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Sex-specific associative learning cues and inclusive fitness benefits in the Seychelles warbler

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Abstract

In cooperative breeding vertebrates, indirect fitness benefits would be maximized by subordinates that accurately assess their relatedness to group offspring and preferentially help more closely related kin. In the Seychelles warbler (*Acrocephalus sechellensis*), we found a positive relationship between subordinate-nestling kinship (determined using microsatellite marker genotypes) and provisioning rates, but only for female subordinates. Female subordinates that helped were significantly more related to the nestlings than were nonhelpers, and the decision to help appears to be based on associative learning cues. High levels of female infidelity means that subordinates cannot trust their legitimacy through the male line, consequently they appear to use the continued presence of the primary female, but not the primary male, as a reliable cue to determine when to feed nestlings. By using effective discrimination, female subordinates are able to maximize the indirect benefits gained within a cooperative breeding system otherwise driven primarily by direct breeding benefits.

Introduction

Cooperative breeding, where some individuals (here referred to as subordinates) assist others in the rearing of offspring, has been well documented in vertebrates (Stacey & Koenig, 1990; Jennions & Macdonald, 1994; Emlen, 1995; Cockburn, 1998; Hatchwell & Komdeur, 2000). The evolution of this apparently altruistic behaviour can be divided into two parts. (i) Why do individuals become subordinates within a group instead of breeding independently? (ii) Why do these subordinates sometimes help raise the nondescendant offspring? Overall, the evidence suggests that ecological constraints/benefits of philopatry (Koenig *et al.*, 1992; Emlen, 1994) prevent independent breeding and persuades subordinates to join groups. However, as helping is not an inevitable consequence of group living, and nonhelping subordinates occur in many cooperative societies (Cockburn, 1998), why subordinates help is still much debated (Emlen, 1995; Cockburn, 1998).

In the majority of cooperative systems, subordinates are philopatric offspring (Emlen, 1995). Consequently, indirect fitness benefits have been suggested to be the primary force driving the evolution of helping (Emlen, 1997). However, the importance of indirect fitness benefits has been questioned for a number of reasons. First, in some systems subordinates preferentially help nonkin individuals (Reyer, 1980; Sherley, 1990; Whittingham *et al.*, 1997). Secondly, in situations where close genetic relationships were thought to exist (i.e. subordinates helping their 'social' parents), shared reproduction and/or extra-group paternity, only revealed by the use of molecular techniques, can result in lower levels of within-group relatedness than originally predicted (Brooker *et al.*, 1990; Mulder *et al.*, 1994; Dickinson & Akre, 1998; Richardson *et al.*, 2002). Thirdly, helping kin may be a by-product of philopatry, rather than an active decision to help based on kinship (Clutton-Brock, 2002).

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Studies have often failed to find a relationship between helper contribution and relatedness (Rabenold, 1985; Du Plessis, 1993; Dunn *et al.*, 1995; Clutton-Brock *et al.*, 2000, 2001). Therefore, helping behaviour may be selected for by direct benefits or, alternatively, may merely be a neutral behavioural response by subordinates to the presence of begging young (Jamieson, 1991).

In situations where there is no positive correlation between helping and relatedness, helping can still provide indirect benefits if subordinates are, on average, more related to recipients than to random individuals in the population. However, indirect fitness benefits would be maximized by individuals that accurately assess relatedness and preferentially help more closely related kin. Subordinates could potentially assess kinship through a variety of mechanisms (reviewed in Komdeur & Hatchwell, 1999); either directly (e.g. through phenotype matching), or indirectly (by using associative learning cues), i.e. only helping offspring raised by parents who fed the subordinate when it was young. Associative learning cues may, however, be unreliable if high levels of extra-group parentage result in low or unpredictable kinship patterns between subordinates and their putative parents. A few studies have shown that subordinates use associative learning cues to determine when to help (Komdeur, 1994a; Hatchwell *et al.*, 2001; Russell & Hatchwell, 2001), but none of these studies have compared direct measures of genetic relatedness against helping levels.

