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Shiny cowbirds share foster mothers but not true mothers in multiply parasitized mockingbird nests

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Abstract Obligate brood parasitic birds, such as cowbirds, evade parental care duties by laying their eggs in the nests of other species. Cowbirds are assumed to avoid laying repeatedly in the same nest so as to prevent intrabrood competition between their offspring. However, because searching for host nests requires time and energy, laying more than one egg per nest might be favoured where hosts are large and can readily rear multiple parasites per brood. Such ‘repeat parasitism’ by females would have important consequences for parasite evolution because young parasites would then incur indirect fitness costs from behaving selfishly. We investigated shiny cowbird (*Molothrus bonariensis*) parasitism of a large host, the chalk-browed mockingbird (*Mimus saturninus*), in a population where over 70 % of the parasitized mockingbird nests receive multiple cowbird eggs. We assessed egg maternity directly, using cameras at nests to film the laying of individually-marked females. We also supplemented video data with evidence from egg morphology, after confirming that each female lays eggs of a consistent appearance. From 133 eggs laid, we found that less than 5 % were followed by the same female visiting the nest to lay again or to puncture eggs. Multiple eggs in mockingbird nests were instead the result of

different females, with up to eight individuals parasitizing a single brood. Thus, while cowbird chicks regularly share mockingbird nests with conspecifics, these are unlikely to be their maternal siblings. Our results are consistent with shiny cowbird females following a one-egg-per-nest rule, even where hosts can rear multiple parasitic young.

Keywords Brood parasitism · Egg morphology · *Mimus saturninus* · *Molothrus bonariensis* · Multiple parasitism · Parental investment · Repeat parasitism

Introduction

Obligate brood parasitic birds do not rear their own offspring but instead lay their eggs in the nests of other species, which thereafter provide all the necessary parental care on the parasite’s behalf. The most important strategic investment that female parasites make in their offspring lies therefore in their decision of where and when to deposit each egg. This includes selection of a suitable host species (Teuschl et al. 1998; Hahn et al. 1999; Langmore and Kilner 2007) and host nest (Soler et al. 1995; Polačiková et al. 2008; Fiorini et al. 2009a; Soler and Pérez-Contreras 2012), and the timing of laying to coincide with the host’s laying period (Davies and Brooke 1988; Ellison et al. 2006; Moskat et al. 2006; Fiorini et al. 2009b). In addition, it is often suggested that females should avoid laying again in nests in which they have already laid (Strausberger and Ashley 2003; Hahn et al. 1999; Trine 2000; Hoover 2003; McLaren et al. 2003; Moskat et al. 2006; Goguen et al. 2011). This is certain to be beneficial in those systems in which the first parasite chick to hatch kills all other young in the nest (e.g. many cuckoos, Davies 2000) or where hosts cannot provide sufficient food to rear more than one parasite at a time (e.g. cowbirds, *Molothrus* sp., using small-bodied hosts, Goguen et al. 2011). It is less clear, however, whether a strict

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one-egg-per-nest rule will be used where multiple parasites can be reared in a single-host brood (Payne 1977; Trine 2000; Rivers et al. 2003). In such cases, local competition between a female's offspring may be less costly than the time and energy required to locate and monitor additional host nests and so favour laying repeatedly in the same nest (hereafter, 'repeat parasitism').

Understanding if and when parasites engage in repeat parasitism is important for understanding not only the trade-offs shaping female-laying strategies but also the behaviour of parasites at the nestling stage (Rivers et al. 2012). Parasite nestlings that share the nest with their siblings or half-siblings would incur indirect fitness costs if they harm their nestmates in the same way as nestlings of non-parasitic species do (Godfray 1995). This could influence the evolution of both begging behaviours (Briskie et al. 1994; Dearborn and Lichtenstein 2002; Hauber and Ramsey 2003; Lichtenstein and Dearborn 2004) and nestling virulence (Kilner et al. 2004; Kilner 2005; Gloag et al. 2012a). For example, relatedness between parasite young in the same nest could help explain why some parasitic species do not show aggression towards their nestmates, despite the potential benefits of eliminating them as competition (Kilner 2005).

