

# Antagonistic antiparasite defenses: nest defense and egg rejection in the magpie host of the great spotted cuckoo

Juan José Soler,<sup>a</sup> Manuel Soler,<sup>a</sup> Tomás Pérez-Contreras,<sup>a</sup> Santiago Aragón,<sup>b</sup> and Anders Pape Møller<sup>c</sup>

<sup>a</sup>Departamento de Biología Animal y Ecología, Facultad de Ciencias, Universidad de Granada, E-18071 Granada, Spain, <sup>b</sup>Ecole Normale Supérieure, Département de Biologie, Laboratoire Signaux et Régulations Endocrines, CNRS EP 119, 46, rue d'Ulm, F-75252 Paris Cedex 05, France, and <sup>c</sup>Laboratoire d'Ecologie, CNRS URA 258, Université Pierre et Marie Curie, 7 quai St. Bernard, Case 237, F-75252 Paris Cedex 05, France

Brood parasites dramatically reduce the reproductive success of their hosts, which therefore have developed defenses against brood parasites. The first line of defense is protecting the nest against adult parasites. When the parasite has successfully parasitized a host nest, some hosts are able to recognize and reject the eggs of the brood parasite, which constitutes the second line of defense. Both defense tactics are costly and would be counteracted by brood parasites. While a failure in nest defense implies successful parasitism and therefore great reduction of reproductive success of hosts, a host that recognizes parasitic eggs has the opportunity to reduce the effect of parasitism by removing the parasitic egg. We hypothesized that, when nest defense is counteracted by the brood parasite, hosts that recognize cuckoo eggs should defend their nests at a lower level than nonrecognizers because the former also recognize adult cuckoos. Magpie (*Pica pica*) hosts that rejected model eggs of the brood parasitic great spotted cuckoo (*Clamator glandarius*) showed lower levels of nest defense when exposed to a great spotted cuckoo than when exposed to a nest predator (a carrion crow *Corvus corone*). Moreover, magpies rejecting cuckoo eggs showed lower levels of nest defense against great spotted cuckoos than nonrecognizer magpies, whereas differences in levels of defense disappeared when exposed to a carrion crow. These results suggest that hosts specialize in antiparasite defense and that different kinds of defense are antagonistically expressed. We suggest that nest-defense mechanisms are ancestral, whereas egg recognition and rejection is a subsequent stage in the coevolutionary process. However, host recognition ability will not be expressed when brood parasites break this second line of defense. *Key words:* brood parasitism, *Clamator glandarius*, coevolution, hierarchical defense strategies, host defense strategies, great spotted cuckoos, magpies, *Pica pica*. [*Behav Ecol* 10:707–713 (1999)]

In general, host–parasite interactions occur at a number of different levels. Parasites first have to search for a host, which may defend itself by concealment or deterrence behavior. Once located, the parasite has to infect the host, which has the possibility of evasion. Even infested hosts may still manage to avoid parasitism by raising an immune response or otherwise evicting the parasite. This hierarchical organization of antagonistic interactions between hosts and parasites may give rise to antagonistic expression of defense at different levels (Hochberg, 1997). Individual hosts that are specialists at avoiding being parasitized may be less capable of defending themselves against parasites at other levels of defense. Although these conclusions make intuitive sense, there is little empirical information available to test for the interactions between different levels of antiparasite behavior of individual hosts and different levels of parasite behavior of individual parasites. Here we report on an experiment that tests the efficiency of nest defense and parasite egg recognition by hosts of a brood parasite. Specifically, we test whether individual hosts that are able to discriminate and reject cuckoo eggs are less efficient at defending their nests against an approaching potential parasite.

Brood parasitism is a reproductive strategy by which parasites lay their eggs in the nest of hosts, which incubate and rear the offspring (Payne, 1977; Rothstein, 1990). This repro-

ductive strategy greatly reduces the reproductive success of hosts (Rothstein, 1990). Hosts have therefore evolved different strategies of defense against brood parasites. The first line of defense is to prevent the parasite from finding or approaching the nest; this is a widespread mechanism of defense in hosts independent of the brood parasite (Payne, 1997). For example, honeyguides (Indicatoridae) are attacked by hosts (Friedmann, 1955; Payne, 1994; Short and Horne, 1985), and cuckoos (Cuculidae) and cowbirds (Icteridae) near the nest of a potential host provoke alarm calls and attacks (Briskie et al., 1992; Davies and Brooke, 1988; Gill et al., 1997; Moksnes et al., 1990, 1993; MacLean and Rhodes, 1991; Payne et al., 1985).

