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DISPERSAL PATTERN OF BLACK-BILLED MAGPIES (*PICA HUDSONIA*) MEASURED BY MOLECULAR GENETIC (RAPD) ANALYSIS

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ABSTRACT.—Black-billed Magpies (*Pica hudsonia*) are a relatively sedentary corvid, with greater dispersal of females than males. To genetically confirm that dispersal pattern, 29 reproductively active adults were captured over two years and were scored for primer-specific random amplified polymorphic DNA (RAPD) bands (53 polymorphic bands in 1996 and 104 in 1997). In both years, we captured more previously banded males than females, and individuals recaptured were more often the heaviest nestling in their brood from the previous years. Genetic distances between all possible adult pairs were calculated and degree of genetic similarity between pair-wise comparisons was assessed using the Mantel test. In the pair-wise comparison of genetic distances, data for both years showed that genetic distances between males were significantly less ($P < 0.05$) than those for other adult combinations (male–female and female–female). Using the same analytical approach, we found that females were no more genetically similar to one another ($P > 0.05$) than were random pairs of adults. Group-distance analysis using the computer package RAPD also showed higher genetic similarity among males than among females. We suggest that a plausible hypothesis to account for the relatively high genetic homogeneity of the breeding male population compared to the female population is that a long-lasting intraclutch dominance hierarchy exists among siblings. That hierarchy is sufficiently long-lasting to influence the breeding population, resulting in alpha males remaining in the natal area, whereas lower ranking birds, including females, are forced to disperse. *Received 13 September 1999, accepted 8 August 2000.*

FEMALE-BIASED natal dispersal occurs in the majority of bird species (Greenwood 1980, Greenwood and Harvey 1982), especially for most species of corvids (Woolfenden and Fitzpatrick 1984, Strickland 1991, Marzluff and Balda 1992), except for White-throated Magpie-Jay (*Calocitta formosa*), which has an extremely male-biased dispersal pattern (Langen 1996a, b). In some cooperatively breeding corvid species with helpers at the nest, such as Florida Scrub-Jay (*Aphelocoma coerulescens*), females disperse substantially farther than males, but neither sex tends to disperse more than a few territories from the birthplace (Woolfenden and Fitzpatrick 1984). A similar pattern occurs in another cooperatively breeding corvid with helpers, Pinyon Jay (*Gymnorhinus cyanocephalus*), where there are more female wanderers (i.e. dispersers in the flock) in spring than males (Marzluff and Balda 1989, 1992). Most females breed in their first year, whereas some males stay and help their parents. Strickland (1991) showed in Gray Jays (*Perisoreus canadensis*), another cooperative breeder (Waite and Strickland 1997) that about two-thirds of the juvenile "stayers" were males.

Black-billed Magpies (*Pica hudsonia*) are a social corvid and are relatively sedentary, with an average dispersal distance of only 1.2 territories reported for populations in England (Birkhead 1991). North American Black-billed Magpies do not have helpers at the nests. Instead, philopatry in magpies seems to be associated with territorial acquisition (Trost 1999). North American Black-billed Magpies are not territorial year round; after fledging, territorial boundaries break down and magpies are seen in large social flocks. Dominance hierarchies are found within those flocks; males are dominant over females, and surprisingly, hatching-year birds are often dominant over adults (Reese and Kadlec 1984, Komers and Komers 1992, Trost and Webb 1997). Hierarchies among young males are strongly linear, whereas female hierarchies are much less stable (Moholt

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1989, Trost and Webb 1997) and may contain triangles (Komers and Komers 1992). Presumably, a dominant male would "squeeze into" the natal territory if its parents or their neighbors were to allow it, but usually such juvenile males are forced to wait for an opening caused by death of a parent or neighbor (Trost 1999). The more submissive siblings, including females, tend to disperse farther, and presumably will only establish a territory if they encounter a reliable food source (Reese and Kadlec 1984, Trost 1999). Such a pattern of sex-biased dispersal leads to specific predictions with regard to the degree of genetic similarity between sexes. Breeding males, due to their tendency to establish territories in or near natal sites, should show higher levels of genetic similarity than breeding females in those same populations.