In the Seychelles warbler (*Acrocephalus sechellensis*) some individuals [normally females: $n = 217$ (88%), Komdeur, 1996; $n = 63$ (68%), Richardson *et al.*, 2002], become subordinates on their natal territory because of a combination of constrained suitable independent breeding opportunities and the benefits of remaining on high quality territories (Komdeur, 1992; Komdeur *et al.*, 1995). These subordinates help raise dependent young, thus improving the reproductive success attributed to the breeding pair (Komdeur, 1994a,b). Recent molecular analysis has shown that joint nesting by subordinate females occurs frequently (Richardson *et al.*, 2001) and that direct benefits through breeding are the predominant benefit gained by subordinate females (Richardson *et al.*, 2002).

However, previous studies have shown that subordinates also help to raise nondescendent offspring and that indirect benefits are gained in Seychelles warblers (Komdeur, 1994a; Richardson *et al.*, 2002). In addition, it is thought that subordinates base their helping decisions on the presence of their parents within the territory (Komdeur, 1994a). However, the efficiency of these cues in maximizing indirect fitness benefits has not been tested using molecular-based coefficients of relatedness. Furthermore, although extra-group maternity does not occur, extra-group paternity is common, occurring in 40% of Seychelles warbler breeding pairs (Richardson *et al.*, 2001). Thus the continued presence of the primary male, in contrast to that of the primary female, will not accurately indicate subordinate-nestling kinship and should not be used as a cue to determine when to help in order to gain indirect benefits.

The present study uses genotyping data from microsatellite markers to determine coefficients of relatedness between individuals in a population of the Seychelles warbler. Whether subordinate Seychelles warblers efficiently maximize the indirect (kin) benefits of cooperative breeding by preferentially feeding related nestlings was tested by using only subordinates that did not gain direct parentage within the territory. It was predicted that subordinates will provision if the primary female, which attended on them as a chick, is still present within the territory, but not if the primary female has changed. However, as subordinates cannot trust their legitimacy through the male line, provisioning is predicted to be independent of the identity of the primary male.

Methods

The population of Seychelles warblers on Cousin Island has been intensively monitored since December 1985. During this time, as nearly all birds within the population have been individually colour-ringed (using a unique combination of three UV-resistant colour rings and a British Trust for Ornithology metal ring) and monitored throughout all breeding seasons, the reproductive history, status and putative pedigree of almost all birds was known.

The present study was on adult warblers observed during the main breeding season (June–September) in the years 1995–98. Approximately 96% of birds were colour-ringed and blood-sampled. Blood samples (*c.* 15 μL) were collected by brachial venipuncture, diluted in 800 μL of 100% ethanol in a screw-cap microfuge tube and stored at room temperature. Sexes were confirmed by PCR analysis (Griffiths *et al.*, 1998). The status of all birds was based upon field observations from the given field season combined with the detailed long-term demographic data available. The 'primary' male and female were defined as the dominant, pair-bonded male and female in a territory, whereas 'subordinates' included all other birds (>8 months old) resident in the territory.

Genotypes were identified for all individuals using microsatellites previously isolated in the Seychelles warbler (Richardson *et al.*, 2000) and the parentage of all nestlings was determined by using the method described by Richardson *et al.* (2001). The software program KINSHIP (Goodnight & Queller, 1999) was used to calculate an individual pairwise relatedness index (r) based on microsatellite loci genotype similarity, for all within the territory dyads of (i) subordinate-nestling (not including direct subordinate parentage), (ii) subordinate-primary female and (iii) subordinate-primary male. The mean subordinate-nestling pairwise relatedness was calculated when there was more than one nestling. The mean pairwise relatedness among all random individuals in the population equalled zero.

Each territory was checked for nesting activity at least once every 2 weeks by following the resident female for 30 min (Komdeur, 1992). The Seychelles warbler produces one clutch per season, which normally consists of just one egg (80%), although two- or three-egg clutches may occur (Komdeur, 1991). Multi-egg clutches may be the result of multiple laying by the dominant female or a result of more than one group female laying within the same nest (Richardson *et al.*, 2001). The provisioning behaviour of colour-ringed birds was observed in 23 different territories containing a breeding pair and at least one subordinate (15 territories contained one subordinate, seven contained two and one contained three). During the nestling period, a minimum of two 90-min observation periods (1 week apart and randomized with respect to time of day) were completed at each nest. The provisioning frequency (feeds per hour) of each bird in the territory was calculated. Subordinates were split into subordinate parents (eight females, one male) and subordinate nonparents (13 females, 10 males). Within nonparent subordinates, 'helpers' provisioned nestlings whereas 'nonhelpers' were never observed provisioning.

