Original article

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Paternity and paternal care in the polygynandrous Smith's longspur

Abstract In species where females copulate with more than one male during a single breeding attempt, males risk investing in offspring that are not their own. In the polygynandrous Smith's longspur (*Calcarius pictus*), females copulate sequentially with one to three males for each clutch of eggs and most of these males later assist in feeding the young. Using multilocus DNA profiling, we determined that there was mixed paternity in $>75\%$ of broods ($n = 31$) but that few offspring ($<1\%$ of 114 nestlings) were sired by males outside the polygynandrous group. Male feeding rate increased significantly with the number of young sired, with males siring four nestlings feeding the brood at double the frequency of males siring only a single nestling. However, male Smith's longspurs appear to show a graded adjustment of paternal care in response to paternity only when other males are available to compensate for reduced care: feeding rate did not vary in relation to paternity when only one male provisioned young at the nest. There was no evidence that males could recognise their own offspring within a brood and feed them preferentially. The number of offspring sired by each male was significantly correlated with the number of days spent copulating with the attending female: on average, a male sired one offspring for every 2 days of copulatory access. If males use their access to females to estimate paternity (and thereby decide on their subsequent level of parental investment), a positive relationship is expected between the amount of female access and the subsequent feeding rate to the nestlings. Nonetheless, male feeding effort was only weakly correlated with female access and more study is needed to determine how males estimate their paternity in a brood.

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Introduction

The ways in which males should optimally allocate parental care has been the subject of a number of theoretical and empirical studies (e.g. Maynard Smith 1978; Werren et al. 1980; Davies et al. 1992; Xia 1992; Westneat and Sherman 1993; Wright and Cotton 1994). Perhaps the most intuitive view holds that investment in offspring should be proportional to parentage, with males more confident of paternity making a greater investment (Winkler 1987; Whittingham et al. 1992; Xia 1992; Westneat and Sherman 1993). For example, dunnock (*Prunella modularis*) females regularly copulate with two males for a single clutch of eggs (Davies 1985). Both males later assist the female in feeding her offspring, with the amount of parental care invested by each male dependent largely on his access to the female during the period of copulation (Burke et al. 1989; Davies et al. 1992). As access to the female is a good predictor of paternity, male dunnocks appear to use this accessibility as a rule governing the subsequent allocation of parental care (Burke et al. 1989). In other words, male dunnocks seem to know how many young they have sired and adjust their feeding effort accordingly. Such a process makes evolutionary sense because selection would be expected to favour individuals that adjust their allocation of parental care in relation to genetic paternity to avoid investing in offspring that were not their own (Burke et al. 1989; Davies et al. 1992).

In this study, we examine the relationship between paternity and paternal care in the Smith's longspur (*Calcarius pictus*), a medium-sized bunting of the North American subarctic tundra. Smith's longspurs are unusual among birds in that females regularly pair and copulate with more than one male for a single clutch of eggs while, at the same time, males pair and copulate

with two or more different females (Briskie 1992, 1993). Both sexes share in raising the young and it is not unusual for several males to jointly provide food to the brood of a single female (Briskie 1993). This mating system is termed polygynandry and has been reported in only a few other passerines to date (e.g. dunnock, Davies 1985; alpine accentor, *P. collaris*, Nakamura 1990). Although polygynandrous mating systems are relatively rare among birds, species such as Smith's longspurs provide ideal subjects for the study of paternity and paternal care as it is clear that males in such systems have direct evidence their partners are simultaneously paired to other males. In contrast, extra-pair matings in most socially monogamous species are clandestine and it is not obvious to human observers that pair males in such species are aware of their mate's behaviour or of the risk of lost paternity.

The relatively high frequency with which female Smith's longspurs pair and copulate with several males means that males risk investing paternal care in offspring sired by other males. Possibly, to reduce this cost and increase the likelihood of paternity, copulation is extraordinarily frequent in Smith's longspurs, averaging > 5 copulations per hour and > 350 copulations for each clutch of eggs (Briskie 1992). This is one of the highest copulation rates recorded for any species, and probably functions to dilute or displace the sperm of rival males (Birkhead et al. 1987). Male longspurs also intensely guard their mates during the fertile period and aggressively chase away intruding males (Briskie 1992). Both mate guarding and frequent copulations appear to be common paternity guards among birds (Birkhead and Møller 1992), but it is less clear what strategies males use to minimise the cost of cuckoldry after the fertile period has ended. In this study, we use multilocus DNA profiling to examine if male Smith's longspurs allocate parental care based upon the paternity they obtained within a brood. Although male longspurs normally provide some assistance to females in feeding offspring, the amount varies greatly from nest to nest and individual females may have help from one to three males to feed her young (Briskie 1993).

