

## Host use strategies of individual female brown-headed cowbirds *Molothrus ater* in a diverse avian community

Bill M. Strausberger and Mary V. Ashley

Strausberger, B. M. and Ashley, M. V. 2005. Host use strategies of individual female brown-headed cowbirds *Molothrus ater* in a diverse avian community. – J. Avian Biol. 36: 313–321.

Although it is well established that brown-headed cowbirds *Molothrus ater* are host generalists, the parasitism strategies of individual females are not well documented. Here we use microsatellite genotyping to characterize host use by individual female cowbirds. Of 205 offspring sampled at a site in northeastern Illinois during one breeding season, we identified or inferred 33 mothers of 141 offspring, with up to 13 (mean 4.72) offspring assigned to individual females. Most (83%) females were host generalists and parasitized up to five host species, indicating that community-wide patterns of parasitism do not result from different individuals specializing on different host species. However, some females (17%) parasitized a single species, suggesting that alternative host-use strategies may exist within a single population. For host generalist females, individuals often parasitized host species that differed in body mass, nest-site placement, and quality, indicating that female cowbirds exhibit extremely flexible host ranges. However, female cowbirds consistently discriminated among potential hosts, for example, by completely avoiding some common, but unsuitable species.

B. M. Strausberger, Zoology Department, The Field Museum, 1400 S. Lake Shore Drive, Chicago IL 60605, USA. M. V. Ashley (correspondence), Department of Biological Sciences (MIC 066), University of Illinois at Chicago, 845 W. Taylor St., Chicago, IL 60607, USA. E-mail: ashley@uic.edu

Obligate brood parasites lay eggs in the nests of other species, relying on these surrogates to incubate eggs and provide parental care. Because potential host species vary greatly in quality (Strausberger and Ashley 1997, Sackmann and Carlos 2003), there may be strong selection for parasites to choose the best surrogate species for raising their offspring (Hamilton and Orians 1965). Host species vary in their physical and physiological capabilities of rearing parasites, and in the effectiveness of their behavioral defenses aimed at thwarting parasitism. The brown-headed cowbird *Molothrus ater* is an extreme host generalist that parasitizes more species than any other avian brood parasite. Although brown-headed cowbirds have been documented parasitizing 226 different host species, a much smaller number of host species regularly rear cowbirds and fewer yet are frequently parasitized (Friedmann 1963, Ortega 1998). Although much is known about host

species-specific parasitism rates, host use by individual female cowbirds remains poorly characterized. Female cowbirds are typically inconspicuous during parasitism and it is almost impossible to directly observe parasitism because there is no a priori way to know which nests will be parasitized. However, distinguishing between community-wide host use practices and that of individual females is critical. Community-wide patterns fail to reveal parasitism strategies of individual females, and whether distinct alternative strategies exist. The use of genetic markers to establish parent/offspring and sibling relationships provides an effective method to identify eggs of individual females, and thus reveal individual host use patterns. This approach has recently proven useful for cowbird studies (Alderson et al. 1999a, b, Hahn et al. 1999, Strausberger and Ashley 2003, Woolfenden et al. 2002, 2003, McLaren et al. 2003).

Here we characterize host use of individual female cowbirds in a diverse avian community located at The Morton Arboretum in northeastern Illinois through the use of DNA microsatellite markers to establish individual relatedness. In a previous study at this site, we described community-wide parasitism patterns, and demonstrated that cowbirds avoid certain species and species assemblages, such as cavity nesters and very large species (Strausberger and Ashley 1997). While microsatellites markers have been extensively used for parentage assignment, their high variability and codominant inheritance also makes it possible to infer sibling relationships among offspring even when parents have not been genotyped, at least in some instances (Emery et al. 2001, Smith et al. 2001, Strausberger and Ashley 2003). Reconstruction of maternally related sib groups allowed us to infer the parasitism behavior of individual females a posteriori, even for females that we did not physically sample. We relied more heavily on sibling group reconstruction than parentage assignment compared to most other cowbird genetic studies (e.g. Fleischer 1985, Alderson et al. 1999a, Hahn et al. 1999, but see Woolfenden et al. 2002, 2003, McLaren et al. 2003). The only other extensive study on host use by individual female cowbirds has been at Delta Marsh, Manitoba, Canada (Alderson et al. 1999a, Woolfenden et al. 2003). Our study site differs substantially from the Delta Marsh site because the avian community is much more diverse. Only three host species account for 97% parasitic eggs at the Manitoba site, whereas nearly a dozen hosts are regularly parasitized at The Morton Arboretum (Strausberger and Ashley 1997). Thus our study provides an important comparison for evaluating variation in individual host use patterns across varying host communities. We evaluate whether individual females at our site are host specific, host generalists, or exhibit intermediate host range strategies.

