Ecological determinants of extrapair fertilizations and egg dumping in Alpine water pipits (*Anthus spinoletta*)

Heinz-Ulrich Reyer, Kurt Bollmann, Alex R. Schläpfer, Angela Schymainda, and Gabriela Klecack
Zoologisches Institut, Universität Zürich, Wintherthurerstrasse 190, CH-8057 Zürich, Switzerland

Behavioral ecology has successfully explained the diversity in social mating systems through differences in environmental conditions, but diversity in genetic mating systems is poorly understood. The difference is important in situations where parents care for extrapair young (EPY) originating from extrapair paternity (EPP), extrapair maternity (EPM), and intraspecific brood parasitism (IBP). In birds, IBP and EPM are rare, but EPP is widespread and highly variable among species and populations. Explanations for this variability are controversial, mainly because detailed ecological information is usually lacking in paternity studies. Here we present results of the first study to identify the ecological determinants of extrapair activities for both sexes of the same species, the water pipit (*Anthus spinoletta*). DNA fingerprints of 1052 young from 258 nests revealed EPP in 5.2% of the young from 12.4% of the nests. EPM and IBP, both involving egg dumping (EDP), each occurred in 0.5% of the young from 1.9% of the nests. Nests with and without EPY could not be distinguished by traits of the breeders and by reproductive success, but they differed with respect to ecology: nests with EPP young were characterized by asynchronous clutch initiation, nests with EPM and IBP young were characterized by higher overlap with neighboring territories and closer proximity to communal feeding sites. We suggest that chance events, resulting from the temporal and spatial distribution of broods, offer a better explanation for the occurrence of extrapair activities than female search for genetic or phenotypic benefits. This possibility of "accidental" extrapair reproduction as an "ecological epiphenomenon" with low potential for selection should also be considered for species other than the water pipit. *Key words: Anthus spinoletta*, breeding synchrony, DNA fingerprinting, extrapair maternity, extrapair paternity, intraspecific brood parasitism, mating system, operational sex ratio, territory overlap, water pipit. [*Behav Ecol 8:534–543 (1997)*]

Present knowledge about the evolution of mating systems and sex-specific parental care is largely based on comparing social associations between males and females under different ecological conditions (Davies, 1991; Emlen and Oring, 1977; Oring, 1982; Reyer, 1994b; Wittenberger, 1979). Social bonds, however, do not necessarily reflect mating combinations and parentage (Gowaty, 1985; Wickler and Seibt, 1983). For example, in many bird species parents care for extrapair young (EPY). These young can originate from extrapair paternity (EPP), extrapair maternity (EPM) and intraspecific brood parasitism (IBP). The former two patterns result from extrapair copulations (EPC) of males and females with individuals other than their social mates, and the latter two result from egg dumping (EDP)—i.e., females laying in foreign nests (Birkhead and Møller, 1992; Petrie and Møller, 1991).

While EPM and IBP are infrequent (reviewed by Andersson, 1984; MacWhirter, 1989; Reyer, 1994a; Yom-Tov, 1980; see also Hartley et al., 1993), EPP is widespread and highly variable among species and populations, affecting between 0% and 60% of nests and young (Birkhead and Møller, 1992). It is generally agreed that this variability reflects individual-, sex-, species-, and environment-specific differences in the trade-off between the benefits from pursuing extrapair activities and the costs from "neglecting" one's own mate, nest, or dependent young (reviews by Birkhead and Møller, 1992; Kempenaers and Dondt, 1993; Petrie and Møller, 1991; Westneat et al., 1990). But how phenotypic and ecological factors influence extrapair activities and which sex is in control is disputed

on the theoretical level and poorly investigated at the empirical level. Some authors suggest that females seek extrapair fertilizations (EPFs) with superior males to obtain genetic and/or phenotypic benefits such as good genes, increased genetic diversity, ensured fertilization, and courtship feeding or paternal care from additional males (e.g., Kempenaers et al., 1992; Weatherhead et al., 1996). Other studies, however, have failed to find clear-cut relationships between EPFs and phenotypic measures of male quality (e.g., Dunn et al., 1994; Westneat, 1990; see also Reyer, 1994a). In terms of ecological factors, decreasing breeding synchrony and/or value of parental care are assumed by some authors to increase (Westneat et al., 1990), and by others to decrease, EPP rates (Stutchbury and Morton, in press; Whittingham et al., 1992). Similarly, some expect EDP to be more frequent in colonial species (Hamilton and Orians, 1965), whereas others expect it to be more common in noncolonial species (MacWhirter, 1989; Yom-Tov, 1980).

Part of the controversy arises from the lack of detailed ecological information in paternity studies (Westneat, 1993). Such information is crucial for understanding the adaptive value and evolution of mating systems and parental care in general, because extrapair activities can markedly alter the fitness costs and benefits calculated from social mating systems. In this study on water pipits (*Anthus spinoletta*), we tried to identify the relative importance of various phenotypic and ecological factors for extrapair activities by relating the extent of EPP, EPM, and IBP to fitness-relevant traits of the breeders and their environment. Based on our results, we suggest that the extrapair mating activities in this species may reflect an "ecological epiphenomenon" of the temporal and spatial distribution of their broods, rather than a specifically selected "mixed reproductive strategy" (*sensu* Trivers, 1972).

MATERIALS AND METHODS

Study area and species

We studied a color-ringed population of water pipits in the Dischma Valley near Davos, Switzerland. The study area of 2.6 km² lies above timberline and extends from the valley floor (1850 m above sea level) into the northeast and southwest slopes up to 2500 m sea level. Vegetation on both slopes is mainly dwarf shrubs (*Rhododendron*, *Juniperus*, *Calluna*, *Vaccinum*), interspersed with grassy and herbaceous associations along brooklets. The valley floor and the upper parts of the northeast slope are dominated by meadows and alpine pastures. The birds feed on arthropods, which they collect both within their breeding territories and in communal feeding sites mainly located in meadows where territory density is lower than in shrubs (Brodmann et al., 1997; Frey-Roos et al., 1995). Breeding occurs between late May and early August in ground nests, built under tussocks and bushes, or in crevices (Rauter, 1995). Average clutch size was 4.5 eggs on both sides of the valley, but nestling survival differed, mainly due to differences in predation by adders (*Vipera berus*), which only occurred on the warmer southwest slope.

During the study period (1990–1992), the proportion of individually color-ringed adults averaged 68%, sex ratios ranged from 1.07 to 1.18 males per female, and territory density ranged from 3.44 to 7.78 males/10 ha, with higher densities on the northeast than on the southwest slope in any one year. The social mating system was predominantly monogamous, with a few cases of simultaneous polygyny and successive polyandry. Averaged over the 3 years, 84% of the territories were occupied by pairs, 11% by unmated males, and 5% by trios of one male and two females (Bollmann, 1995; Schläpfer AR, unpublished data).

Field methods

Regular observations on foraging, territorial (singing, displaying, fighting), and reproductive behavior (courtship, mate guarding, nest building, egg laying, incubation, and feeding) started when the first males had settled in the study area. Throughout the breeding season, we visited each section of the area once every 2–4 days, depending on the breeding stage. We monitored nests daily when hatching or fledging was expected. Locations of nests and of birds tending them (hereafter called putative or social parents) were recorded on a map (scale 1:2500), and territory size and shape was determined by the minimum concave polygon method (Clutton-Brock et al., 1982) using the program MiniCad+ (version 4; Diehl Grafsoft Inc., Elliot City, Maryland, USA). Prey biomass in territories and communal feeding sites was measured twice per season by sweep netting, based on a 50×50 m grid system put over a topographical map of the study area (1:5000). The first prey sampling period (3–21 June) covered the time of first clutches; the second period (14–30 July) represents the time of replacement and second clutches. Further details on study area, birds, behavioral observations, and measurement of ecological variables are given by Bollmann et al. (1997), Brodmann et al. (1997), Frey-Roos et al. (1995), and Rauter and Reyer (1997).