Dispersal of birds or mammals from natal sites typically is quantified by using direct methods (Payne 1991, Stenseth and Lidicker 1992) such as banding, or mark and recapture. A potential bias of direct methods is the low probability of finding marked individuals once they leave the population (Peacock 1997). According to Koenig et al. (1996), chance of detecting dispersal events in an area larger than the diameter of the study area is virtually zero using direct methods. To compensate for those limitations, recent dispersal studies have used genetic methods as well as direct banding or marking and recapturing (Dobson 1994, Horn et al. 1996, Peacock 1997).

In our study, we explored the genetic relatedness of reproductively active adult Black-billed Magpies using random amplified polymorphic DNA (RAPD) markers developed by Williams et al. (1990) and Welsh and McClelland (1990). The RAPD technique of examining genotypes has been used to identify species and to study variation within and among populations (e.g. Haig et al. 1994, Horn et al. 1996). This modification of the polymerase chain reaction (PCR) amplifies segments of genomic DNA that are primer-specific and reproducible, but of varying lengths. Amplification primers, which are 10 nucleotides long with random sequences, bind to homologous bases along both strands of DNA, and the PCR-like protocol replicates the intervening base sequence. The variable lengths of amplified DNA are inherited in a Mendelian fashion (Williams et al. 1990) and thus can be used for genetic analysis (Hadrys and Schierwater 1992, Grosberg et al. 1996). Here, primer-specific amplified DNA bands of reproducible lengths are considered to be attributes, just as any other genetically-determined trait, with the assumptions that (1) marker alleles from different loci do not comigrate to the same position on a gel; and (2) each locus can be treated as a two-allele system, with only one of the alleles per locus—the dominant allele—being amplifiable by the PCR reaction (Lynch and Milligan 1994).

We have used that technique to determine the pattern and extent of RAPD variation within a natural, freely ranging population of magpies. In particular, we asked whether or not the genetic similarity among breeding males was greater than that among breeding females. If males are more genetically similar to each other than are females, that would support the hypothesis of female-biased dispersal in magpies. A higher degree of genetic similarity among males would suggest that more males than females had remained in their natal area; and that females are coming from other areas, which implies that females tended to disperse farther to breeding sites than did males.

STUDY AREA AND METHODS

Study area.—Our study area is located about 45 km southeast of Pocatello, 6 km north of McCammon, Idaho, USA (42°4'N, 112°10'W). Breeding magpies were captured from late February to late June 1996 and 1997 in this study area, which is approximately 1.6 km^2 and is roughly bisected by the Portneuf River. On the east side of the river are scattered pastures and farm houses, and on the west side is an old lava flow covered with sagebrush (*Artemisia tridentata*). Nesting trees on both sides of the river are mostly black hawthorn (*Crataegus* sp.), Russian olive (*Elaeagnus angustifolia*), and Utah junipers (*Juniperus osteosperma*). Banding data for nestlings, including weight measurements prior to fledging, have been obtained on this study site annually since 1989.

Capturing breeding magpies.—Breeding Black-billed Magpies are extremely wary birds and tend to avoid unusual objects, making them one of the most difficult birds to capture (Alsager et al. 1972, Scharf 1985). That is especially true of territorial pairs. Wang and Trost (2000) found three reliable trapping methods for successfully capturing breeding magpies during different stages of their breeding cycle. In the early season while adults were still foraging in flocks and when food was scarce, we used a square, walk-in trap with a funneled entrance on the

TABLE 1. Number of recaptured male magpies in each nestling weight rank in 1996 and 1997.

	Rank of male by weight in the nest				
Year	1	2	3	4	5
1996	6	1	1	0	0
1997	7	2	1	1	1

floor. During the egg-laying period, we used a modified bal-chatri trap (Berger and Mueller 1959), traditionally used for raptors, with a live female magpie inside. During the feeding and chick-rearing period, we used a modified mist net with a live-mounted Great Horned Owl (*Bubo virginianus*) as a decoy. The gender of the captured bird was determined by the methods of Reese and Kadlec (1982), and Gerstell and Trost (1997), or by the presence of an incubation patch in females.