In this study, we investigated whether the brood parasitic shiny cowbird (*M. bonariensis*) engages in repeat parasitism of a common host, the chalk-browed mockingbird (*Mimus saturninus*). The shiny cowbird-mockingbird system is an interesting one in which to investigate this aspect of female-laying behaviour for several reasons. First, parasitism intensities of mockingbird nests are very high throughout Argentina, where they have been most studied (Hudson 1874; Salvador 1984; Fraga 1985; Mason 1986; Sackmann and Reboreda 2003; Fiorini et al. 2009b). At the site of the present study, over 70 % of all the parasitized nests are done so multiply, with an average of three eggs per nest (Gloag et al. 2012b). Egg distribution indicates that a nest's existing parasite egg load does not influence the likelihood of receiving further parasite eggs (Gloag et al. 2012b), but it is unknown whether one or more females are responsible for eggs in the same nest. Secondly, intrabrood competition between a female's offspring is relatively low when parasitizing mockingbirds. Shiny cowbirds are generalists that use hosts that vary in size from smaller to larger than themselves (adult body masses: hosts 12–80 g, cowbird 40–45 g, Friedmann and Kiff 1985). While small hosts can struggle to rear more than one cowbird per brood, larger hosts generally do not (Post and Wiley 1977; Fraga 1985; Mermoz and Reboreda 1994; Trine 2000). Chalk-browed mockingbirds are amongst the largest of the shiny cowbird's hosts (70–75 g) and are capable of fledging multiple parasites from the same nest (Salvador 1984; Fraga 1985), provided that not all of their own eggs survive to hatch (Fiorini et al. 2009b). There could though be a different drawback for repeat parasitism in this system. Female cowbirds reduce their

host's clutches by puncturing holes in eggs when visiting nests (Gloag et al. 2013), so any female returning to a nest in which she had previously laid would need to either selectively target only the eggs of mockingbirds and other cowbirds when puncturing or risk destroying her own egg or eggs.

In the brown-headed cowbird (*Molothrus ater*), another generalist parasite that uses hosts both larger and smaller than itself, the incidence of repeat parasitism has been reported to vary between populations (Alderson et al. 1999; Hahn et al. 1999; McLaren et al. 2003; Ellison et al. 2006; Rivers et al. 2012) from negligible (Hahn et al. 1999) to high (over 40 % of the parasitized nests containing multiple eggs of the same female, Rivers et al. 2012). Such variation is consistent with female nest-use strategy being flexible and responding to the characteristics of the host population, though whether host size or host species is important remains unclear. These previous studies assessed cowbird egg maternity by genetic analyses, usually by sampling egg tissue (Alderson et al. 1999; Hahn et al. 1999; McLaren et al. 2003; Ellison et al. 2006; Rivers et al. 2012), but we use, for the first time, a direct method that avoids both destroying the eggs and the risk of overlooking maternal half-siblings. We positioned infrared microcameras above nests and filmed egg laying by individually marked female cowbirds. These video data were supplemented with comparisons of egg morphology of cowbird eggs in the same nest. At the population level, shiny cowbird eggs are highly variable in size, shape, background colour, spot colour, and spotting pattern, but individual females have long been proposed to lay eggs of a consistent appearance (Hudson 1874; Lyon 1997). Using sets of eggs from our marked females, we tested this assumption and subsequently referred to egg morphology when female identity could not be established from video recordings (15 % of the total eggs). We also examined the likelihood that observed rates of repeat parasitism arise from active preferences by females, rather than by chance.

A note on terminology

In studies of brood parasitic birds, multiple parasitism is widely used to refer to the laying of two or more eggs in a single host nest, but, as yet, there is no consensus term to refer specifically to multiple parasitism by the same female. We use 'repeat parasitism' in this study, as we feel it is the most apt and concise of the terms available. It should be noted, however, that some studies of avian brood parasitism have used 'repeated parasitism' to refer to a different occurrence: the parasitism of multiple nesting attempts of a given host pair (i.e. parasitism that is repeated from the host's perspective rather than the parasite's (Hauber et al. 2004; Hoover et al. 2006).