For banding, weighing, measuring and blood sampling, adult birds were caught in mist nets, and chicks were taken from the nest when 8–9 days old. After puncturing the bird's brachial vein with a sterile syringe needle (no. 20, 0.4×20 mm), 25–50 µl blood was collected in a heparinized capillary tube and immediately transferred to a 1.2-ml Nunc cryotube, where it was suspended in 1 ml buffer (0.15 M NaCl, 15 mM trisodium citrate, 10 mM EDTA; pH 7.0). At the end of each day we placed all tubes in a cool box where they were stored for some days or weeks until they could be taken to the laboratory and frozen at −20°C.

DNA fingerprinting

After thawing the blood samples at room temperature, we extracted the DNA from blood cells using the salt extraction method described by Signer (1988). DNA concentration was balanced for all lanes with the aid of a photospectrometer. Then 8 µg of DNA was digested overnight with 4 U of the restriction enzyme *Hinf*I at 37°C and electrophoresed through a 20×25 cm 0.8% agarose gel (type I, Low EEO; Sigma, St. Louis, Missouri, USA) in a 1x TBE buffer (0.089 M Tris, 0.089 M borate, 2 mM EDTA, 0.5 µg/ml ethidium bromide). To minimize effects of distortion in the gel, we always ran offspring adjacent to the lanes of their putative parents, and three to four marker lanes were evenly distributed over the gel. As markers we initially used DNA molecular weight markers II and III (Boehringer, Mannheim, Germany) but later switched to the DNA Analysis Marker System (Life Technologies), which gives a finer resolution.

After running at 35 V for about 70 h, while the electrophoresis tank was standing in a waterbath cooled to 6°C, the gel was put in 0.25 M HCl for 15 min to cut long bands, then soaked in 0.5 M NaOH, 1 M NaCl for 30 min, and finally neutralized in 1 M Tris-HCl, 3 M NaCl for another 30 min. Thereafter, the DNA was bound to nylon filters (Pall Biodyne 1.2 µm) by crosslinking and baking at 80°C for 3 h. Filters were then hybridized overnight at 42°C in a solution containing 50% deionized formamide, 10x Denhardt's solution, 0.1% SDS, 5% PEG 6000, 0.9 M NaCl, 0.05 M natrium phosphate pH 7.0, 0.0005 M EDTA, and either Jeffreys' 33.15 or 33.6 as a radioactively labeled probe (Random Primer DNA labeling kit, Boehringer). We washed filters twice for 10 min each at 64°C in 1x SSC and then exposed them to X-ray film for 1–3 days to produce autoradiographs. For rehybridizing with a second probe, filters were shaken a few min with 0.1% boiling SDS, allowed to cool to 40°C, and then rinsed in 2x SSC.

Scoring fingerprints

Initially, we scored fingerprints visually by overlaying autoradiographs with acetate sheets and marking all bands with a permanent marker, using different colors for maternal, paternal, joint, and novel bands (Bruford et al., 1992). Later, we analyzed fingerprints as described by Freeland et al. (1995) and Pöldmaa et al. (1995): autoradiographs were scanned into a computer, location and intensity of bands was analyzed with GelReader (version 2.05, National Center for Supercomputing Applications, Champaign, Illinois, USA) and checked for errors through visual comparison of original and scanned images. In both the visual and the computerized analysis, we scored in the range of 2.3–20 kb, which yielded a mean number (± SD) of 27.2 (± 6.2) bands for probe 33.15 and 21.6 (± 3.1) bands for probe 33.6. Bands of two individuals were considered identical when their intensities differed less than twofold and their centers were within 0.5 mm or 1.25% of the molecular weight (Bruford et al., 1992; Freeland et al., 1995). When both scoring techniques were applied to the same autoradiograph, results from parentage analyses (see below) agreed well, and so did results from Jeffreys' probes 33.15 and 33.6. Therefore, we used the computerized analysis, based on probe 33.15, as our standard method of determining parentage. When this yielded ambiguous results, we added in descending order scoring by eye, a new computer analysis based on probe 33.6, and running a new gel.
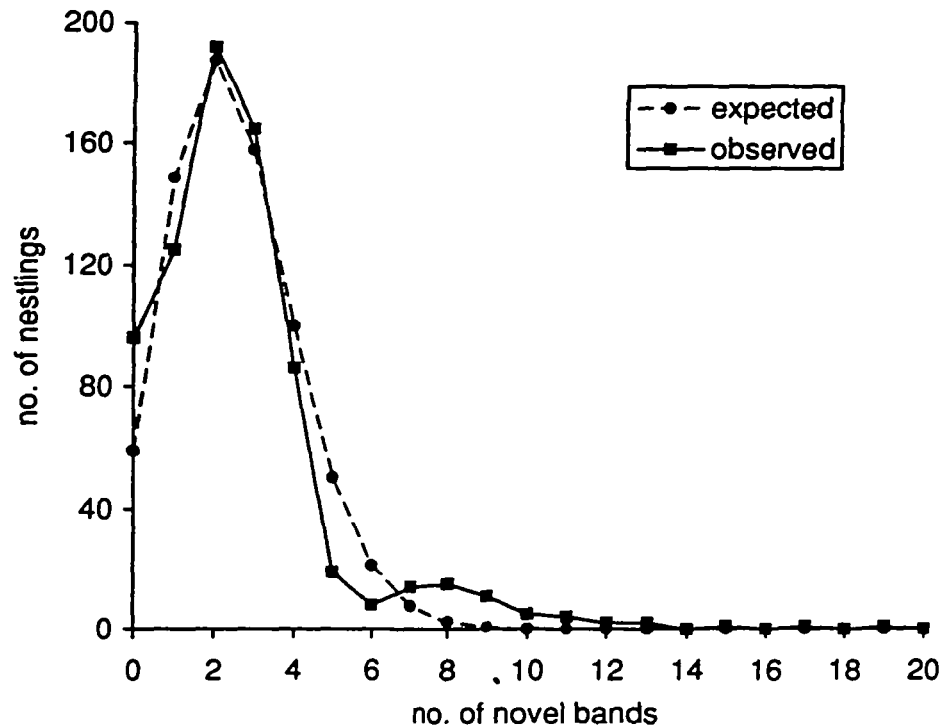
Figure 1
Frequency distribution of observed and expected numbers of novel bands in water pipit nestlings. The observed distribution is based on 734 nestlings from 179 families; the expected distribution is a Poisson distribution, calculated from the observed probabilities.

Parentage analysis

We analyzed parentage and relatedness through band-sharing coefficients (D) and, where fingerprints from both social parents were available, the number of novel bands (i.e., offspring bands unmatched by parental bands; see Westneat, 1990). Band sharing was calculated from the equation $D = [(N_{ab}/N_a) + (N_{ab}/N_b)]/2$, where N_a and N_b are the total number of bands for individuals a and b, respectively, and N_{ab} is the number of bands with similar intensity and similar electrophoretic mobility that individuals a and b have in common (Bruford et al., 1992). We considered nestlings to be EPY when their number of novel bands was > 5 and/or their band-sharing coefficient (D) with the putative parent was < 0.35. The threshold of five novel bands is higher than that of two to three bands used in most other studies because our computer analysis classified more bands as "novel" than visual screening did. However, our thresholds for distinguishing between pair and extrapair young were derived as in other studies (for details, see Hasselquist et al., 1995; Westneat, 1990, 1993): we compared the actual frequency distribution of novel bands with an expected Poisson distribution (Figure 1), calculated under the assumption that different numbers of novel bands arise from random causes such as mutation, scoring errors, or impure DNA. For one to four novel bands, observed and expected distributions did not differ ($x^2 = 6.04$, df = 3, $p > .10$), suggesting that random causes are a sufficient explanation. Beyond four novel bands, however, the two distributions diverge (e.g., for one to five bands: $x^2 = 25.50$, df = 4, $p < .001$) due to the bimodal nature of the observed distribution. This indicates that more novel bands arise from EPF or EDP. Because the two modes were not entirely distinct, we used the distribution of D as a second source of information about parentage. The combination of both distributions resulted in two distinct groups representing related and unrelated young, respectively (Figure 2).