Methods

Smith's longspurs were studied in three small populations (neighbourhoods) near the town of Churchill, Manitoba, Canada from 1989 to 1993. Each neighbourhood consisted of 8 to 19 birds, all of which were colour-banded for individual identification. In this area, breeding sites are restricted to disjunct patches of open, sedge tundra bordering the northern edge of the boreal treeline. Adults arrive on the breeding grounds in early June; clutch initiation starts by mid-June and most young have fledged by mid-July. Clutch size ranges from three to five eggs but clutches of four are the most frequent. Females alone build the nest and incubate, but both sexes feed the young. Only one brood is raised per year and renesting after nest failure is uncommon (Briskie 1993).

Over the course of our study, we monitored the mating behaviour of 23 female and 30 male longspurs, making daily observations of birds as they interacted with other individuals in the study area. Each day we located every female and recorded the identity of the male with which she was copulating at that time. Although female Smith's longspurs usually pair and mate with two or three males for a single nesting attempt, on any given day, copulations are usually restricted to a single male (Briskie 1992). For example, shortly after arriving on the breeding grounds, a female first pairs and mates with one male (here called the primary male) for 3–7 days (mean $\pm$ SE $= 4.18 \pm 0.42$ days, $n = 11$), during which time that male has exclusive access to the female. The female then switches to a second (or secondary) male and copulates with him for a further 1–6 days (mean $\pm$ SE $= 2.67 \pm 0.43$ days, $n = 12$), during which time he has exclusive access to her. Occasionally, a female may even mate with a third (or tertiary) male, although in the two such cases we observed, all copulations with the tertiary male occurred on a single day. Most copulation behaviour ceases upon laying of the penultimate egg, which coincides with the day the last egg in the clutch is fertilised (see Briskie 1992 for a detailed description and discussion of copulation behaviour of this species). There was no evidence that males in polygynandrous groups were relatives (unpublished data).

We used the total number of days over which a male copulated with a female as an index of his access to her for mating. Thus, a male that copulated repeatedly with a female over a period of 5 days was assumed to have greater access than a male that copulated with a female over only 2 days. We also calculated a second index of female access based only on the number of days over which a male was copulating with a female during the egg-laying period (defined as starting on the day the first egg was laid and ending with the laying of the last egg). In most birds, eggs are laid at daily intervals and fertilized about 24 h before they are laid; thus a male may be better able to directly assess the number of eggs he sires through copulations that occur during this period by observing the egg-laying stage of his mate (see Hatchwell and Davies 1992 for an example of this behaviour in dunnocks). Indeed, male Smith's longspurs were observed inspecting nests on several occasions and often accompanied females to the nest during the act of egg-laying.

After copulation activity ceased, we located the nests of all females and monitored their progress until the nest failed or the young fledged. At 5 days after hatching (young fledge at 7 or 8 days of age), the amount of parental care performed by each male (as measured by feeding frequency) was recorded at each of 19 nests. Feeding frequency was observed from a blind 5–10 m from the nest and set up at least 24 h prior to the start of

observations (to allow time for habituation). Each nest was watched for 5–10 h (mean ± SE = 5.9 ± 0.5 h) between 0600 and 2300 hours CDT. For each feeding trip, the identity of each adult visitor was recorded. The average number of feeding trips per hour was used as an estimate of a male's level of investment in parental care. Six males (out of 25) were observed feeding at more than one nest (either in the same year or in different years), but we considered each brood/sire combination to be independent units. Our conclusions are not changed by using data from each male for only the first nest at which he was studied. At four nests, we also marked nestlings on the head with a unique pattern of ink blotches for individual recognition. We then recorded which nestlings were fed by each of the attending males. Our objective here was to determine if males were preferentially feeding their own offspring within a brood.

When the nestlings were 5–6 days of age, we collected about 100 μl of their blood by jugular or brachial venipuncture. Blood samples from adults were collected either at the time of banding, usually early in the breeding season, or during the nestling period. In the latter cases, adults were trapped on the nest, bled and released at least 24 h prior to the start of any feeding observations. Banding and blood collection did not cause any mortality or nest desertion and most adults returned to feeding nestlings within 10–20 min of being released. Blood samples were added immediately to 1 ml of Queen's lysis buffer (Seutin et al. 1991) and stored at 4°C until processed for DNA profiling up to several months later.

Multilocus DNA profiles were made and analysed using standard techniques that we have described in detail elsewhere (Weatherhead et al. 1994; Põldmaa et al. 1995). Briefly, we extracted (phenol:chloroform) DNA from the blood samples, then digested 15 μg samples of DNA with a restriction enzyme. We used the restriction enzyme *Mbo*I for profiles made from 1989 to 1991 samples but we switched to *Alu*I for the 1992–1993 samples because it improved the readability of DNA profiles and the number of bands (DNA fragments) that were scorable (Table 1). We ran 5 μg of the enzyme-digested DNA (with a lambda DNA internal size marker) through an agarose gel until fragments smaller than 1–2 kb had migrated off the gel. For each family, all nestlings, the attending female, all attending males and any other male neighbours were loaded onto the same gel to facilitate analysis. DNA fragments were transferred to Immobilon-N membranes by Southern blotting. Membranes were then hybridised sequentially to minisatellite probes and then a lambda DNA probe (used to detect lambda internal size marker bands), all of which were labelled with [^{32}P]-dCTP. We probed all profiles with Per, Jeffreys 33.15 and lambda. DNA profiles for 12 nests gave equivocal results for one or more nestlings, so these were also probed with Jeffreys 33.6 (seven nests) or new gels were produced using the enzyme *Hae*III and probing with Per and Jeffreys 33.15. In each case, the additional DNA profiles allowed us to resolve the paternity assignments.