Host defense of the nest is not only used against brood parasites. There are examples where cowbird hosts behave similarly toward predators and intruders (Neudorf and Sealy, 1992; Ortega and Cruz, 1991; Uyehara and Narins, 1995), and even typical egg-rejecter species do not discriminate between predators and brood parasites (Bazin and Sealy, 1993; Neudorf and Sealy, 1992; Robertson and Norman, 1976). However, some cowbird hosts apparently recognize the brood parasite because these hosts respond most intensely to cowbirds (*Molothrus ater*) during the laying stage (Folkers and Lowther, 1985; Neudorf and Sealy, 1992; Smith et al., 1984), which is the time when most nests are parasitized (Rothstein, 1975). Some European cuckoo (*Cuculus canorus*) hosts also show higher levels of defense against cuckoos than against predators (Duckworth, 1991). The most clear-cut experiment showing that nest defense has evolved in response to brood parasitism demonstrated that yellow warblers (*Dendroica petechia*)

Address correspondence to J. J. Soler. E-mail: jsolerc@goliat.ugr.es.

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in areas of sympatry with cowbirds responded more intensely toward a cowbird model than did warblers in areas of allopatry (Briskie et al., 1992). However, as many brood parasites are also nest predators (Moksnes et al., 1990; Neudorf and Sealy, 1992; Bazin and Sealy, 1993), host nest defense against brood parasites could have arisen in response to predation pressure. If hosts recognize heterospecifics both as predators and brood parasites, they should show a higher level of nest defense than if hosts only recognize heterospecifics as predators.

Once the first line of defense has been broken, hosts use other defense tactics against brood parasitism such as recognition and rejection of parasitic eggs (e. g., Briskie et al., 1992; Davies and Brooke, 1989; Kelly, 1987; Moksnes et al., 1990; Rothstein, 1975; Soler and Møller, 1990; Takasu et al., 1993). A few experimental studies have shown that the ability of host recognition of parasitic eggs differs among populations of the same host species, depending on the duration of sympatry between host and parasite (Briskie et al., 1992; Soler and Møller, 1990; Soler et al., 1999a), supporting the hypothesis that this kind of host defense has evolved as a response to brood parasitism.

Both host defense against adult brood parasites and recognition of parasitic eggs could have evolved in response to brood parasite selection pressures. Moksnes et al. (1990) hypothesized that bird species that have been involved in a co-evolutionary process with the European cuckoo (detected by their ability to recognize and reject foreign eggs from their nests, as well as cuckoo eggs being mimetic to those of the host) should also be able to recognize the potential brood parasite as an enemy. That is, an increase of the two kinds of host defense tactics, rather than being antagonistic, should be of selective advantage for hosts. In accordance with that prediction, Moksnes et al. (1990) found a significant positive correlation between level of rejection and level of aggression among host species. Moreover, Briskie et al. (1992), by studying allopatric and sympatric populations of two different hosts of the brown-headed cowbird (*Molothrus ater*), found evidence supporting the hypothesis that recognition of cowbirds and their eggs evolved as adaptations to counter cowbird parasitism and not some other selection pressure. Thus, based on this point of view, it could be predicted that selection will favor an increase in defenses against adult brood parasites and recognition of parasite eggs, although this prediction has never been tested at the level of individual hosts.

However, both host defense against adult brood parasites and recognition of parasitic eggs by hosts are costly, and those costs could influence predictions about the evolution of host defenses. Nest defense by the host is costly in terms of time, energy, and risk of being injured by the brood parasite, mainly when the brood parasite is larger than the host. Nest defense by the host could also imply additional costs such as increased detectability of the nest for the brood parasite (nesting-cue hypothesis; Robertson and Norman, 1976, 1977), and increased exposure of the nest due to host pursuit of the male brood parasite, allowing the brood parasite female to parasitize the nest (a strategy adopted by the great spotted cuckoo, *Clamator glandarius*; Alvarez and Arias de Reyna, 1974). Furthermore, host recognition of parasitic eggs is sometimes costly because hosts make recognition errors and eject one or more of their own eggs rather than the egg of the brood parasite (Davies and Brooke, 1988; Davies et al., 1996 Marchetti, 1992) or because the host breaks some of its own eggs when trying to eject a parasitic egg (Davies and Brooke, 1988; Rohwer et al., 1989). Due to such costs, when an individual host is sufficiently efficient using one defense tactic against parasitism, other tactics may be less efficient due to their associated costs. For a given cost associated with host defenses