RESULTS

Types and numbers of extrapair young

We performed paternity and maternity analysis for 1052 young from 258 nests. Based on our criteria for identifying EPY (> 5 novel bands and/or D< 0.35; Figure 2), we found 65 young (6.2%) in 42 nests that were unrelated to one or both parents (Figure 3). In 32 (= 12.4 %) of these nests, 55 young (range 1–4 per nest, median = 1) had been sired by a male other than the nest owner (EPP). In each of the other 10 nests (3.9%), 1 young originated from an egg laid by another female. In five of these cases the dumped egg had been fertilized by the male attending the nest (EPM); in the other five cases the young was unrelated to both nest owners (IBP). Mean band-sharing coefficients (± SD) between EPY and their foster parents were 0.219 (± 0.073; range 0.044–0.316) for males and 0.220 (± 0.045; range 0.158–0.295) for females. These values differ neither between them nor from background levels of band-sharing among presumed unrelated adults (0.236 ± 0.070; range 0.106–0.351; all $p > .20$, t test). Band-sharing coefficients between nest-attending adults and the remaining (i.e., their own) young were 0.495 (± 0.084; range 0.336–0.681) and 0.504 (+ 0.087; range 0.335–0.778) for males and females, respectively. These values also did not differ significantly ($p > .10$)

Tests for potential artifacts

Linkage
Calculated band-sharing coefficients will be inflated if bands do not assort independently (Amos et al., 1992). To test for independence, we conducted a segregation analysis on one water pipit family consisting of both parents and their 10 full-sib nestlings from two successive broods of the same year. All 22 maternal and 23 paternal bands revealed by the 33.15 probe were absent in at least one offspring, and thus apparently heterozygous. With the exception of one maternal allelic
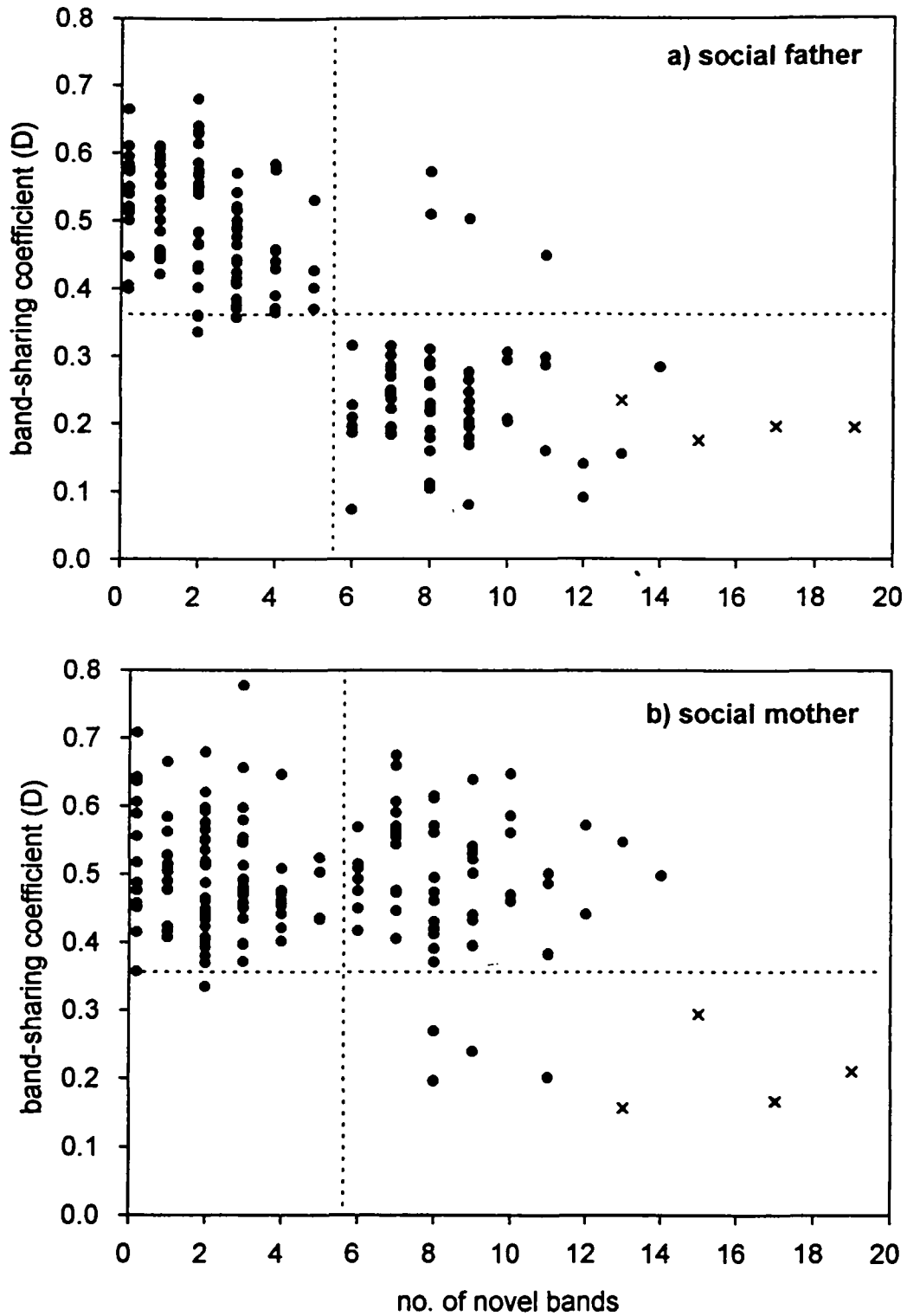
Figure 2
Relationship between number of novel bands in nestlings and the corresponding band-sharing coefficients (D) with (a) the social father and (b) the social mother. The dashed lines indicate the criteria for excluding parentage (see Methods). For clarity, only nestlings from families with at least one extrapair young have been plotted. Dots to the left of and above the dashed lines are from genetic young of both social parents; dots to the right of the vertical dashed lines indicate extrapair paternity when below the horizontal line in panel a and above the horizontal line in panel b; they indicate extrapair maternity (EPM) when below the line in panel b and above the line in panel a. In both panels, the crosses to the right of and below the lines refer to intraspecific brood parasitism (IBP), with neither male nor female related to the young. Of the five EPM and five IBP cases found in this study, only four each could be plotted because in the other two cases the social father was not fingerprinted, and thus the number of novel bands not known.
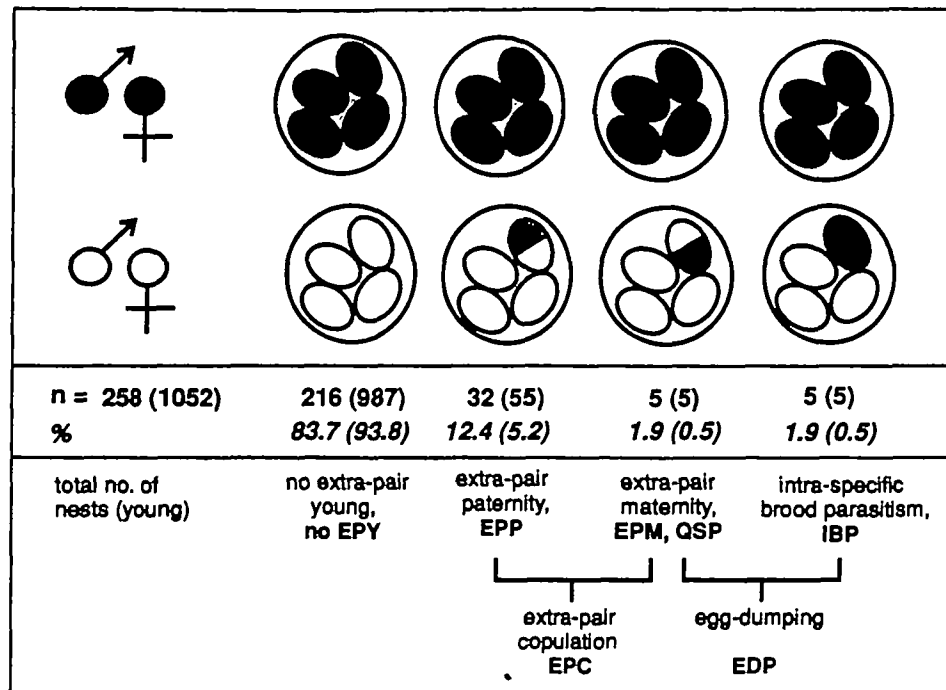
Figure 3
Types and frequencies of extra-pair activities in water pipits. Egg shadings indicate the genetic parents.