Methods

Trapping and marking

This study was conducted in pampas grasslands at Reserva El Destino, Buenos Aires Province, Argentina (35°08'S, 57°23' W) during the 2010–2011 breeding season (October–January). We trapped 55 female cowbirds during spring–summer 2011, using walk-in funnel traps baited with millet. We banded cowbirds with unique colour-ring combinations but found that ring colours were difficult to distinguish in video recordings made during the low light of pre-sunrise when cowbird egg laying occurs (Gloag et al. 2013). Thus, each cowbird was also given a unique head mark by decolouring the distal end of head feathers with hair bleach to facilitate individual identification, as shown in Fig. 1 and the video Online Resource 1. We hereafter refer to these birds as 'painted females'. Re-trappings and video data of painted females at nests indicated that head marks persisted for 12 weeks of the study. In addition, a number of cowbirds ringed in previous years were active at the site, such that



Fig. 1 Examples of three 'painted' female shiny cowbirds, which were individually marked by dyeing the distal 5 mm of head feathers blond. Each female was given a unique pattern, allowing her identification from the infrared nest cam footage

the presence/absence of rings could sometimes be used to distinguish between two unpainted females.

Nest filming and monitoring

We located mockingbird nests by searching trees within known breeding territories. Mockingbirds lay a clutch of four eggs, and most egg laying by cowbirds coincides with mockingbird laying (Fiorini et al. 2009b). At 125 nests located during or prior to mockingbird laying, we suspended a 'nest cam' (Handykam 420 CCD colour microcamera) in the vegetation above the nest connected by cables to a digital video recorder (Lawmate PVR1000 or PVR500 ECO) at the base of the tree. Nests were filmed between twilight and dusk, each day until at least the end of the first day of the mockingbird's incubation period or until the nest was predated or abandoned. Nests were considered abandoned if parents were not observed at the nest during a recording day. We filmed both laying visits, which include egg puncturing as well as laying and occur mostly prior to sunrise, and non-laying visits during which cowbirds attempt to puncture eggs and occur between sunrise and sunset (Gloag et al. 2013). We checked the contents of nests each day and labelled new cowbird eggs with a permanent marker. As a record of egg morphology, each egg was removed from the nest and photographed in the field using a Canon SX230 camera against a common brown-cloth backdrop, away from strong light. Clutches were sometimes manipulated, at this time, by removal or addition of eggs for a concurrent study on egg-puncturing behaviour (VDF et al., unpublished data), but the eggs that were laid by the painted females were always immediately returned to the nest, allowing us to assess whether a female attacked her own egg if she made a return visit (either laying or non-laying).

Video analysis and egg category assignment

We assessed repeat parasitism rates by female shiny cowbirds based only on those nests that were active until at least the first day of incubation and were therefore likely to have attracted their full complement of parasitism (Fiorini et al. 2009b). For each cowbird egg laid in these nests we asked the following: Did the female that laid this egg return to parasitize the nest again? Nest cam footage was usually sufficient to answer this question based on the head marks, rings and/or day of laying of any subsequent females (each female can lay only one egg per day). If we failed to capture an egg-laying behaviour on film however, we instead compared the egg's morphology with that of existing eggs in the nest to determine whether repeat parasitism could be excluded. Five scorers naive to the study's aims were asked to classify eggs from photographs into one of six categories that represented variation in spotting pattern, using the panel shown in Fig. 2. These categories represent only arbitrary divisions that serve well to discriminate between eggs



Fig. 2 a–f Six categories that span typical variation in spotting pattern of shiny cowbird eggs at our fieldsite. Where the identity of females could not be known from nest cam footage, their eggs were assigned to category

by human scorers, based on photographs taken in the field. The photos in this reference panel were modified to give all eggs a common size and shape

at our field site and do not capture the full diversity of shiny cowbird egg spotting patterns. Scorers were presented with a sequence of digital images of eggs from nests in which an unidentified female laid, interspersed with eggs laid by painted females (700-ppi resolution). We first checked whether the eggs of the same female were consistently assigned to the same category within and between the scorers. Following from this, we considered that two eggs that did not attract any scores in the same category by any of the five scorers were sufficiently different that the same female could not have laid them. Combined data from videos and egg morphology therefore gave us an upper estimate of the occurrence of shiny cowbird repeat parasitism in our sample of eggs, for which we calculated the confidence interval (95 %) of this proportion. All confidence intervals we report were calculated using the exact method (Zar 1999).