Parentage was assigned by scoring the bands on autoradiograms made from the radioactively labelled membranes. Autoradiograms were digitised and scored using a microcomputer. The number and molecular size of DNA fragments (see Table 1) in the 3.5- to 21.2-kb range were determined using GelReader (version 2.05) software. We used both band-sharing coefficients (BSCs) and novel bands to assess paternity (Table 1). For each nestling, we initially assigned paternity to any male with BSC > 0.40 and ≤ 1 novel band, as in our previous studies using this technique (e.g. Weatherhead et al. 1994; Põldmaa et al. 1995). In the few cases where more than one male fitted this criterion, or the BSC was relatively high (> 0.40) and there was more than one novel band, we ran additional profiles as described above. DNA profiling and scoring of bands was done independently of data on the birds' copulation and parental behaviour. In total, we determined the parentage of 31 broods and 114 nestlings.

Because of small sample sizes and significant departures from normality for most variables, we used non-parametric tests for most statistical analyses. To show trend lines on graphs we plotted cubic splines (Schluter and Nychka 1994); this is a non-parametric regression method that makes no assumptions about linearity and normality. We used one-tailed tests for all analyses of

Table 1 Summary of DNA profiling analyses. Data are presented as the mean ± SE, followed by the range, and the sample size in parentheses for the combined results from Per and Jeffrey's 33.15 probes. Only the first profile made for each individual is counted here – paternity analysis in some cases was based on additional DNA profiles, particularly when the band-sharing coefficient between nestling and putative sire was < 0.40 and there was > 1 novel band (see text)

	Enzyme	
	*Alu*I	*Mbo*I
Number of bands		
Sires	36.8 ± 1.5, 24–47 (19)	28.1 ± 1.6, 18–38 (14)
Mothers	35.6 ± 1.8, 23–47 (15)	28.4 ± 2.3, 14–43 (17)
Nestlings	31.4 ± 0.9, 12–42 (54)	27.9 ± 1.9, 9–42 (57)
Band-sharing coefficients		
Nestling-sire	0.59 ± 0.01, 0.43–0.75 (48)	0.55 ± 0.02, 0.21–0.79 (33)
Nestling-mother	0.58 ± 0.01, 0.40–0.76 (54)	0.58 ± 0.01, 0.26–0.80 (56)
Novel bands	0.15 ± 0.08, 0–3 (48)	0.41 ± 0.15, 0–3 (34)

the correlation between paternity and paternal care because a positive relation was expected both from theory (Westneat and Sherman 1993) and from other empirical studies (Davies et al. 1992). Means are given ±1 SE throughout. Nest loss due to predators and poor weather meant that sample sizes for paternity success, feeding frequency and observations of copulation behaviour are not always the same.

Results

Social and genetic mating system

Most Smith's longspurs formed mating partnerships with two or more members of the opposite sex. Of 23 colour-banded females followed over the copulation period, 17 (73.9%) formed partnerships and copulated with two different males, and 2 (8.7%) with three different males: only 4 (17.4%) females were observed to copulate with a single male. On average, females formed polyandrous associations with 1.9 (range 1–3) males for each clutch of eggs. Males similarly formed polygynous associations with one to three different females: of 30 colour-banded males followed over the copulation period, 9 (30.0%) copulated with only one female, 19 (63.3%) copulated with two females and 2 (6.7%) copulated with three females. Males were seen copulating with 1.8 (range 1–3) females on average, although the copulation success of males was probably underestimated as they were less easily followed than females. No unmated males or females were found in the three neighbourhoods we studied.

DNA profiling demonstrated that most Smith's longspur broods were of mixed paternity. We found that only 7 of 31 broods (22.6%) were sired by a single male. In contrast, 20 broods (67.7%) were sired by two males, and 4 broods (12.9%) by three different males. The number of sires per brood averaged 1.90 (range 1–3). The proportion of broods with mixed paternity (77.4%; 24 of 31 broods) was very similar to that expected from the frequency of social polyandry by females (82.6%; 19 of 23 females), indicating that observed mating associations in this species parallel the underlying genetic mating system. No conspecific brood parasitism was detected.