and one paternal linked band pair, all bands showed independent inheritance patterns. This, plus the fact that the numbers of bands scored per individual are high, ranges of D for related and unrelated offspring are clearly separated (see above), and paternity analyses from 33.15 and 33.6 probes give identical results, all indicate that the probability of false paternity conclusions is negligible (see Amos et al., 1992).

Pseudoreplication
Because 54 adults occurred more than once in the set of 258 nests, we tested for effects of pseudoreplication by comparing the incidence of EPY in individuals across subsequent broods. In no case did individuals of either sex show a consistent tendency to have EPY in successive broods within or between years (all $p \geq .245$, Fisher's Exact tests). We therefore used nests as independent units.

Incomplete fingerprints
For 79 of the 258 nests, fingerprints were not available for one of the social parents, usually because the birds could not be caught. In these cases, relatedness between nestlings and the fingerprinted adult had to be determined by band-sharing coefficients alone. To test whether this affected paternity and maternity exclusion, we compared the proportion of nests with and without EPY between the 79 pairs where only band sharing could be used and the 179 pairs where the number of novel bands was available. The result was far from being significant ($\chi^2 = 0.055$, df = 1, $p = .815$).

Observer interference
Disturbance of birds around the time of copulation and egg laying may result in reduced territorial defense, mate guarding, or nest attendence and thus increase the chances for EPCs and EDP (Yezerinac et al., 1995). We therefore tried not to catch breeders during this critical period, although this could not always be avoided (Figure 4). To test for potential effects of interference, we compared the proportion of nests with and without EPY between parents which were caught,

measured, marked, and blood sampled between days −10 and +5 around clutch initiation and parents caught outside this period or not at all in the respective year. Again, there was no significant effect ($\chi^2 = 0.000$, df = 1, $p = 1.000$). In the above tests for potential artifacts, population effect sizes (i.e., differences between actual and hypothetical distributions) were very small. Consequently, the power of rejecting a false null hypothesis with $p < .05$ was < 0.07 in all cases (power analyses; calculated according to Cohen, 1988).

Biological determinants

Factors considered
We believe that the above tests ensure that the recorded incidences of EPY represent biological facts rather than methodological artifacts. Our search for the most important biological determinants of extrapair activities (see Introduction) was based on three groups of variables that have been shown in other studies to be correlated with fitness: quality traits of (1) the breeders, (2) their environment, and (3) measures of reproductive success.

(1) Among the breeder traits, we considered five variables: (a) feather length of the eighth primary as a measure for body size (Jenni and Winkler, 1989), (b) body condition expressed by weight/(tarsus length)^3, (c) age (1, 2, and $\geq$ 3 years), (d) experience (none, one, or both breeders experienced), (e) social mating status (monogamous versus polygynous groups), and (f) survival to the next year (no, yes).

(2) Among the potential ecological determinants, we included six variables that affect competition for mates, predation pressure, and availability of food. In terms of competition, we considered (a) territory density per hectare, (b) percentage of territory overlap with all neigbors, and (c) laying synchrony, measured by the total number of nests in the study area where egg laying began between day −2 and day + 2 around clutch initiation in the focal nest (Westneat, 1992). In terms of predation pressure, we compared (d) two valley sides, one (northeast) with no adders, the other (southwest) with high adder densities. Food availability was expressed by (e)
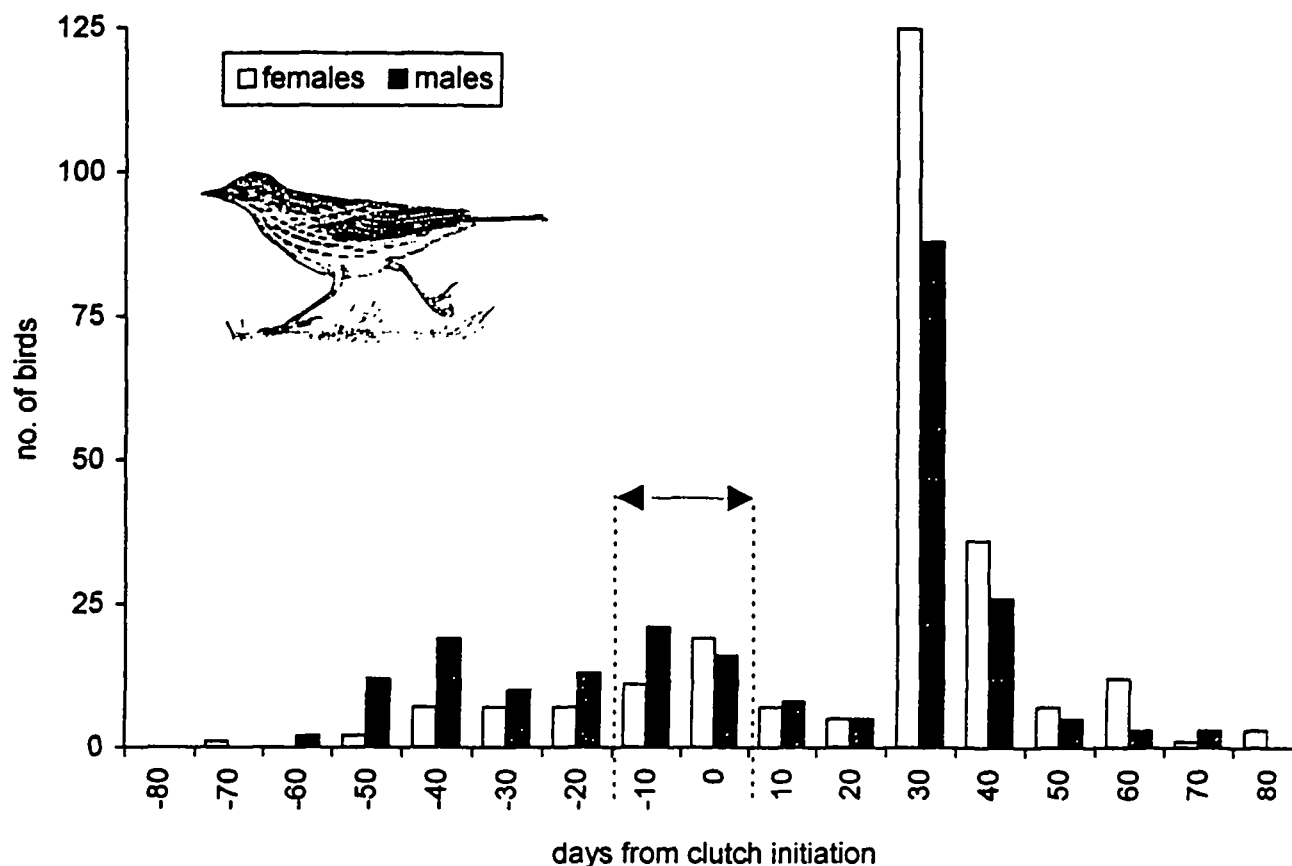
Figure 4
Number of females (open bars) and males (hatched bars) caught from 80 days before to 80 days after clutch initiation (= day 0). The critical period where disturbance through catching, measuring, banding, and blood sampling can potentially affect the rate of extrapair copulations and egg dumping (days −10 to +5) is marked by the arrow between the dotted lines. Pipit drawing by F. Weick from Glutz von Blotzheim and Bauer (1985), with permission.