Although mockingbirds do not reject the majority of cowbird eggs, they may reject unspotted white cowbird eggs (i.e. Fig. 2a; Sackmann and Reboresda 2003). It is possible that some eggs were missed therefore as a result of mockingbird rejection, though this is unlikely given that most egg lays were filmed and the incidence of white eggs at our fieldsite is low (see “Results”).

Assessment of female cowbirds’ nest selection mechanism

It is interesting to determine whether female parasites actively target or avoid already parasitized nests or if observed rates of repeat parasitism result from some process that does not take previous parasitism into account. One such process could be for females to choose randomly each day amongst a set of nests that they know to be at the right stage for parasitism, inclusive of nests they have already parasitized. In this case, the rate of repeat parasitism will be inversely proportional to the number of nests known to be available to a given female. The fact that this ‘null’ model refers to the set of nests known to the female, and not to those objectively present in her home range, makes it difficult to determine whether a given rate of repeat parasitism is caused by active or passive mechanisms. However, it is possible to approach the issue by calculating the number of laying-stage nests that females would need to be monitoring simultaneously to yield a given rate of repeat parasitism in the absence of any active preference for, or

avoidance of, self-parasitized nests. If this number of nests is unrealistically high, then the null hypothesis (i.e. that no active bias exists) loses plausibility.

To do this calculation, we defined the following variables: observed number of cases of repeat parasitism in our sample (x), the number of laying-stage host nests known to each female on a given day (N), and the number of days that nests in our sample remained in the laying stage after parasitism (i.e. the number of days they remained available for repeat parasitism, d). We then computed the probability (P) of observing x cases of repeat parasitism if females were displaying no selectivity within their set of known nests.

Results

We filmed a total of 311 cowbird visits to mockingbird nests of which 136 were by painted females ($n=18$ different females). Based on all visits filmed subsequent to our last female marking ($n=177$), the painted females represented 44 % of nest visits (confidence interval (CI)=37–52 %), giving an estimate of 41 individual females targeting mockingbird nests within our study area (i.e. $18/0.44$, CI=35–49 females).

Do individual shiny cowbirds lay eggs of a consistent appearance?

We filmed 179 laying visits, including 67 by painted females ($n=13$ individuals, 1–13 eggs per female). Twenty-five of these egg lays by painted females occurred on the same day as another egg lay in the same nest (meaning that we could not be certain which egg belonged to which female), and two eggs were laid by females that were not filmed laying further eggs. This left the remaining 40 eggs to be used to assess within-individual consistency in egg morphology ($n=8$ individuals, 2–8 eggs per female). As illustrated in Fig. 3, eggs of a single female were similar to one another in appearance. When five scorers were asked to assign eggs to the categories of Fig. 2, they either uniformly assigned all of the eggs belonging to a single female into a single category ($n=6$ females) or nearly did so ($n=2$ females, both of which had a single egg placed into a different category by two of the five scorers). No two eggs laid by the same female approached our