## Methods

### Sample collection

In 1995, we collected samples from cowbird eggs or nestlings (combined as "offspring") by conducting systematic searches for all bird nests located within an approximately 133 ha section of The Morton Arboretum in northeastern Illinois (Strausberger and Ashley 1997). Less than 100 ha provided habitat suitable for birds to nest, and these areas were extensively searched. The study site consists of a variety of habitats, including lawn, grassland, forest edge, and forest. To prevent biased sampling toward species having the easiest nests to spot, we used both visual and audio cues to identify breeding birds and their territory spacing patterns. The area was searched every three days. Approximately 1,550 total hours were spent in searching by BMS and two

field assistants. We increased our search efforts for species whose nests are more difficult to find such as ground nesters (Strausberger and Ashley 1997). All cowbird eggs and nestlings were removed from parasitized nests at the time of discovery. Previous work at this site suggested that there were approximately 60 to 70 actively laying female cowbirds in our study area (Strausberger and Ashley 2003). To maintain clutch composition, eggs were replaced with plaster-of-Paris eggs painted to resemble cowbird eggs.

We focused our sampling on offspring rather than adults to reduce the potential of influencing the breeding and parasitism behavior of adults at our site. However, we obtained a partial sample of adults by using baited Potter traps placed at different locations throughout the study site. Prior to releasing trapped adults, we drew blood and placed a unique color-combination of bands on their legs.

### Microsatellite analyses

DNA from offspring or adults was obtained from ~3–15  $\mu$ l of blood, plucked feather, or blood from shafts (Taberlet and Bouvet 1991), or the embryos from partially developed eggs. We incubated freshly laid eggs to promote tissue growth prior to DNA extraction (Strausberger and Ashley 2001). Eggs were dissected by using a razor blade to remove the top portion of the shell and a pipette was used to remove the embryo (Strausberger and Ashley 2001). Additional mothers were sampled from the residual DNA that remained on the surface of their egg shells (see Strausberger and Ashley 2001). We used six microsatellite primer pairs for genotyping; three loci (Ma $\mu$  10, Ma $\mu$  25, Ma $\mu$  29) are described in Alderson et al. (1999b), two loci (Ma $\mu$  101 and Ma $\mu$  104) are described in Strausberger and Ashley (2001), and one (Ma $\mu$  102) is described in Strausberger and Ashley (2003). The forward primer for each locus was fluorescently labeled and analyzed on an ABI 373-A DNA automated sequencer using GeneScan software (Applied Biosystems).

### Data Analysis

Allele frequencies, heterozygosities, tests of Hardy-Weinberg equilibrium, null allele calculations and exclusion probabilities were performed using CERVUS 1.0 (Marshall et al. 1998). Two different approaches, maternity assignment and sib-group reconstruction, were used both independently and together to reconstruct sets of offspring that belong to individual females. For parentage analysis, a likelihood approach implemented in CERVUS 1.0 was used to assign genotyped mothers to their putative offspring. The CERVUS program was developed for paternity assignment, but its use here to

assign maternal parents is a novel but appropriate application. Details of parentage analysis are given in Strausberger and Ashley (2003). Sib-group reconstruction was performed using a likelihood approach implemented using Kinship 1.2 (Goodnight and Queller 1999), also detailed in Strausberger and Ashley (2003). Comparing offspring genotypes with the Kinship program enabled us to determine full-sibling relationships among offspring without genotyping parents. When assigning offspring to full-sib groups, we required that the value of each pair-wise comparison of individual offspring have a  $\geq 0.99$  probability of being full-sibs. We then examined the genotypes of all members of putative sib groups to confirm relationships. When a maximum of four alleles was found across all loci, a full-sib relationship was assigned to the group. Some full-sibling groups may have shared a parent (and thus be half-sibs), but our data was insufficient to confidently infer half-sibling relationships unless one parent was genotyped. In such cases, full-sibling groups represent the minimum number of offspring for any given mother. However, in cases where we had two half-sib groups that shared a genotyped parent, we were often able to reconstruct the other two parental genotypes (Strausberger and Ashley 2003).

We defined host generalists and host specialists as female cowbirds that parasitized  $>1$ , or one host species, respectively. All results are given as mean  $\pm$  SD.

## Results

Despite our extensive field searches, some cowbird offspring undoubtedly went unsampled due to cowbird egg rejection and predation. Rejecter species were parasitized at the site (Strausberger and Ashley 1997, Strausberger and Burhans 2001), and depredation rates are high at our site (Schmidt and Whelan 1999). As a result, the reported cowbird parasitism frequencies and the number of offspring assigned to a female should be considered as minimum values (see also Strausberger 1998). During the 1995 cowbird breeding season, we found a total of 373 nests from 29 altricial species (Table 1). The most common hosts were indigo bunting *Passerina cyanea*, northern cardinal *Cardinalis cardinalis*, red-winged blackbird *Agelaius phoeniceus*, and song sparrow *Melospiza melodia*, while the nests of eastern bluebird *Sialia sialis*, barn swallow *Hirundo rustica*, mourning dove *Zenaida macroura*, and common grackle *Quiscalus quiscula* were never parasitized.

