

Kate L. Durrant · Jane M. Hughes

Differing rates of extra-group paternity between two populations of the Australian magpie (*Gymnorhina tibicen*)

Received: 15 December 2003 / Revised: 16 August 2004 / Accepted: 15 November 2004 / Published online: 13 January 2005
© Springer-Verlag 2005

Abstract Extra-pair paternity (EPP) is a common feature of the mating systems of many birds. The rate of EPP may vary between species, races and populations. A comparison of extra-group paternity (EGP) rates was made between two races of a group-living passerine, the Australian magpie (*Gymnorhina tibicen*), to determine if similar mating systems were being employed. The two populations had similar social structure, but differed in group size and dispersal. It was predicted that dispersal differences would have a profound effect on the rate of EGP between the populations, as the population with the lower rate of dispersal and higher chance of breeding with a close relative would engage in EGPs more frequently. Eight microsatellite loci were used to determine parentage in the white-backed Australian magpie (*G. t. tyrannica*). The rate of EGP was found to be 44%. Dispersal rates were estimated from observational data. Over half of the juvenile magpie cohort from the previous breeding season left the territorial group. These results contrast sharply with the results found by other researchers in a population of western Australian magpies (*G. t. dorsalis*). In this population, 82% EGP is recorded and dispersal of juveniles is close to nil. The results indicate that dispersal rate is a potentially important predictor of rates of extra-group fertilisations between populations of this species, and suggest that females maximise their reproductive output by avoiding breeding with close kin.

Keywords Australian magpie · Dispersal · Extra-group paternity · Inbreeding avoidance · Microsatellites

Introduction:

Extra-pair (EPC) or extra-group copulations (EGC) are a feature of many avian mating systems (Griffith et al. 2002). Benefits to the male bird are well established; they gain cost-free genetic offspring (Ligon 1999). However, the reasons for females seeking copulations outside the pair bond are not so obvious. Females may seek direct benefits such as food from courtship feeding, male parental care, or territorial space, but when the birds are already permanently territorial and group living these benefits are less important (Double and Cockburn 2000). Indirect benefits such as increased genetic diversity (Petrie and Kempenaers 1998), ‘good genes’ (Kempenaers et al. 1992; Hasselquist et al. 1996; Sheldon et al. 1997), or enhanced genetic compatibility (Zeh and Zeh 1996, 1997) may become more important in a female bird’s mate choice.

The genetic diversity hypothesis states that a female seeks EPCs primarily to enhance the genetic diversity of her brood. In a review of variation in rates of extra-pair fertilisation (EPF), evidence was found that in closely related species, or races or populations of the same species, the group with the higher level of genetic variability sustained higher rates of EPF (Petrie and Kempenaers 1998). Under the genetic diversity hypothesis, females may seek EPCs as a routine ‘bet hedging’ strategy to maximise genetic diversity across their broods.

It has also been found that females that are genetically similar to their social mate have more failed breeding efforts (Bensch et al. 1994; Kempenaers et al. 1996). These individuals may be genetically similar to their social mate because the two are closely related. Many researchers have concluded that females mate multiply primarily to avoid the negative effects of inbreeding (Brooker et al. 1990; Pusey and Wolf 1996; Stockley et al. 1993; Tregenza and Wedell 2002), and to improve the heterozygosity of offspring (Blomqvist et al. 2002; Foerster et al. 2003).

The ‘good genes’ hypothesis states that females engage in EPCs in order to obtain the high quality genes of

Communicated by M. Soler

K. L. Durrant (✉) · J. M. Hughes
Australian School of Environmental Studies,
Griffith University,
Box 145, QLD 4111 Nathan, Australia
e-mail: durrantk@si.edu
Tel.: +1202 6734648
Fax: ++1202 6734781

preferred males for their offspring. For example, Kempenaers et al. (1992) found that male blue tits (*Parus caeruleus*) that received a high number of visits from extra-territorial females also had fewer extra-pair young in their broods, and consequently suffered less lost paternity. These preferred males survived better, recruited more young and were larger than males that lost paternity.

The genetic compatibility hypothesis suggests that females engage in EPFs when the male they select has a genotype more compatible to their own than their social mate does. This may prevent intragenomic conflict which would lead to non-viable embryos or increased mortality of young (Zeh and Zeh 1996, 1997; Jennions 1997). Females may adopt this strategy routinely as a form of 'bet hedging' or possibly after failed breeding attempts with their social mate. Males that are incompatible with one female may be compatible with another, thus each male's suitability to a particular female depends upon her own genotype, and male compatibility should be randomly distributed in a panmictic population.

In a recent review, the rate of extra-pair paternity (EPP) has been recorded as varying between 0 and 72% across all passerine species, with an average frequency of 11.1% of offspring being the result of EPP (Griffith et al. 2002). Differences in the rate of EPFs within and between species have been attributed to several causes. By comparing the rates of EPFs in two populations of a species living in different social contexts, information can be gathered about what is the most important indirect benefit (or benefits) for the female seeking an extra-pair mating. A comparison between populations can shed light on the constraints, genetic or otherwise, placed upon the females in each population and the strategies they are following in order to maximise their reproductive output. Australian magpies (*Gymnorhina tibicen*) offer an ideal opportunity for testing these hypotheses as there are several very closely related races living in populations with different social structures. A comparison between populations of two of the races is not likely to be affected by phylogenetic bias.

The Australian magpie comprises seven races occurring on the Australian continent, all of which are morphologically distinct (Schodde and Mason 1999). Generally, these large passerines live in pairs or groups on permanent all-purpose territories (Carrick 1963). Some populations are known to breed cooperatively (Hughes et al. 1996), while other populations with multiple-adult groups breed plurally (more than one female nesting mated with the same or different males) (Hughes et al. 2003). In some populations, there is a non-territorial floater flock (Hughes et al. 1996). Magpies will vigorously defend their territory, with all group members participating in aggression towards intraspecific intruders (Carrick 1963; Farabaugh et al. 1992).

A population of Australian magpies (*G. t. dorsalis*) from south-western Australia (Guildford population) was studied over three breeding seasons between 1996 and 1998 (Hughes et al. 2003). Using eight microsatellite loci to determine parentage, the rate of extra-group paternity

(EGP) was assessed as 82%, currently the highest known rate of EPP in any passerine species. The western magpies (*G. t. dorsalis*) live in groups of 2–12 adults, and young usually do not disperse from the natal territory, instead remaining there to eventually become breeders (Carrick 1972; Hughes et al. 2003). Lack of dispersal from the natal territory would be expected to lead to high levels of inbreeding within territories, but Hughes et al. (2003) found no evidence of inbreeding, and attributed it to the high level of EGPs. In this scenario, by seeking EGPs females would avoid inbreeding and increase the genetic variation of their brood.

There is indirect evidence that eastern magpies may have a different dispersal pattern to western birds, and evidence of gene flow over long distances among eastern races suggests they may disperse more than western races (Baker et al. 2000, 2001). Differing dispersal rates could have a profound influence on EGP rates between races.