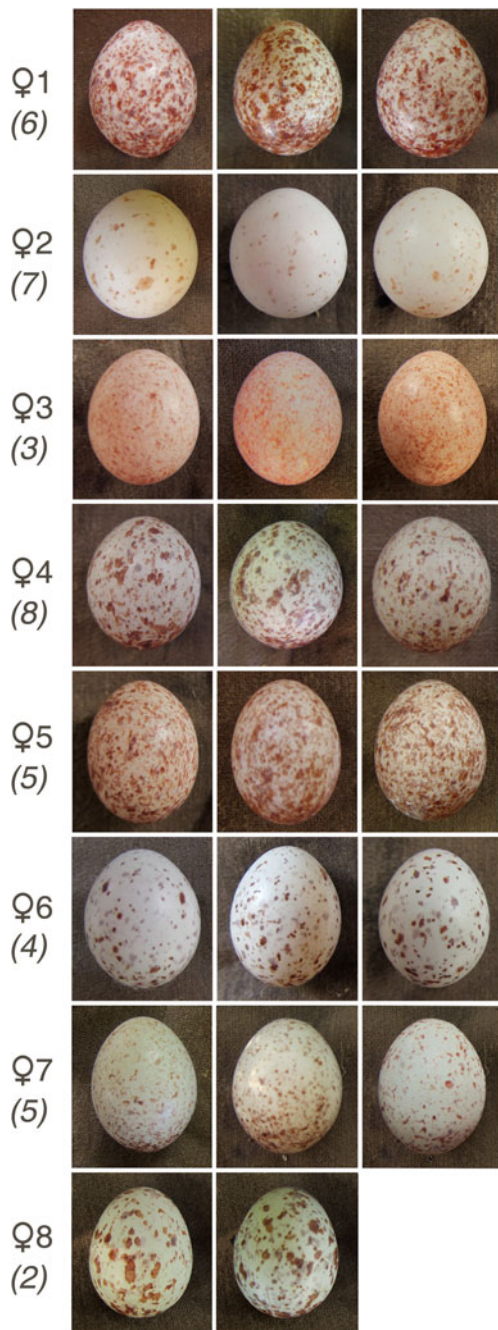


Fig. 3 Examples of eggs laid by each of eight painted shiny cowbird females on filmed laying visits to chalk-browed mockingbird nests. Sample sizes of filmed egg lays in which egg assignment was unambiguous are given in parentheses for each female and ranged from 2 to 8. Eggs laid by the same female were very similar, to the human eye, in colour, spotting pattern, shape and size. When scored into our categories (see main text), no two eggs by the same female were uniformly scored into different categories

criterion for ‘different females’, indicating that this criterion was very unlikely to lead us to underestimate the true repeat parasitism rate. That is, for any pair of same-female eggs, the observed rate at which all five scorers uniformly placed one egg in one category and the other egg in another category was

zero (0 in 94 egg pairs, CI=0–0.038 %). Category frequencies based on all photographed eggs are given in Table 1.

Do shiny cowbirds return to mockingbird nests that they have already parasitized?

We did not film any of our painted females returning to lay again in a nest that they had already parasitized (0 returns from 67 lays by 13 different painted females), though we did film painted females laying eggs on consecutive days ($n=9$ occasions) in different nests and at 1-day intervals ($n=6$ occasions) in different nests. Individual painted females were also observed parasitizing successive nesting attempts of the same mockingbird breeding pairs ($n=7$ occasions).

Sixty nests remained active until the start of the incubation period or beyond, of which 52 were parasitized (87 %). Amongst the parasitized nests, 15 were singly parasitized and 37 were multiply parasitized (71 %, ranging from two to eight eggs per nest; Fig. 4, and see Online Resource 1 for an example of a recording). We filmed the majority of cowbird egg lays in these nests ($n=113$ of 133, 85 %) with the remaining eggs assessed from photographs only ($n=20$, 15 %). Amongst those egg lays that were not filmed, some white eggs could have been missed because they were rejected by mockingbirds before we could photograph them, but such omissions would have been very few. White eggs represented 13 of the 179 filmed cowbird egg lays (7 %), and only seven of these were rejected prior to our morning nest checks (4 % of the total filmed lays; mean latency to reject=88 min after cowbird laying, range 27–185 min).

For 126 of all 133 eggs laid (95 %), we were able to confirm that the female that laid the egg did not return to lay again in the same nest. Thus, different females were typically responsible for eggs in multiply parasitized nests, as illustrated in Fig. 4, with up to eight different females laying in the same nest. For the remaining 5 % of the eggs in our sample (7 of 133), we did not have sufficient data to either confirm or refute that a later egg laid into the same nest belonged to the same female. This occurred where an egg lay in the nest was not filmed, and at least one other egg in the same nest was assigned the same category. Five per cent (CI=2.1–10.5 %)