against a brood parasite, selection should favor individuals with a high level of nest defense or those with a high level of egg-recognition ability, but not those with intermediate levels of the two kinds of defense, as this would constitute a case of disruptive selection. A failure in nest defense against brood parasites and subsequent successful parasitism may imply complete reproductive failure for nonrecognizers of cuckoo eggs, and they should therefore defend their nests at a higher level than host individuals with a fine-tuned recognition ability (low costs associated with egg-recognition errors) because recognizers could later remove the parasitic egg from its nest. That will be the case if parasites, by successfully laying an egg in a host nest, do not cause additional costs (such as breakage or removal of a host egg) for hosts that later reject the brood parasite egg. Thus, because parasites destroy or eat some host eggs when parasitizing a host nest (Rothstein, 1990; Sealy, 1992; Soler et al., 1997) and also act as nest predators (Arcese et al., 1996; Bazin and Sealy, 1993; Moksnes et al., 1990; Neudorf and Sealy, 1992; Soler et al., 1995), a certain level of nest defense might still be appropriate even in egg-recognizer hosts.

Counteradaptations of brood parasites against host nest-defense make the scenario even more complex. Some brood parasites use the "host distraction strategy" of the host, whereby the male brood parasite attracts the attention of the host away from their nest, providing female brood parasites with easy access to the host nest (Arias de Reyna, 1998). This strategy has been detected in brood parasites of the genus *Clamator*; as well as in several African Cuculinae (Arias de Reyna, 1998). Thus, adoption of a distraction strategy when laying has the additional cost of increasing nest accessibility for the cuckoo female, thereby increasing the probability of being parasitized.

In the present study we tested the hypothesis that individual hosts that recognize and reject parasitic eggs (rejecter individuals) should show a lower level of nest defense against brood parasites than nonrecognizers (acceptor individuals). The reason for this is that recognizers have the possibility of reducing the effects of parasitism, even after failed nest defense, which is counteracted by the distraction strategy of the brood parasite. This hypothesis implies that hosts that recognize parasitic eggs should also recognize adult parasites. To test this hypothesis, we studied magpies (*Pica pica*) parasitized by the great spotted cuckoo in southern Spain and recorded information on the level of magpie defense of nests against live great spotted cuckoos and carrion crows (*Corvus corone*) (the main predator of magpie nests; Soler, 1990), as well as the rejection behavior of the magpies. Great spotted cuckoos use the distraction strategy to facilitate female access to the host nest. When a female is about to lay, the male flies around the magpie nest, singing loudly, provoking an attack by both male and female magpies; meanwhile, the female great spotted cuckoo approaches the magpie nest silently and inconspicuously as soon as the nest owners leave the nest and lays her egg in only 2–3 s (Alvarez and Arias de Reyna, 1974; Arias de Reyna et al., 1982). In this context, it is not beneficial for magpies to chase the great spotted cuckoo by abandoning their nests.

We tested the following predictions: (1) If hosts discriminating against parasitic eggs are able to recognize parasitic adults, they should demonstrate a higher level of nest defense when exposed to a predator than when exposed to a brood parasite because of the associated costs of nest defense. (2) Because both egg recognition and nest defense are costly for hosts, we predicted that magpies recognizing cuckoo eggs should show a lower level of defense against a great spotted cuckoo than nonrecognizer magpies. The basis for this prediction is that nest defense is not very efficient in this species (see above), and recognizers have the opportunity of reducing

the effect of parasitism after the cuckoo has laid its egg. (3) Both recognizer and nonrecognizer magpies should defend their nests at a similar level when a potential nest predator is perching close to their nests because both categories of magpies suffer from nest predators, and recognition of a foreign egg does not provide efficient protection against predation.

## MATERIALS AND METHODS

### Study areas

During the breeding seasons 1994–1995, we carried out egg-recognition experiments and tests of nest defense (see below) in a magpie population in Guadix, in southeastern Spain. In 1995, we avoided testing nest defense in magpie territories already used in 1994 [many magpies were unbanded and thus not individually identifiable; however, magpies have high territorial fidelity (Birkhead, 1991), and an individual using a territory one year is likely to use the same territory in subsequent years]. During 1995, we also conducted egg-recognition and nest defense experiments in a magpie population in Doñana National Park, in southwestern Spain. Because magpie rejection behavior during replacement clutches has been demonstrated to be mediated by the predatory behavior of its brood parasite, the great spotted cuckoo (Soler et al., 1999b), we did not use magpie replacement clutches for the experiments.