In order to make a comparison of EGP rates between races of the Australian magpie, a population of the white-backed magpie (*G. t. tyrannica*) was studied in western Victoria, south-eastern Australia. White-backed magpies have not been studied in great detail before the present study, and detailed demographic information is non-existent. If the inbreeding avoidance/ increased heterozygosity hypotheses account for EGP seeking behaviour in this species, then the level of natal dispersal and the level of inbreeding in the territories would become important factors. If both populations display a lack of natal dispersal, leading to socially close relatives breeding within territories, then levels of EGP would be the same. If dispersal capabilities were significantly different, everything else being equal, it should be reflected in differing levels of EGP. If the major reason for females engaging in EGP was to obtain 'good genes' from their extra-group males, males detected as responsible for an EGP would be expected to have territorial fledglings of their own, or males that were never cuckolded would be expected to be found as EGP males in other territories. Also, rates of EGP would be expected to be similar between populations. Similarly, if genetic incompatibility was a major factor, rates of EGP could be expected to be similar between populations if they are also genetically similar. Given the close relationship between the races, the males in each population may be expected to have the same or similar incidence of genetic incompatibility with their social mates.

The aims of this study were to: (1) describe the social structure in a population of the white-backed magpie including the rate at which male and female juveniles remained in the natal territory; (2) determine the rate of EGP in this population; and (3) to compare EGP rates with those of western magpies in an attempt to discriminate between the major EPF hypotheses.

Methods

Study site and field methods

Thirty-two contiguous magpie territories were studied on a rural property in Rowsley, 50 km west of Melbourne, Victoria, Australia (37°43'S, 144°24'E). All territorial magpies were caught in a walk-in trap baited with grated cheese. On two occasions, in 2000 and 2002, decoy trapping was employed. An adult male, either a flock bird or a territorial male from a two-male territory was captured and placed inside a small cage. This cage was placed inside the larger walk-in trap. Territorial magpies were attracted to the 'intruding' male and entered the trap in order to attack the decoy, whereupon they were removed by researchers observing from a short distance. Each bird was bled from the claw and banded with a unique colour combination and an Australian Bird and Bat Banding Scheme (ABBBS) serial band in accordance with ABBBS regulations. Blood samples were stored in lysis buffer and kept at -20 to -80°C.

Twenty-one of the 32 territories produced fledglings in at least one of the three breeding seasons between 2000 and 2002 ($n=79$). Territory membership and information on breeding activity was determined during censuses conducted between June and December each year. Each individual territory was watched between 2 and 3 times per week. Watches were 20 min in duration, in the morning when the birds were most active. Binoculars and a spotting scope were used to make detailed observations. The presence or absence of members of a territorial group was noted, as well as any disputes within the group or with neighbouring groups. Breeding activity such as which female was nesting, and the order of female nesting in multiple female groups was recorded. Nests were observed intensively as fledging became imminent. Magpie nestlings that appear well developed and spend time perched on the edge of the nest are usually about to fledge within the next 1–2 days. Fledglings were caught by hand, bled and banded on the day they fledged from the nest, or very soon thereafter. This reduced the chance of a fledgling straying from its true natal territory to virtually zero. Nestlings that were ejected from the nest early and died under the nest tree were also sampled by amputating a toe and storing it in lysis buffer.

Microsatellite analysis

Eight microsatellite loci were analysed, using primers developed in the Molecular Ecology Laboratory, Griffith University (Hughes et al. 2003), with a new locus employed, 208 (Sequence: Forward: 5' 7TC AGA AAG ACC TAG TTG GTG C, Reverse: 5' GCC TAG TTG AGG TTT TCA AAT G, repeat sequence: (CT)₁₃G (CT)₁₀GT(CT)₆, annealing temperature: 57°C), instead of using Hughes et al.'s (2003) locus 119a. Total genomic DNA was isolated from blood samples using standard phenol-chloroform extraction. PCR amplification of nDNA occurred in 12.5 µl reactions for each primer set comprising: 1.25 µl of 10x *Taq*. polymerase buffer, 2 mM MgCl₂, 10 mM of dNTP's, 10 µl of forward and reverse primers, 0.25 U of Biotech *Taq*. polymerase, and 50–100 ng of DNA template. For each microsatellite locus, the following thermocycler program was used: denature at 94°C followed by 40 cycles at 94°C for 30 s, annealing temperature (Hughes et al. 2003) for 30 s, 72°C for 30 s, finishing with an extension step of 72°C for 30 s. When complete, samples were held at 4°C. Microsatellite product with formamide added was denatured and cooled immediately on ice before being run on a 5% polyacrylamide gel on a GelScan 2000 DNA analyser (Corbett Research). Products were sized across and between gels using a commercial standard (ABI PRISM Tamra 350). In some cases, entire territorial groups were run on the same gel to assist in scoring allele sizes, in other cases, a homemade ladder was employed, which was made from the combined product of a number of individuals. Twenty-four individuals were run per gel, with four lanes of Tamra, and an additional three lanes of homemade ladder, if used.

Genetic sexing

Juvenile magpies cannot be sexed by plumage characteristics and were genetically sexed according to the methods of Griffiths et al. (1996). The primers used were P2 and P8 as described in Griffiths et al. (1996), and the test simultaneously amplified homologous sections of the CHD-Z and CHD-W genes. Male birds are the homogametic sex (ZZ) and females are heterogametic (ZW), so the test amplified a single section for males and two sections for females. Purified DNA underwent PCR in the following thermocycler conditions: denature at 94°C for 1.30 min followed by 45 cycles at 94°C for 30 s, 54.5°C (annealing temperature) for 30 s, 72°C for 30 s, finishing with an extension step of 72°C for 7.00 min. Samples were then held at 4°C until required. PCR product was electrophoresed through a 3% agarose gel in 0.5×T.B.E. buffer, and the results visualised under a UV light. Each gel included a known male and female sample from adult magpies that had been sexed by plumage characteristics. Male birds produced a single band, and female birds produced two bands.

Statistical analysis

Basic allelic statistics were calculated, including: F_{IS} values (calculated using GENEPOP; Raymond and Rousset 1995), allele frequencies, expected and observed heterozygosities, exact tests for deviation from Hardy-Weinberg expectations, and exclusion probabilities using CERVUS 2.0 (Marshall et al. 1998). Using the simulation function of the program, a typing error rate of 1% was assumed and 75% of the candidate parents in the population were claimed as sampled to account for neighbouring groups of birds that were not included in the study. This ensured that conservative critical Delta scores were used in the confidence testing of parentage assignments. Confidence levels of 80% are sufficient to make accurate estimates of parentage, including distinguishing between close relatives as potential fathers (Slate et al. 2000). Parentage was assigned first within territories (the social group defending a particular parcel of land), with all adults being candidate parents, then if maternity or paternity could not be assigned, fledglings were screened against all adults within the sampled population. Maternity was detected first and accepted if the female had a confidence level (C.L.) at or above 80%, then the group was re-analysed with the assigned mother as the 'Known Parent', males that received a C.L. of 80% or above were accepted as fathers. Occasionally CERVUS could not detect mothers, but stated there were zero or very few mismatches between fledglings and putative parents. These individuals were rechecked by hand, each gel re-assessed visually, and any errors corrected. These highly matching females were then taken as the known mother. If a father was detected from outside the social group holding the territory at or above the 80% C.L., it was termed a case of extra-group paternity (EGP), cases where mothers were detected outside the social group were labelled extra-group maternity.