Sample collection and DNA isolation.—Blood samples were collected from all captured adults and nestlings in each year of the study. To collect blood samples for DNA extraction, we used an ethanol patch to swab the skin over the point at which the brachial vein crossed the elbow on the ventral surface of the wing, then punctured the vein with a 23-gauge needle held at a shallow angle and collected 100 μL of blood in sterile 1.5 mL microcentrifuge tubes. Samples were immediately put on ice, carried back to the laboratory in a cooler, and stored at −70°C until DNA was extracted. Neosporin was applied to the wound after the blood was collected and the bird was held for at least 3 min before releasing. That blood sampling technique did not appear to cause undue injury or death to any of the captured magpies.

Two methods of DNA extraction were used. The first method followed the standard protocol of phenol–chloroform–isoamyl alcohol extraction (Sambrook et al. 1989). A 100 μL blood sample was suspended in SET buffer (0.15 M NaCl, 0.05 M Tris-HCl, 1 mM EDTA; pH 8.0) and was stored at −60°C. Later, 15 μL proteinase K (10 mg/mL) and 0.3 mL 10% SDS were added to the thawed sample and incubated overnight at 37°C. Samples were extracted three times with equal volumes of phenol (saturated with 10 mM Tris, 1 mM EDTA, pH 7.5)–chloroform–isoamyl alcohol (25:24:1) followed by one chloroform–isoamyl alcohol (24:1) extraction. Approximately two and a half volumes of cold 100% ethanol, and 1/10 volume of 7.5 M ammonium acetate was added to precipitate DNA. The DNA pellet was washed with 80% ethanol and resuspended in sterile distilled water.

The second method used the G NOME Whole Blood DNA Isolation Kit (Bio 101, Vista, California). DNA concentration in the extracted samples was quantified by using GeneQuant II DNA/RNA calculator (Pharmacia Biotech, Cambridge, United Kingdom).

RAPD PCR procedure.—DNA was diluted to optimal concentrations that were later used for the RAPD amplification of each individual sample. Carefully optimizing DNA concentrations ensures reproducible RAPD amplification runs. To determine the DNA concentration that gave the clearest and most consistent amplification results, four doubling dilutions of each sample in the range of 5 to 40 ng/μL were amplified. Once the optimal DNA concentration was chosen, it was then considered to be the "working" dilution and was later used in all further RAPD reactions. RAPD reactions were set up as described by Williams et al. (1990). Basically, a primer-specific master mix of reagents was prepared and then aliquoted into individual 0.2 mL microreaction, thin-walled polypropylene PCR tubes (USA/Scientific, Ocala, Florida) in a final volume of 10.5 μL containing 1 × PCR buffer; 1.5 mM $MgCl_2$; 0.5 μM of 10 mer primers (Operon Technologies, Alameda, California); 100 μM each of dATP, dCTP, dGTP, and dTTP; and 0.5 units of *Taq* DNA polymerase (Promega, Madison, Wisconsin). Ten ng of DNA for each individual was used per reaction.

Amplification started with an initial cycle of 2.5 min at 94°C, 1 min at 35°C, and 2 min at 72°C; following 45 cycles of 1 min at 94°C, 1 min at 35°C, and 2 min at 72°C, then ended with 10 min elongation at 72°C. Thermal cycles were performed overnight on GeneAmp PCR System 2400 (Perkin Elmer, Foster City, California). Amplification products were electrophoresed on a combination of a total 2% gel, containing 0.8% agarose (Promega) and 1.2% synergel (Diversified Biotech, Boston, Massachusettes), at 5 to 10 v/cm. Gels were stained with ethidium bromide (2.5 μg/500 mL) for 20 min, and destained in water for 30–60 min. Stained gels were photographed while illuminated with an ultraviolet light.

Scoring and analyzing bands.—Only amplifications that produced clear, crisp bands were analyzed, whereas those primer amplifications that resulted in a smeared pattern were not considered for analysis.

TABLE 2. Average genetic similarity of pairwise comparisons between each sex and between random pairs.