For 13 broods we knew the status of all males (primary, secondary, tertiary) from their copulation behaviour. At one of these nests, only a single male copulated with the female and he sired all four offspring. At the remaining 12 nests, males shared paternity with up to two other males: of 44 offspring in these nests, primary males sired 26 (59.1%), secondary males 16 (36.4%) and tertiary males 2 (4.5%) of the nestlings. On average, primary males sired 2.31 ± 0.33 nestlings (range 1–4) per brood, secondary males sired 1.33 ± 0.26 nestlings (range 0–3) per brood, and the two tertiary males each sired only one nestling per brood ($n = 2$ broods).

Only one possible instance of "extra-pair" paternity was observed among the 114 nestlings sampled: at one nest in 1992, two males were observed copulating with the female but one of the two eggs was fertilized by a third male that had not been observed to copulate. The home range of this third male overlapped completely with that of the female and he was often observed in the area of the female so it is possible that we missed seeing copulations with him. In all other nests, young were sired by males that were known to have previously paired and copulated with the female for at least 1 day.

The probability of mixed paternity did not vary significantly with brood size (2×4 contingency table analysis, $\chi^2 = 2.21$, $P = 0.53$, $df = 3$). Fourteen out of 20 broods of four (70%) and 5 out of 6 broods of three (83.3%) were of mixed paternity (all 3 broods of 2 and both broods of five nestlings were also of mixed paternity). In broods of four with mixed paternity, primary males were much more likely to sire the first- (100% of 8 nests) or second-hatched (62.5%) young in a brood than either the third- (12.5%) or fourth-hatched (0%) offspring. In contrast, secondary and tertiary males never sired the first-hatched young (0% of 8 nests) but often sired the second-hatched (37.5%) young, and usually sired most of the third- (87.5%) and fourth-hatched (100%) young. This intrabrood pattern of paternity corresponds to the sequential pattern of copulation behaviour (Briskie 1992), suggesting that eggs are usually fertilised by the male copulating with the female on the day of ovulation.

Paternity and paternal care

The mother and from one to three males (mean = 1.53 ± 0.14 males, $n = 19$ nests) fed the offspring in each nest where feeding was observed. Two or more males were observed feeding nestlings at 9 (47.4%) of these nests. Most males that copulated with a female returned later to provision the brood: of 39 sire/brood combinations (25 different males at 19 nests), 29 (74.4%; 21 different males at 19 nests) were later observed feeding young. The number of nestlings sired per brood by males that did feed (1.90 ± 0.22 nestlings, $n = 29$ sires) did not differ significantly from the number sired by males that did not feed (1.50 ± 0.22 nestlings, $n = 10$ sires; Mann-Whitney test: $U = 120$, $n = 29,10$, $P = 0.39$), nor did the total number of days that a male copulated with the female differ significantly between males that fed (3.27 ± 0.48 days, $n = 15$ sires) and those that did not feed (3.00 ± 0.95 days, $n = 5$; $U = 35.5$, $n = 15,5$, $P = 0.86$) at nests where they were sires. As brood size did not differ significantly between sires that fed (3.72 ± 0.13 nestlings, $n = 29$) and those that did not (3.50 ± 0.31, $n = 10$; $U = 125.5$, $n = 29,10$, $P = 0.44$), it is unlikely that males failed to provide feeding assistance because of a reduced need in smaller broods. Males were more likely to feed nestlings when they were primary sires (10 of 11, 90.9%) than when they were

secondary sires (6 of 10, 60%) but this difference is not significant (Fisher exact test, $P = 0.15$).

When all males (regardless of mating status) are considered, feeding rate to the brood was positively and significantly correlated with the number of offspring sired (Fig. 1, Table 2) – males that sired four nestlings in a brood fed at twice the rate (mean = 5.52 feeds/h, $n = 3$) of males that sired only one (mean = 2.76 feeds/h, $n = 18$). Male feeding rate as a proportion of both the total feeding rate (all males and the female) and the feeding rate of just the males at a given nest were also significantly positively correlated with the number of chicks sired (Table 2). These relationships between paternity and paternal care were essentially the same when only those males that actually fed at the nest are considered (Table 2). Likewise, male feeding rate and percent of feeds by each male were significantly related to the proportion of chicks sired within a brood (Table 2).

At 10 of the 19 nests at which feeding was observed, only one male delivered food to the brood even though seven of these broods had mixed paternity. At those nests,
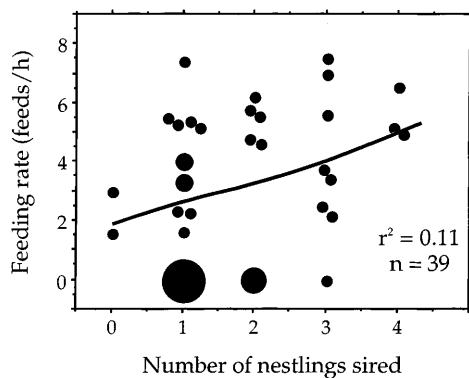
Fig. 1 Relationship between number of nestlings sired and feeding rate (feeds/h) in male Smith's longspurs. Each male was watched for 5–10 h. All males, regardless of mating status are shown in this figure. *Size of symbols* reflects the number of coincident data points and some data points are staggered horizontally to improve clarity. The *plotted line* is a cubic spline (lambda = 10); as in ordinary least-squares regression, r^2 indicates the proportion of variation accounted for by the spline model