The six microsatellite loci used had from 7 to 27 alleles per locus, with gene diversity ( $H_e$ ) ranging from 0.53 to 0.92. The combined parental exclusion probability exceeded 0.99. None of the six loci showed significant deviations from Hardy-Weinberg expectations, and there was no significant linkage across loci.

A total of 205 cowbird offspring were found. Of these, 175 were successfully genotyped. We also captured, sampled, and genotyped eight adult female cowbirds, three of which were assigned offspring. We genotyped nine additional mothers from DNA we obtained from the shells of the eggs that they laid (Strausberger and Ashley 2001), making a total of 17 genotyped adult females, of which 12 were assigned offspring.

Using a combination of parentage assignment, sib-group assignment, and parental genotype reconstruction, we identified or inferred the mothers of 141 offspring (Table 2). Of these, 60 offspring were assigned to one of the 12 genotyped mothers. An additional 81 offspring could be assigned to another 21 'inferred' mothers based on sib analysis in combination with reconstruction of parental genotypes (for details see Strausberger and Ashley 2003). Assuming each full sibling group had a different mother, the combined maternity, sib-group analysis, and genotype reconstruction identified a maximum of 33 female cowbirds reproducing at our study site. Using a similar approach, we identified a maximum of 32 fathers, three from direct sampling and 29 'inferred' from reconstructed genotypes.

Female cowbirds had from 1–13 offspring each (mean  $4.72 \pm 2.83$ ). Twenty-nine of the 33 mothers had multiple offspring assigned to them and thus provided information on parasitism strategies of these individual females. Of the 29 mothers having more than one offspring, 24 (83%) parasitized multiple species ( $2.34 \pm 0.94$ , range 1–5; Table 2).

We examined several host characteristics to assess whether individual female cowbirds parasitize hosts that differ in their substantially in their attributes, or, alternatively, preferentially parasitize similar host species. Individual cowbirds parasitized hosts with body masses that varied three-fold, ranging from about 12 g for chipping sparrows *Spizella passerina* to 45 g for northern cardinals (Dunning 1993). Twelve (41.4%) females parasitized both hosts nesting on the ground, such as many song sparrows, field sparrows *Spizella pusilla* and rufous-sided towhees *Pipilo erythrophthalmus* as well as hosts that nested above the ground (Table 2). Ten (34.5%) females parasitized both acceptor and rejecter species (Tables 1 and 2). Together, our findings demonstrate that individual cowbirds frequently parasitize species that vary widely in their attributes.

Of the 29 females with multiple offspring, five (17%) were host specific, but only two of these were assigned more than two eggs each. The parasitic behavior of these two females, however, was interesting. Female "Unknown 12W" parasitized two field sparrow nests with two eggs each, and "Unknown 14W" parasitized four different red-winged blackbird nests with one egg each (Table 2). Mothers having multiple offspring ( $n=29$ ) parasitizing 1, 2, 3, 4, and 5 species had a mean ( $n$ ) or

Table 1. Nests and parasitism frequency for each species during the 1995 breeding season.

Species	Parasitized nests (n)	Total (%)	Cowbird offspring (n)
Northern cardinal <i>Cardinalis cardinalis</i>	33	46 (71.7)	60
Red-winged blackbird <i>Agelaius phoeniceus</i>	18	41 (43.9)	26
Indigo bunting <i>Passerina cyanea</i>	28	35 (80.0)	49
Song sparrow <i>Melospiza melodia</i>	15	20 (75.0)	26
Orchard oriole <i>Icterus spurius</i>	3	3 (100)	4
House finch <i>Carpodacus mexicanus</i>	4	6 (66.7)	4
Rufous-sided towhee <i>Pipilo erythrophthalmus</i>	2	2 (100)	2
Eastern bluebird <i>Sialia sialis</i>	0	10 (0)	0
Common grackle <i>Quiscalus quiscula</i>	0	10 (0)	0
Mourning dove <i>Zenaida macroura</i>	0	20 (0)	0
Barn swallow <i>Hirundo rustica</i>	0	13 (0)	0
House wren <i>Troglodytes aedon</i>	0	9 (0)	0
Tree swallow <i>Tachycineta bicolor</i>	0	3 (0)	0
Black-capped chickadee <i>Parus atricapillus</i>	0	3 (0)	0
Bobolink <i>Dolichonyx oryzivorus</i>	0	5 (0)	0
Eastern meadowlark <i>Sturnella magna</i>	0	5 (0)	0
Grasshopper sparrow <i>Ammodramus saviarum</i>	0	4 (0)	0
Red-eyed vireo <i>Vireo olivaceus</i>	6	6 (100)	7
Common yellowthroat <i>Geothlypis trichas</i>	0	1 (0)	0
Field sparrow <i>Spizella pusilla</i> <sup>a</sup>	6	14 (42.9)	10
Chipping sparrow <i>Spizella passerina</i> <sup>b</sup>	3	11 (27.3)	4
Yellow warbler <i>Dendroica petechia</i> <sup>c</sup>	0	1 (0)	0
Brown thrasher <i>Toxostoma rufum</i> <sup>d</sup>	2	11 (18.2)	3
Gray catbird <i>Dumetella carolinensis</i> <sup>d</sup>	4	25 (16.0)	7
Baltimore oriole <i>Icterus galbula</i> <sup>d</sup>	2	7 (28.6)	3
Blue jay <i>Cyanocitta cristata</i> <sup>d</sup>	0	8 (0)	0
Eastern kingbird <i>Tyrannus tyrannus</i> <sup>d</sup>	0	2 (0)	0
Cedar waxwing <i>Bombycilla cedrorum</i> <sup>d</sup>	0	1 (0)	0
American robin <i>Turdus migratorius</i> <sup>d</sup>	0	51 (0)	0
Total	126	373 (33.8)	205