prey index of a territory, measured in milligrams dry weight per 10 net sweeps (Brodmann et al., 1997), and (f) minimum distance of the nest to the nearest communal feeding site outside the territory.

(3) Reproductive success was measured by (a) initial clutch size, (b) the proportion of nestlings hatching, and (c) the proportion of nestlings fledging. Survival of young to the next year, a better measure for fitness, could not be considered statistically because only 7.6% of the males and 3.0% of the females returned to their natal area (Bollmann, 1995, Schläpfer AR, unpublished data).

Identification of relevant factors
In a first set of analyses, we related the numbers of nests with and without EPY to categorical variables (variables 1c–f; 2a,d). In terms of breeder traits, we found no significant differences with age, experience, and mating status (all $p \geq .589$; Fisher's Exact or chi-square tests, all power values < 0.09); for survival there was even a tendency for cuckolded males to survive better than noncuckolded ones ($\chi^2 = 2.780$, df = 1, $p = .095$; power = 0.40). In terms of ecological conditions, proportions of nests with EPP and EDP did not differ between predator rich and predator poor valley sides ($\chi^2 = 1.493$, df = 2, $p = .493$; power = 0.02) nor did they vary with territory density ($r_s = ^-0.593$, $n = 6$, ns; Spearman rank correlation calculated from 3 years with separate data from the northeast and southwest side of the valley).

In a second step, we performed discriminant analyses to test whether nests with and without EPY differed in the continuous variables (variables 1a,b, 2b,c,e,f, 3a–c). For these analyses, we pooled data from EPM and IBP nests into one category of EDP nests and performed three separate discriminant analyses: the first analysis included variables representing quality measures of male and female breeders (feather length and condition index), the second analysis tested for the effects of ecological factors in and around their territories (synchrony, territory overlap, distance to communal feeding grounds, and prey index), and the third analysis considered reproductive success (clutch size, percent hatching and percent fledging). Splitting EPM and IBP and including all continuous variables in a single discriminant analysis would have resulted in too many variables for the available sample sizes.

Nests with and without EPY differed neither in breeder quality nor in reproductive success. This was true for both the multivariate tests (Wilk's lambda for analyses 1 and 3: $p = .314$ and $p = .837$, respectively) and the univariate F tests for individual variables (all $p \geq .176$). Nests with and without EPY could be separated, however, by ecological conditions (Wilk's lambda for analysis 2: $p = .006$). EPP nests were surrounded by fewer nests with synchronous laying than nests without EPP (Figure 5a), but they did not differ with respect to territory overlap and distance from communal feeding sites (Figure 5b,c). Conversely, territories with EPM/IBP nests showed more overlap with neigboring territories and were closer to
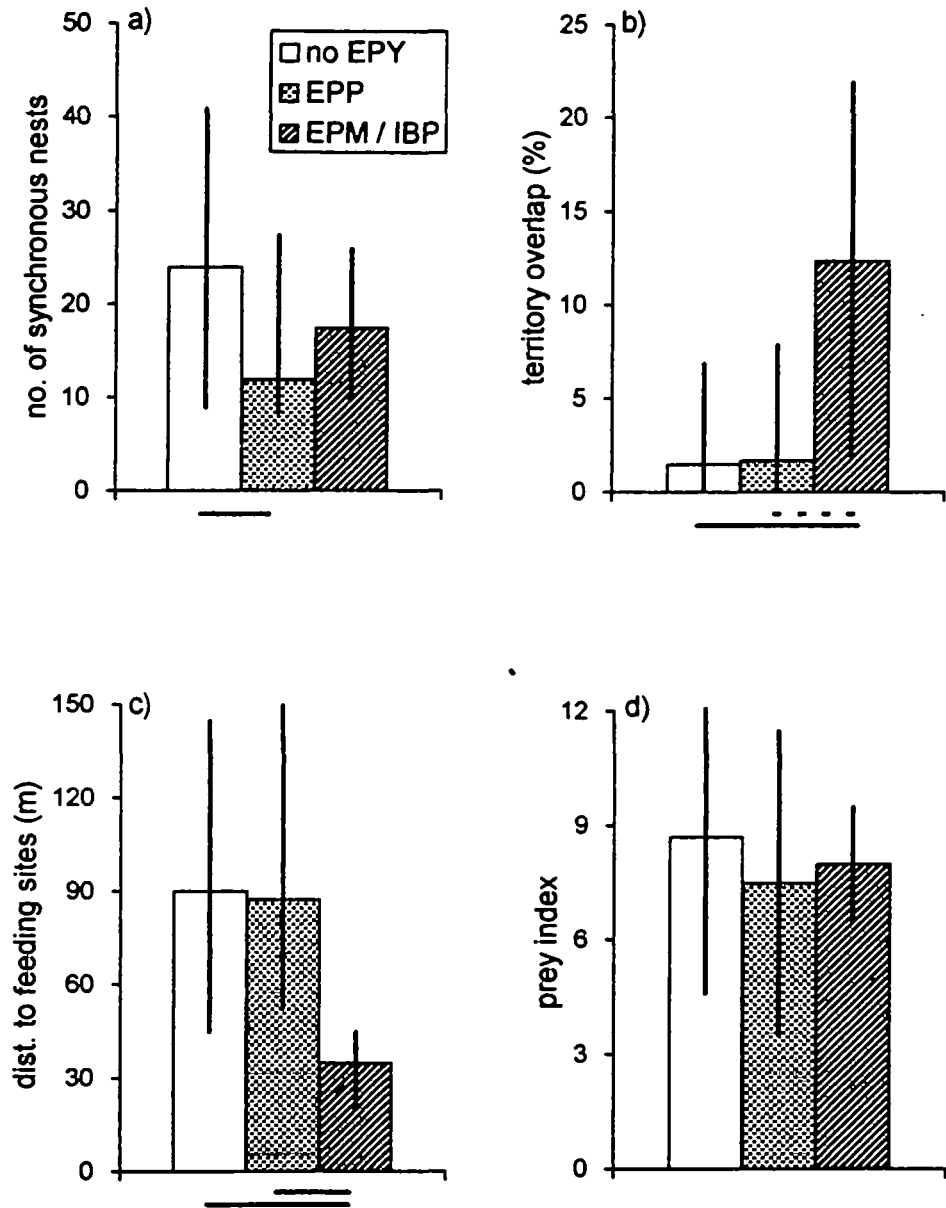
Figure 5
Extrapair activities in relation to ecological conditions. Differences between nests without any extrapair young (EPY; open bars) and nests containing either extrapair paternal (EPP; stipled bars) or extrapair maternal (EPM/IBP; hatched bars) young are shown with respect to (a) temporal and (b, c) spatial location of the brood and to (d) food quality of the territory. Bars and vertical lines represent means and SDs in panel d, but medians and interquartile ranges in panels a–c because these three variables were not normally distributed. Consequently, they were log-transformed before entering into the discriminant analysis. Results from Scheffe's pairwise comparisons in univariate ANOVAs are indicated below the bars: broken line, $p < .10$; solid line, $p < .05$.

