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ORIGINAL ARTICLE

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Frequency and timing of extrapair fertilisation in the polyandrous red phalarope (*Phalaropus fulicarius*)

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Abstract In sequentially polyandrous bird species, where females mate with different males in succession during a single breeding season, sperm stored by females can occasionally lead to extrapair fertilisations (EPFs) in clutches cared for by the second and subsequent males. Thus, we predicted that in red phalaropes (*Phalaropus fulicarius*) – a sex-role-reversed, sequentially polyandrous, arctic breeding shorebird – EPFs would occur more frequently in clutches laid later in the breeding season. We used multilocus DNA profiling to examine the frequency and timing of EPFs in a population of red phalaropes breeding in the Canadian high arctic. Using a technique to determine parentage without maternal DNA, we inferred that 6 of 70 chicks in 18 broods resulted from EPFs – one extrapair chick in each of 6 broods. These results were supported by a further analysis using microsatellite DNA. As predicted, broods containing EPFs hatched from clutches laid significantly later in the season than did broods containing no EPFs. The difference in median hatch dates between broods with and without EPFs was 9.0 days, or 38% of the entire egg-laying period in that season. For the whole breeding season, we estimated that 6.5% of chicks were sired by extrapair males, which is similar to extrapair paternity estimates for other sex-role-reversed birds, but relatively low compared to the majority of socially monogamous species studied so far.

Key words *Phalaropus* · Polyandry · Sex role reversal · DNA profiling · Sperm storage · Paternity

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Introduction

Males in socially polyandrous bird species might be expected to have fewer opportunities to obtain extrapair paternity (EPP) because, unlike males in most socially monogamous species, they perform all of the incubation and parental care duties. Nonetheless, recent studies have suggested that males in polyandrous matings might gain some EPP via more cryptic means than the usual pursuit of extrapair copulations. For example, in an isolated population of red-necked phalaropes (*Phalaropus lobatus*) breeding in Iceland, males appeared to avoid pairing with females who had already laid a clutch (Whitfield 1990). Whitfield (1990) argued that males behaved this way to protect their paternity; females which had previously laid clutches could potentially store sperm from their previous mates and use it to fertilise some of the eggs laid while consorting with their second mates. Since frequent copulation is the only known way for a male to minimise the likelihood of cuckoldry through stored sperm (Birkhead et al. 1987; Møller and Birkhead 1989), Whitfield's hypothesis was supported by the observation that males consorting (albeit briefly) with females who had already laid a clutch that season tended to copulate at twice the rate of males paired with females working on their first clutches.

Oring et al. (1992) further investigated sperm storage in polyandrous shorebirds by determining the parentage of spotted sandpiper (*Actitis macularia*) clutches using DNA profiling. Unlike phalaropes, spotted sandpipers have a polyandrous mating system based on resource defence: females defend multipurpose territories and attract up to four males with whom they lay separate clutches sequentially. Like phalaropes, however, male spotted sandpipers usually perform all of the parental care duties, and can often be mated to a female who has already laid a clutch for a different mate during the same breeding season. Indeed, Oring et al. (1992) found that extrapair fertilisations (EPFs) were significantly more frequent in second and subsequent pairings in spotted sandpipers,

and that for 10 of 11 EPFs detected, the cuckolding male appeared to be an earlier mate of the female that season. Behavioural data suggested that the primary source of the EPFs in these clutches was stored sperm rather than surreptitious extrapair copulations (Oring et al. 1992).

More recently, however, Delahanty et al. (1998) found no evidence of extrapair fertilisations in 17 clutches of the sex-role-reversed Wilson's phalarope (*P. tricolor*). In this species, occasional EPFs from stored sperm might be expected because females are sometimes serially polyandrous (Colwell and Jehl 1994), though it is unclear whether any of the clutches studied by Delahanty et al. (1998) were from polyandrous females.