Simple exclusion-based estimates of parentage were also employed in order to make a direct comparison with the results of Hughes et al. (2003). In this method, CERVUS was used to detect the most closely related adults to the offspring in question, first within territories, then outside if no parents were found. Those adults with zero or a maximum of one mismatch between parental and offspring alleles were assigned as parents. All mismatches that were only one microsatellite repeat unit out were checked on the original gel to ensure they were true mismatches and not scoring errors.

A chi-squared test was employed to determine if the number of males in a territory had an effect on the number of extra-group young produced. An index of relatedness (r) between all individuals within territories and within the whole population was calculated in RELATEDNESS 5.0.8 (Queller and Goodnight 1989). The categories examined were relatedness of all males, all females, all males and females averaged across all territories and within territories, and all juveniles. Positive relatedness scores indicate that two individuals are more related than expected by chance if the two genotypes were randomly selected. Negative scores mean they are

Table 1 Demographic information on the Rowsley population of white-backed Australian magpies (*Gymnorhina tibicen tyrannicus*). Individuals refer not only to adults within a territorial group, but also sub-adults between one year and three years of age. Fledglings born within the timeframe of this study were not included in this analysis

Territorial group category <i>n</i> =32	Males <i>n</i> =41	Females <i>n</i> =63	Sub-adults <i>n</i> =21
Zero bird groups	n/a	n/a	56.3%
Single bird groups	71.9%	18.8%	25%
Two bird groups	28.1%	65.6%	15.6%
Three bird groups	0%	15.6%	3.1%
Average number of birds	1.3	2.0	0.7
Average number of adults		3.3	
Average number of individuals		3.9	

less related than expected by chance. Relatedness scores of $r \approx 0.25$ indicate half sibling or uncle-nephew relationships, and scores of $r \approx 0.5$ indicate full sibling or parent-offspring relationships (Queller and Goodnight 1989).

Relatedness scores were also calculated for the social breeding pair in each group that produced fledglings and related to the level of EGP in the group over three seasons. A Spearman's correlation was used to examine the relationship between relatedness and level of EGP. Then a two-tailed *t*-test was used to compare the relatedness of social pairs with EGP below 0.5 across three seasons with the relatedness of social pairs with EGP above 0.5 across three seasons. The average within-group relatedness of adults belonging to a territorial group was compared between the two populations (white-backed and western), using a *t*-test. Finally, the average relatedness between the breeding female and the males available to her within a territory were compared between the two populations, using a *t*-test.

Results:

Demographic data

All territorial residents were captured and sampled ($n=221$), including all within-group putative parents, with the exception of a single adult female. The fledglings from her group (DH) were excluded from further parental analyses. However, the genotypes of the two DH males that were sampled were included in the searches for extra-group fathers for the rest of the study population. The complete sampling of all the remaining territorial groups was confirmed by observational data and record-keeping. White-backed magpies live in permanent territorial groups in the Rowsley population. There was a range of two–six individuals per group with an average of 3.3 adults per group, usually two females and one male, although there were nine multiple male groups (Table 1). Where there was more than one female, usually only the dominant one built a nest and incubated eggs, but occasionally the subordinate female also bred in a separate nest after the first female had started incubating. The breeding season was highly asynchronous, lasting between June and November each year with young fledging from mid-September until the end of November.

Dispersal rates were calculated from observational data for two seasons (Table 2). A fledgling that was re-sighted in its natal territory after 1 year, (i.e. the beginning of the

Table 2 Dispersal rates for 1-year-old and 2-year-old magpies for two breeding seasons. Dispersed refers to birds that were not re-sighted in their natal territory 1 year after fledging or after the commencement of observations in the case of 2-year-old sub-adults. All sub-adults were genetically sexed

Season	Age	Sex	Dispersed
Dec 2000 – August 2001	1-year-olds	Males (<i>n</i> =10)	0.60
		Females (<i>n</i> =7)	0.57
		Both sexes	0.59
	2-year-olds	Males (<i>n</i> =7)	0.57
		Females (<i>n</i> =14)	0.14
		Both sexes	0.29
Dec 2001 – August 2002	1-year-olds	Males (<i>n</i> =14)	0.64
		Females (<i>n</i> =29)	0.62
		Both sexes	0.63
	2-year-olds	Males (<i>n</i> =3)	0.33
		Females (<i>n</i> =3)	0.66
		Both sexes	0.57

next breeding season) was considered not to have dispersed. Fledglings that had not been re-sighted in their natal territory could either have died in the interim or dispersed out of their natal territory. The average rate of loss of 1-year-old birds from the natal territory was 0.61 over two seasons. Between the first and second seasons, the rate of dispersal of 2-year-old birds was 0.29, and dispersal was male biased (0.57 of male 2-year-olds compared to 0.14 of females left their natal territories). Dispersal of fledglings born in the 2000 breeding season between the 2001 and 2002 seasons (2-year-old birds) was 0.57. Within the study area a non-territorial flock of magpies was observed in 2000 and 2001. The flock numbered between 20 and 70 individuals, contained both sexes and sub-adult as well as adult birds. On one occasion a banded sub-adult bird that disappeared from a territorial group was re-sighted some weeks later feeding within the flock.

Parentage analysis

The eight microsatellite loci used in the parentage analysis were highly variable, with a range of allele numbers from 4 to 22, and when used in conjunction, correctly excluded non-parents in 99.8% of cases (Table 3). After Bonferroni correction, none of the loci deviated from Hardy-Weinberg expectations.