Table 1 The proportion of shiny cowbird eggs found in chalk-browed mockingbird nests at our study site that were scored into each of six categories indicating variation in spotting pattern according to Fig. 2 and the 95 % confidence intervals of these proportions

	Category a	Category b	Category c	Category d	Category e	Category f
% Total	7.1	6.8	10.4	9.1	39.8	26.8
% CI	4.5–10.6	4.3–10.2	7.2–14.3	6.1–12.8	34.3–45.5	22–32.2

$n=309$

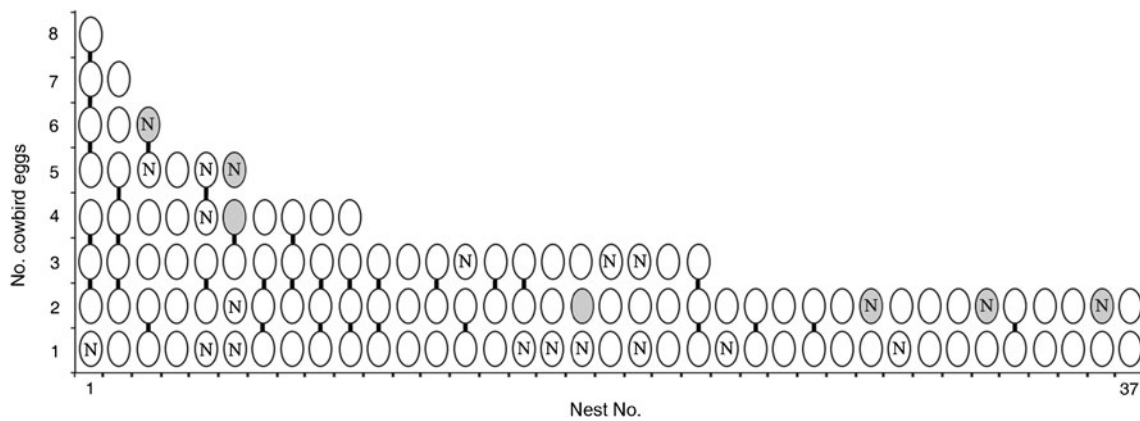


Fig. 4 Cowbird eggs ($n=118$) received by 37 multiply parasitized mockingbird nests. *White eggs* indicate that the female that laid the egg had not previously parasitized the nest. *Grey eggs* indicate equivocal cases, where it was not possible to confirm whether or not the female repeat parasitized the nest. Most egg lays were filmed

allowing direct assessment of female identity based on head marks or the presence/absence of rings. Egg lays marked 'N' were not filmed and were compared with other eggs in the same nest based on egg morphology only. Eggs connected by *black bars* were laid on the same day

is therefore the upper estimate for the observed repeat parasitism rate, and given that there were multiple females in our population laying in each egg category (Fig. 3, Table 1), the true incidence is likely to be lower.

We also filmed 132 non-laying visits to nests, including 69 by painted females ($n=15$ individuals, 1–17 visits per female). Most painted females made just one non-laying visit per nest ($n=49$), though some made two such visits at the same nest ($n=10$). Amongst those nests that remained active for the duration of mockingbird laying, 55 % of the painted females' non-laying visits were followed by parasitism (17 of 31; mean latency between final non-laying visit and laying 1.35 ± 0.15 days, range 1–3 days), and a further 10 % were followed by an unfilmed egg lay that matched her egg category ($n=3$). We found no evidence, however, that shiny cowbirds return to make non-laying visits at active nests that they have previously parasitized. On two occasions, a painted female did return to a nest where she had laid but which the mockingbirds had since abandoned. In both cases, she attacked her own egg and, in one case, successfully punctured it.

Do shiny cowbirds actively avoid repeat parasitism of mockingbird nests?

Figure 5 shows the probability (P) of observing little or no repeat parasitism in our sample as a function of the number of available host nests if female cowbirds selected which nest to parasitize at random from a list of remembered nests, rather than actively avoided nests in which they had already laid. Females would need to be selecting each day from a pool of at least 55 candidate nests for the probability of observing no cases of repeat parasitism to be ≤ 0.05 and from a pool of at

least 13 available nests for the probability of observing 7 or fewer cases to be ≤ 0.05 .