Territory quality, subordinate age, subordinate sex, the number of offspring and the number of subordinates present on a territory were also included in the analysis of provisioning behaviour as these factors have previously been shown to influence helping (Komdeur, 1991; Komdeur, 1994b). Territory quality (T) has been shown to be dependent on insect prey availability and was determined using the method of Komdeur (1994b). Subordinate age was determined by using either the long-term demographic data available or by using eye colour. Juvenile birds (<8 months old) have grey eyes and adult birds have chestnut brown eyes (Komdeur, 1991).

A larger sample ($n = 40$) of all nonparent subordinate birds observed on Cousin between 1997 and 2000 was collated to test whether relatedness to a primary male or female in the territory was a good indicator of relatedness to nestlings, i.e. subordinate-nestling relatedness correlated with (a) subordinate-primary female relatedness, or (b) subordinate-primary male relatedness.

Statistical analysis

T-tests were used to assess differences in mean provisioning levels. Paired *t*-tests were used to control for possible differences between territories when comparing the provisioning levels of subordinates with primary birds. Univariate linear regressions were initially used to determine the relationship between provisioning and subordinate-nestling relatedness for both sexes. General linear model analysis was used to test the effects of multiple independent variables on the provisioning frequency of nonparent subordinate Seychelles warblers. In the multivariate analyses, the minimum adequate model is presented following parsimonious streamlining (the stepwise backwards removal of nonsignificant terms) of the original model. Tests were performed using SPSS 10.7 (SPSS Inc., 1999). All tests are two-tailed and corrected for continuity or tied ranks as appropriate. Unless otherwise stated, mean values are given as ±SD.

Results

There was no significant difference between the sexes in the amount of provisioning by primary birds (Table 1). Subordinates of both sexes were grouped together in the initial analysis of the provisioning data. Provisioning by individuals within each status class was compared against the primary female within the territory using paired sample *t*-tests (Table 1). There was a slight nonsignificant difference in the provisioning by subordinate parents compared with primary female (Table 1); however, the sample size for subordinate parents was low in this test. The amount of provisioning by nonparent helpers was significantly less than that of the primary female, but was not significantly different from that of subordinate parents (paired *t*-tests: $t = 0.28$, d.f. $= 21$, $P = 0.78$; Table 1). Combining subordinate helpers and subordinate parents provided a more powerful test, which indicated that subordinates that help (both parents and nonparents) provision nestlings less than the primary female in their territory (subordinate vs. primary female: 5.44 ± 3.10 vs. 7.82 ± 2.87; paired *t*-tests: $t = 3.43$, d.f. $= 22$, $P = 0.002$). In addition, when separated by sex, female subordinates that helped (both parents and nonparents) provisioned significantly more than did their male counterparts (6.31 ± 3.11 vs. 3.45 ± 2.10, respectively; independent *t*-test: $t = 2.21$, d.f. $= 21$, $P = 0.038$).

Subordinate parents always provisioned and were, therefore, excluded from subsequent analyses. The relationship between provisioning level and pairwise relatedness to group offspring was investigated for female and

Table 1 Provisioning by primary and subordinate Seychelles warblers. For male primary birds and subordinate birds, levels of provisioning were compared against the feeding rate of the primary female in the territory. Statistical significance was assessed by paired sample *t*-test.

Status	*n*	Feeds per hour	Compared to primary female (*t*)
Primary birds			
Female	23	7.30 ± 3.09	
Male	23	6.82 ± 2.92	0.68^{ns}
Subordinates			
Parents	9	5.20 ± 3.23	2.25^{**}
Nonparent helpers	14	5.59 ± 3.12	2.50^{*}
Nonparent nonhelpers	9	0.00	–

ns, not significant.
$^{*}P < 0.05$; $^{**}P < 0.1$.