communal feeding grounds (Figure 5b,c) but did not differ from unaffected nests with respect to synchrony (Figure 5a). Thus, EPP was influenced by the location of the brood in time (synchrony), and EDP was influenced by its location in space (territory overlap and distance to communal feeding sites). Territory quality in terms of prey density did not affect the occurrence of EPY (Figure 5d).

The temporal component of EPP is further illustrated in Figure 6. While synchrony decreases from breeding period 1 to 4, the proportion of nests with EPP increases and is higher for second than for first clutches ($p = .029$; Fisher's Exact test, one tailed). In only one case did the female change mates between her first and second brood, making sperm storage an alternative explanation to EPP. The spatial aspect of EDP is further supported by our finding that among the 10 EDP nests, 80% were directly bordering the communal feeding places, whereas among the other 248 nests, only 44% were bordering communal feeding places ($p = .024$; Fisher's Exact test, one tailed).

DISCUSSION

Egg dumping

With 1.9% of the nests and 0.5% of the young affected, proportions of EPM and IBP in water pipits seem to exceed values for most other territorial bird species. Although application of molecular techniques is revealing more species in which EPM and IBP are found (Barber et al., 1996; Birkhead et al., 1990; Gowaty and Karlin, 1984; Otter, 1996), rates are typically low and do not change the general conclusion that EDP is much rarer than EPP. The overall rareness of EDP in birds has been explained through selection on both parents to reject unrelated eggs, rather than selection on fathers alone in the case of EPP (Petrie and Møller, 1991). This explanation, however, is unlikely to hold for two reasons. First, evidence for the ejection of intraspecific parasitic eggs is weak (Andersson, 1984; Petrie and Møller, 1991). Second, counterselection on both parents is not true for EPM, and yet EPM is even rarer than IBP.
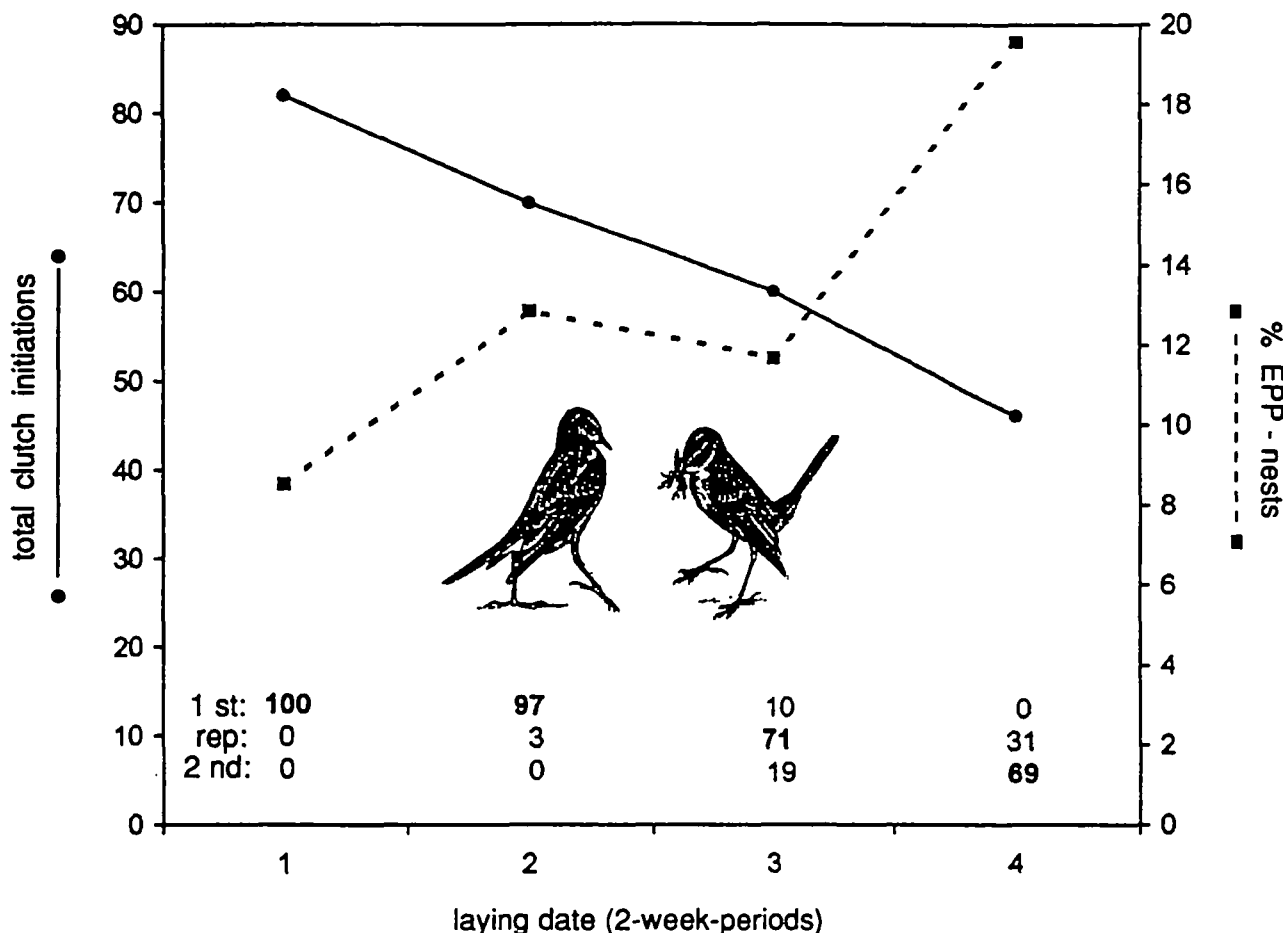
Figure 6
Number of clutches initiated (solid line) and proportion of clutches affected by extrapair paternity (broken line) in relation to laying date broken down into four periods of 2 weeks each, starting 1 May. Numbers above the x-axis show the percentages of first (top row), replacement (middle), and second clutches (bottom), calculated from 221 of the 258 nests for which clutches could be reliably assigned to one of the three categories. The clutch type dominating in the respective period is shown in bold. Pipit drawing by F. Weick from Glutz von Blotzheim and Bauer (1985), with permission.

It is more plausible to assume that EDP is rare because females of territorial species normally do not have knowledge of and access to other nests. Water pipits, however, do not restrict their activities to their territories, but regularly forage in overlapping territories and shared feeding sites. Such visits will increase a female's chances of locating and accessing a suitable nest for deliberate or accidental egg dumping (see Andersson, 1984). These visits will also increase the probability that an egg, fertilized by the male of that other territory, will later be laid in his nest. This is probably a rare event and may be partly responsible for the extreme scarcity of EPM in territorial birds (Birkhead et al., 1990), as opposed to colonial and waterfowl species, from which most cases of EDP are known (Brown and Brown, 1988; Emlen and Wrege, 1986; Lank et al., 1989; McRae and Burke, 1996; Morton et al., 1990; Weigmann and Lamprecht, 1991; Wrege and Emlen, 1987).