there was no significant relationship between feeding rate and the number of offspring sired ($r_s = 0.14$, one-tailed $P = 0.34$, $n = 10$). However, at 3 of the 10 nests, the male was the sole father as well (i.e. the male and female were a monogamous pair). If these 3 nests are excluded, the feeding rate of solitary males with broods of mixed paternity was also not significantly correlated with the number of offspring sired ($r_s = 0.42$, one-tailed $P = 0.16$, $n = 7$). Thus male longspurs do not appear to adjust their level of feeding relative to their actual paternity if they are the only male attendant at a nest.

The actual and proportional (of all adults tending a brood) feeding rate of primary males increased significantly with the number of chicks sired in a brood ($r_s = 0.55$ and 0.58, one-tailed $P = 0.04$ and 0.035, respectively, $n = 11$). However, neither the actual nor the proportional feeding rate of secondary males was significantly correlated with the number of nestlings sired (in both cases $r_s = 0.07$, one-tailed $P = 0.42$, $n = 10$). Our sample sizes are quite small for all of these comparisons and so the power of the tests is limited. Nonetheless, primary males seem to be more likely to adjust the level of parental care in relation to paternity than secondary males.

Do males preferentially feed their own offspring within a brood? We individually marked nestlings in four broods and recorded the identity of all males feeding each of the young. In each case, all males were observed feeding every nestling within a brood, and there was no obvious preferential feeding of any young (Table 3; contingency table analyses, $\chi^2 = 0.33$–6.31, all P-values > 0.20, $n = 4$ broods). Thus, although males adjust their feeding behaviour according to how many young they have sired in a given brood, they do not appear to be able to recognise their own offspring in these nests.

Paternity and female access

To adjust parental care in relation to paternity, males need some way of evaluating their fertilisation success.

Table 2 Relationship between various measures of male parental care and the number and proportion of chicks that they sired in a nest. All Spearman rank correlations (r_s) were done on individual sire/brood combinations (n) at 19 nests. All P values are one-tailed because we were testing the hypothesis that parental care increases with paternity (see text)

	All males			Only males that fed		
	r_s	P	n	r_s	P	n
Number of chicks sired vs sire's:						
feeds/h	0.32	0.02	39	0.36	0.03	29
percent of total feeds/h	0.27	0.05	38	0.25	0.10	29
percent of feeds by males/h	0.35	0.02	38	0.37	0.03	29
Proportion of chicks sired vs sire's:						
feeds/h	0.27	0.03	39	0.34	0.04	29
percent of total feeds/h	0.21	0.10	38	0.25	0.10	29
percent of feeds by males/h	0.31	0.03	38	0.40	0.02	29

Table 3 Two examples of food distribution within Smith's long-spur broods. Data are the total number of feeds to each nestling (designated *1–4*) by each male. All three sires at nest *90-01* fed the young, while only the primary sire at nest *91-01* was observed feeding. Offspring sired by each male are *italicised*. Nest 90-01 was observed for 9.7 h, nest 91-01 for 9.3 h. There was no significant variation in the distribution of food among nestlings within each brood (nest 90-01: $\chi^2 = 6.31$, $df = 6$, $P = 0.39$; nest 91-01: $\chi^2 = 0.33$, $df = 3$, $P = 0.95$)

Nest	Male	Nestling			
		1	2	3	4
90-01	Primary	*11*	9	10	11
	Secondary	3	1	*6*	7
	Tertiary	13	13	11	*9*
91-01	Primary	9	7	*8*	9
	Secondary	0	0	0	*0*

One possibility is that males use their access to the female during the copulation period as an index of subsequent paternity (Burke et al. 1989). As expected, the number of nestlings sired was significantly and positively correlated with the total time spent copulating with a female (Fig. 2a; $r_s = 0.53$, $P = 0.009$, $n = 26$). On average, a male sired approximately one chick for every 2 days that he copulated with a female (Fig. 2a).

A few males, however, were not successful in fertilising even a single egg, despite copulating with a female for up to 3 days (Fig. 2). Since eggs are fertilised ap-proximately 24 h before they are laid, we compared male fertilisation success in relation to female access during the egg fertilisation period. The number of offspring sired was strongly correlated with the total number of days spent copulating with a female during her egg fertilisation period (Fig. 2b; $r_s = 0.70$, $P = 0.0005$, $n = 26$). For every day that a male copulated with a female during this period, he sired about one more offspring, on average (Fig. 2b). Male access to the female during her egg fertilisation period was clearly the best predictor of paternity. Despite this, two males did not sire any young even though they copulated with a female during this period, and three males fertilised a single offspring each even though all of their copulations preceded the egg fertilisation period by more than 24 h (Fig. 2b).