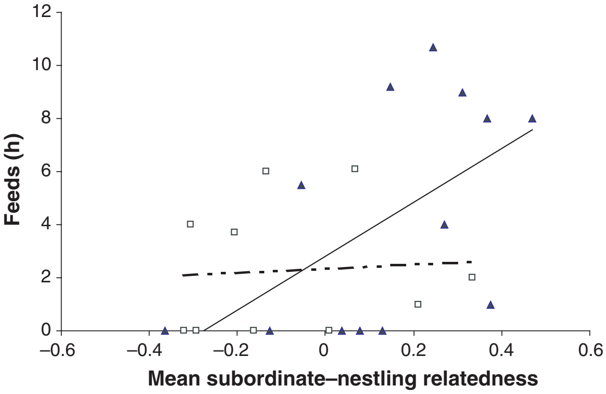

Fig. 1 Provisioning by female (filled triangles) and male (empty squares) nonparent subordinates in relation to subordinate-nestling relatedness (r). There was a positive relationship between provisioning and subordinate-nestling relatedness for female subordinates (solid line) but not for male subordinates (broken line).

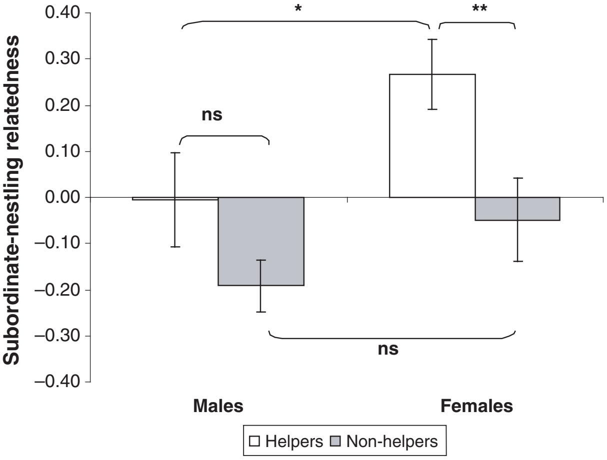

Fig. 2 The relatedness (r) of female and male subordinates to helped or nonhelped nestlings. Bars indicate mean ± SE. $^{*}P < 0.05$, $^{**}P < 0.01$.

male nonparent subordinates (Fig. 1; the distribution of relatedness values for female and male nonparent subordinates is also shown in this figure). For female nonparent subordinates there was a positive relationship between provisioning and subordinate-nestling relatedness ($F_{1,11} = 4.82$, $P = 0.05$, $r^2 = 0.31$; Fig. 1), but not for male subordinates ($F_{1,8} = 0.04$, $P = 0.86$, $r^2 = 0.00$). In a general linear model analysis (Table 2), only subordinate-nestling relatedness was found to positively influence subordinate provisioning, whereas territory quality, subordinate sex, number of nestlings, number of subordinates or subordinate age did not help explain variation in provisioning.

We then compared the subordinate-nestling relatedness of those nonparent subordinates that provisioned with those that did not. Female helpers were significantly more related to nestlings than were female nonhelpers (0.27 ± 0.16, $n = 8$ vs. −0.05 ± 0.20, $n = 5$, respectively; $t = 3.13$, d.f. = 11, $P = 0.01$; Fig. 2). No significant difference in relatedness to nestlings was found between male helpers and male nonhelpers (0.01 ± 0.25, $n = 6$ vs. −0.19 ± 0.15, $n = 4$, respectively; $t = 1.33$, d.f. = 8, $P = 0.22$; Fig. 2). Only female helpers were significantly related to nestlings [female helper-nestling relatedness vs. nonrelated (zero): 0.27 ± 0.16 vs. zero; one-sample t-test: $t = 4.64$, d.f. = 7, $P < 0.002$]. All other subordinate nonparent categories had negative mean relatedness indices (Fig. 2). Finally, female helpers were significantly more related to nestlings than were male helpers (0.27 ± 0.16, $n = 8$ vs. −0.01 ± 0.25, $n = 6$, respectively; $t = 2.47$, d.f. = 12, $P = 0.03$).

Table 2 Factors influencing the provisioning frequency of nonparent subordinate Seychelles warblers. Variables highlighted in bold were left in the minimal adequate model after stepwise removal of nonsignificant variables. The analysis was repeated after replacing 'number of subordinates' with 'number of provisioning adults', thus excluding nonhelping subordinates; however this change did not have a significant effect on the model.