Year	Between males	Between females	Between random pairs
1996 (n = 29)	0.657 ± 0.099	0.559 ± 0.129	0.580 ± 0.121
1997 (n = 29)	0.625 ± 0.125	0.537 ± 0.120	0.572 ± 0.129

TABLE 3. Genetic distances among breeding magpies in 1996. Individuals are identified by banding numbers and sex (m or f).

	051m	126m	325m	146m	075m	460m	330m	224m	271m	036m	063m	331m	472m	701m
051m														
[a]126m	0.355													
325m	0.440	0.467												
[a]146m	0.484	0.444	0.467											
[a]075m	0.412	0.282	0.333	0.333										
460m	0.379	0.294	0.357	0.529	0.243									
330m	0.448	0.471	0.500	0.529	0.297	0.312								
224m	0.394	0.211	0.375	0.368	0.171	0.167	0.278							
[a]271m	0.429	0.250	0.471	0.350	0.256	0.316	0.316	0.143						
[a]036m	0.400	0.314	0.448	0.429	0.211	0.333	0.273	0.189	0.282					
063m	0.486	0.250	0.471	0.500	0.302	0.368	0.421	0.238	0.273	0.231				
331m	0.333	0.316	0.438	0.474	0.317	0.278	0.444	0.250	0.286	0.243	0.238			
472m	0.429	0.350	0.471	0.350	0.349	0.368	0.474	0.286	0.273	0.282	0.273	0.143		
701m	0.538	0.419	0.520	0.484	0.412	0.379	0.448	0.333	0.429	0.333	0.371	0.333	0.371	
[a]267m	0.529	0.333	0.394	0.487	0.381	0.351	0.459	0.220	0.209	0.316	0.256	0.220	0.209	0.353
034m	0.471	0.385	0.455	0.436	0.333	0.351	0.405	0.220	0.209	0.263	0.302	0.171	0.163	0.353
215m	0.355	0.333	0.400	0.333	0.282	0.294	0.412	0.211	0.250	0.257	0.350	0.263	0.200	0.419
045f	0.548	0.556	0.400	0.500	0.436	0.471	0.647	0.474	0.450	0.543	0.650	0.474	0.500	0.613
335f	0.500	0.680	0.444	0.520	0.630	0.565	0.652	0.556	0.517	0.739	0.714	0.538	0.500	0.810
343f	0.438	0.405	0.419	0.405	0.300	0.371	0.486	0.282	0.268	0.333	0.415	0.282	0.268	0.500
332f	0.400	0.440	0.444	0.440	0.556	0.565	0.652	0.556	0.448	0.652	0.500	0.615	0.571	0.905
333f	0.391	0.429	0.429	0.500	0.400	0.385	0.538	0.333	0.375	0.462	0.484	0.448	0.419	0.667
349f	0.353	0.436	0.515	0.333	0.381	0.405	0.459	0.366	0.395	0.368	0.442	0.366	0.349	0.412
354f	0.448	0.353	0.357	0.471	0.459	0.375	0.625	0.389	0.368	0.455	0.421	0.444	0.421	0.586
336f	0.484	0.500	0.600	0.500	0.487	0.471	0.647	0.474	0.500	0.486	0.550	0.474	0.400	0.548
337f	0.543	0.450	0.588	0.450	0.442	0.474	0.579	0.429	0.455	0.436	0.455	0.429	0.364	0.600
461f	0.588	0.436	0.636	0.590	0.476	0.514	0.568	0.463	0.581	0.421	0.442	0.415	0.442	0.529
462f	0.407	0.375	0.385	0.562	0.429	0.267	0.533	0.353	0.444	0.419	0.389	0.294	0.333	0.481
513f	0.250	0.517	0.478	0.586	0.500	0.407	0.481	0.484	0.455	0.500	0.576	0.419	0.455	0.667

[a] Previously banded as nestlings.