Discussion

Using direct evidence from videos of cowbird egg laying and indirect evidence from egg morphology, we show that shiny cowbirds rarely, if ever, return to lay a second egg in a mockingbird nest that they have already parasitized. The high incidence of multiple parasitism in this host arises instead

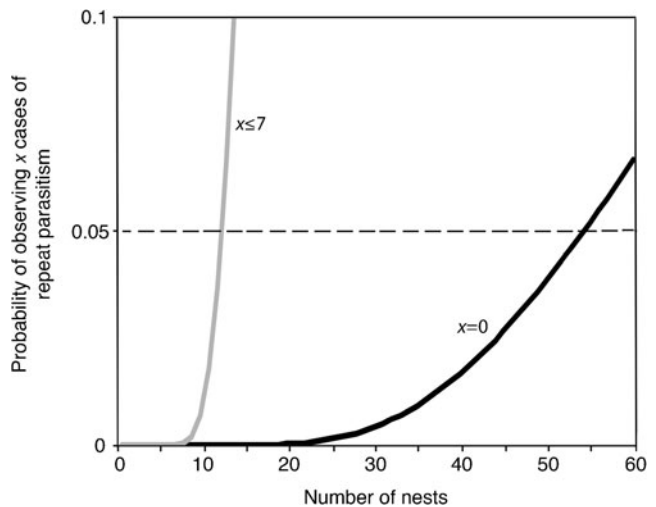


Fig. 5 The probability (P) of observing x cases of repeat parasitism by shiny cowbirds in our sample of 133 eggs laid into chalk-browed mockingbird nests if cowbirds selected nests each day at random from a list of N nests (i.e. if there was no active avoidance of repeat parasitism). The *black line* indicates $x=0$ (our lowest estimate of observed repeat parasitism rate), and the *grey line* indicates $x \leq 7$ (our upper estimate of observed repeat parasitism rate)

from the activity of multiple cowbird females, with between two and eight different individuals laying in the same nest.

A low rate of repeat parasitism might result from females remembering a large list of suitable nests and choosing randomly amongst them, rather than active avoidance of nests in which they have already laid. For the former scenario to account for our results, we calculate that each female would need to be choosing from between 13 and 55 candidate nests each day. Certainly, the higher end of this estimate exceeds what we would consider to be realistic. A female cowbird's area of activity is in the order of 70 ha, wherein there would be no more than four to five mockingbird nests in the laying stage on any day (R. Scardamaglia, personal communication). The lower end of the estimate approaches plausibility provided that females use several additional hosts within their territories and do not prefer mockingbirds. Keeping in mind though that this lower estimate of nest number derives from a particularly conservative upper estimate of repeat parasitism rate (where every egg of inconclusive origin was assumed to represent repeat parasitism), we consider that a null model of random nest selection by females is not strongly supported. That is, females are most likely actively recalling and avoiding nests that they have parasitized.

Our results are consistent with shiny cowbirds employing a one-egg-per-nest rule irrespective of a host's ability to rear multiple cowbird chicks. Whether or not such a rule is adaptive for cowbirds will depend on several factors. By restricting themselves to just one egg per nest, females must make a greater per egg investment in nest searching than if they reused each nest they found. They also, however, escape any reduction in reproductive success as a result of offspring in the same nest competing with one another for food, a cost which may be high in small hosts (Kattan 1997; Tuero et al. 2007; Goguen et al. 2011) though lower in large hosts such as mockingbirds (Post and Wiley 1977; Fraga 1985; Mermoz and Reboreda 1994; Trine 2000). Also important will be the ability of females to adjust their egg-breaking behaviour selectively. Cowbird eggs are thicker-shelled than host eggs but can still be broken during puncture attacks (Mermoz and Ornelas 2004; Gloag et al. 2012b). Thus, unless females were able to modify their puncturing behaviour on return visits to nests, repeat parasitism would involve the risk of a female damaging her own previously laid egg or eggs. Even if repeat parasitism was a good strategy for shiny cowbirds laying in mockingbird nests, it could be that its avoidance is maintained by selection across hosts. If individual cowbird females regularly parasitize both small and large hosts but use a common laying strategy in all cases, their strategy may not be optimized within each host but instead bring a net benefit on average across host species (Gloag et al. 2012a). The extent to which individual females specialize on particular host species or host types is poorly understood. Mahler et al. (2007) proposed that females show partial host fidelity based on haplotype divergences, though