The Doñana and Guadix magpie populations showed a similar degree of rejection of model cuckoo eggs (rejection rate: Doñana = 43.5%,  $N = 23$ ; Guadix, 1994–1995 = 58.6%,  $N = 29$ ;  $\chi^2 = 1.18$ ,  $df = 1$ ,  $p = .28$ ), but the parasitism rate in Guadix was higher than in Doñana (parasitism rate: Doñana = 11.5%,  $N = 52$ ; Guadix = 51.8%,  $N = 85$ );  $\chi^2 = 22.5$ ,  $df = 1$ ,  $p = .00001$ ; data from 1994; Soler et al., 1999a). The main difference potentially affecting the experiments in the two magpie populations is that the carrion crow is the main nest predator in Guadix, but not in Doñana, where crows are absent. However, other corvid species such as jackdaws (*Corvus monedula*) and ravens (*Corvus corax*) occur in Doñana. Nonsignificant differences appeared between magpies from Guadix and Doñana in level of nest defense against the experimental carrion crow [overall score of defense (see below), Guadix: mean = 5.1, SE = 1.5,  $N = 7$ ; Doñana: mean = 7.7, SD = 0.3,  $N = 8$ ; see Results]. In any case, because ecological factors could affect the level of nest defense (Tolonen and Korpimäki, 1995), we adjusted the values of nest defense by magpies in different areas and years by subtracting the mean value from the value for each pair. We report the results using both adjusted and unadjusted values.

### Egg-recognition experiments

At the beginning of the breeding season, we searched systematically for magpie nests. We regularly visited the nests, and when a nest contained at least one egg, we added a mimetic cuckoo model egg. Previous experiments have shown that the rejection probability does not depend on the timing of introduction of the model egg during the laying sequence of the magpie (Soler JJ et al., unpublished data). Model eggs were made by filling molds of great spotted cuckoo eggs with plaster of Paris. Once dry, the model was removed from the mold and painted with a color similar to the background of great spotted cuckoo eggs. Subsequently, we added brown spots with a distribution and size resembling those of real cuckoo eggs. Finally, the model egg was covered with a thin layer of lacquer, which simulates the sheen of real cuckoo eggs. The mass of model eggs was similar to the mass of real cuckoo eggs (see Soler and Møller, 1990). Between 3 and 5 days later (a suffi-

cient time to record rejection; Soler and Møller, 1990), we revisited the nests and scored the magpies as acceptors if the mimetic model was still in the nest, or as rejecters if the model egg was no longer present or the nest was abandoned.

### Nest-defense experiment

During 1992, we raised two different great spotted cuckoo chicks from magpie nests and a carrion crow chick. All nestlings were close to fledging when they were brought to the lab. We kept the great spotted cuckoos and the carrion crow in an aviary for 2 years, training them to eat. They were also trained to perch quietly if tied in order to prevent escape. When the birds were 2 years old and accustomed to perching quietly, we used them for the magpie nest defense experiment. We used live birds because in a previous attempt magpies did not defend their nests against stuffed birds.

The experiment consisted of randomly placing a great spotted cuckoo or the carrion crow tied on a perch (1.5–5 m high) about 1 m from a magpie nest and recording for a maximum of 1 h (after we detected magpies) the defensive behavior of the magpie pair. We observed behavior from a car when possible, or from a hiding place situated 25–100 m from the nest. We measured (1) duration of latency to approach by magpies, using a stopwatch; when no magpie appeared during the experiment, we assumed 60 min; (2) whether no, one, or two magpies defended the nest; (3) distance to the bird presented; when no magpie appeared during the experiment we assumed 100 m; (4) the number of times that a magpie approached the presented bird scolding or attacking; (5) the number of scolding calls by the magpie; and (6) whether magpies physically attacked the presented bird. If magpies attacked the bird, and there was a risk of injury (five of the nine cases where we detected physical contact), we terminated the experiment and removed the bird to prevent injury. We recalculated data from experiments lasting <1 h so they equaled rates per 60 min. The experiments were carried out simultaneously with the egg-recognition experiment when there were magpie eggs in the nest and incubation had already started. In most cases magpies were incubating when we approached the nest to perform the experiments. They flew away when we were close to the nest, and then we introduced the model egg into the magpie nest and placed the adult cuckoo close to the magpie nest. Thus, when the magpies returned to the surroundings of the nest, they first noticed the presence of the adult cuckoo close to the nest, but could not know that a model cuckoo egg was in the nest. Moreover, no magpie entered their nest during the nest-defense experiment, and the results from the nest-defense experiment thus do not depend on magpies knowing whether their nest had been parasitized, nor on magpie detection of the adult cuckoo.