In the present study of EPFs in the red phalarope (*P. fulicarius*), our objectives were: (1) to determine the rate of EPFs in a sex-role-reversed species using DNA profiling and (2) to test the hypothesis that males nesting later in the breeding season were more likely to be cuckolded than earlier-nesting males. Red phalaropes, like the other two species in this genus, have a sex-role-reversed mating system, described by Emlen and Oring (1977) as female access polyandry. In this rare social system (Oring 1982), females compete directly for access to males, and males perform all of the incubation and parental care duties. Females typically desert their mates shortly after clutch completion and seek out new males with whom they attempt to mate and lay additional clutches (Schamel and Tracy 1977; Cramp and Simmons 1983). Although the mating system is characterised as polyandrous, the degree to which female red phalaropes have more than one mate in a breeding season has not been well studied. Many females appear to acquire only one mate per season (Cramp and Simmons 1983), but in at least two studies, marked females have been observed to be serially polyandrous: 4 of 11 females in Alaska (Schamel and Tracy 1977), and 3 of 6 females in Iceland (Whitfield 1995).

Based on the findings of Oring et al. (1992), we predicted that any EPFs occurring in our red phalarope population would be more likely to occur toward the end of the breeding season (see also Valle 1994). Red phalaropes have a short egg-laying season (e.g. 25 days in this study), so females mating polyandrously will tend to be over-represented among birds nesting toward the end of the season (Schamel and Tracy 1977; Whitfield 1995). If these females store sperm from previous mates (or from copulations carried out in failed attempts to acquire mates; e.g. Whitfield 1990), then later-nesting males should be cuckolded more frequently. Similarly, females nesting near the beginning of the season will undoubtedly be laying their first clutches and so EPFs due to stored sperm should be rare.

Methods

Field studies

We studied a breeding population of red phalaropes during the summer of 1993 on Igloolik Island, Northwest Territories, Canada (69°24′ N, 81°49′ W). Igloolik Island has typical high-arctic habitats comprising 103 km^2 of raised beaches, vegetated slopes and marshy sedge meadows (Forbes et al. 1992). Summers at Igloolik are short and cool, with various species of arctic-nesting birds arriving to breed in early June and typically leaving the island by mid-August (Forbes et al. 1992; J. Dale and R. Montgomerie, unpublished data).

Red phalaropes begin nesting a few days after they arrive on the breeding grounds (Schamel and Tracy 1987). In 1993, we studied birds breeding on approximately 5 km^2 of tundra, marshy meadows and ponds mainly within the study area delineated by Forbes et al. (1992). We saw the first phalaropes on 6 June and the first clutch was completed on 12 June ($n \pm 38$). In all, we found 43 nests and studied 35 of these in detail – the remainder were either preyed upon before clutch completion or we were unable to catch the attending male.

Since many nests were found after clutch completion, we were unable to record most egg-laying dates directly. Thus, we used hatch date (when the first chick of a clutch hatched) to estimate the date of clutch completion. In 6 nests where we knew the dates of clutch completion and where the eggs were incubated almost entirely (≥16 days) in an incubator (see below), the incubation period was either 18 or 19 days [mean incubation period $\pm$ 18.8 $\pm$ 0.41 (SD) days, $n \pm 6$]. This is similar to incubation periods observed in three other field studies: 20.1 $\pm$ 2.7, 19.7 $\pm$ 1.2 and 18.3 $\pm$ 0.6 days (Schamel and Tracy 1987). Thus we calculated the date of clutch completion as 19 days prior to hatch date.

Since female red phalaropes typically desert their mates soon after completing a clutch (Schamel and Tracy 1977; Ridley 1979; Cramp and Simmons 1983; Whitfield 1995; J. Dale and R. Montgomerie, unpublished data), we were only able to catch females during the short period immediately prior to or during egg-laying. Consequently, we chose not to catch females because of potential disturbance to natural patterns of mating, egg fertilisation and clutch completion. Thus, unlike most other studies using multilocus DNA profiling, we analysed paternity without reference to maternal DNA (see below).

We caught incubating males at their nests with cloverleaf nest-traps and collected 50–100 µl of blood from their brachial vein. Blood samples were stored in lysis buffer (Seutin et al. 1991) at 4 °C until DNA extraction, up to 8 months later. After banding and releasing the male, we collected the eggs (invariably four) in his nest and replaced them with dummy eggs made from self-hardening clay coloured with acrylic paints to resemble real eggs. The collected eggs were individually labelled and placed in an incubator (model RX2 ROLL-X; Lyon Electric Company) maintained at constant dry- (37.5 °C) and wet-bulb (29.4 °C) temperatures. Eggs were automatically turned every 45 min throughout incubation. Hatching eggs were isolated in separate incubator compartments. We banded chicks immediately after they hatched and extracted 30 µl of blood from their femoral vein. Chicks were then released at the nests of foster males still incubating artificial eggs. In all cases, males began brooding the chicks immediately.