Extrapair fertilizations

For male water pipits, the spatial separation between breeding territories and feeding sites can make simultaneous defense of territories and foraging mates incompatible. This creates opportunities for EPCs, especially later in the season when the ratio of sexually active males to fertilizable females ("inclusive

operational sex ratio"; Westneat et al., 1990) increases, because more and more males have completed their own breeding attempts and, consequently, no longer need to guard their own mates or feed their young (cf. Figure 6). As a consequence, asynchronous breeding increases a male's risk of being cuckolded.

But what are the characteristics of those birds that achieve EPFs? To what extent are extrapair activities in water pipits chance events, representative of mixed reproductive strategies of breeders (sensu Trivers, 1972), or reflective of alternative mating strategies of birds that failed to breed? These questions cannot be answered unless the biological parents have been identified. For two reasons, this identification is difficult in our study. First, about 50% of the pipits' foraging trips lead to communal feeding places (Frey-Roos et al., 1995), thus increasing the number of potential candidates far beyond the immediate neighbors that were responsible for extrapair activities in some other studies (Gibbs et al., 1990; Hasselquist et al., 1995; Møller, 1989; Weatherhead et al., 1996). Second, among the candidates, no blood samples are available from 32% of the adults within and all birds around the study area, including the floaters that stay only briefly on their way to higher altitudes, where breeding occurs later.

Which sex is in control of EPFs?

Related to the question of who suffers and who gains from extrapair activities is another question: Which sex is in control of EPFs? In water pipits, the mating behavior is too secretive to allow detailed observations on whether females seek, tolerate, or reject EPCs. Yet, even without such observations and without identifying the genetic parents of EPY, there are several reasons to conclude that females do not seek EPFs for genetic or phenotypic benefits.

1. A female should only opt for EPFs if her fitness is not reduced, in case her male responds by withdrawing his paternal care (cf. "constrained female hypothesis"; Gowaty, 1996). In water pipits, however, unassisted females raise significantly fewer young (Bollmann, 1995; Schläpfer AR, unpublished data).

2. For female-controlled EPF, a positive relationship between synchrony and EPP rates has been predicted (Stutchbury and Morton, in press). We found a negative relationship (Figure 6), suggesting that the occurence of EPCs is better explained by the inclusive operational sex ratio than by female solicitation.

3. No obvious benefits from pursuing EPCs were detected for female water pipits. Females with EPP young were not courtship-fed more and did not recruit additional help at the nest.

4. Choice of superior fathers is unlikely, partly because the number of EPY per brood was low (median = 1), and partly because cuckolded males did not differ from uncuckolded ones in size, condition, age, experience, and social mating status or in prey abundance within their territories.

5. Search for genetic diversity among offspring is not encouraged, because in nests with more than one EPY, all had been sired by the same male.

6. Fertility insurance seems to be unnecessary because unhatched eggs, an indicator of reduced male fertility (Sittman et al., 1966; van Noordwijk and Scharloo, 1981), were rare, and their proportion (4%) did not differ between nests with and without EPY (see also Hasselquist et al., 1995; Lifjeld et al., 1993; Wetton and Parkin, 1991).

7. Finally, in the few cases of EPM, copulating with another male may have increased the female's chances of getting access to his nest (Petrie, 1986), but these cases account for only 9% of all 60 extrapair fertilizations.

Thus, it seems likely that female water pipits do not actively seek the majority of EPFs but merely accept copulation attempts from other males because "the costs and benefits of being inseminated by an extrapair male lie between those associated with either resistance or solicitation" (Westneat et al. 1990: 358). This is the situation where ecological determinants of EPF are most likely to be detected because they affect the male mating strategy without being confounded by female control.

The ecological epiphenomenon hypothesis

We conclude with the working hypothesis that extrapair reproduction in male and female water pipits mainly reflects chance events, arising from the spatial and temporal distribution of nests and feeding sites. The ability to make use of the arising chances can be assumed to have evolved, but variance in reproductive success, and hence the opportunity for sexual selection, in such opportunistic extrapair activities are likely to be much lower than in cases of predictable, trait-related variance (see Kempenaers et al., 1992; Morton et al., 1990; Yezerinac, 1995). This possibility of "accidental" extrapair reproductive activities as an ecological epiphenomenon with low potential for selection should also be considered for

other species, especially where extrapair events are rare. As shown in this study, the precise nature of the ecological determinants may differ between the sexes.

REFERENCES

Amos W, Barrett JA, Pemberton JM, 1992. DNA fingerprinting: parentage studies in natural populations and the importance of linkage analysis. Proc R Soc Lond B 249:157–162.

Andersson M, 1984. Brood parasitism within species. In: Producers and scroungers (Barnard CJ, ed). London: Chapman and Hall; 195–228.

Barber CA, Robertson RJ, Boag PT, 1996. The high frequency of extrapair paternity in tree swallows is not an artefact of nestboxes. Behav Ecol Sociobiol 38:425–430.

Birkhead TR, Møller AP, 1992. Sperm competition in birds. London: Academic Press.

Birkhead TR, Burke T, Zann R, Hunter FM, Krupa AP, 1990. Extrapair paternity and intraspecific brood parasitism in wild zebra finches Taeniopygia guttata, revealed by DNA fingerprinting. Behav Ecol Sociobiol 27:315–324.

Bollmann K, 1995. The mating systems of the alpine water pipit in a variable environment: ecological, demographic and fitness aspects (PhD dissertation). Zürich: University of Zürich.

Bollmann K, Reyer H-U, Brodmann PA, 1997. Territory quality and reproductive success: can alpine water pipits Anthus spinoletta assess the relationship reliably? Ardea (in press).

Brodmann PA, Reyer H-U, Bollmann K, Schläpfer A, Rauter C, 1997. The importance of food quantity and quality for reproductive performance in alpine water pipits (Anthus spinoletta). Oecologia 109: 200–208.

Brown CR, Brown MB, 1988. Genetic evidence of multiple parentage in broods of cliff swallows. Behav Ecol Sociobiol 23:379–387.

Bruford MW, Hanotte O, Brookfield JFY, Burke T, 1992. Single-locus and multi-locus DNA fingerprinting. In: Molecular genetic analysis of populations: a practical approach (Hoelzel AR, ed). Oxford: Oxford University Press; 225–269.

Clutton-Brock TH, Guiness FE, Albon SD, 1982. Red deer: behavior and ecology of two sexes. Edinburgh: Edinburgh University Press.

Cohen J, 1988. Statistical power analysis for the behavioral sciences. Hillsdale, New Jersey: Lawrence Erlbaum Associates.

Davies NB, 1991. Mating systems. In: Behavioural ecology, 3rd ed (Krebs JR, Davies NB, eds). Oxford: Blackwell; 263–294.

Dunn PO, Whittingham LA, Lifjeld JT, Robertson RJ, Boag PT, 1994. Effects of breeding density, synchrony, and experience on extrapair paternity in tree swallows. Behav Ecol 5:123–129.

Emlen ST, Oring LW, 1977. Ecology, sexual selection and the evolution of mating systems. Science 197:215–223.

Emlen ST, Wrege PH, 1986. Forced copulations and intra-specific parasitism: two costs of social living in the white-fronted bee-eater. Ethology 71:2–29.

Freeland JR, Hannon SJ, Dobush G, Boag PT, 1995. Extra-pair paternity in willow ptarmigan broods: measuring costs of polygyny to males. Behav Ecol Sociobiol 36:349–355.