In magpies, scolding rate has been found to be highly positively correlated with propensity to attack and therefore is a reliable indicator of the birds' willingness to defend their nests (Redondo and Carranza, 1989; Röell and Bossema, 1982). We performed a logistic regression analysis (maximum likelihood method) with magpie attack of the bird as the dependent variable and the other five response variables as independent variables. When removing latency time to approach by magpies, the fit of the regression model improved significantly (difference in log-likelihood from the previous model = 10.2,  $p = .0014$ ), and thus we did not use this variable in the final regression model because variance explained by this variable was already explained by the others. The overall model was highly significant (Table 1), and we used the equation from the logistic regression [attack = 2; no attack = 1; probability of attack =  $-3.2 + 3.9(\text{no. of magpies attacking}) - 1.8(\text{distance of magpies}) - 0.02(\text{no. of times that}$

Table 1

Results of the logistic regression model between attack (yes or no) as dependent variable and number of magpies attacking ("magpies"), distance of magpies ("distance"), number of times that magpies approached ("approach"), and number of magpie scolding calls ("scolding") as independent variables

	Constant	Magpies	Distance	Approach	Scolding
Parameter estimate	-3.21	3.86	-1.82	-0.02	0.21
SE	3.18	3.27	1.03	0.18	0.34
$t_{35}$	-1.01	1.18	-1.77	-0.13	0.62
$p$	.32	.25	.09	.90	.53

-2 log-likelihood = 4.01,  $\chi^2 = 38.6$ ,  $df = 4$ ,  $p < .0000001$ .

magpies approached) + 0.21(no. of magpie scolding calls); see Table 1] to generate a score of the level of defense of each magpie pair. We used the results from this equation for each magpie pair as the value of level of defense. Finally, we added 200 to the resulting value and divided by 25 to have values of level of defense ranging from 0 to 10. Hence we obtained a single measure of level of defense related to the probability of physical attack, thereby avoiding problems with multivariate analyses using non-normalized variables.

### Statistical analyses

The level of magpie defense was not normally distributed, and it was impossible to transform the variable to obtain an approximately normal distribution. Therefore, we used non-parametric statistical tests following Siegel and Castellan (1995). All tests were two-tailed.

## RESULTS

### Egg-recognition tests

We performed 40 egg-recognition tests (13 in Doñana, 11 in Guadix 1994 and 16 in 1995). On average 57.5% ( $N = 40$ ) of the cuckoo-model eggs were rejected from magpie nests (rejection rates: Doñana, 46.2%,  $N = 13$ ; Guadix 1994, 81.8%,  $N = 11$ ); 1995, 50.0%,  $N = 16$ ; comparison between populations and years,  $\chi^2$  tests, ns; Table 2). None of the experimental nests was abandoned in Doñana, two nests were abandoned in Guadix in 1994 and none in 1995.

### Nest-defense experiments

All 40 magpie nests used for the egg-recognition tests were used for the nest-defense experiment. We performed the experiment using randomly chosen great spotted cuckoos in 25 nests (5 in Doñana, 20 in Guadix: 4 in 1994 and 16 in 1995), and using the carrion crow in the remaining 15 nests (8 in

Doñana and 7 in Guadix 1994). Both magpie populations showed nest defense against the great spotted cuckoo and the carrion crow (see Table 2 for values of nest-defense variables). There were no significant differences between levels of defense among magpies in Doñana and Guadix when the stimulus was a great spotted cuckoo (Kruskal-Wallis ANOVA,  $H_2$ ,  $N = 25 = 1.13$ ,  $p = .57$ ), or a carrion crow (Kruskal-Wallis ANOVA,  $H_1$ ,  $N = 15 = 0.33$ ,  $p = .56$ ; Table 2). Moreover, we found no significant difference in the response of magpies (recognizers or nonrecognizers) to the two cuckoos used in the experiment (Mann-Whitney  $U$  test,  $z < 1.5$ ,  $p > .15$ ).

In general, magpies showed a similar level of defense when exposed to a carrion crow or to a great spotted cuckoo (carrion crow: 6.57, SE = 0.73,  $N = 15$ ; great spotted cuckoo: 5.58, SE = 0.60,  $N = 25$ ; Mann-Whitney  $U$  test,  $z = 1.35$ ,  $p = .18$ ), which was more clear when controlling for variation between populations and years (carrion crow: -0.40, SE = 0.69,  $N = 15$ ; great spotted cuckoo: -0.24, SE = 0.61,  $N = 25$ ; Mann-Whitney  $U$  test,  $z = 0.63$ ,  $p = .53$ ).