DNA profiling

We used multilocus DNA profiling to evaluate paternity in a sample of the broods studied. Hatch dates for the 38 nests from which we could accurately estimate the date of clutch completion occurred over a 25-day period and had a slightly bimodal distribution, with a larger peak during the earlier half of this period (Fig. 1). To test the hypothesis that clutches laid later in the season were more likely to contain extrapair offspring, we selected for DNA profiling all 7 clutches from the latter half of the egg-laying period and a random sample of 11 clutches from the first half (Fig. 1). These clutches contained a total of 72 eggs but we were unable to extract DNA from 2 apparently infertile eggs in one clutch from the early nesting subsample.

To prepare DNA profiles, we used standard procedures that we have published in detail elsewhere (Smith et al. 1991; Hill et al. 1994; Weatherhead et al. 1994), initially digesting the DNA with the enzyme *Alu*I and probing the gels with Jeffreys 33.15 and 33.6

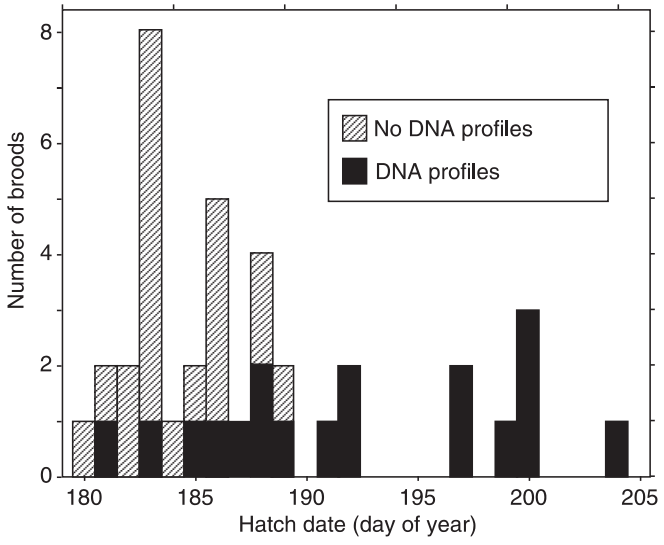

Fig. 1 Hatch dates for 38 red phalarope clutches found on Igloolik Island during 1993. A subsample (*dark bars*) of these broods that had a roughly even distribution of hatch dates was chosen for DNA profiling. 1 July is day 182

probes. DNA from one clutch (see Results) was also digested with *Hae*III and probed with the same probes.

Because we did not have DNA from mothers, our method of scoring DNA profiles for paternity analysis differs slightly from those used in most other studies using multilocus DNA profiling. Normally, unique bands are defined as those present in a chick but not present in either of its putative parents (e.g. Smith et al. 1991). In this study, however, we defined unique bands as those present in a chick's profile that were not present in the profiles of either its putative father or any of its siblings. From each chick's multilocus DNA profile, we used both bandsharing coefficients, or *D*-scores (Wetton et al. 1987), and the number of unique bands to identify possible extrapair chicks (i.e. arising from either EPFs or intraspecific brood parasitism, ISBP).

Bands on autoradiographs were initially scored using Gelreader (version 2.5) on a Macintosh microcomputer (see Hill et al. 1994 for methods) to determine *D*-scores. We then compared DNA profiles by hand (see Smith et al. 1991 for methods) to determine the number of unique bands. Bands revealed by each probe were assumed to be independent and were thus pooled for the determination of both *D*-scores and the number of unique bands (Burke and Bruford 1987; Westneat 1990).

To validate the results from multilocus profiles, we analysed microsatellite DNA of all individuals in our sample of 18 families (males and chicks). For microsatellite analysis we used the methods and the PAT MP2-43 microsatellite probe described in detail in Otter et al. (1998).

Parentage analysis

In our previous studies (e.g. Smith et al. 1991; Hill et al. 1994), we have found that extrapair chicks generally had *D*-scores (to putative father or mother) less than 0.30–0.40 and more than 10% of their bands were unique (i.e. absent from either putative parent). Up to 10% of a chick's bands are expected to be scored as unique due to mutation and sampling error alone (Hill et al. 1994), so a few unique bands are expected even when a chick is compared to its genetic parents.