A similar correlation between female access and fertilisation success was observed when males of different mating status were considered separately. Although the number of nestlings per brood that were sired by primary and secondary males was not significantly related to the total number of days that they copulated with the female ($r_s = 0.30$ and 0.42, $P = 0.34$ and 0.16, $n = 11$ and 12, respectively), the number of offspring sired by both primary and secondary males was significantly and strongly correlated with days copulating during her egg fertilisation period ($r_s = 0.73$ and 0.77, $P = 0.02$ and 0.01, $n = 11$ and 12, respectively). We observed the copulation of only two tertiary males during our study and both copulated for a single day. In both instances, this day fell during the egg fertilisation period and each male sired a single young in that female's clutch. Thus, the relation between female access and a male's success at fertilisation appeared similar for all males, regardless of mating status.
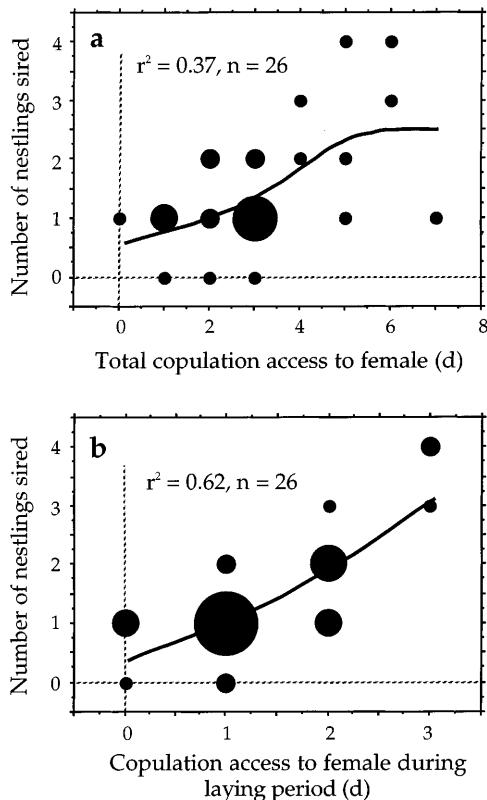
Fig. 2a,b Male fertilisation success in Smith's longspurs in relation to the duration of copulatory access with the female. In both figures, the plotted lines are cubic splines (lambda = 10). **a** Relationship between total number of days a male spent copulating with a female and the subsequent number of young sired. **b** Number of offspring sired in relation to the number of days a male spent copulating with a female during the laying period

Female access and paternal care

If males use their copulation access to the female as an index of subsequent paternity, then a positive correlation is expected between the number of days that they copulated with the female and their feeding investment. However, the relationship between male feeding rate and both the total number of days copulating ($r_s = -0.10$,

$P = 0.67$, $n = 20$) and the number of days copulating during the egg-laying period ($r_s = 0.30$, $P = 0.19$, $n = 20$; Fig. 3) were not significant. The relation between feeding rate and the number of days copulating during egg-laying was significant ($r_s = 0.85$, $P = 0.04$, $n = 7$; Fig. 3) only for primary males who actually copulated during the egg-laying period. Thus there is some evidence that males may use access to the female as an index to paternity, and therefore to adjust their level of paternal care.
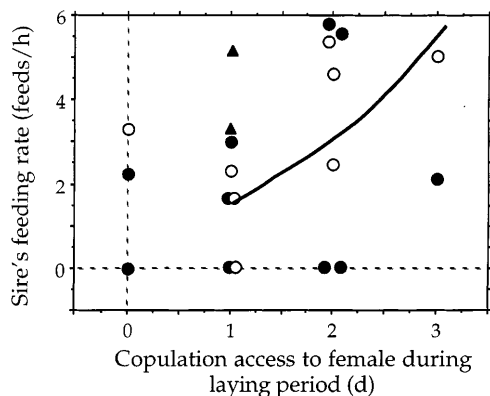
Discussion

Smith's longspurs have one of the highest rates of mixed paternity of any species. Over 75% of broods in our study population were sired by two or more males: about 13% of broods even contained offspring that had been sired by three different males. As males provide approximately half of all parental feeding effort (Briskie 1993, unpublished data), selection should favour males that avoid allocating investment in offspring that are not their own. Our observations of male feeding behaviour suggest that this is indeed the case. Both the absolute frequency of feeding and the proportion of male feeds increased with the number of young sired in a brood. At a given nest, males that sired four young delivered food at double the rate of males siring only a single chick. These results suggest male longspurs are able to measure their fertilisation success and adjust their parental investment accordingly – but how does a male know how many young he has sired?