From the microsatellite analysis, territorial mothers of fledglings could be definitely assigned in 92.4% of cases examined (Table 4). In no cases of extra-group maternity was a mother found to come from another territory included in the study. Paternity could not be assigned within a territory in 44.3% of cases. Where the father was from outside the territorial group, he was detected in another territory within the study population in 11.4% of cases, and 50% of the time this father was a resident in a close or neighbouring territory. In 5% of cases a fledgling was not related to any territorial adults, and was termed an 'egg-dump'. Two males in two territories were never cuckolded (M21 PC: five fledglings; M17 HHH: two fledglings) over the three breeding seasons. Two of the EGP fathers iden-

Table 3 Locus statistics for microsatellites used in parentage analysis. *Ho* Observed heterozygosity, *He* expected heterozygosity, *Exc P1* probability of excluding the first parent correctly, *Exc P2* probability of excluding the second parent correctly, *HWE* Hardy-Weinberg Equation ($\alpha=0.05$). Mean number of alleles per locus: 15.78, Mean expected heterozygosity: 0.778, Total exclusionary power, first parent: 0.98, Total exclusionary power, second parent: 0.99

Locus	N	k	Ho	He	F _{IS} *	Exc P1	Exc P2	HWE**
43a	221	20	0.796	0.871	0.0860	0.59	0.74	NS
67c	221	4	0.489	0.447	-0.0940	0.10	0.22	NS
112	221	21	0.891	0.911	0.0215	0.70	0.82	NS
115a	221	16	0.769	0.837	0.0813	0.51	0.67	NS
115b	221	22	0.946	0.908	-0.0414	0.69	0.81	NS
201a	221	13	0.584	0.658	0.1136	0.26	0.43	NS
206b	221	14	0.778	0.791	0.0157	0.42	0.59	NS
208	221	17	0.679	0.688	0.0129	0.32	0.51	NS

Mean number of alleles per locus: 15.78, Mean expected heterozygosity: 0.778, Total exclusionary power, first parent: 0.98, Total exclusionary power, second parent: 0.99

* (Weir and Cockerham 1984)

** After Bonferroni correction

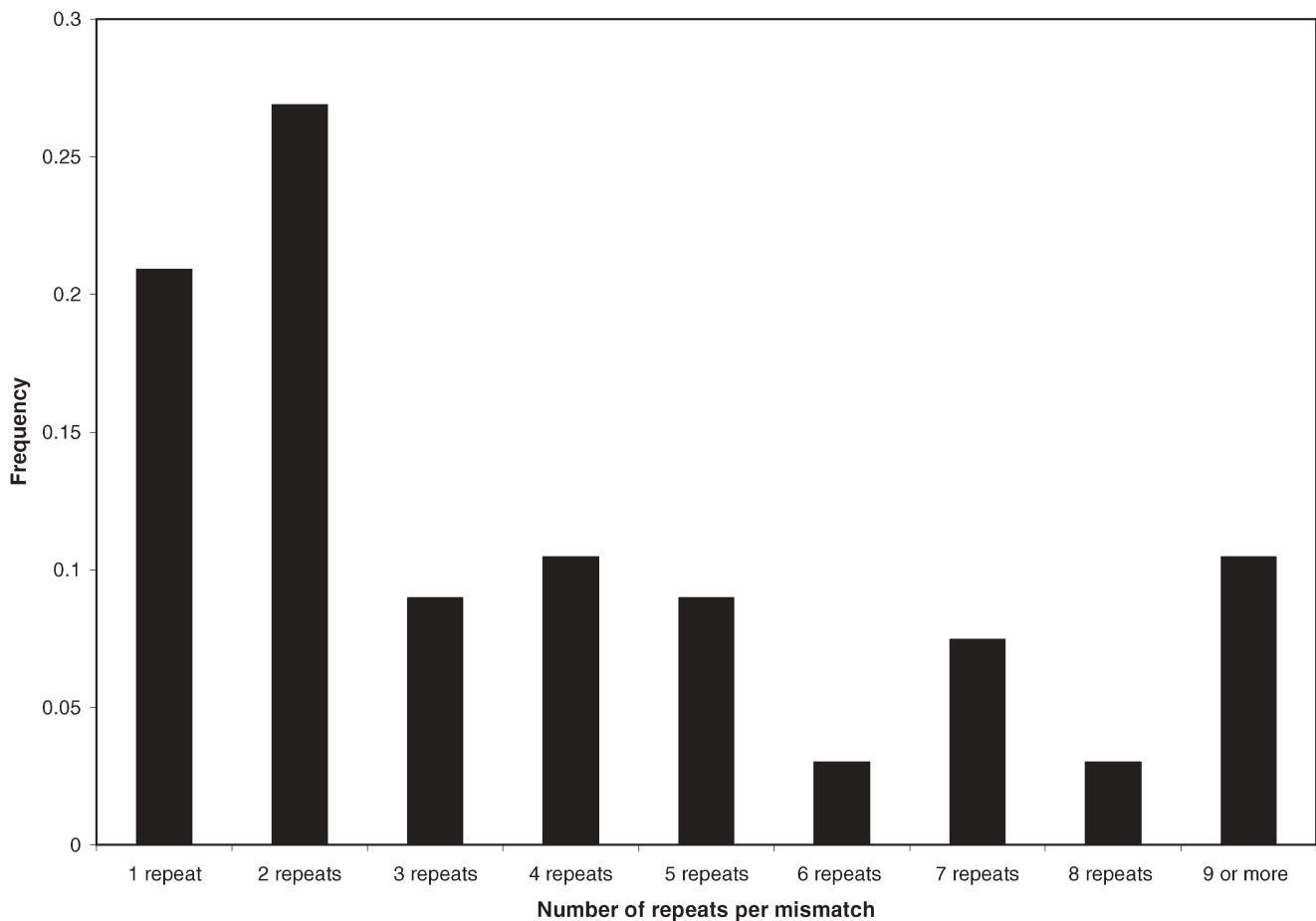


Fig. 1 Frequency of the number of microsatellite repeat units that constituted the degree of each mismatch between excluded territorial fathers and their putative offspring, simple exclusion analysis only

tified were themselves cuckolded within their home territories, (M9 CT=0.75, M3 BE=1.00), the other two identified EGP fathers did not have any fledglings produced in their respective territories during the course of the study. One of these birds (M5 AS) moved into a new territory every year, and no fledglings were produced in any of his 'home' territories for the duration of the study.

Analysing the data using exclusionary methods yielded similar results and the two methods of assessing parentage gave highly concordant results. Maternity could be assigned within the social group in 93.7% of cases, extra-group paternity occurred in 38% of cases. Exclusionary analysis revealed a few extra fathers within groups that were not assigned using CERVUS alone, after every

Table 4 Territorial groups of white-backed Australian magpies in the Rowsley population that produced fledglings during the 2000–2002 breeding seasons. Underlined birds are genetic parents, birds in the *Extra-group parent* column are all extra-group fathers, and birds in the *Male* or *Female* columns in bold type were found to be parents using the exclusionary method of assessing parentage. The

(n1) denotes a territorial female who incubated the first or only nest for that season and (n2) denotes a territorial female who incubated the second nest in the season. Fledglings that have a nesting female recorded in its row came from that nest. This data was unavailable for all fledglings, particularly where two broods fledged within a short space of time, or nests were undiscovered