In the present study, however, unique bands in a chick's profile could also have come from the mother, whose DNA we did not analyse. To minimise this problem, we scored the number of unique bands by comparing each chick to all of its siblings (as well as its putative father) in an attempt to identify the majority of maternal bands. Since each chick shares about 50% of its bands with its mother, and the majority of bands segregate independently (e.g. Burke and Bruford 1987; Burke et al. 1989; Birkhead et al. 1990), about 0.5^{n-1} of the mother's bands in any one chick from a brood of n chicks will be scored as unique, even though they came from the mother. Thus in a brood of four chicks from the same mother, about 13% of each chick's maternal bands (i.e. 6.5% of all its bands) would be scored as unique for this reason. We therefore initially identified chicks as extrapair if $>17\%$ (10% due to mutation and sampling error plus 7% due to maternal bands scored as unique) of their bands were unique and their chick-putative father *D*-score was <0.35.

Since extrapair chicks can be the result of either ISBP (when females lay their eggs into other females' nests) or EPF, it is important to distinguish between these two factors. If an extrapair chick arises from ISBP, most of its paternally and maternally inherited bands will appear as unique bands (Westneat 1990). Thus, ISBP chicks should have a higher percentage of unique bands than those resulting from EPFs. To determine the expected percent of unique bands in ISBP chicks, we simulated ISBP by comparing the DNA profile of a chick from one clutch to the DNA profiles of the male and three of four chicks in a different clutch on the same autoradiograph. We pooled the results from 16 different such ISBP simulations, using no chicks previously identified as extrapair. If any of the extrapair chicks in our sample was a result of ISBP, we expected it to have a percentage of unique bands similar to the observed percentages in the group of simulated ISBP chicks. If the chick resulted from an EPF, we expected it to have fewer unique bands than the simulated ISBP chicks.

To analyse paternity from the microsatellite data, we followed the procedure outlined in Otter et al. (1998). Based on Jamieson (1994), we determined, from the frequency distribution of alleles in fathers ($n=18$), that we could detect, on average, about 33% of extrapair chicks by analysing paternity using this single locus.

Results

Multilocus DNA profiles

Our analysis of multilocus DNA profiles revealed 13–29 [mean = 20.7 ± 3.1 (SD), $n = 88$] bands/individual for the Jeffreys 33.15 probe and 14–32 (mean = 21.5 ± 3.8, $n = 88$) bands/individual for the Jeffreys 33.6 probe. For both probes combined, there were 29–55 (mean = 42.2 ± 5.7, $n = 88$) scorable bands on the DNA profiles of an individual chick or male.

Of the 70 chicks from 18 broods examined, 6 (1 in each of 6 different broods) appeared to be extrapair – each had chick-putative father *D*-scores <0.35 as well as large percentages ($\geq 20\%$) of unique bands in their DNA profiles (Fig. 2). We were unable to identify the real fathers of any of these extrapair chicks from their DNA profiles

We used two statistical analyses to further evaluate the identification of extrapair chicks. First, we calculated the 99% confidence limit (CL) for the *D*-scores of chicks that had only zero or one unique band ($n = 23$) under the assumption that these were clearly not extrapair (Westneat 1993; Hasselquist et al. 1995). These chicks had a mean chick-father *D*-score of 0.486 ± 0.06 (SD), giving a lower 99% CL of 0.333 (Fig. 2). Second, we compared the *D*-scores of the 6 extrapair chicks with the