Independent variables	F	P-value
Territory quality	0.010	0.922
Subordinate sex	0.026	0.875
Number of nestlings	0.121	0.733
Number of subordinates	0.970	0.339
Subordinate age	0.702	0.414
Subordinate-nestling relatedness	**7.561**	**0.011**

d.f. = 1,21; $r^2 = 0.214$.

We tested whether the continued presence of either the primary male or female in the territory was a good indicator of subordinate-nestling relatedness. When entered together in a multiple regression analysis, subordinate-primary male relatedness did not predict subordinate-nestling relatedness and, was removed from the model ($F_{2,37} = 2.11$, $P = 0.16$), whereas subordinate-primary female relatedness did ($F_{1,38} = 27.37$, $P < 0.001$).

Did nonparent female subordinates adjust their provisioning according to the continued presence of either their putative mother or putative father within the territory? In a multiple regression analysis, the presence of the putative father had no effect on female subordinate provisioning ($F_{2,10} = 0.02$, $P = 0.88$), but the presence of the putative mother did ($F_{2,10} = 12.53$, $P = 0.005$, $R^2 = 0.56$). There was no interaction effect. Figure 3 shows that female subordinates provisioned significantly more when the primary female that fed them as chicks was still present in the territory (presence vs. absence of putative mother: 7.57 ± 2.44, $n = 6$ vs. 1.43 ± 3.60 $n = 7$; t-test: $t = 3.71$, d.f. = 11, $P = 0.003$). Levels of provisioning did not alter with the presence/absence of the primary male (presence vs. absence of

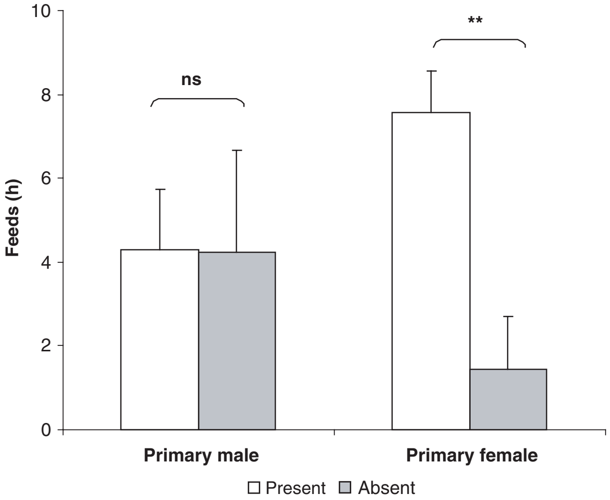

Fig. 3 Provisioning by female subordinate (nonparents) in relation to the continued presence of their putative parents. Bars indicate mean ± SE. $^{**}P < 0.01$.

Table 3 Variables predicting the rate of provisioning by female, nonparent, subordinate Seychelles warblers. The presence of the primary female was the only variable left in the minimal adequate model after stepwise removal of nonsignificant variables.

Explanatory variable	*F*	*P*-value
Presence of primary female	**13.72**	**0.003**
Subordinate-primary female relatedness	0.062	0.810
Subordinate-nestling relatedness	2.137	0.174

d.f. = 1,11; $r^2 = 0.55$.

putative father: 4.30 ± 4.89, $n = 4$ vs. 4.24 ± 4.29 $n = 9$; $t = 0.02$, d.f. = 11, $P = 0.98$). Male subordinates were not included in this analysis as no link was detected between provisioning and male subordinate-nestling relatedness (see above).

Finally, female subordinates were investigated further to determine which cues determined provisioning levels. The continued presence of the primary female explained the level of provisioning better than subordinate-nestling relatedness or subordinate-primary female relatedness (Table 3).

Discussion

We investigated the effect of relatedness between nestlings and subordinates on provisioning behaviour in the cooperative breeding Seychelles warbler. Molecular techniques were used to determine genetic relatedness, thereby incorporating the effects of reproductive sharing and extra-pair parentage on within-group kinship patterns.