Frey-Roos F, Brodmann PA, Reyer H-U, 1995. Relationships between food resources, foraging patterns, and reproductive success in the water pipit, Anthus sp. spinoletta. Behav Ecol 6:287–295.

Gibbs HL, Weatherhead PJ, Boag, PT, White BN, Tabak LM, Hoysak DJ, 1990. Realized reproductive success of polygynous red-winged blackbirds revealed by DNA markers. Science 250:1394–1397.

Glutz von Blotzheim U, Bauer KM, 1985. Handbuch der Vögel Mitteleuropas, vol. 10/II. Wiesbaden: Aula-Verlag.

Gowaty PA, 1985. Multiple parentage and apparent monogamy. Ornithol Monogr 37:11–21.

Gowaty PA, 1996. Field studies of parental care in birds. In: Advances in the study of behaviour (Snowdon CT, Rosenblatt JS, eds). New York: Academic Press; 476–531.

Gowaty PA, Karlin AA, 1984. Multiple maternity and paternity in single broods of apparently monogamous eastern bluebirds (*Sialia sialia*). Behav Ecol Sociobiol 15:91–95.

Hamilton WJ, Orians GH, 1965. Evolution of brood parasitism in altricial birds. Condor 67:361–382.

Hartley IR, Shepherd M, Robson T, Burke T, 1993. Reproductive success of polygynous male corn buntings (*Miliaria calandra*) as confirmed by DNA fingerprinting. Behav Ecol 4:310–317.

Hasselquist D, Bensch S, von Schantz T, 1995. Low frequency of extrapair paternity in the polygynous great reed warbler, *Acrocephalus arundinaceus*. Behav Ecol 6:27–38.

Jenni L, Winkler R, 1989. The feather length of small passerines: a measurement for wing length in live birds and museum skins. Bird Study 36:1–15.

Kempenaers B, Dhondt AA, 1993. Why do females engage in extrapair copulations? A review of hypotheses and their predictions. Belg J Zool 123:93–103.

Kempenaers B, Verheyen GR, Van den Broeck M, Dondt AA, 1992. Extra-pair paternity results from female preference for high-quality males in the blue tit. Nature 357:494–496.

Lank DB, Cooch EG, Rockwell RF, Cooke F, 1989. Environmental and demographic correlates of intraspecific nest parasitism in lesser snow geese *Chen caerulescens caerulescens*. J Anim Ecol 58:29–45.

Lifjeld JT, Dunn PO, Robertson RJ, Boag PT, 1993. Extra-pair paternity in monogamous tree swallows. Anim Behav 45:213–229.

MacWhirter RB, 1989. On the rarity of intraspecific brood parasitism. Condor 91:485–492.

McRae SB, Burke T, 1996. Intraspecific brood parasitism in the moorhen: parentage and parasite-host relationships determined by DNA fingerprinting. Behav Ecol Sociobiol 38:115–129.

Møller AP, 1989. Intraspecific nest parasitism in the swallow *Hirundo rustica*: the importance of neighbours. Behav Ecol Sociobiol 25:33–38.

Morton ES, Forman I, Braun M, 1990. Extra-pair fertilizations and the evolution of colonial breeding in purple martins. Auk 107:275–283.

Oring LW, 1982. Avian mating systems. In: Avian biology, vol. 6 (Farner DS, King JR, Parkes KC, eds). Orlando, Florida: Academic Press; 1–92.

Otter K, 1996. Female reproductive strategies and reliable male signals in the black-capped chickadee (PhD dissertation). Kingston: Queen's University.

Petrie M, 1986. Reproductive strategies of male and female moorhens (*Gallinula chloropus*). In: Ecological aspects of social evolution (Rubenstein DI, Wrangham R, eds). Princeton, New Jersey: Princeton University Press; 43–63.

Petrie M, Møller AP, 1991. Laying eggs in others' nests: Intraspecific brood parasitism in birds. Trends Evol Ecol 6:315–320.

Pöldmaa T, Montgomerie R, Boag P, 1995. Mating system of the cooperatively breeding noisy miner *Manorina melanocephala*, as revealed by DNA profiling. Behav Ecol Sociobiol 37:137–143.

Rauter C, 1995. Environmental factors, parental care and reproductive performance in the water pipit (*Anthus spinoletta*) (PhD dissertation). Zürich: University of Zürich.

Rauter C, Reyer H-U, 1997. Incubation patterns and foraging effort of female water pipits (*Anthus sp. spinoletta*) in relation to temperature, nest site, food availability and incubation feeding. Ibis 139 (in press).

Reyer H-U, 1994a. 'Do-si-do your partner': Report on the annual conference of the BOU. Ibis 136:110–112.

Reyer H-U, 1994b. Ecology and evolution of mating systems. Verh Dtsch Zool Ges 87:97–113.

Signer E, 1988. DNA fingerprinting: improved DNA extraction from small blood samples. Nucl Acid Res 16:7738.

Sittman K, Abplanalp B, Fraser RA, 1966. Inbreeding depression in Japanese quails. Genetics 54:371–379.

Stutchbury BJ, Morton ES, in press. The effect of breeding synchrony on extra-pair mating systems in songbirds. Behaviour.

Trivers RL, 1972. Parental investment and sexual selection. In: Sexual

selection and the descent of man (Campbell B, ed). Chicago: Aldine; 136–179.

van Noordwijk AJ, Scharloo W, 1981. Inbreeding depression in an island population of the great tit. Evolution 35:674–688.

Weigmann C, Lamprecht J, 1991. Intraspecific nest parasitism in bar-headed geese, *Anser indicus*. Anim Behav 41:677–688.

Weatherhead PJ, Montgomerie R, Gibbs HL, Boag PT, 1996. The cost of extra-pair fertilizations to female red-winged blackbirds. Proc R Soc Lond B 258:315–320.

Westneat DF, 1990. Genetic parentage in the indigo bunting: a study using DNA fingerprinting. Behav Ecol Sociobiol 27:67–76.

Westneat DF, 1992. Nesting synchrony by female red-winged blackbirds: effects on predation and breeding success. Ecology 73:2284–2294.

Westneat DF, 1993. Polygyny and extrapair fertilizations in eastern red-winged blackbirds (*Agelaius phoeniceus*). Behav Ecol 4:49–60.

Westneat DF, Sherman PW, Morton ML, 1990. The ecology and evolution of extra-pair copulations in birds. Curr Ornithol 7:331–369.

Wetton JH, Parkin DT, 1991. An association between fertility and cuckoldry in the house sparrow, *Passer domesticus*. Proc R Soc Lond B 245:227–233.

Whittingham LA, Taylor PD, Robertson RJ, 1992. Confidence of paternity and male parental care. Am Nat 139:1115–1125.

Wickler W, Seibt U, 1983. Monogamy: an ambiguous concept. In: Mate choice (Bateson P, ed). Cambridge: Cambridge University Press; 33–50.

Wittenberger JF, 1979. The evolution of mating systems in birds and mammals. In: Handbook of behavioral neurobiology: social behavior and communication (Master P, Vandenburgh J, eds). New York: Plenum Press; 271–349.

Wrege P, Emlen ST, 1987. Biochemical determination of parental uncertainty in white-fronted bee-eaters. Behav Ecol Sociobiol 20:153–160.

Yezerinac SM, Weatherhead PJ, Boag PT, 1995. Extra-pair paternity and the opportunity for sexual selection in a socially monogamous bird (*Dendroica petechia*). Behav Ecol Sociobiol 37:179–188.

Yom-Tov Y, 1980. Intraspecific nest parasitism in birds. Biol Rev Cambr Philos Soc 55:93–108.