Species that frequently reject parasitism <sup>a</sup>(Strausberger and Burhans 2001), <sup>b</sup>(Ortega and Ortega 2001, Strausberger unpubl. data), <sup>c</sup>(Sealy 1996), <sup>d</sup>(Rothstein 1975).

total of  $2.8 \pm 1.10$  (5),  $3.83 \pm 1.47$  (12),  $5.8 \pm 3.08$  (10), 6 (1), and 13 (1) offspring each, respectively. Across these mothers, there was a significant positive correlation between the number of offspring produced and the number of host species used ( $r_s = 0.523$ ,  $n = 29$ ,  $P = 0.005$ ).

## Discussion

Although cowbirds as a species are known to be host generalists, there has been very little information available regarding host use by individual females. By identifying the mothers or, alternatively, full-siblings offspring groups, we were able to ascertain individual host-use strategies. This is in contrast to the large number of field studies reporting parasitism frequencies on different avian hosts by the cowbird population as a whole. Here, we have focused on the question of host choice by *individual* females.

Most individual cowbirds (83%) at our study site parasitized multiple species, indicating that host-generalism is the most common parasitism strategy. This value is similar to that reported by Woolfenden et al. (2003), 77%, at Delta Marsh. Consequently, community-wide patterns of cowbird parasitism do not generally result from individuals specializing on different host species. A

previous study found a lack of genetic differentiation among cowbirds raised by different host species (Gibbs et al. 1997), but such differentiation would require host specialization persisting on an evolutionary time scale. Our findings indicate that even over the course of a single breeding season, female cowbirds generally do not limit parasitism to a single host species. Not only did individual females typically parasitize multiple species, they were often generalists with regard to host's size, nesting site, and quality as surrogate parent. Our study covered only one breeding season, but Woolfenden et al. (2003) report that cowbirds also use different host species in different years. Thus flexibility in cowbird host use appears to be the common pattern, and may in part explain the success of cowbirds in exploiting temporally and spatially varying host communities.

We found only two females that laid more than two eggs each into nests of a single host species, but they raise the possibility of host specialization by a minority of female cowbirds at our site. One female (Unknow 14W, Table 1) parasitized red-winged blackbirds in two habitat patches separated by more than 1 km. The other individual (Unknown 12W) parasitized only field sparrows, a species that deserts all, or nearly all, early-stage nests following an encounter with an adult cowbird (Strausberger and Burhans 2001). This particular individual seemingly avoided detection and had all four of

Table 2. Results of relatedness analysis and host use of female brown-headed cowbirds.