About half of the primers tested did not produce interpretable data. Photographs of interpretable gels were scored as 1 (band present) or 0 (band absent) for each individual bird. Both distance and similarity matrices were generated from the raw data matrix of 1s and 0s by using the computer program, RAPD, written by Vera Ford (unpubl. University of California at Davis). This program only considers band matches in the estimation of distance and similarity values (Wolfe et al. 1998). On the basis of the shared bands between each pair of individuals and the total number of bands amplified in each individual, the program will calculate genetic similarity between each pair by:

$$S_{xy} = 2n_{xy}/(n_x + n_y) \quad (1)$$

where n_x and n_y are numbers of bands amplified in individuals x and y, respectively, and n_{xy} is number of bands shared by those two individuals. Pairs with high scores are most similar genetically.

All possible pairwise genetic distance values (Nei and Li 1979) were also calculated by:

$$D_{xy} = 1 - S_{xy} = 1 - [2n_{xy}/(n_x + n_y)] \quad (2)$$

Because distance measures are the inverse of similarity, smaller numbers are associated with more genetically similar individuals, whereas larger numbers suggest genetic dissimilarity.

The genetic-distance matrix was then subjected to a Mantel test (Schnell et al. 1985) to assess degree of genetic similarity among breeding adults (Wang 1999). A Mantel test was used because a given individual was pair multiplied with every other individual, thus multiple pair-wise comparisons were not independent data points. A Mantel test takes into account that lack of independence when testing for significant associations between particular groups (Schnell et al. 1985). For the test, as detailed in Wang (1999), we must compute the test statistic Z, as well as its permutational variance, standard error, and expected value. The expected value is subtracted from the test statistic and the result divided by the standard error to provide a t-value that can be compared against a t-distribution with infinite degrees of freedom (i.e. the standard normal distribution). Thus, test significance of t is tested against a standard normal distribution ($Z = \pm 1.96$, $\alpha = 0.05$).

TABLE 3. Extended.

267m	034m	215m	045f	335f	343f	332f	333f	349f	354f	336f	337f	461f	462f	513f
0.095														
0.282	0.231													
0.385	0.385	0.444												
0.538	0.538	0.583	0.429											
0.250	0.200	0.243	0.351	0.333										
0.538	0.615	0.500	0.619	0.400	0.417									
0.379	0.379	0.259	0.417	0.391	0.185	0.391								
0.429	0.381	0.333	0.590	0.556	0.350	0.481	0.333							
0.405	0.459	0.294	0.471	0.545	0.371	0.364	0.440	0.459						
0.538	0.487	0.389	0.667	0.538	0.405	0.615	0.379	0.436	0.353					
0.442	0.442	0.350	0.700	0.643	0.415	0.571	0.290	0.395	0.421	0.250				
0.429	0.429	0.487	0.692	0.778	0.450	0.704	0.467	0.381	0.568	0.436	0.209			
0.371	0.371	0.375	0.562	0.545	0.333	0.545	0.280	0.371	0.333	0.312	0.333	0.371		
0.562	0.500	0.379	0.586	0.474	0.400	0.474	0.273	0.438	0.407	0.310	0.394	0.500	0.200	

RESULTS

Captured magpies.—There were more male (33 in 1996, 36 in 1997) than female (22 in 1996, 25 in 1997) magpies captured in both years, but the difference was not significant (1996, $\chi^2 = 2.20$, df = 1, $P > 0.05$; 1997, $\chi^2 = 1.98$, df = 1, $P > 0.05$). However, among previously banded birds in the samples, significantly more males (8 in 1996, 12 in 1997) were captured than females (1 in each year) that were banded as nestlings in previous years (1996, $\chi^2 = 5.44$, df = 1, $P < 0.05$; 1997, $\chi^2 = 9.31$, df = 1, $P < 0.05$). Additionally, on the basis of a weight hierarchy measured at fledging time, there were 75% first-ranking birds in 1996, and 58% first-ranking birds in 1997 (Table 1). Those patterns suggest that heavier, and thus more dominant, male magpies in their broods remained in their natal area.