### Relationship between nest defense and egg recognition ability

Magpies that rejected cuckoo eggs showed a significantly lower level of defense against great spotted cuckoos than against carrion crows (Mann-Whitney  $U$  test,  $z = 2.03$ ,  $N = 12$ ,  $N = 11$ , respectively,  $p = .04$ ; data adjusted for differences between areas and years, Mann-Whitney  $U$  test,  $z = 1.96$ ,  $N = 12$ ,  $N = 11$ , respectively,  $p = .05$ ). However, these differences disappeared when considering only nonrejecter magpies (Mann-Whitney  $U$  test,  $z = 0.57$ , cuckoos,  $N = 13$ , crows,  $N = 4$ ,  $p = .57$ ; data adjusted for differences between areas and years, Mann-Whitney  $U$  test,  $z = 0.79$ , cuckoos,  $N = 13$ , crows,  $N = 4$ ,  $p = .43$ ). These tests suggest that magpies, which recognize cuckoo eggs, also recognize adult cuckoos, in accordance with the first prediction.

Moreover, rejecter-magpie behavior was significantly differ-

Table 2

Summary statistics for rejection behavior of experimental magpie nests and defensive behavior<sup>a</sup>

Presented bird	Magpie behavior	$N$	Latency		Magpies		Distance		Approach		Scolding		Defense		Defense-corrected		Attacks %
			Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE	
Cuckoo	Acceptors	13	17.6	3.5	1.3	0.1	11.2	3.8	11.5	5.8	5.8	3.2	7.3	0.2	1.4	0.2	41.7
	Rejecters	12	39.3	6.6	0.7	0.2	58.4	12.7	1.0	0.4	0.9	0.6	3.7	1.0	-2.1	1.0	0.0
Crow	Acceptors	4	9.3	3.3	2.0	0.0	4.0	1.4	4.0	1.8	4.8	2.1	7.9	0.1	1.2	0.1	33.3
	Rejecters	11	21.0	6.7	1.1	0.2	29.4	12.7	6.7	5.3	21.6	16.1	6.1	0.9	0.11	0.9	27.3

<sup>a</sup> Measured as latency to approach, number of magpies defending the nest (0, 1, or 2), distance from the presented cuckoo or crow, number of times that any magpie approached the presented bird, number of magpies giving scolding calls, overall score of defense, the defense score corrected for differences between areas and years, and the percentage of magpie nests where physical attack of the presented bird occurred.

ent from that of nonrecognizer magpies when exposed to a brood parasite. In accordance with the second prediction, magpies that recognized cuckoo eggs showed a lower level of defense than did nonrecognizer magpies (Mann-Whitney  $U$  test,  $z = 2.83$ ,  $N = 12$ ,  $N = 13$ , respectively,  $p = .0047$ ; Table 2), even when controlling for variation between populations and years (Mann-Whitney  $U$  test,  $z = 2.50$ , rejecter,  $N = 12$ , acceptor,  $N = 13$ ,  $p = .012$ ). However, these differences between magpies rejecting and accepting cuckoo eggs were not present when exposed to a carrion crow close to their nests (Mann-Whitney  $U$  test,  $z = 0.65$ ,  $N = 11$ ,  $N = 4$ , respectively,  $p = .51$ ; Table 2), even when controlling for variation between populations and years (Mann-Whitney  $U$  test,  $z = 0.00$ , rejecter,  $N = 11$ , acceptor,  $N = 4$ ,  $p = 1$ ). Therefore, magpies that rejected cuckoo eggs defended their nests less aggressively against the cuckoos, but equally strongly against carrion crows, compared to magpies that accepted cuckoo eggs.

The results did not change when we removed from the analyses two nests that magpies abandoned (both nests were tested with carrion crow and classified as recognizers; level of defense of recognizer magpies against great spotted cuckoo and carrion crow was: Mann-Whitney  $U$  test,  $z = 2.77$ ,  $p = .005$ ; data adjusted for differences between areas and years,  $z = 2.27$ ,  $p = .023$ ; level of defense of recognizer and nonrecognizer magpies when exposed to a carrion crow, Mann-Whitney  $U$  test,  $z = 0.15$ ,  $p = .88$ ; data adjusted for differences between areas and years,  $z = 0.62$ ,  $p = .54$ ).