Mother	Offspring	Host species <sup>a</sup>	Nest placement <sup>b</sup>
<b>Maternity assignments to genotyped females</b>			
5910BW2-S <sup>a</sup>	5263B	Northern cardinal	Above ground
	763B	Northern cardinal	Above ground
	672C	Indigo bunting	Above ground
	6262A	Northern cardinal	Above ground
	5910B	Northern cardinal	Above ground
	5101B	Northern cardinal	Above ground
5112AW-S	51715B	Northern cardinal	Above ground
	5101C	Northern cardinal	Above ground
	5112A	Northern cardinal	Above ground
	51714A	Song sparrow	Ground
	51714B	Song sparrow	Ground
	5259A	Northern cardinal	Above ground
5111AW-S	5263A	Northern cardinal	Above ground
	6191C	Indigo bunting	Above ground
	5111A	Northern cardinal	Above ground
75W-B <sup>b</sup>	596B	Northern cardinal	Above ground
	5125A	Northern cardinal	Above ground
593CW-S	688A	Northern cardinal	Above ground
	688B	Northern cardinal	Above ground
	754C	Indigo bunting	Above ground
	593C	Northern cardinal	Above ground
6293W-B	672B	Indigo bunting	Above ground
	751A	Rufus-sided towhee	Ground
	5124A	Northern cardinal	Above ground
	5166A	Northern cardinal	Above ground
	6261A	Indigo bunting	Above ground
594AW-S	762A	Indigo bunting	Above ground
	763A	Northern cardinal	Above ground
	594A	Song sparrow	Above ground
	594B	Song sparrow	Above ground
	626B	Red-winged blackbird	Above ground
766W-B	6222A	Red-winged blackbird	Above ground
	52610B	Northern cardinal	Above ground
	599A	Northern cardinal	Above ground
	52610A	Northern cardinal	Above ground
	5312B	Red-winged blackbird	Above ground
	694A	Indigo bunting	Above ground
	6113A	House finch	Above ground
	6115A	Indigo bunting	Above ground
	6131C	Indigo bunting	Above ground
	6171A	Indigo bunting	Above ground
	761A	Northern cardinal	Above ground
	775A	Song sparrow	Ground
	7115A	Northern cardinal	Above ground
	7183B	Indigo bunting	Above ground
	5910BW1-S	5191A	Orchard oriole
5192A		Red-winged blackbird	Above ground
5198A		Red-winged blackbird	Above ground
5269A		Red-winged blackbird	Above ground
6206A		Orchard oriole	Above ground
5910AW1-S	5910A	Northern cardinal	Above ground
	5168A	Song sparrow	Ground
	7182A	Field sparrow	Above ground
	7182C	Field sparrow	Above ground
	6131A	Indigo bunting	Above ground
	7183A	Indigo bunting	Above ground
5233AW-S	5233A	Song sparrow	Ground
	615A	Northern cardinal	Above ground
	678A	Indigo bunting	Above ground
594CW-S	594C	Song sparrow	Above ground
<b>Mothers inferred by sib-group analysis</b>			
Unknown 1W	51711B	Song sparrow	Ground
	51711C	Song sparrow	Ground
	754B	Indigo bunting	Above ground
Unknown 2W	593A	Northern cardinal	Above ground
	593B	Northern cardinal	Above ground
Unknown 3W	673A	Brown thrasher	Above ground
	673B	Brown thrasher	Above ground
	6112C	Northern cardinal	Above ground

Table 2 (Continued)

Mother	Offspring	Host species <sup>a</sup>	Nest placement <sup>b</sup>
Unknown 4W	753B	Gray catbird	Above ground
Unknown 5W	6113B	House finch	Above ground
Unknown 6W	5254B	Red-winged blackbird	Above ground
	5312A	Red-winged blackbird	Above ground
	6912A	Northern cardinal	Above ground
	6213B	Red-winged blackbird	Above ground
	6221A	Red-winged blackbird	Above ground
Unknown 7W	6222B	Red-winged blackbird	Above ground
Unknown 8W	5171C	Song sparrow	Ground
	677A	Indigo bunting	Above ground
	677B	Indigo bunting	Above ground
	6813A	Song sparrow	Ground
	6918D	Indigo bunting	Above ground
	6192A	Indigo bunting	Above ground
	6202A	Indigo bunting	Above ground
	6282A	Song sparrow	Ground
	711A	Indigo bunting	Above ground
	771A	Chipping sparrow	Above ground
	Unknown 9W	651A	Indigo bunting
661A		Indigo bunting	Above ground
661B		Indigo bunting	Above ground
672A		Indigo bunting	Above ground
6153A		Indigo bunting	Above ground
6191A		Indigo bunting	Above ground
6191B		Indigo bunting	Above ground
6193A		Rufus-sided towhee	Above ground
6201B		Orchard oriole	Above ground
711B		Indigo bunting	Above ground
7141A		Indigo bunting	Above ground
Unknown 10W	5265A	Northern cardinal	Above ground
	623A	Song sparrow	Ground
	623B	Song sparrow	Ground
Unknown 11W	599B	Northern cardinal	Above ground
	5112B	Northern cardinal	Above ground
	5155A	Northern cardinal	Above ground
	5237A	Northern cardinal	Above ground
	5259C	Northern cardinal	Above ground
	6910A	House finch	Above ground
	6911A	Chipping sparrow	Above ground
6911B	Chipping sparrow	Above ground	
Unknown 12W	5123A	Field sparrow	Ground
	5123B	Field sparrow	Ground
	689A	Field sparrow	Above ground
	689B	Field sparrow	Above ground
Unknown 13W	5245B	Northern cardinal	Above ground
	775B	Song sparrow	Ground
	7182B	Field sparrow	Above ground
Unknown 14W	5122A	Red-winged blackbird	Above ground
	5141A	Red-winged blackbird	Above ground
	51712A	Red-winged blackbird	Above ground
	52616A	Red-winged blackbird	Above ground
Unknown 15W	52610C	Northern cardinal	Above ground
	6152C	Indigo bunting	Above ground
	6284A	Northern cardinal	Above ground
Unknown 16W	5179A	Gray catbird	Above ground
	5231A	Northern cardinal	Above ground
	5245A	Northern cardinal	Above ground
	52610D	Northern cardinal	Above ground
	682A	Indigo bunting	Above ground
Unknown 17W	753A	Gray catbird	Above ground
	51711A	Song sparrow	Ground
	754A	Indigo bunting	Above ground
Unknown 18W	5259B	Northern cardinal	Above ground
	678B	Indigo bunting	Above ground
	6201A	Orchard oriole	Above ground
Unknown 19W	51715C	Northern cardinal	Above ground
	6217A	Baltimore oriole	Above ground
Unknown 20W	684A	Indigo bunting	Above ground
	6131B	Indigo bunting	Above ground