We found that investment in provisioning varied greatly between the different members of the cooperative groups. Subordinate parents of both sexes appear to accurately assess when they are parents and always provisioned. However, for subordinates that did not gain parentage, provisioning was positively correlated to their relatedness to the dependent nestlings, although this was the case only for female subordinates and not for male subordinates (who also provisioned less in general). Female subordinates that helped were significantly related to nestlings (mean $r = 0.27$), whereas female nonhelpers were not (mean $r = -0.05$). So, within a territory, nonparent female subordinates do appear to adjust their provisioning effort according to their kinship to nestling(s). For male subordinates neither helpers nor nonhelpers were significantly related to nestlings, nor was there a significant difference in the relatedness of the two groups. However, the results must be treated with caution, as male helping is rare in this system and, consequently, the sample sizes for male subordinates were relatively small. We did not detect a link between the level of provisioning by subordinates and any of the other factors tested.

It is possible that some nonparent female subordinates may provision because they incorrectly judged themselves to be parents, for example, if they had laid an egg that was ejected subsequently by the primary female, or that did not hatch, or that was predated. However, no ejection of eggs has been reported in this species, even when false eggs were put into the nest (Komdeur, 1999). Furthermore, it has previously been shown, using the same data set as in this study (Richardson *et al.*, 2002), that there was no relationship between the probability of being a co-breeder and the relatedness of the subordinate to the territorial female. So, even if some subordinate females did lay eggs that did not hatch, we would not expect such failures to be biased towards individuals who were more related to the remaining nestling(s). Failed co-breeding attempts would not, therefore, undermine the result that subordinates help when more related to the nondirect offspring. It has also been suggested that in cooperative breeding systems subordinates may be coerced into helping as a 'payment of rent' for staying on the territory (Gaston, 1978), but there is no evidence to suggest that this occurs in Seychelles warblers (i.e. no observations of aggression towards, or eviction of, nonhelping subordinates; D. Richardson & J. Komdeur, unpublished data). Furthermore there is no reason to think that only helpers who are closely related to the nestlings would be coerced.

The results described here and in another paper (Richardson *et al.*, 2003), indicate that kinship was the major determinant of provisioning levels in female (but probably not in male) nonparent subordinate Seychelles warblers. Subordinates have been shown to help more closely related kin preferentially in some other cooperatively breeding species (Curry, 1988; Emlen & Wrege, 1988; Russell & Hatchwell, 2001); however, many other studies have shown that this is not a universal trait

(Reyer, 1980; Dunn *et al.*, 1995; Cockburn, 1998; Clutton-Brock *et al.*, 2000).

For indirect benefits to accrue to the subordinate, there must be both relatedness between the donor (i.e. the subordinate) and the recipient (i.e. the nestlings), and benefits to the recipient (Hamilton, 1964). In Seychelles warblers, the amount of food brought to the nestling determines fledging success and first-year survival (Komdeur, 1991), and the presence of helpers increases the number of young fledged on a territory (Komdeur, 1994b). During the present study, the number of fledglings produced on a territory increased significantly (by 17%, even after excluding direct parentage) for each subordinate present (Richardson *et al.*, 2002). Therefore, the preferential provisioning of related young by female subordinates will increase the number of related offspring produced and so maximize the indirect benefits of cooperative breeding. Male subordinates, which provision less than females and which are not normally related to the nestlings they do provision, do not appear to help in order to gain indirect fitness benefits. However, firm biological conclusions are hard to draw, as male helper sample sizes are low.

If male subordinate Seychelles warblers do not gain indirect benefits, then why do they help? Helping may allow a male subordinate to ingratiate itself within a group (or with a female), and thus gain the various direct benefits open to nonrelated subordinates (reviewed in Cockburn, 1998). Previous work on the Seychelles warbler has shown that some male subordinates (budders) may reside on a territory in order to bud off part of that territory in which to breed independently (Komdeur & Edelaar, 2001). In addition, subordinate males may occasionally gain direct benefits through within-group paternity, although this is rare compared with female subordinate within-group maternity (Richardson *et al.*, 2001, 2002). The benefits of direct breeding opportunities, possibly combined with an inbreeding avoidance mechanism, could explain why males often become subordinates on non-natal territories (five of 11 subordinate males in this study). The less philopatric nature of male subordinates can in turn, explain why male helpers do not gain indirect benefits, although it is difficult to unravel the direction of this relationship. The present study supports the idea that the benefits of cooperative breeding may differ between the sexes (Cockburn, 1998), but further work is needed to uncover the causes and consequences of male helping behaviour in this species.