Territory	Breeding season	Fledglings	Females	Males	Extra-group parent
AS	2000	J1	F1(n1), F2, F3	M1	
	2001	J2	<u>F1</u> (n1), F2, F3	<u>M1</u>	
BAC	2001	J3	<u>F4</u> (n1), F5	<u>M2</u>	
	2002	J4	<u>F4</u> (n1), F5	<u>M2</u>	
BE	2001	J5	<u>F6</u> (n1)	M3	
BPP	2000	J6	<u>F7</u> (n1), F8	<u>M4</u>	
	2001	J7	<u>F7</u> (n1), F8	<u>M4</u>	<u>M5</u> AS
CJ	2000	J8	<u>F9</u> (n1)	<u>M6</u> , M7	
	2002	J9	<u>F9</u> (n1)	<u>M6</u> , M7	
CT	2000	J10	<u>F10</u> (n1)	<u>M8</u> , <u>M9</u>	
	2001	J11	<u>F10</u> (n1)	<u>M8</u> , <u>M9</u>	
	2002	J12	<u>F10</u> (n1)	<u>M8</u> , <u>M9</u>	
	2001	J13	<u>F10</u>	<u>M8</u> , <u>M9</u>	
FW	2001	J14	<u>F11</u> (n1), F12	M10	
		J15	<u>F11</u> (n1), F12	M10	
		J16	<u>F11</u> , <u>F12</u> (n2)	M10	
	2002	J17	<u>F11</u> (n1), F12	<u>M10</u>	
		J18	<u>F11</u> (n1), F12	<u>M10</u>	
GK01	2001	J19	<u>F13</u> , F14	M11	
		J20	<u>F13</u> (n1), F14	M11	
		J21	<u>F13</u> (n1), F14	M11	
	2002	J22	<u>F13</u> (n1), F14	M11	
HB	2000	J23	<u>F15</u> (n1), F16	M12, M13	
	2002	J24	<u>F15</u> (n1), F16, F17	<u>M14</u> , M15	
		J25	<u>F15</u> (n1), F16, F17	<u>M14</u> , M15	
HHH	2002	J26	<u>F18</u> , F19(n1)	M16, <u>M17</u>	
		J27	F18, F19(n1)	M16, <u>M17</u>	
JD	2001	J28	F20(n1), F21	<u>M18</u>	
		J29	<u>F20</u> (n1), F21	<u>M18</u>	
		J30	F20 (n1), F21	M18	
		J31	F20, <u>F21</u> (n2)	M18	
	2002	J32	F20(n1), F21	M18	
KN	2000	J33	<u>F22</u> (n1)	<u>M19</u>	
		J34	<u>F22</u> (n1)	<u>M19</u>	
		J35	<u>F22</u> (n1)	<u>M19</u>	
	2002	J36	<u>F22</u> (n1)	<u>M19</u>	
LB	2000	J37	<u>F23</u> , <u>F24</u> (n1)	<u>M20</u>	
		J38	F23, <u>F24</u> (n1)	<u>M20</u>	
		J39	F23, <u>F24</u> (n1)	<u>M20</u>	
	2001	J40	<u>F23</u> , <u>F24</u> (n1)	<u>M20</u>	
		J41	F23, <u>F24</u> (n1)	<u>M20</u>	
		J42	F23(n2), F24	<u>M20</u>	
		J43	F23(n2), F24	<u>M20</u>	
	2002	J44	<u>F23</u> , <u>F24</u> (n1)	<u>M20</u>	
		J45	F23(n2), F24	<u>M20</u>	
		J46	<u>F23</u> (n2), F24	<u>M20</u>	
PC	2001	J47	<u>F25</u> (n1), F26, F27	<u>M21</u>	
		J48	<u>F25</u> (n1), F26, F27	<u>M21</u>	
	2002	J49	<u>F25</u> (n1), F26, F27	<u>M21</u>	
		J50	<u>F25</u> (n1), F26, F27	<u>M21</u>	
		J51	<u>F25</u> (n1), F26, F27	<u>M21</u>	
RR	2000	J52	<u>F5</u> (n1), F28, F29	<u>M22</u>	
	2001	J53	<u>F28</u> (n1), F29	<u>M22</u>	
	2002	J54	<u>F28</u> (n1), F29	<u>M22</u>	
SD	2001	J55	<u>F30</u> (n1), F31	<u>M23</u>	
		J56	<u>F30</u> (n1), F31	<u>M23</u>	
		J57	<u>F30</u> (n1), F31	<u>M23</u>	
	2002	J58	<u>F30</u> , <u>F31</u> (n1)	<u>M23</u>	<u>M9</u> CT
		J59	F30, <u>F31</u> (n1)	M23	
		J60	<u>F30</u> (n2), F31	M23	
		J61	<u>F30</u> (n2), <u>F31</u>	M23	<u>M3</u> BE
SS	2000	J62	F32(n1), <u>F33</u>	M24	
	2001	J63	F32(n1), F33	M24	
	2002	J64	<u>F32</u> (n1), F33	M24	

Table 4 (continued)

Territory	Breeding season	Fledglings	Females	Males	Extra-group parent
SW	2001	J65	F34(n1), F35, F36	<u>M25</u>	
		J66	F34(n1), F35, F36	<u>M25</u>	
		J67	F34, F35(n2), <u>F36</u>	<u>M25</u>	
		J68	F34, F35(n2), F36	<u>M25</u>	
		J69	F34, <u>F35</u> (n2), F36	<u>M25</u>	
	2002	J70	F34(n1), F35	<u>M25</u>	
		J71	<u>F34</u> , F35(n2)	<u>M25</u>	
TR	2001	J72	<u>F34</u> , F35(n2)	<u>M25</u>	<u>M26</u> LBG
		J73	<u>F37</u> (n1), F38	M27, M28	
VE	2000	J74	<u>F37</u> (n1), F38	M27, M28	
		J75	<u>F39</u> (n1), F40	M29	
WWF	2002	J76	<u>F39</u> (n1), F40	<u>M29</u>	
		J77	<u>F39</u> (n1)	<u>M29</u>	
	2001	J78	<u>F41</u> (n1), F42	M30, M31	
		J79	<u>F41</u> (n1), F42	M30, M31	

mismatch between putative parents (as assigned by CERVUS), territorial putative parents (gained from observational data) and offspring was checked on the original gel. Paternal exclusions of putative fathers from within territories averaged 3.3 mismatches per father / offspring comparison, and were an average of 3.8 microsatellite repeat units out (Fig. 1), indicating that they were true genotype mismatches and not gel scoring errors.

Table 4 shows all territorial groups that produced fledglings, all putative parents and genetic parents where possible. Multiple male groups were uncommon, just six of all territories that produced young during the course of the study, but paternity was shared between both males on one occasion. There was no significant relationship between the number of males in a group and the number of extra-group young detected within that group ($\chi^2=0.829$, $P=0.3625$). There were three apparent cases of egg-dumping where an unknown female had laid an egg in another's nest. On one occasion, a sub-adult bird (later genetically sexed and found to be female) was identified as the mother of a fledgling within a territory that already had an adult female nesting.