Table 2 (Continued)

Mother	Offspring	Host species <sup>a</sup>	Nest placement <sup>b</sup>
Unknown 21W	752A	Field sparrow	Ground
	753C	Gray catbird	Above ground
	5173A	Red-winged blackbird	Above ground
	5173B	Red-winged blackbird	Above ground

<sup>a</sup>S indicates that the maternal DNA was extracted from eggshells.

<sup>b</sup>B indicates that maternal DNA was extracted from blood samples of captured cowbirds.

her eggs accepted by field sparrows. However, findings from the Delta Marsh study (Woolfenden et al. 2003) raise caution against inferring host specialization from small samples or from single seasons. There, for example, one female laid all of her 13 eggs in redwing blackbird nests during one season, but parasitized another host in a subsequent year.

Incomplete sampling appears to strongly influence conclusions regarding cowbird breeding behavior and parasitism and thus raises a cautionary note for drawing conclusions from limited sampling. For example, Alderson et al. 1999a reported that at Delta Marsh, approximately half the female cowbirds parasitized a single host, and that most individuals were monogamous. However, more extensive sampling of adults at Delta Marsh revealed much higher levels of host generalism (Woolfenden et al. 2003). Although our study covers only one breeding season, we assigned a relatively high number of eggs to individual females (4.7) and our results should provide increased confidence in the conclusions regarding host generalism in cowbirds.

The parasitism patterns we observed suggest that cowbirds' host discrimination behavior is largely adaptive. Cowbirds completely avoided four species that are likely unsuitable hosts, mourning doves, barn swallows, common grackles and eastern bluebirds. Mourning doves and barn swallows have special structural and behavioral adaptations for food exchange between adult and nestlings that likely prevent successful rearing of cowbirds (Friedmann 1929, 1963, Wiley 1988). Common grackles are poor cowbird hosts due mainly to their large size (Peer and Bolinger 1998). Although there has been at least one report of eastern bluebirds rearing cowbirds (Woodward 1979), all bluebird nests we found were placed in cavities. At our study site, cowbirds consistently avoided nests in cavities, even though we determined that cowbirds could fit through nest-entrance holes (Strausberger and Ashley 1997). Cavity nesting may discourage cowbird parasitism for a variety of reasons (Friedmann 1963, Kattan 1997).

Considering that most individual female cowbirds were host generalists, they 'misaid' a remarkably low proportion of eggs in nests of unsuitable hosts, suggesting an innate ability to recognize such species. If host avoidance were a learned behavior, we would expect some parasitism of unsuitable species by inexperienced individuals. It is unlikely that unsuitable host species

completely prevented parasitism because even highly aggressive species are often successfully parasitized (Strausberger and Horning 1998, Strausberger 2001). Female cowbirds consistently and nearly completely avoid such unsuitable host species such as mourning doves, common grackles and barn swallows throughout their range (Friedmann 1963, Friedmann et al. 1977, Strausberger and Ashley 1997, Peer and Bollinger 1997, Peer and Bollinger 1998). On the other hand, the occasional parasitism of house finches is a case that appears to be maladaptive and possibly reflects limitations in cowbirds' host avoidance. The seed diet of the house finch is unsuitable for rearing cowbirds (Kozlovic et al. 1996). However, cowbirds and house finches *Carpodacus mexicanus* were, until relatively recently, allopatric in the eastern United States, raising the possibility that insufficient time has passed for avoidance behavior to have evolved or spread through the cowbird population (Kozlovic et al. 1996).

Another finding that seemingly runs counter to the adaptive nature of host-choice was parasitism of species that frequently reject parasitism and thus are apparently unsuitable hosts. All five rejector species that were parasitized at our site occasionally to frequently accepted cowbird eggs (Strausberger and Burhans 2001, BMS unpubl. data). Unfortunately, data on overall rates of parasitism of rejectors (and thus rates of acceptance) is difficult to obtain. Parasitism of rejector species may be advantageous to cowbirds adept at defeating host-rejection defense (e.g. Unknown 12W), or cowbirds skilled at identifying host pairs more likely to accept parasitism. Categorical classification of 'rejector' species as poor cowbird hosts appears over-simplistic as it fails to consider individual variation within a species.