Genetic relatedness of breeding magpies.—Forty-one Operon 10 mer primers were used to amplify the DNA from a sample of 29 reproductively active magpies (17 males and 12 females) from 1996 and 29 reproductively active birds (18 males and 11 females) from 1997. Of those primers, 12 produced scorable gels, yielding 53 polymorphic bands among breeding birds in 1996 and 104 bands in 1997. An average of 8.6 polymorphic bands was produced per primer.

Average similarity among breeding males was higher than that among females and that between random pairs in both years (Table 2), indicating that males were more genetically similar to each other than were females. However, average genetic distance between all possible pairs was 0.420 ± 0.121 (SD) for breeding birds in 1996, and 0.428 ± 0.130 (SD) for breeding birds in 1997, which are representative of an overall randomly mating population.

The genetic distance matrix for each year (Tables 3 and 4) produced by the RAPD program was subjected to the Mantel test (Schnell et al. 1985), which showed that reproductively active males were significantly more genetically sim-

TABLE 4. Genetic distances among breeding magpies in 1997. Individuals are identified by banding numbers and sex (m or f).

	711m	719m	712m	586m	209m	146m	706m	472m	580m	738m	462m	610m	063m	331m
711m														
719m	0.185													
712m	0.127	0.193												
[a]586m	0.265	0.176	0.269											
209m	0.222	0.250	0.228	0.098										
[a]146m	0.277	0.224	0.265	0.111	0.125									
706m	0.478	0.375	0.469	0.209	0.292	0.286								
472m	0.296	0.250	0.298	0.216	0.214	0.265	0.292							
580m	0.345	0.298	0.310	0.308	0.298	0.240	0.429	0.228						
738m	0.259	0.214	0.263	0.255	0.321	0.306	0.417	0.250	0.228					
462m	0.351	0.316	0.316	0.371	0.436	0.455	0.484	0.487	0.385	0.333				
610m	0.283	0.236	0.286	0.400	0.382	0.417	0.489	0.309	0.286	0.345	0.500			
[a]063m	0.276	0.267	0.311	0.309	0.333	0.358	0.462	0.267	0.213	0.300	0.429	0.288		
331m	0.258	0.281	0.231	0.356	0.313	0.370	0.464	0.313	0.323	0.281	0.467	0.302	0.324	
[a]701m	0.424	0.475	0.387	0.429	0.377	0.362	0.509	0.443	0.452	0.443	0.628	0.467	0.446	0.420
599m	0.569	0.623	0.593	0.625	0.585	0.565	0.733	0.547	0.481	0.547	0.622	0.654	0.614	0.574
705m	0.444	0.429	0.368	0.451	0.429	0.435	0.500	0.393	0.333	0.393	0.500	0.418	0.433	0.438
[a]381m	0.377	0.333	0.302	0.424	0.419	0.467	0.536	0.429	0.406	0.333	0.421	0.387	0.403	0.412
726f	0.447	0.429	0.388	0.511	0.542	0.609	0.619	0.510	0.480	0.429	0.429	0.375	0.434	0.444
513f	0.393	0.379	0.379	0.444	0.439	0.527	0.490	0.379	0.390	0.414	0.556	0.404	0.323	0.397
210f	0.393	0.414	0.414	0.519	0.544	0.564	0.608	0.448	0.458	0.345	0.514	0.404	0.484	0.365
461f	0.556	0.447	0.500	0.381	0.447	0.366	0.282	0.404	0.458	0.447	0.533	0.522	0.529	0.527
718f	0.296	0.286	0.263	0.294	0.286	0.292	0.417	0.286	0.333	0.357	0.436	0.309	0.333	0.313
713f	0.300	0.323	0.270	0.333	0.290	0.283	0.444	0.290	0.302	0.323	0.455	0.344	0.394	0.314
737f	0.439	0.458	0.400	0.444	0.390	0.469	0.529	0.390	0.400	0.390	0.550	0.448	0.460	0.373
563f	0.439	0.424	0.400	0.444	0.390	0.440	0.490	0.390	0.333	0.390	0.512	0.448	0.397	0.433
336f	0.541	0.429	0.500	0.483	0.492	0.527	0.600	0.429	0.375	0.460	0.591	0.419	0.403	0.465
337f	0.418	0.333	0.414	0.462	0.509	0.469	0.592	0.439	0.448	0.368	0.487	0.464	0.443	0.446
591f	0.680	0.654	0.692	0.826	0.760	0.765	0.830	0.804	0.765	0.680	0.706	0.673	0.698	0.614