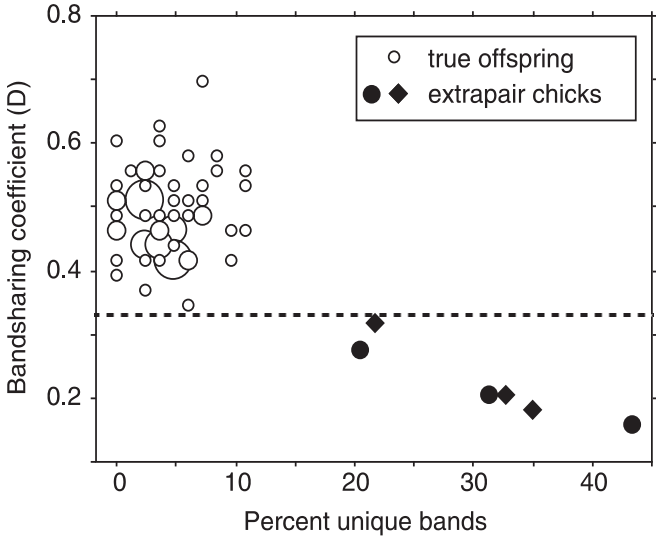

Fig. 2 Bandsharing coefficients and percent unique bands from DNA profiling analysis of 70 chicks and their putative fathers. The lower 99% confidence limit for bandsharing coefficients for 23 chicks that were identified as the true offspring of their putative fathers and had only zero or one unique band (see Results) is shown by the *horizontal dashed line*. Extrapair chicks were all initially identified using multilocus DNA profiling; the three extrapair chicks indicated by *diamonds* were confirmed with microsatellite profiling

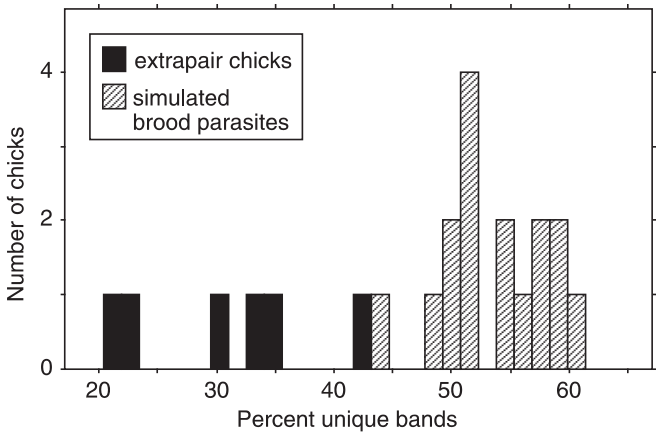

Fig. 3 Frequency distribution of the percentage of unique bands for 6 chicks identified as extrapair and 16 simulated brood parasites

D-scores of 24 pairs of presumably unrelated individuals. As expected, pairs of unrelated individuals had low bandsharing coefficients (mean = 0.186 ± 0.030, range: 0.133–0.244, n = 24 dyads), and this distribution was not significantly different from that of the *D*-scores of the identified extrapair chicks (mean = 0.218 ± 0.060, range: 0.165–0.318, n = 6 male-chick dyads; t-test, t = 1.28, P = 0.21). We conclude from these analyses that our identification of extrapair chicks was probably correct.

We double-checked the status of the extrapair chick with the highest *D*-score (0.318, Fig. 2) by analysing additional DNA profiles using the enzyme *Hae*III (see Methods). These new profiles revealed 33–45 bands/individual (n = 5), and the chick in question had D = 0.233 with respect to its putative father compared to D = 0.318 from its original profiles. Furthermore, the *D*-scores between each of its siblings and the putative father remained high (0.436, 0.482 and 0.554 vs 0.556, 0.474 and 0.520; *D*-scores from *Hae*III vs *Alu*I gels, respectively). Based on the evidence from these additional DNA profiles, we concluded that this chick was indeed an extrapair offspring.

The percentage of unique bands in any extrapair chick (mean = 30.6%, range = 20.4–42.6%, n = 6) was lower than that of any simulated brood parasite (mean = 53.7%, range = 43.6–60.5%, n = 16; Fig. 3). This suggests that the 6 extrapair chicks resulted from EPFs rather than ISBP. Moreover, all nests in our sample contained four eggs. When ISBP has been suspected in other populations of phalaropes, this suspicion has been based on the observation of clutches larger than four (Bent 1927; Congreve and Freme 1930; Lovenskiold 1964; Bengston as cited in Kistchinski 1975; Cramp and Simmons 1983; Colwell and Jehl 1994). In addition, egg size and eggshell patterns (characters commonly used to identify ISBP; e.g. Jackson 1992; Robertson et al. 1992; Lyon 1993; Thomas et al. 1989) were very consistent within clutches (based on precise size measurements and a standardised photograph of each clutch; J. Dale and R. Montgomerie, unpublished data). Thus, we conclude that 6 chicks were the result of EPFs, and that there were no cases of ISBP occurring in our sample of 18 broods.

Microsatellite DNA profiles

In our sample of 88 individuals (70 chicks plus 18 adult males), the microsatellite probe detected seven alleles (121, 123, 125, 127, 129, 131 and 159 bp) with respective frequencies of 0, 3, 20, 9, 2, 1 and 1 in 18 presumably unrelated fathers and 1, 21, 55, 28, 13, 1 and 1 in their 70 chicks. In all, 44% of male parents and 71% of chicks were heterozygous at this locus.