In contrast to the lack of parasitism on unsuitable hosts, we found that the parasitism rates of frequently used host species ranged widely (43.9–80.0%). Such large variation of parasitism rates for common hosts at different sites has been reported elsewhere (Robinson et al. 1995). Such variation is consistent with a learned or environmental component to cowbirds' host selection. Parasitism of newly available species indicates that cowbirds' host selection strategy enables them to quickly adapt to disturbed or novel habitats compared to more Neophobic specialists (see Greenberg 1984, Altshuler and Nunn 2001, Warren et al. 2001). Cowbirds have greatly benefited from human-induced landscape

changes following European settlement, increasing in number and expanding their range, and successfully parasitizing previously allopatric species (Robinson et al. 1995). Our study demonstrates that this can be attributed, in large part, to remarkably flexible and adaptive host use by individual female cowbirds.

*Acknowledgements* – This work was supported in part by a grant from the National Science Foundation. We thank Kevin A. Feldheim, Kenneth L. Jones, and Michael S. Webster for their assistance and suggestions in the lab. David E. Springer's editorial suggestions added greatly to manuscript preparation. Additionally, comments from Joel S. Brown, Christopher J. Whelan, Scott K. Robinson, and Douglas F. Stotz enhanced the manuscript greatly. We thank The Morton Arboretum, especially Christopher P. Dunn and Dave Cascarano, for access and assistance in the field. This work was conducted in part while M.V.A. was a Sabbatical Fellow at the National Center for Ecological Analysis and Synthesis, a Center funded by National Science Foundation, the University of California, and the Santa Barbara campus. This research was completed in partial fulfillment of the requirements for the doctoral degree (to BMS) at the Graduate College of the University of Illinois at Chicago.