In the present study, we show that for subordinate nonparent females the continued presence of the putative mother explained provisioning better than actual relatedness between subordinate and nestlings (Table 3). This indicates that subordinate females use their putative mother (i.e. any female who provisioned the subordinate when it was a nestling) as the cue to determine provisioning levels, as opposed to directly measuring their kinship to the nestlings. Further evidence that subordinates use associative learning cues comes from cross-fostering experiments performed as part of another study (J. Komdeur & D.S. Richardson, unpublished data). Subordinates raised by foster-parents had the same chance of helping nestlings, as did subordinates raised by their social parents. Four cross-fostered females were observed as nonparent subordinates in subsequent years. In all cases, the original primary female was still present in the territory and in three of these cases the subordinate provisioned the nest.

As predicted, the presence of the putative father was not used as a cue to determine provisioning levels. This is logical in an evolutionary sense as the correlation between the subordinate-primary male relatedness and subordinate-nestling relatedness was greatly reduced as a result of the high frequency of extra-pair paternity found in this species (Richardson *et al.*, 2001). Subordinate-nestling pairs produced under the same primary parents have a 72% (85% × 85%) chance of having the same mother (85% of chicks are produced by the primary females; Richardson *et al.*, 2001). However, as only *c.* 60% of the nestlings are produced by the primary male (Richardson *et al.*, 2001), then only *c.* 36% (60% × 60%) of these pairs are expected to have the same genetic father.

A previous study on Seychelles warblers assessed the effect of kinship on the provisioning behaviour of subordinates (Komdeur, 1994a). However, without the use of molecular techniques to determine parentage and relatedness, this study could not take into account co-breeding by subordinates, nor could it accurately test whether this strategy resulted in the efficient maximizing of indirect benefits. Komdeur's study correctly identified that subordinates used associative learning, but suggested that it was based on the continued presence of both putative parents within the territory. It now appears that the results from this earlier study, reporting that helpers could distinguish between sibs and half-sibs, might have been an artefact caused by helpers only assisting their mothers. Other studies have also shown that subordinates preferentially help their parents (Jones *et al.*, 1991; Dickinson *et al.*, 1996) and that helping may be directed through associative learning cues (Hatchwell *et al.*, 2001; Russell & Hatchwell, 2001). Importantly, the present study (described here and in Richardson *et al.*, 2003) is the first to show that this learning mechanism could have evolved to be focused only on one sex (the female attendant at the nest), the sex to which the subordinates are reliably related.

The benefits that subordinates gain through direct breeding are substantially higher than the indirect benefits of cooperative breeding for both sexes of subordinate Seychelles warblers (Richardson *et al.*, 2002). However, here we show that female subordinates are able to allocate their investment to maximize the indirect benefits they do gain. So, whereas ecological constraints coupled with direct benefits may be the main selection pressure behind becoming a subordinate, provisioning by female nonparent subordinates appears to be selected through indirect benefits. This study confirms that the evolution of vertebrate cooperative breeding systems must be addressed in two parts. (i) Why do individuals become subordinates? (ii) Why do these subordinates help? One could argue that the Seychelles warblers' cooperative breeding system may have evolved along the following path: after the species was restricted to small discrete islands, ecological constraints (i.e. limited good quality habitat) and the initial benefits of philopatry (i.e. increased survival) led to individuals becoming subordinates within their natal group. Occasional direct breeding could then have reinforced the benefits of group living for subordinate females, but led male subordinates to disperse to non-natal territories to gain direct breeding benefits while avoiding inbreeding. Helping to provision related offspring might have evolved in nonparent female subordinates later, as a way of making the most of their situation by maximizing indirect benefits. However, this is mere speculation, as we do not yet have the evidence to determine which benefits, direct breeding or indirect kin benefits, evolved first.

Recent studies have provided evidence for direct (Cant & Field, 2001; Clutton-Brock *et al.*, 2001) or indirect benefits (Russell & Hatchwell, 2001) in different systems and have fuelled the argument over their importance in the maintenance of cooperative breeding (Emlen, 1995; Cockburn, 1998). The evidence from the Seychelles warbler (Richardson *et al.*, 2002, 2003 and the present study) supports the idea that both direct and indirect (breeding) benefits may be important within a cooperative breeding system.

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