Relatedness between all territorial males in the white-backed population was -0.0247 on average (SE=0.0063, 95% CI=0.0145), average relatedness between all females was -0.0122 (SE=0.0027, 95% CI=0.0062), and between all males and females the average relatedness was -0.0152 (SE=0.0028, 95% CI=0.0064). The average relatedness of males and females within territories was 0.0541 (SE=0.0144, 95% CI=0.0284). The average relatedness between all juveniles in the white-backed population was 0.0032 (SE=0.0026, 95% CI=0.0060).

There was no significant correlation between the relatedness coefficient of the social pair within a territory and the rate of EGP observed for that group over three seasons [Spearman's $R_{20}=0.038$, $P=0.875$ (2-tailed)]. A two-tailed t -test between the relatedness of social pairs with EGP below 0.5 over three breeding seasons, and the relatedness of social pairs with EGP above 0.5 was also not significant ($t_{7,13}=2.201$, $P=0.840$).

The average relatedness scores between all adult males and females within territories for both populations (white-backed mean $r_{111}=0.0604$, SE mean=0.0301; western mean $r_{68}=0.0918$, SE mean=0.0595) were not significantly different [$t=-0.470$, $P=0.644$ (2-tailed)]. The average relatedness scores between the breeding female and the males available to her within a territory for both populations (white-backed mean $r_{71}=0.0087$, SE mean=0.0299; western mean $r_{50}=0.0868$, SE mean=0.0589) were also not significantly different [$t=-1.182$, $P=0.254$ (2-tailed)].

To check that the difference in the observed rate of EGP's between the two populations [white-backed (p_1) vs western (p_2)] was significant, a test for differences in proportions based on the normal approximation to the binomial distribution was used. The sample sizes in both populations (white-backed $n=79$, western $n=43$; Hughes et al. 2003) were sufficiently large to allow such an approach. The results of the two-tailed z -test indicated that the difference in EGP rate between the two populations was significant ($z=4.086$, $P<0.001$).

Discussion

Demographics of the white-backed magpie

The social system of white-backed magpies was extremely variable, single pairs defended a territory and bred successfully, while other groups in the population contained multiple adults, employed plural breeding strategies and even appeared to breed cooperatively at times. The major difference in the social structure of the white-backed when compared to the western magpie is the number of birds occupying a territory and the rate of dispersal of the young. While white-backed magpies live in groups averaging 3.3 adults, western magpies live in groups averaging 4.9 adults (Hughes et al. 2003). More than half the 1-year old and an average of 43% of the 2-year-old subadults of the white-backed population dispersed or disappeared from the natal territory before the

next breeding season commenced. It should be noted that the parentage of the 2-year-old birds dispersing between 2000 and 2001 was not established, as they were not sampled in the year they fledged. Many of these young birds probably joined the flock of non-territorial magpies that was observed within the study area. In contrast, young western magpies rarely disperse from the natal territory, and remain there until death or a breeding opportunity arises, and there is no non-territorial flock (Robinson 1956; Hughes et al. 2003).

The EGP rate of 44% in the white-backed magpie population is high when compared to the average EPP rate of 11% calculated across all passerine species (Griffith et al. 2002). Despite the complex social structure of the population, the extreme territoriality, and the fact that intra-specific intruders are severely punished, EGPs are commonplace. Hughes et al. (2003) suggested that magpies may engage in EGP-seeking behaviour in the pre-dawn hours, similar to the behaviour that was discovered after radio-tracking female superb fairy wrens (*Malurus cyaneus*) (Double and Cockburn 2000), but this is yet to be tested.

Comparison of EGP rates between white-backed and western magpies

The large difference observed in the incidence of EGP between the two populations of magpies is interesting given the overall similarity of the two races. Both races live in multiple adult social groups, displaying extreme territoriality. In a race with limited dispersal of young, one would expect a high level of relatedness within territories and a positive relationship between territory proximity and relatedness, but this was not the case. Genetic data from western magpies showed that males and females from within a territory were more closely related than males and females from different territories, but that there was no relationship between the geographic proximity of males or females and their relatedness (Hughes et al. 2003). In white-backed magpies, the mean relatedness between territorial males and females in the Rowsley population was also positive, while the mean value between all males and females in the population was negative. This indicates that there may be some offspring who remain in the natal territory after reaching adulthood.

The inbreeding avoidance hypothesis predicted that a female would seek an EGP when her social mate was more genetically similar to her than an extra-group male. Females seeking EGPs to enhance genetic diversity and avoid the effects of inbreeding depression within their brood is a reasonable explanation for the EGP phenomenon in the western magpie. In this population, the low level of dispersal of juveniles, coupled with extreme aggression towards intra-specific intruders could be expected to lead to a situation where first or second order relatives are breeding with each other. As described above, the population does not show excessively high levels of relatedness within territories. Hughes et al. (2003) suggest

high levels of EGP in their population may be an inbreeding avoidance strategy, designed to avoid the negative effects of genetic similarity between the parents. This has been suggested for other passerine species such as tree swallows (*Tachycineta bicolor*) (Kempnaers et al. 1999), and for group-living species such as the superb fairy-wren (Cockburn et al. 2003), and splendid fairy-wren (*Malurus splendens*) (Brooker et al. 1990).

The 'good genes' hypothesis can be discounted as the most important factor given the large difference in EGP rates between the populations. Rates of EGP were expected to be the same or similar in both populations as females would seek to maximise their reproductive output by mating with a high quality male outside the pair bond. There is no reason to suggest that one population has a different distribution of high quality males than the other, or that in such closely related races, the characteristics of a high quality male would differ. In the white-backed population, the same male was not detected as siring more than one EGP fledgling within a group, or in any other group. Male great reed warblers (*Acrocephalus arundinaceus*) that sired extra-pair young sired multiple young within that group (Hasselquist et al. 1996). Also, male white-backed magpies that were never cuckolded, contrary to predictions for the 'good genes' hypothesis, were never detected as the males responsible for any EGP fledglings produced. Likewise, the males that were detected as EGP fathers were males that were either cuckolded on their home territory, or never produced any territorial fledglings of their own. One male was even unable to maintain a permanent territory from year to year. These males do not fit the general image of high quality males such as male yellow warblers (*Dendroica petechia*) (Yezerinac and Weatherhead 1997), and male red-winged blackbirds (*Agelaius phoeniceus*) (Weatherhead and Boag 1995) which managed to both gain EPPs and maintain some paternity rights at home. However, it has been suggested that among fairy-wrens (*Malurus* spp.) extra-pair and within-pair paternity may not always be positively correlated, as the presence of helpers allows the dominant male to leave parental duties and seek extra-group mating opportunities, but also means he will lose paternity in his home territory to intruder males (Green et al. 1995). In order to reject this hypothesis with confidence, further studies that investigate the relative quality and future reproductive success of EGP and non-EGP young should be attempted. This study relied on sampling those individuals that fledged from the nest, causing a potential bias in the sample if EGP young had some sort of competitive advantage at the nestling stage. Unfortunately, sampling nestlings is costly, logistically difficult and was outside the scope of the present study. All Australian magpies nest high in very tall trees (30 m above ground), and will defend the nest site vigorously. Because of this, sampling of nestlings has not been attempted as yet.