this would not preclude some degree of individual generalism. Indeed, during work for the present study, we made opportunistic recordings of cowbird parasitism at five nests of the house wren (*Troglodytes aedon*), a small-bodied host, and one of these featured a painted female that had previously been filmed laying eggs in mockingbird nests.

Lyon (1997) and Kattan (1997) proposed that shiny cowbirds did not engage in repeat parasitism when targeting a medium-sized host (chestnut-capped blackbird, *Chrysomus ruficapillus*, 32–40 g) and a small host (house wren, 12–14 g), respectively. In those studies, individual females were not identified, but eggs in the same nests were assigned as belonging to different females on the basis of differences in their colour or spotting pattern. Our findings support the idea that shiny cowbird egg morphology can be a useful tool for assessing patterns of host use by confirming that the eggs of a given shiny cowbird female are very similar in appearance, as is true for other brood parasites that exhibit high variability in egg appearance at the species level (Dufty 1983; Langmore et al. 2007; Moksnes et al. 2008; Spottiswoode and Stevens 2011). Importantly though, egg morphology alone can serve only to exclude repeat parasitism and not to affirm it, as while eggs that look very different can be safely assumed to belong to different females, those that look very similar do not necessarily belong to the same female. The reason why shiny cowbird eggs are so variable in their appearance remains unexplained. In other systems, variation in egg morphology has been associated with mimicry of host eggs as a means to evade detection and rejection by hosts, with each female's eggs best matching a particular host egg appearance (Davies and Brooke 1988; Spottiswoode and Stevens 2011). Amongst those shiny cowbird hosts studied to date, however, selective rejection of cowbird eggs is rare, with the exception of a handful of hosts that reject the unspotted white morph (Segura and Reboreda 2012). Furthermore, shiny cowbird eggs do not have host-specific distributions with respect to spotting pattern (De la Colina et al. 2011), with all variants found in all hosts, and this is not consistent with host defence as the selective pressure maintaining egg variability.

It remains possible that shiny cowbirds will repeat parasitize nests under some conditions, such as when the supply of host nests is low. When this is the case, the effort required to find nests will be greater and the chances of successfully locating one lower, such that searching costs are more likely to outweigh any disadvantages of laying more than once in a nest. In brown-headed cowbirds, a high density of parasites has been proposed to decrease nest availability by increasing competition for host nests (Rivers et al. 2012), but the same is probably not true for shiny cowbirds. Females are no less likely to lay in nests that already contain cowbird eggs (Kattan 1997; Gloag et al. 2012b) and, unlike brown-headed cowbirds, frequently share their breeding territories with other females (Ortega 1998). Instead, the principal consequence of a

high parasite density for shiny cowbird females is the increased likelihood that their eggs will share the nest with those of other females. We found 89 % of all cowbird eggs laid into mockingbird nests at our study site occurred in multiply parasitized nests. From the nestling's perspective, it is worth noting that multiple parasitism by different females could still introduce inclusive fitness costs for selfish behaviour if nests were parasitized by females mated to the same male or by related females. Indeed, even if unrelated, the presence of other cowbirds in the same nest might have implications for nestling behaviours by changing the composition of the brood in which parasites must compete. In mockingbirds, conspecific nestmates represent less competition for food than the larger host young, so whether multiple parasitism increases or decreases the intensity of intrabrood competition for cowbirds reared by mockingbirds will depend on whether conspecifics are replacements for host young (i.e. one or more host eggs are broken for each cowbird egg added) or additional to them. We propose these, and other implications of multiple parasitism for the ecology and evolution of brood parasites are a fertile area for future research.

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Ethical standards This research was conducted in accordance with relevant Argentinean regulations (Law of Conservation of Wild Fauna), under the permit issued to JCR, University of Buenos Aires.

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