, M. C., Orr, K. & Dawson, R. J. G.** 1998. A simple DNA test to sex most birds. *Molecular Ecology*, **7**, 1071–1075.
- Hasselquist, D., Bensch, S. & Von Schantz, T.** 1996. Correlation between male song repertoire, extra-pair paternity and offspring survival in the great reed warbler. *Nature*, **381**, 229–232.
- Hatchwell, B. J., Russell, A. F., Ross, D. J. & Fowlie, M. K.** 2000. Divorce in cooperatively breeding long-tailed tits: a consequence of inbreeding avoidance? *Proceedings of the Royal Society of London, Series B*, **267**, 813–819.
- Haydock, J., Koenig, W. D. & Stanback, M. T.** 2001. Shared parentage and incest avoidance in the cooperatively breeding acorn woodpecker. *Molecular Ecology*, **10**, 1515–1525.
- Hughes, J. M., Mather, P. B., Toon, A., Ma, J., Rowley, I. & Russell, E.** 2003. High levels of extra-group paternity in a population of Australian magpies *Gymnorhina tibicen*: evidence from microsatellite analysis. *Molecular Ecology*, **12**, 3441–3450.
- Jennions, M. D. & Petrie, M.** 2000. Why do females mate multiply? A review of the genetic benefits. *Biological Reviews*, **75**, 21–64.
- Kempenaers, B., Verheyen, G. R., Van den Broeck, M., Burke, T., Van Broekhoven, C. & Dhondt, A. A.** 1992. Extra-pair paternity results from female preference for high-quality males in the blue tit. *Nature*, **357**, 494–496.
- King, B. R.** 1980. Social organization and behaviour of the grey-crowned babbler *Pomatostomus temporalis*. *Emu*, **80**, 59–76.
- Koenig, W. D. & Dickinson, J.** 2004. *Ecology and Evolution of Cooperative Breeding in Birds*. Cambridge: Cambridge University Press.
- Koenig, W. D. & Haydock, J.** 2004. Incest and incest avoidance. In: *Ecology and Evolution of Cooperative Breeding in Birds* (Ed. by W. D. Koenig & J. Dickinson), pp. 142–156. Cambridge: Cambridge University Press.
- Koenig, W. D., Haydock, J. & Stanback, M. T.** 1998. Reproductive roles in the cooperatively breeding acorn woodpecker: incest avoidance versus reproductive competition. *American Naturalist*, **151**, 243–255.
- Legge, S. & Cockburn, A.** 2000. Social and mating system of cooperatively breeding laughing kookaburras (*Dacelo novaeguineae*). *Behavioral Ecology and Sociobiology*, **47**, 220–229.
- Lynch, M. & Ritland, K.** 1999. Estimation of pairwise relatedness with molecular markers. *Genetics*, **152**, 1753–1766.
- McRae, S. B.** 1996. Family values: costs and benefits of communal nesting in the moorhen. *Animal Behaviour*, **52**, 225–245.
- Marshall, T. C., Slate, J., Kruuk, L. E. B. & Pemberton, J. M.** 1998. Statistical confidence for likelihood-based paternity inference in natural populations. *Molecular Ecology*, **7**, 639–655.
- Mulder, R. A., Dunn, P. O., Cockburn, A., Lazenby-Cohen, K. A. & Howell, M. J.** 1994. Helpers liberate female fairy-wrens from constraints on extra-pair mate choice. *Proceedings of the Royal Society of London, Series B*, **255**, 223–229.

- Peakall, R. & Smouse, P. E.** 2006. GenAIEx 6: genetic analysis in Excel. Population genetic software for teaching and research. *Molecular Ecology Notes*, **6**, 288–295.
- Piper, W. H. & Slater, G.** 1993. Polyandry and incest avoidance in the cooperative stripe-backed wren of Venezuela. *Behaviour*, **124**, 227–247.
- Reyer, H. U., Bollmann, K., Schlapfer, A. R., Schymainda, A. & Klecack, G.** 1997. Ecological determinants of extrapair fertilizations and egg dumping in Alpine water pipits (*Anthus spinolella*). *Behavioral Ecology*, **8**, 534–543.
- Richardson, D. S., Jury, F. L., Blaakmeer, K., Komdeur, J. & Burke, T.** 2001. Parentage assignment and extra-group paternity in a cooperative breeder: the Seychelles warbler (*Acrocephalus sechellensis*). *Molecular Ecology*, **10**, 2263–2273.
- Sorenson, M. D.** 1992. Why is conspecific nest parasitism more frequent in waterfowl than in other birds – comment. *Canadian Journal of Zoology*, **70**, 1856–1858.
- Tarvin, K. A., Webster, M. S., Tuttle, E. M. & Pruett-Jones, S.** 2005. Genetic similarity of social mates predicts the level of extrapair paternity in splendid fairy-wrens. *Animal Behaviour*, **70**, 945–955.
- Templeton, M. T.** 1955. Babbler mating with two hens. *Emu*, **55**, 190.
- Whittingham, L. A., Dunn, P. O. & Magrath, R. D.** 1997. Relatedness, polyandry and extra-group paternity in the cooperatively-breeding white-browed scrubwren (*Sericornis frontalis*). *Behavioral Ecology and Sociobiology*, **40**, 261–270.
- Woolfenden, G. E. & Fitzpatrick, J. W.** 1990. Florida scrub jays: a synopsis after 18 years of study. In: *Cooperative Breeding in Birds: Long-term Studies of Ecology and Behavior* (Ed. by P. B. Stacey & W. D. Koenig), pp. 241–266. Cambridge: Cambridge University Press.
- Zink, A. G.** 2000. The evolution of intraspecific brood parasitism in birds and insects. *American Naturalist*, **155**, 395–405.