In polyandrous and polygynandrous groups of the dunnock, a male's access to a female during the copulation period provides a good prediction of his subsequent paternity (Burke et al. 1989; Davies et al. 1992). Through a series of removal experiments, Davies et al.

Fig. 3 Relationship between feeding rate of male Smith's longspurs and their copulation access during the egg-laying period. Primary sires are shown as *open circles*, secondary males as *closed circles* and tertiary males as *triangles*. The *plotted line* is a cubic spline (lambda = 10) through data for primary males that copulated during the egg-laying period ($r^2 = 0.56$, n = 7). Some data points are staggered horizontally to improve clarity

(1992) demonstrated that when two males copulate with a female and both help at her nest, there is a clear causal link between a male's share of mating access, his paternity success and his subsequent level of paternal care. Those males with access to the female during the egg-laying period (i.e. when eggs were being fertilised) were more likely to sire offspring than males without access, and the amount of access was positively related to both paternity and paternal care. Davies et al. (1992) suggest that males use their access to the female as an indirect method to predict their paternity, and that this is then used to adjust feeding behaviour accordingly. In Smith's longspurs, we also found that paternity was positively correlated with male access, particularly during the laying period. On average, a male longspur could expect to sire one chick for every 1–2 days spent copulating with a female. Nonetheless, we found a positive relationship between male access and the level of paternal care only for primary males. This result may simply reflect our relatively small sample size, but it is also possible that males use other cues to estimate paternity success, such as copulation frequency or female solicitation behaviour. Although understanding the exact mechanism by which males assess paternity will require an experimental approach in which access and other variables can be controlled (e.g. Wright and Cotton 1994), our results agree with the findings of Davies et al. (1992) that male access during the laying period is particularly important in determining paternity.

Previous studies on the effect of varying paternity on parental care have been equivocal. In some species, males seem to reduce parental care in response to either perceived or real reductions in their paternity (e.g., reed buntings *Emberiza schoeniclus*, Dixon et al. 1994; European starlings *Sturnus vulgaris*, Wright and Cotton 1994), but in most other species no effect has been observed (e.g. tree swallows *Tachycineta bicolor*, Lifjeld et al. 1993; Whittingham et al. 1993; purple martins *Progne subis*, Wagner et al. 1996; yellow warblers *Dendroica petechia*, Yezerinac et al. 1996). In red-winged blackbirds (*Agelaius phoeniceus*), males in one population reduced parental care in response to lost paternity (Weatherhead et al. 1994), while in another population no effect was found (Westneat 1995). For males to respond to changes in paternity appropriately in any of these species, they must have the ability to assess that their paternity has changed. In dunnocks, and perhaps Smith's longspurs, males may use their access to the female during the copulation period as an index to their subsequent paternity. In contrast, it is not clear what cues males in socially monogamous species might use to assess paternity. Extra-pair copulations in many species are notoriously clandestine and usually take place in the absence of the pair male, suggesting that most may go undetected by the partner. Westneat (1993) found only a weak correlation between paternity in red-winged blackbirds and the frequency of within-pair copulations and no relationship between paternity and a number of behavioural cues that could potentially be used by males

to assess paternity (e.g., frequency of extra-pair courtships, number of chases by extra-pair males). Thus, it is possible that males in many species simply cannot with certainty ascertain their paternity and so do not reduce paternal care in response to a decrease in paternity.

An alternative view for why some males may not adjust their investment in response to paternity was proposed by Whittingham et al. (1992) and Westneat and Sherman (1993). They suggest that since optimal parental effort depends on the relative costs and benefits of parental behaviour, differences in the shape of these cost/benefit curves could lead to differing responses to paternity changes. For example, if increasing parental effort has a dramatic increase on the survival of the offspring, but only a minimal cost, then even large reductions in paternity may not be enough to favour a reduction in the optimal level of parental care. Such a situation seems likely in those species of birds where parental feeding greatly increases the survival of offspring and where the cost to males in terms of adult survival or lost opportunities of seeking additional mates are low. Under these conditions, the lack of a male response to reductions in paternity is viewed not as misdirected parental care but rather as the optimal strategy given the current cost/benefit trade-offs.

Such conditions may also apply to those Smith's longspur and dunnock males that were unassisted by other males and did not show any reduction in parental care in response to decreased paternity (this study; Davies and Hatchwell 1992; Davies et al. 1992). In other words, if a male reduces his parental care at a nest in which he has lost some paternity but there are no additional males to compensate, then it may pay him to continue investing at a high level despite some of this investment being wasted on raising non-kin. This situation-dependent behaviour would seem to be especially important in species in which males are unable to differentiate their own offspring from those of other males and so would place their young at risk if they unilaterally decreased parental care. Further work is needed, however, to determine whether similar constraints in other species can account for the apparent variety of responses males show to variation in paternity.