Genetic incompatibility as the major factor in EGP-seeking behaviour was predicted to lead to similar levels of EGP in both populations given their close relationship. This clearly is not the case. However, it may still be a

factor in that some females may engage in extra-pair copulations as a 'bet-hedging' strategy to avoid the negative effects of intragenomic conflict. Similarly, mating to maximise genetic diversity across a brood may also be utilised as a 'bet hedging' strategy. In the absence of data concerning hatching failure and nestling survival, these hypotheses are difficult to test. Genetic incompatibility and genetic diversity may not be the sole or even major explanations for the EGP phenomenon within this species given the importance of inbreeding avoidance for at least one of its populations, but may still be elements that require further investigation.

The best explanation for high levels of extra-group paternity in the western magpie appears to be females seeking EGP to ensure maximum heterozygosity and avoid the effects of inbreeding depression in their offspring. This may account for the incredibly high rate of EGP within this race. However, white-backed magpies still have a relatively high rate of EGP themselves when compared to other passerines. They have moderate dispersal rates of juveniles, and these dispersing young are able to enter a flock system. Presumably, when a breeding opportunity appears, either a neighbouring territorial bird or one of these flock-living birds fills it. There is gene flow created from these dispersal events and movements between territories. Inbreeding should not be an issue; however, not all juvenile white-backed magpies disperse out of their natal territory. The positive mean relatedness values between territorial males and females indicate that some adult offspring may still be present in the natal territory. Just as in the western magpies, some juveniles will remain on the natal site to await either death or a breeding opportunity. There is still a risk, albeit much reduced, of breeding with a close relative and suffering the negative effects of inbreeding. Additionally, all males were sampled in a contiguous network of territories in this population, yet few EGP fathers were detected within the sample, indicating that females may be mating with males that reside outside the study area, and are potentially less likely to be closely related to them. Inbreeding avoidance may be an important factor to explain why white-backed magpies still engage in extra-group copulations regularly.

Although inbreeding avoidance may be a factor, it must be stated that no correlation was found between the relatedness scores of social breeding pairs in the white-backed population, and the average within-group rate of EGP for those pairs. Also, mean relatedness of territorial adults did not differ significantly between the two populations. However, logically, a difference in the average relatedness scores within territories between populations would not be expected. In the western population, extremely high levels of EGPs may act to balance out the potential effect on relatedness coefficients of very low juvenile dispersal, and related individuals breeding with each other. In the white-backed population, where there are moderate levels of juvenile dispersal, lower rates of EGPs may act to keep relatedness scores at an equivalent level to those found in the western population. In both populations, relatively high levels of EGPs appear to have

acted successfully to avoid the negative effects of inbreeding between socially related individuals, and this is reflected in low, but still positive, mean relatedness coefficients within territories.

Taken together, this indicates that it is quite likely that more than one factor influences EGP-seeking behaviour in the Australian magpie, and that perceived social relationships are more important to the individual bird in making mating decisions than actual genetic relatedness. Perceived social relationships were important to the helping decisions of Seychelles warblers (*Acrocephalus sechellensis*) (Komdeur 1994). While there was a relationship between relatedness and helping effort, it was also found that helpers were more likely to help those that fed them at the nest when they were young than those that were equally related, but had never fed them (Komdeur 1994). Birds that fed nestlings would normally be close kin, but not necessarily.

It is acknowledged that this comparison is between only two populations (of different races) due to the time-consuming, labour-intensive nature of the work. There is, however, a lack of intraspecific analyses of the variation in traits like EGP in the literature. Intraspecific variation must first be addressed before looking at interspecific variation, and more broad-based hypotheses. Sampling a population of the black-backed race of magpies (*G. t. tibicen*) is further testing the hypothesis presented here. Black-backed magpies live on the eastern side of the continent and hybridise with white-backed magpies in central Victoria (Schodde and Mason 1999). In north-eastern Australia, they live almost exclusively in pairs, and before the start of each breeding season, violently harass the previous year's offspring until they leave the natal territory (personal observation; R. Kallioinen, personal communication). Thus, juvenile dispersal is close to 100%. If dispersal rate within a population is a major predictor of EGP activity in this species, and given the behaviour of the black-backed race, a lower rate of EGP would be predicted.

Evidence from another group-living species, the red-cockaded woodpecker (*Picoides borealis*), which displays very low levels of EGP (Haig et al. 1994), shows that high levels of female dispersal occurs when there are related males left on the natal territory (Daniels and Walters 2000). Dispersal was thought to be influenced by the costs of inbreeding. Dispersal behaviour itself remains largely a mystery for many species, as the study of it relies upon long-term gathering of data on colour-banded populations. But with the recent availability of suitable genetic markers it is now feasible to investigate medium and long-distance dispersal patterns (e.g. Baker et al. 2001; Hansson et al. 2003). Why Australian magpies have differing rates of dispersal in different populations, and exactly how this affects other aspects of the population's and species' life history, aside from influencing behavioural traits such as extra-group copulations, remains to be explored further. Dispersal capability and actual dispersal rate may have a much greater effect on avian behaviour and mating systems than was previously thought.

Acknowledgements: Field assistance was ably provided by Graham Durrant, Chris Hepper, and Alicia Toon. Thank you to the Manly and Lord families for allowing access to their land. Thanks also to Jing Ma, Jill Shephard, and Alicia Toon for guidance in the laboratory. Mark Ponniah and an anonymous reviewer gave helpful comments on this manuscript. K.L.D. was supported by an Australian Postgraduate Award. Trapping was undertaken with permission from the Department of Natural Resources, Victoria, banding was carried out under license from the Australian Bird and Bat Banding Scheme, and the Griffith University Ethics Committee approved the study.