Using the microsatellite locus, 3 of the 70 chicks were identified as extrapair and all three had been identified as extrapair by multilocus fingerprinting (Fig. 2), including the extrapair chick with the highest *D*-score. The remaining 67 chicks all had at least one allele in common with their putative father and could thus not be excluded, including 3 of the chicks identified as extrapair with multilocus fingerprinting. Since we expected (see Methods) to detect only one-third of the extrapair chicks using this microsatellite locus, this analysis clearly supports our assessment of paternity using multilocus profiling.

Timing of EPFs

The 6 broods containing extrapair chicks hatched from clutches laid significantly later than the 12 broods in

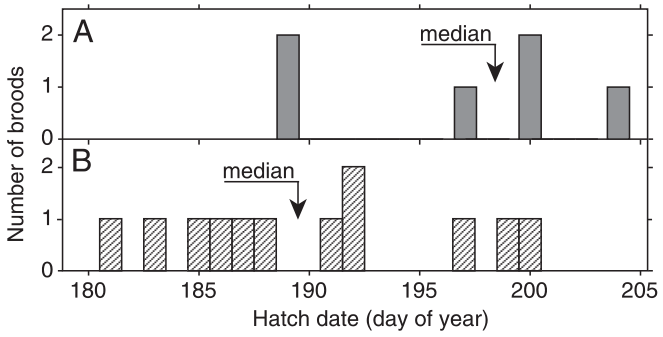

Fig. 4 Hatch dates for broods with ($n = 6$) (**A**) and without ($n = 12$) (**B**) extrapair chicks. *Arrows* indicate medians of each distribution

which all chicks were sired by the incubating male (Mann-Whitney test, $U = 56.5$, one-tailed $P = 0.027$, $n = 12,6$; Fig. 4). Since our hypothesis about the timing of EPFs makes the specific prediction that later-nesting males were more likely to be cuckolded (Oring et al. 1992; Valle 1994; this study), we used a one-tailed statistical test for this analysis. The median hatch date of clutches with extrapair chicks was 9.0 days later than clutches in which no EPFs were detected (Fig. 4). A delay in median hatch date of 9.0 days is 38% of the observed range of hatch dates that year ($n = 35$ clutches).

Frequency of EPFs in the population

The frequency of EPP in our study was 8.6% of chicks (6 EPFs in 70 chicks) and 33% of broods (EPFs in 6 of 18 broods). However, our subsample of broods used for DNA profiling was biased toward late clutches (Fig. 1) and later clutches contained EPFs significantly more frequently than early clutches (Fig. 4). Correcting for this sampling bias, we estimate the population level of EPP at 6.5% of chicks and 25.4% of clutches contained EPFs.

Discussion

The EPFs detected in this study could have occurred either through extrapair copulations that we did not see or through stored sperm from previous mates. To distinguish between these two factors, one would need paternity data from all clutches of polyandrous females plus information on their copulation behaviour (Oring et al. 1992). We do not have this information but we believe that the EPFs we detected were more likely the result of stored sperm. For example, in continuous focal observations of birds identified as paired (Cramp and Simmons 1983), we saw no extrapair copulations during either the 1993 (17 copulations observed) or 1994 (41 copulations) field seasons. Furthermore, paired male and female red phalaropes almost always stayed within 10 m of each other (J. Dale and R. Montgomerie, unpublished data), so it would be difficult for females to actively seek copulations from extrapair males. That extrapair copulations are generally rare in phalaropes is further suggested by studies on the other two species in the genus: in Wilson's phalaropes, Colwell and Jehl (1994) found no evidence for extrapair copulations and in red-necked phalaropes, Reynolds (1987) observed only two extrapair copulations in approximately 80 copulations. Thus, to explain the preponderance of EPFs in red phalaropes during the latter part of the 1993 breeding season (Fig. 4), we argue that females who fertilised their eggs with extrapair males did so via stored sperm either from previous mates or from males that made earlier, unsuccessful pairing attempts (e.g. Whitfield 1990). When female birds mate sequentially with different males within a breeding season, EPFs from stored sperm may occur regularly (Valle 1994). Oring et al. (1992) have shown that female spotted sandpipers fertilise eggs from sperm stored from previous mates, Owens et al. (1995) have argued the same phenomenon might obtain in the sex-role-reversed dotterel (*Charadrius morinellus*), and our results suggest that cuckoldry through sperm storage occurs occasionally in red phalaropes. In such species, any influence of natural selection on nesting dates will be augmented by sexual selection favouring those males who breed as early as possible so as to protect their own paternity and to gain EPFs in their mate's subsequent broods. Male red-necked phalaropes, for example, discriminate against females who have previously mated, apparently to prevent cuckoldry through sperm storage (Whitfield 1990).