Although our observations suggest that male Smith's longspurs seem to know roughly how many offspring they had sired in a given nest, we did not find any evidence to show that they feed these young preferentially. Instead, each male fed every nestling within a brood about equally. This pattern has also been found in dunnocks (Burke et al. 1989), red-winged blackbirds (Westneat et al. 1995) and western bluebirds (*Sialia mexicana*; Leonard et al. 1995) and suggests that male (and perhaps female) birds are generally unable to recognise their own offspring. From the perspective of the male, this can clearly be maladaptive, as he will often feed nestlings that are not his own offspring. However, from the perspective of both the nestlings and the female, such a situation may be in their best interests (Davies et al. 1992). For example, it would not pay a nestling to advertise its paternity if this meant it would be ignored by some male attendants at the nest. Similarly, it would not be to the advantage of the female to have the paternity of her offspring revealed if it lead to the neglect of some nestlings. Thus, males may simply be unable to identify their own offspring and so feed the entire brood.

It is possible that males could avoid investing in non-kin by using some phenotypic trait or "green beard" to identify their own offspring, but such traits are thought to be easily open to cheating and unlikely to evolve (Hamilton 1964; Dawkins 1989). There is however, a non-phenotypic trait that male Smith's longspurs could use to identify their own offspring based on hatching sequence. As primary males are the first to copulate with a female, they sired all of the first-laid (and hatched) eggs, and most of the second-laid eggs. In contrast, secondary and tertiary males never sired any of the first-laid eggs, but almost always sired the third- and fourth-laid eggs. Although hatching asynchrony was slight in most broods, a size hierarchy based on hatching sequence (and therefore paternity) developed within a day or two of hatching (unpublished data). Thus, for a primary male, a good rule of thumb should be to feed only the oldest two nestlings in a brood as these are most likely to have been sired by him, while secondary (or tertiary) males should be expected to favour the smallest offspring of the brood as these are the only ones that they are likely to have sired. Nonetheless, our observations of food allocation within a brood show that this potential rule of thumb is not exploited by male longspurs, and therefore some paternal care is often invested in non-kin.

The majority of male Smith's longspurs helped to feed the nestlings they sired. As male parental care greatly increases the survival of offspring in a number of species (e.g. Bart and Tornes 1989), including Smith's longspurs (unpublished data), why did some males not help at all? In the dunnock, males only fed if they had mated with the female during the egg-laying period (and therefore when eggs were being fertilised; Burke et al. 1989; Davies et al. 1992). Our results suggest that a lack of male care in Smith's longspurs was not the result of a failure to gain access to a female or paternity. All males that did not feed nestlings sired from one to three young in those broods, and this did not differ significantly from the paternity success of males that did feed offspring. In the alpine accentor, males did not feed offspring if the period of nestling feeding coincided with the copulation period of other females in their polygynandrous group (Hartley et al. 1995). Instead, males preferred to mate-guard and copulate with a fertile female over feeding young in a previously hatched brood (Davies et al. 1995). It is unlikely that a similar process happens in Smith's longspurs as the short subarctic summer limits clutch initiation (including renests) to a 7–13 day period in mid-June (Briskie 1993). This means that clutch initiation (and therefore the availability of fertile females) never coincides with the period in which nestlings are

available for feeding (early to mid-July). Thus, male longspurs do not forego the feeding of nestlings to pursue additional mating opportunities. At present, we have no explanation for why some male Smith's longspurs do not help with parental care.

If male longspurs are unable to obtain additional matings during the nestling stage, then what benefits do they receive by reducing their investment in a brood in which they have few genetic offspring? On the one hand, it would seem worthwhile for males to continue feeding such broods at the maximal rate so that their own chicks grow fast and fledge in good condition. This may be particularly important in species such as Smith's longspurs, where males cannot distinguish their own offspring and so may place the entire brood at risk by reducing parental care. However, feeding offspring entails more costs than simply lost opportunities for acquiring additional mates. For example, searching for extra prey may expose males to greater risks of predation or increase the energetic costs of foraging at a time when both sexes are also preparing for the autumn migration. Indeed, most males examined during the nestling stage were already moulting their primaries and rectrices (unpublished data), suggesting that they are under severe time constraints at this time of year. Presumably, by reducing feeding effort at nests where they have low paternity, males can invest this time and energy into moult and premigratory fattening. It is possible that males in particularly poor condition may even stop parental care altogether and that this might account for the lack of feeding by some individuals (see above). On the other hand, when a male is the sole male providing food at a nest, we found that feeding rate did not vary significantly with paternity. As reduced paternal care at such nests would likely place some or all of the brood at risk, solitary males may be unable to trade off decreased paternal feeding against increased investment in moult or fattening. As with most studies of paternity and paternal care, experimental studies will be required to isolate the effects of male quality on the relationship between paternity and male investment (Westneat and Sargent 1996; Kempenaers and Sheldon 1997). Nonetheless, our results suggest that in at least some species such as Smith's longspurs, some males do compensate for lost paternity by reducing investment in offspring and thereby minimise the cost of cuckoldry.

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