References:

- Baker AM, Mather PB, Hughes JM (2000) Population genetic structure of Australian magpies: evidence for regional differences in juvenile dispersal behaviour. *Heredity* 85:167–176
- Baker AM, Mather PB, Hughes JM (2001) Evidence for long-distance dispersal in a sedentary passerine, *Gymnorhina tibicen* (Artamidae). *Biol J Linn Soc* 72:333–343
- Bensch S, Hasselquist D, Von Schantz T (1994) Genetic similarity between parents predicts hatching failure: nonincestuous inbreeding in the great reed warbler? *Evolution* 48:317–326
- Blomqvist D, Andersson M, Küpper C, Cuthill IC, Kis J, Lanctot RB, Sandercock BK, Székely T, Wallander J, Kempenaers B (2002) Genetic similarity between mates and extrapair parentage in three species of shorebirds. *Nature* 419:613–615.
- Brooker M, Rowley I, Adams M, Baverstock PR (1990) Promiscuity: an inbreeding avoidance mechanism in a socially monogamous species? *Behav Ecol Sociobiol* 26:191–199
- Carrick R (1963) Ecological significance of territory in the Australian magpie, *Gymnorhina tibicen*. *Proc Int Ornithol Congr* 13:740–753
- Carrick R (1972) Population ecology of the Australian black-backed magpie, royal penguin, and silver gull. Population ecology of migratory birds: a symposium. U.S. Department of the Interior Wildlife Research Report 2:41–99
- Cockburn A, Osmond HL, Mulder RA, Green DJ, Double MC (2003) Divorce, dispersal and incest avoidance in the cooperatively breeding superb fairy-wren *Malurus cyaneus*. *J Anim Ecol* 72:189–202
- Daniels S, Walters JR (2000) Inbreeding depression and its effects on natal dispersal in red-cockaded woodpeckers. *Condor* 102:482–491
- Double M, Cockburn A (2000) Pre-dawn infidelity: females control extra-pair mating in superb fairy-wrens. *Proc R Soc Lond B* 267:465–470
- Farabaugh SM, Brown ED, Hughes JM (1992) Cooperative territorial defense in the Australian magpie, *Gymnorhina tibicen* (Passeriformes, Cracticidae), a group-living songbird. *Ethology* 92:283–292
- Foerster K, Delhey K, Johnsen A, Lifjeld JT, Kempenaers B (2003) Females increase offspring heterozygosity and fitness through extra-pair matings. *Nature* 425:714–717
- Green DJ, Cockburn A, Hall ML, Osmond H, Dunn PO (1995) Increased opportunities for cuckoldry may be why dominant male fairy-wrens tolerate helpers. *Proc R Soc Lond B* 262:297–303
- Griffith SC, Owens IPF, Thuman KA (2002) Extra pair paternity in birds: a review of interspecific variation and adaptive function. *Mol Ecol* 11:2195–2212
- Griffiths R, Daan S, Dijkstra C (1996) Sex identification in birds using two CHD genes. *Proc R Soc Lond B* 263:1251–1256
- Haig SM, Walters JR, Plissner JH (1994) Genetic evidence for monogamy in the cooperatively breeding red-cockaded woodpecker. *Behav Ecol Sociobiol* 34:295–303
- Hansson B, Bensch S, Hasselquist D (2003) A new approach to study dispersal: immigration of novel alleles reveals female-biased dispersal in great reed-warblers. *Mol Ecol* 12:631–637.
- Hasselquist D, Bensch S, von Schantz T (1996) Correlation between male song repertoire, extra-pair paternity and offspring survival in the great reed warbler. *Nature* 381:229–232.
- Hughes JM, Hesp JDE, Kallioinen R, Kempster M, Lange CL, Hedstrom KE, Mather PB, Robinson A, Welbourne MJ (1996) Differences in social behaviour between populations of the Australian magpie *Gymnorhina tibicen*. *Emu* 96:65–70
- Hughes JM, Mather PB, Toon A, Ma J, Rowley I, Russell E (2003) High levels of extra-group paternity in a population of Australian magpies (*Gymnorhina tibicen*): evidence from microsatellite analysis. *Mol Ecol* 12:3441–3450
- Jennions MD (1997) Female promiscuity and genetic incompatibility. *Trends Ecol Evol* 12:251–253
- Kempenaers B, Verheyen GR, Van den Broeck M, Burke T, Van Broeckhoven C, Dhondt AA (1992) Extra-pair paternity results from female preference for high-quality males in the blue tit. *Nature* 357:494–496
- Kempenaers B, Adriaensen F, Van Noordwijk AJ, Dhondt AA (1996) Genetic similarity, inbreeding and hatching failure in blue tits: are unhatched eggs infertile? *Proc R Soc Lond B* 263:179–185
- Kempenaers B, Congdon B, Boag P, Robertson RJ (1999) Extrapair paternity and egg hatchability in tree swallows: evidence for the genetic compatibility hypothesis? *Behav Ecol* 10:304–311
- Komdeur J (1994) The effect of kinship on helping in the cooperative breeding Seychelles warbler (*Acrocephalus sechellensis*). *Proc R Soc Lond B* 256:47–52
- Ligon JD (1999) *The Evolution of Avian Breeding Systems*. Oxford University Press, Oxford
- Marshall TC, Slate J, Kruuk LEB, Pemberton JM (1998) Statistical confidence for likelihood-based paternity inference in natural populations. *Mol Ecol* 7:639–655
- Petrie M, Kempenaers B (1998) Extra-pair paternity in birds: explaining variation between species and populations. *Trends Ecol Evol* 13:52–58
- Pusey A, Wolf M (1996) Inbreeding avoidance in animals. *Trends Ecol Evol* 11:201–206
- Queller DC, Goodnight KF (1989) Estimating relatedness using genetic markers. *Evolution* 43:258–275
- Raymond F, Rousset M (1995) Population genetics software for exact tests and ecumenicism. *J Heredity* 86:534–543
- Robinson A (1956) The annual reproductive cycle of the magpie, *Gymnorhina dorsalis* Campbell, in south-western Australia. *Emu* 56:235–336
- Schodde R, Mason IJ (1999) *The Directory of Australian Birds: Passerines*. CSIRO, Melbourne
- Sheldon BC, Merilä J, Qvarnström A, Gustafsson L, Ellegren H (1997) Paternal genetic contribution to offspring condition predicted by size of male secondary sexual character. *Proc R Soc Lond B* 264:297–302
- Slate J, Marshall T, Pemberton J (2000) A retrospective assessment of the accuracy of the paternity inference program CERVUS. *Mol Ecol* 9:801–808
- Stockley P, Searle JB, MacDonald DW, Jones CS (1993) Female multiple mating behaviour in the common shrew as a strategy to reduce inbreeding. *Proc R Soc Lond B* 254:173–179
- Tregenza T, Wedell N (2002) Polyandrous females avoid costs of inbreeding. *Nature* 415:71–73
- Weatherhead PJ, Boag PT (1995) Pair and extra-pair mating success relative to male quality in red-winged blackbirds. *Behav Ecol Sociobiol* 37:81–91
- Weir BS, Cockerham CC (1984). Estimating F-statistics for the analysis of population structure. *Evolution* 38:1358–1370
- Yezerinac SM, Weatherhead PJ (1997) Extra-pair mating, male plumage coloration and sexual selection in yellow warblers (*Dendroica petechia*). *Proc R Soc Lond B* 264:527–532
- Zeh JA, Zeh DW (1996) The evolution of polyandry I: intragenomic conflict and genetic compatibility. *Proc R Soc Lond B* 263:1711–1717
- Zeh JA, Zeh DW (1997) The evolution of polyandry II: post-copulatory defences against genetic incompatibility. *Proc R Soc Lond B* 264: 69–75