## References

- Alderson, G. W., Gibbs, H. L. and Sealy, S. G. 1999a. Determining the reproductive behavior of individual brown-headed cowbirds using microsatellite DNA markers. – *Anim. Behav.* 58: 895–905.
- Alderson, G. W., Gibbs, H. L. and Sealy, S. G. 1999b. Parentage and kinship analyses in an obligate brood parasitic bird, the brown-headed cowbird (*Molothrus ater*) using microsatellite DNA markers. – *J. Hered.* 90: 182–190.
- Altshuler, D. L. and Nunn, A. M. 2001. Observational learning in hummingbirds. – *Auk* 118: 795–799.
- Dunning, J. B. 1993. *CRC Handbook of avian body masses*. – CRC Press, Boca Raton, Florida.
- Emery, A. E., Wilson, I. J., Craig, S., Boyle, P. R. and Noble, L. R. 2001. Assignment of paternity groups without access to parental genotypes: multiple mating and developmental plasticity in squid. – *Mol. Ecol.* 10: 1265–1278.
- Fleischer, R. C. 1985. A new technique to identify and assess the dispersion of eggs of individual brood parasites. – *Behav. Ecol. Sociobiol.* 17: 91–99.
- Friedmann, H. 1929. The cowbirds: a study of the biology of social parasitism. – Charles C. Thomas, Springfield.
- Friedmann, H. 1963. Host relations of the parasitic cowbirds. – *Bull. U.S. Natl. Mus.* 223: 1–196.
- Friedmann, H., Kiff, L. F. and Rothstein, S. I. 1977. A further contribution to knowledge of the host relations of the parasitic cowbirds. – *Smithson. Contrib. Zool.* 235: 1–75.
- Gibbs, H. L., Miller, P., Alderson, G. W. and Sealy, S. G. 1997. Genetic analysis of brown-headed cowbirds *Molothrus ater* raised by different hosts: data from mtDNA and microsatellite DNA markers. – *Mol. Ecol.* 6: 189–193.
- Goodnight, K. F. and Queller, D. C. 1999. Computer software for performing likelihood tests of pedigree relationship using genetic markers. – *Mol. Ecol.* 8: 1231–1234.
- Greenberg, R. 1984. Differences in feeding neophobia in the tropical migrant wood warblers *Dendroica castanea* and *D. pennsylvanica*. – *J. Comp. Physiol.* 98: 131–136.
- Hahn, D. C., Sedgwick, J. A., Painter, I. S. and Casna, N. J. 1999. A spatial and genetic analysis of cowbird host selection. – *Stud. Avian Biol.* 18: 204–217.
- Hamilton, W. J. and Orrians, G. H. 1965. Evolution of brood parasitism in altricial birds. – *Condor* 67: 361–382.
- Kattan, G. H. 1997. Shiny cowbirds follow the “shotgun” strategy of brood parasitism. – *Anim. Behav.* 53: 647–654.
- Kozlovic, D. R., Knapton, R. W. and Barlow, J. C. 1996. Unsuitability of the house finch as a host of the brown-headed cowbird. – *Condor* 98: 253–258.
- Marshall, T. C., Slate, J., Kruuk, L. E. B. and Pemberton, J. M. 1998. Statistical confidence for likelihood-based paternity inference in natural populations. – *Mol. Ecol.* 7: 639–655.
- McLaren, C. M., Woolfenden, B. E., Gibbs, H. L. and Sealy, S. G. 2003. Genetic and temporal patterns of multiple parasitism by brown-headed cowbirds (*Molothrus ater*) on song sparrows (*Melospiza melodia*). – *Can. J. Zool.* 81: 281–286.
- Ortega, C. P. 1998. Cowbirds and other brood parasites. – University of Arizona Press, Tucson.
- Ortega, C. P. and Ortega, J. C. Effects of brown-headed cowbirds on the nesting success of chipping sparrows in southwest Colorado. – *Condor* 103: 127–133.
- Peer, B. D. and Bolinger, E. K. 1997. Explanations for the infrequent cowbird parasitism on common grackles. – *Condor* 99: 151–161.
- Peer, B. D. and Bolinger, E. K. 1998. Rejection of cowbird eggs by mourning doves: a manifestation of nest usurpation? – *Auk* 115: 1057–1062.
- Robinson, S. K., Rothstein, S. I., Brittingham, M. C., Petit, L. J. and Grzybowski, J. A. 1995. Ecology and behavior of cowbirds and their impact on host populations. – In: Martin, T. E. and Finch, D. M. (eds). *Ecology and management of neotropical migratory birds*. Oxford University Press, New York, pp. 428–460.
- Rothstein, S. I. 1975. An experimental and teleonomic investigation of avian brood parasitism. – *Condor* 77: 250–271.
- Sackmann, P. and Rebores, J. C. 2003. A comparative study of shiny cowbird parasitism of two large hosts, the chalk-browed mockingbird and the rufous-bellied thrush. – *Condor* 105: 728–736.
- Schmidt, K. A. and Whelan, C. J. 1999. Effects of exotic *Loicera* and *Rhammas* on songbird nest predation. – *Cons. Biol.* 13: 1502–1506.
- Sealy, S. G. 1996. Evolution of host defenses against brood parasitism: implications of puncture-ejection by a small passerine. – *Auk* 113: 346–355.
- Smith, B. R., Herbinger, C. M. and Merry, H. R. 2001. Accurate partition of individuals into full-sib families from genetic data without parental information. – *Genetics* 158: 1329–1338.
- Strausberger, B. M. 1998. Temporal patterns of host availability, brown-headed cowbirds parasitism, and parasite egg mass. – *Oecologia* 116: 267–274.
- Strausberger, B. M. 2001. The relationship of habitat and spatial distribution of nests with brown-headed cowbird parasitism of red-winged blackbirds. – *Wilson Bull.* 113: 129–133.
- Strausberger, B. M. and Ashley, M. V. 1997. Community-wide patterns of parasitism of a host “generalist” brood-parasitic cowbird. – *Oecologia* 112: 254–262.
- Strausberger, B. M. and Ashley, M. V. 2001. Eggs yield nuclear DNA from egg-laying females, their embryos and offspring. – *Conserv. Genet.* 2: 385–390.
- Strausberger, B. M. and Ashley, M. V. 2003. Breeding biology of brood parasitic cowbirds characterized by parent-offspring and sib-group reconstruction. – *Auk* 120: 433–445.
- Strausberger, B. M. and Burhans, D. E. 2001. Nest desertion by field sparrows and its possible influence on the evolution of cowbird behavior. – *Auk* 118: 770–776.
- Strausberger, B. M. and Horning, M. E. 1998. Response of nesting song sparrows (*Melospiza melodia*) and red-winged blackbirds (*Agelaius phoeniceus*) to models of parasitic cowbirds and nonthreatening towhees. – *Bird Behav.* 12: 71–78.
- Taberlet, P. and Bouvet, J. 1991. A single plucked feather as a source of DNA for bird genetic studies. – *Auk* 108: 959–960.

- Warren, M. S., Hill, J. K., Thomas, J. A., Asher, J., Fox, R., Huntley, B., Roy, D. B., Telfer, M. G., Jeffcoate, S., Harding, P., Jeffcoate, G., Willis, S. G., Greatorex-Davies, J. N., Moss, D. and Thomas, C. D. 2001. Rapid response of British butterflies to opposing forces of climate and habitat change. – *Nature* 414: 65–69.
- Wiley, J. W. 1988. Host selection by the shiny cowbird. – *Condor* 90: 289–303.
- Woolfenden, B. E., Gibbs, H. L. and Sealy, S. G. 2002. High opportunity for sexual selection in both sexes of an obligate brood parasitic bird, the brown-headed cowbird (*Molothrus ater*). – *Behav. Ecol. Sociobiol.* 52: 417–425.
- Woolfenden, B. E., Gibbs, H. L., Sealy, S. G. and McMaster, D. G. 2003. Host use and fecundity of individual female brown-headed cowbirds. – *Anim. Behav.* 66: 95–106.
- Woodward, P. W. 1979. Brown-headed cowbird parasitism on eastern bluebirds. – *Wilson Bull.* 91: 321–322.
- (Received 6 October 2003, revised 2 June 2004, accepted 8 August 2004.)