[a] Previously banded as nestlings.

ilar in both years than were adults in general (1996, Mantel $t = 3.796$, $P < 0.05$; 1997, Mantel $t = 2.271$, $P < 0.05$). In addition, reproductively active females were no more genetically similar to one another than were random pairs of adults (1996, Mantel $t = -0.582$, $P > 0.05$; 1997, Mantel $t = -0.611$, $P > 0.05$).

DISCUSSION

RAPD analysis.—Available data suggest that in many taxa RAPD-PCR can easily generate hundreds of highly polymorphic and independent markers, the majority of which behave as neutral Mendelian alleles (Hadrys et al. 1992, Grosberg et al. 1996). In the present study, the RAPD technique proved to be a reliable method for evaluating polymorphism within this magpie population. We are aware of the limitations of RAPD-PCR technique. Being PCR-based, the principal limitations of RAPD fingerprinting arise from its sensitivity to reaction conditions (Hadrys et al. 1992), and slight changes in the conditions may affect the reproducibility of amplification products (Williams et al. 1990, Carlson et al. 1991). The technique is sensitive to (1) the shape of the temperature profile, (2) the type of polymerase used, (3) Mg^{2+} concentration, and (4) *Taq* or DNA concentration (Hadrys et al. 1992). In the lab, we optimized conditions and ran all the reactions in the same thermal cycler. Once the appropriate DNA dilution, the optimal polymerase concentration, and thermal cycles were selected, the random primers that could produce reproducible, readable bands on the gel were used to screen numerous individuals rapidly and relatively inexpensively without any prior DNA sequence information. This proved to be an important advantage of this technique for our study.

Another difficulty of the RAPD-PCR method is the possibility of comigration. An assumption of the use of the RAPD technique is that the amplified fragments are unique; that is, that the procedure does not amplify two distinct fragments that comigrate on gels because of similar

TABLE 4. Extended.

701m	599m	705m	381m	726f	513f	210f	461f	718f	713f	737f	563f	336f	337f	591f
0.690														
0.443	0.623													
0.410	0.667	0.400												
0.532	0.739	0.478	0.367											
0.429	0.673	0.345	0.246	0.273										
0.464	0.709	0.382	0.275	0.418	0.281									
0.423	0.773	0.447	0.455	0.707	0.520	0.520								
0.377	0.698	0.429	0.323	0.417	0.368	0.404	0.362							
0.313	0.559	0.290	0.343	0.509	0.387	0.387	0.321	0.258						
0.344	0.607	0.356	0.460	0.469	0.414	0.379	0.480	0.424	0.262					
0.375	0.643	0.288	0.375	0.520	0.356	0.424	0.400	0.356	0.292	0.323				
0.441	0.667	0.460	0.420	0.527	0.406	0.438	0.481	0.397	0.333	0.394	0.424			
0.484	0.593	0.509	0.460	0.673	0.552	0.483	0.500	0.509	0.429	0.567	0.533	0.375		
0.600	0.692	0.692	0.489	0.600	0.545	0.535	0.689	0.714	0.673	0.630	0.538	0.684	0.593	

size (Hadrys et al. 1992). We attempted to minimize that problem by using a combination of agarose and synergel instead of agarose gel alone, which greatly increased the resolution of band separation (Levitan and Grosberg 1993). The scored arrays of presence or absence of primer-specific amplified bands of particular length could be used to identify individuals as unique, thus allowing comparisons of genetic similarity among individuals (Grosberg et al. 1996, Horn et al. 1996).