## DISCUSSION

Magpies that recognized cuckoo eggs defended their nests against great spotted cuckoos at a lower level than did nonrecognizers, suggesting antagonistic expression of these two kinds of host defense against brood parasites. A potential explanation for this result is that magpies recognizing cuckoo eggs in general showed an overall low level of defense. However, when a carrion crow perched close to magpie nests, both rejecters and nonrejecters of cuckoo eggs defended their nests at a similar level, suggesting that the two categories of hosts were able to raise a similar level of defense against a potential nest predator.

To the best of our knowledge this is the first experiment investigating the interaction between two levels of antiparasite defense. Modeling has suggested that different hierarchical levels of host defense should be negatively related (Hochberg, 1997). All previous studies of host defense against parasites have either analyzed avoidance or evasion behavior (Fineblum and Rausher, 1995; Kraaijeveld and van Alphen, 1995; Mauricio et al., 1997; Sokolowski et al., 1997). However, these findings were not based on observations of the same individual hosts, and thus we cannot be certain whether host defenses were traded against each other.

If host defense tactics are complementary, as suggested here, this could explain the lack of recognition of parasite eggs in some host species and populations (see reviews in Payne, 1997; Rothstein, 1990) and why egg recognizer and nonrecognizer phenotypes are present in a host population: a paradigm of brood parasitism studies (e.g. Takasu, 1998).

Magpies that recognize cuckoo eggs showed a low level of nest defense when exposed to a great spotted cuckoo, possibly because this brood parasite also acts as a nest predator (Soler et al., 1995; Soler et al., 1997); in the case of nest predation, recognizer magpies have no advantage over nonrecognizers. Moreover, we can assume that the nest of the host had already been located by the cuckoo because the great spotted cuckoo perched close to the magpie nest (Gill et al., 1997). In this case, host defensive behavior does not imply an increase in nest detectability for the cuckoo, and nest defense by the host

should therefore be highly beneficial (Gill et al., 1997). In previous papers (Soler, 1990; Soler et al., 1996, 1998a) we found that the main damage caused by cuckoos was a result of egg destruction during egg laying (magpie eggs hatched in parasitized nests = 1.3, in unparasitized nests = 5.0; Soler et al., 1996), rather than competition with the cuckoo chick (magpie chicks dying in parasitized nests = 0.7, in unparasitized nests = 1.5; Soler et al., 1996). Thus, magpies do not recover the cost of being parasitized by removing the parasitic egg from the nest, although this increases breeding success. Selection should therefore increase the first line of defense, preventing cuckoos access to the magpie nest, and this would be predicted to be independent of the egg recognition ability of the host. That would be the case if the benefit of nest defense exceeds its costs. However, the strategy adopted by the great spotted cuckoo when parasitizing magpies ("distraction strategy"), by implying additional costs, counteracts the defense of nests by magpies. In this context, it is not beneficial for magpies that recognize great spotted cuckoo adults to chase them by abandoning their nests because this behavior increases nest accessibility for cuckoo females.

In accordance with this suggestion, magpies that recognize cuckoo eggs do not defend their nests by chasing the male great spotted cuckoo since the level of nest defense in our experiment is related to the probability of chasing a cuckoo. They stay close to their nests and maintain the advantage of being able to discover the female cuckoo attempting to enter the nest. This observation could provide magpies with information on the probability that a cuckoo egg will be in the nest. The magpie could subsequently carefully search the nest for cuckoo eggs, as suggested for hosts of the European cuckoo (Davies and Brooke, 1988; Davies et al., 1996; Moksnes and Røskaft, 1989; Moksnes et al., 1993). Although previous experiments on magpie recognition of cuckoo eggs demonstrated that the presence of an adult cuckoo close to magpie nest did not increase the probability of cuckoo egg rejection (Soler et al., in press), the relevant cue for recognizer magpies could be the visual cue of the great spotted cuckoo entering or leaving the nest, which was not controlled in that experiment.

If magpies that recognize cuckoo eggs are able to recognize adult cuckoos, they should defend their nests at a lower level when exposed to a brood parasite than when exposed to a potential nest predator. Nest defense against great spotted cuckoo adults would increase the risk of parasitism during a momentary absence from the nest. In accordance with this scenario, magpies that rejected cuckoo eggs had a significantly lower level of nest defense against great spotted cuckoos than against carrion crows, whereas this difference was absent in nonrejecter magpies. This suggests that magpies that recognize cuckoo eggs also recognize adult cuckoos. However, these results do not imply that magpies that did not recognize cuckoo eggs did not recognize adult cuckoos, because nest defense against brood parasites is the only defense available for nonrecognizer magpies.