Oring et al. (1992) argued that female spotted sandpipers might use sperm storage as a means of passively gaining fertilisations from high-quality mates – an alternative to actively seeking extrapair copulations from superior males (e.g. Kempenaers et al. 1992). Are early nesting red phalarope males superior to late nesters? Early nesting males may indeed be of higher quality because they have been able to migrate early to the far north, have withstood the harsh conditions of the high-arctic spring, and have recovered from the long spring migration sooner than later-arriving males. In the closely related red-necked phalarope, older birds arrived earlier on their breeding grounds than did 1-year-olds (Hildén and Vuolanto 1972), and this is probably true of red phalaropes as well (Cramp and Simmons 1983). We also found that early nesting males tended to have larger bill and tarsus lengths and brighter bill coloration than late-nesting males (R. Montgomerie and J. Dale, unpublished data). These patterns suggest that first mates of polyandrous females will often be older and healthier than second mates. Age and health could reflect a genetic component of male quality if, for instance, older and healthier males are more likely to have better genes for defence against current pathogens. Polyandrous females which fertilise some of the eggs in their second clutch with the sperm of their first mate could accrue

offspring that are genetically more suitable for future survival and reproduction (see Hasselquist et al. 1996) than those sired by their second mate.

Our estimate of 6.5% EPP in the population of red phalaropes on Igloolik Island is similar to EPP rates of other sex–role-reversed bird species. Delahanty et al. (1998) reported 0% EPP in Wilson's phalaropes, Owens et al. (1995) estimated 4.6% EPP in dotterels, Emlen et al. (1998) estimated 7.5% EPP in wattled jacana (*Jacana jacana*) and Oring et al. (1992) observed 8.6% EPP in spotted sandpipers. All of these EPP rates are quite low, however, in comparison to those observed in most socially monogamous species of birds, where EPP averages 15–20% across species and is sometimes as high as 30% [e.g. indigo buntings (*Passerina cyanea*; Westneat 1990) and tree swallows (*Tachycineta bicolor*; Dunn et al. 1994)] or even $>50\%$ [e.g. American robins (*Turdus migratorius*; R. Montgomerie, unpublished data) and reed buntings (*Emberiza schoeniclus*; Dixon et al. 1994)].

Why is EPP in red phalaropes so low? In theory, female phalaropes should have some control of the levels of EPP in their clutches (Birkhead and Møller 1993) but males probably have little direct control over EPP levels in the broods they care for. In species with biparental care, males could potentially respond to EPP by decreasing the amount of paternal care they invest in a brood (Møller and Birkhead 1993; Dixon et al. 1994). However, in polyandrous species, where males provide all the care, this option is available only by deserting the entire brood. If the male deserts his clutch early, he has an excellent chance of renesting, at least in red phalaropes, because the operational sex ratio usually becomes strongly female biased by the middle of the egg-laying season, when the number of females seeking second mates increases dramatically (Schamel and Tracy 1977; J. Dale and R. Montgomerie, unpublished data). Deserted early nesting females, on the other hand, have more difficulty renesting because of the heightened intrasexual competition for a dwindling number of males not already tending clutches that occurs later in the season. This sexual asymmetry in the costs of desertion provides the male with some indirect control over the level of EPP. Provided that males have some means of assessing their risk of being cuckolded, the threat of clutch desertion could constrain the females to sacrifice the possible benefits of mixed paternity in their broods and thus keep levels of EPP low (Delahanty et al. 1998; Emlen et al. 1998). In high-arctic breeding birds such as red phalaropes, deserting males do suffer increased costs toward the end of the breeding season because the rapidly approaching winter limits future nesting opportunities. Later-nesting males should therefore be more reluctant to desert their clutches even when they are uncertain of their paternity. This may explain why later-nesting red phalaropes have significantly more extrapair chicks in their broods.

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