The number of primer-specific bands amplified in our study was higher than that reported by others for other species (Huff et al. 1993, Horn et al. 1996). That is likely a reflection of the higher resolution of the combined agarose and synergel gel, rather than any inherent property of the DNA of Black-billed Magpies. The average genetic distances between all possible pairs of adult magpies (0.420 from 1996 data, and 0.428 from 1997 data) were greater than those reported by Horn et al. (1996) within reproductively active Magpie Geese (*Anseranas semipalmata*). That suggests there is more genetic diversity in our population of freely ranging adult birds than there was in the breeding goose population (Horn et al. 1996), or that the dispersal capabilities of Black-billed Magpies are greater than those in Magpie Geese.

Sibling relatedness.—In fall and following winter after fledging, there were always more after-hatching-year males than females among recaptured magpies (C. Trost pers. obs.), although that difference was not significant in our study. That could be due to female avoidance of concentrations of aggressive males, as demonstrated in a caging experiment (Trost and Webb 1997). However, significantly more hatching-year males from known nests than females were recaptured, which suggests that more females than male magpies dispersed from their natal area. Our molecular data confirmed that result, showing a higher genetic similarity among males than females. That is partially explained by the long lasting intraclutch dominance hierarchy that exists among

siblings. Young females and subordinate males are forced to disperse because of those hierarchies. A linear dominance-hierarchy exists among hatching-year male Black-billed Magpies (Trost and Webb 1997, Trost 1999), but not among females (Moholt 1989). Baeyens (1979) noticed this phenomenon among chicks from within one nest, and Trost and Webb (1997) extended it to nonrelated males among nests. Young males are highly aggressive when foraging, dominating in almost 75% of the observed interactions among hatching-year and adult magpies; they simply overwhelmed other magpies that were not part of their social hierarchy (Trost and Webb 1997). Komers and Komers (1992) also reported that juvenile male magpies dominate adults irrespective of sex or size differences during feeding. Dominant young males will also take risks around predators (Moholt and Trost 1989, Stone and Trost 1991), presumably to show off social status, or to gain social recognition (Zahavi 1995). A study on the Gray Jay (Strickland 1991) also showed that the "stayers" were dominant juveniles that forced out the "leavers," and about two-thirds of the stayers were males.

Because of those behaviors, young male magpies have increased access to territories and resources after the breeding season. Young subordinate males presumably disperse because they are unlikely to obtain a mate (or any other limited resources) in the presence of their dominant male siblings. Males might also disperse because there are more males than females in winter flocks (Trost and Webb 1997). If those dispersing subdominant males find a reliable food supply, such as a dead cow, they will establish a territory and breed (Knight 1988). However, males may rarely benefit from dispersing because they are likely to encounter territories with few females just as on their home territory. Dominant males (heaviest nestlings) tend to stay in their natal or nearby area (reflected from our recapture data, see Table 1) and wait for an opportunity to usurp a potential territorial opening. Thus, dominant sons are most likely individuals to be sampled with their fathers and their nearby brothers of lesser status, accounting for the increase in the relatedness among males in a given breeding population.

Fewer previously banded females than males were captured at our study area, which suggests that females disperse more often and over greater distances than males. Because female magpies are usually the limiting sex, more females breed in their first year than males (Birkhead 1991). Why then do most females disperse? Trost (1999) suggests that females are forced to leave their natal area due to aggression by their dominant male siblings. From our field observation and capture data, we found that after-hatching-year males are likely to accept females that are not their siblings in their flock. Emigration from the natal area may also be beneficial, because females would broaden their pool of prospective mates, thus increasing their chance of breeding. Finding a compatible mate, not just any mate, is an important determinant of an individual's fitness (Marzluff and Balda 1992, Black 1996). Our male-biased recapture data suggest that if most females came from some other population, they would likely establish themselves in flocks where the males were not their siblings.

The evolution of dispersal in birds and mammals has been investigated primarily using mark–recapture data or observations of marked animals (Koenig et al. 1996, Peacock 1997). An implicit assumption of such studies is that direct methods permit the researcher to collect an unbiased sample of measurements of dispersal distance. Use of molecular markers avoids that assumption and gives us an even clearer understanding of the relatedness of breeding Black-billed Magpies and a more accurate picture of the dynamics of genetic relatedness within the population. Our study confirms greater emigration by females, which suggests that male natal philopatry leads to female-biased dispersal in magpie breeding systems.

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