These results could suggest that host nest defense could have evolved simply in response to predation pressure because many brood parasites are also nest predators (Bazin and Sealy, 1993; Moksnes et al., 1990; Neudorf and Sealy, 1992). That is, at an early stage in the brood parasite-host association, all magpies may have behaved aggressively toward the brood parasite, but brood parasites developed the distraction strategy to counteract magpies defending their nests. After evolution of egg rejection behavior, these birds could have lost aggressive reactions toward the brood parasite because the costs exceeded the benefits. Therefore, these two groups of magpies could represent two different stages of the defense against cuckoos.

In a previous paper, Soler et al. (1995) demonstrated that

great spotted cuckoos depredated magpie nests where cuckoo eggs had disappeared and hypothesized that, by this mafia-like behavior, cuckoos force magpies to accept cuckoo eggs during replacement clutches. This is a cuckoo strategy to counteract magpie rejection behavior. This cuckoo mafia behavior makes the scenario in which magpie defense tactics develop even more complex. If magpie rejection of cuckoo eggs implies nest predation by cuckoos, breeding success of magpies would increase by turning off egg-rejection behavior by instead investing in nest defense. This scenario is just the opposite of that described above where egg rejection would be the most beneficial host strategy against the brood parasite. Thus, because great spotted cuckoos are able to counteract not only magpie nest defense (by the distraction strategy), but also egg rejection (by the cuckoo mafia behavior), it could be predicted that natural selection, rather than selecting for magpies displaying antagonistic antiparasite defense, should select for magpies increasing both nest defense and egg rejection against the great spotted cuckoo.

Soler et al. (1995) hypothesized that recognizer magpies should reject cuckoo eggs during the first breeding attempt even when there is a risk of rejecter nests being depredated by the cuckoo because such magpies can produce replacement clutches. A magpie should change from rejection to acceptance of a cuckoo egg only in replacement clutches because magpies have no further opportunity for reproduction that year (Soler et al., 1995). This hypothesis has recently been tested (Soler et al., 1999b), and rejecter magpies during their first breeding attempt changed to accept experimental model eggs during replacement clutches after having their first clutches experimentally depredated. However, this occurred only in magpie plots with a high parasitism rate (>50%), but not in a plot with a low parasitism rate (<10%), suggesting that the mafia mechanism only operates in areas with a high risk of parasitism (Soler et al., 1999b). Local change in the distribution of great spotted cuckoos occurs continuously, with parasitism showing spatially structured cyclic changes (Soler et al., 1998b). For example, in Calahorra, a plot of the Guadix magpie population, parasitism rate has decreased from around 55% in 1992 to 0% in 1996 (Soler et al., 1998a, 1999b). Such rapid local change in parasitism may prevent magpie acceptance of cuckoo eggs from going to fixation. Because the probability for a rejecter magpie nest being revisited and depredated by a cuckoo is particularly high for replacement clutches, magpies should learn to accept a cuckoo egg in replacement clutches, but only in areas where the great spotted cuckoo is abundant (Soler et al., 1999b). Accordingly, magpie rejection behavior is not affected by the mafia mechanism during the first breeding attempt because the rejection rate in plots with a low parasitism rate did not differ from that of plots with a high parasitism rate during the first breeding attempt, but highly significant differences appeared in replacement clutches (Soler et al., 1999b).

To perform the experiment described in this article, we did not use replacement clutches, and thus magpie rejection behavior is unlikely to have been affected by the mafia mechanism. However, different results should be predicted if performing the same experiment in areas of high parasitism rate and using replacement clutches because the cost of cuckoo egg rejection by magpies is then considerably increased. In this case, recognizer magpies should turn off their recognition ability and invest in nest defense if that decreases nest accessibility for cuckoos. This predicted result for replacement clutches is in accordance with the hypothesis because, at the individual level, different costs are associated with defense strategies at different times during the breeding season (first and replacement clutches). We predict that individual hosts modulate their behavior by using one of the two defense strat-

egies based on their associated costs and benefits. At least in the case of magpies, natural selection should favor individuals with a high level of recognition ability, but also with a high level of nest defense, because the same individual should efficiently use one or the other defensive strategy depending on external conditions (first or replacement clutches, high or low risk of parasitism). Therefore, the antagonistic expression of magpie antiparasite defenses detected in the present study is not the result of disruptive selection, as hypothesized, but the result of phenotypic plasticity in the expression of antiparasite defense tactics mediated by a learning process of different costs associated with different defense tactics at different times.

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