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# Female and male plumage brightness correlate with nesting failure in azure-winged magpies *Cyanopica cyanus*

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Animals may assess the quality of other individuals by using information that is contained in elaborate traits. We investigated the degree of sexual dimorphism in structural blue plumage coloration and the potential signal value of these traits in the azure-winged magpie *Cyanopica cyanus*. We predicted that in this species blue coloration should signal individual quality in both sexes since both females and males invest significantly in caring for offspring. Males have more saturated UV/blue coloration than females and blueness decreased from moulting to reproduction. Males and females did not mate assortatively for blue coloration although they did in relation to body size and condition. Blue colour did not correlate with adult body size or condition. However, nest predation decreased with female and male brightness. Our results suggest that blue coloration may potentially be used to assess parental qualities by potential mates in both sexes of the azure-winged magpie.

Bird coloration is produced by chemical pigments (carotenoids and/or melanins), or by the physical interaction of the light with feather microstructure (i.e. structural colours; Hill and McGraw 2006). Structural colours are broadly distributed throughout Aves (Prum 2006), and there is growing evidence that their expression might be condition-dependent (reviewed in Hill 2006). Most studies, however, were carried out on male structural coloration while neglecting female coloration (see Siefferman and Hill 2005).

The azure-winged magpie is a small colonial corvid in which males appear to invest a considerable effort in reproduction by feeding incubating females and chicks (Cramp and Perrins 1994). Males are only slightly larger than females although both sexes are highly coloured with blue and chestnut as main colours (Cramp and Perrins 1994). To the human eye structural blue is the predominant colour on wing and tail which is displayed during the whole year in both sexes.

The first aim of this study was to identify relationships between structural blue coloration and sex and age in the azure-winged magpie. Second, we investigated whether coloration may signal individual quality in both sexes under the assumption that mate quality assessment will be equally advantageous for females and males (see above). In the azure-winged magpie only a small fraction of the nests in a population escape to predation, since about 80% of the nests are depredated (Cruz and Valencia 2004). Therefore, coloured traits might provide conspecifics with information

on the quality of potential partner in terms of predator avoidance.

## Methods

### Study site and data collection

The study was carried out in a colony of azure-winged magpies during four consecutive years (2002–2005), at Valdesequera, in the province of Badajoz, Spain (39° 03'N, 6° 48'W) (see details in Valencia et al. 2006).

Every year we performed an intensive capture using a specially designed trap before reproduction (Valencia et al. 2006). A total of 143 birds were captured and marked. Sex of breeders was always assigned according to their behaviour, being scored as females those individuals that incubated the eggs and brooded the young (Valencia et al. 2003). Age was assigned using moult extension, following Cruz et al. (1992) as: juveniles, birds born in the previous year, and adults, birds older than one year.

We searched for nests at the beginning of the breeding season by inspecting every tree in the study area. Nests were marked and checked at least once every two days to record laying date, clutch size and number of fledglings. Second clutches can occur in our population (Cruz and Valencia 2004), and thus be confounded with replacement clutches after a predation event, therefore only first clutches were considered in the analyses. Early monitoring of every nest

allowed us to identify the members of the breeding pair before the chick hatched. About 50% of nests in our population present a helper (Valencia et al. 2003). Helpers usually join the breeding group after the chicks hatched and only occasionally feed the incubating female (Valencia et al. 2006), thus it is always possible to differentiate them from the male breeder. Observations for every nest were made from a hidden position with a telescope for at least 1h every two days which allowed us to identify 41 females and 45 males as breeders. Only in 9 nests we were able to capture the two members of the breeding pair. Thus, we performed separately our analyses on males and females when looking at the relationships between blue plumage coloration and reproductive attributes. Although some birds were recaptured between years, we only used data for the first capture in the analyses.

### **Body size and condition**

We used tarsus length as a measure of body size. We also measured body mass using a Pesola balance ( $\pm 0.5$  g) and used the residuals of a regression of body mass on tarsus length as an index of body condition.

### **Plumage colour**

Upon capture, we collected 2 secondary wing covert feathers from each individual. Feather samples were plucked from the same location on all birds. At the time of data colour collection, feathers of each individual were placed on black paper in a fashion that mimicked the way the feathers lay on the bird. We quantified plumage reflectance in the range 300–700 nm using a spectrometer (DH 2000, Ocean Optics Europe) using a bi-furcated micron fiber optic probe at a  $45^\circ$  angle from the feather surface and illuminating an area of  $1 \text{ mm}^2$ . Using OOIBase, a spectra acquisition software package, we recorded 10 spectra relative to a standard white reference (WS-2) sequentially and then averaged the spectra to reduce electrical noise. This process was repeated three times; the probe lifted and replaced on the feather sample each scan. We then averaged the three spectra for each individual. We summarized reflectance data by calculating three different standard descriptors of reflectance spectra: brightness, hue and chroma. Brightness is the summed reflectance from 300–700 nm. UV-blue hue corresponds to the wavelength at which the maximum peak of reflectance is reached within the ultraviolet-blue waveband (300–500 nm). UV-blue chroma was the ratio between the spectral reflectance in the ultraviolet-blue waveband (300–500 nm) and the reflectance of the entire spectrum (300–700 nm).

Recent findings suggest that structural plumage coloration is not a static trait and that changes in feather coloration may occur after moulting (Örnborg et al. 2002, Delhey et al. 2006). To account for this potential source of bias, we classified feathers in relation to the period in which they were collected as “freshly” moulted feathers, when collected from birds trapped from October to February, and “late” moulted feathers when collected from birds trapped from March to the reproduction. We subsequently introduced period as a fixed term in all our

models to control for possible seasonal changes in plumage coloration.

### **Statistical analysis**

The influence of age and sex on structural blue coloration was tested by running generalized linear mixed models (Mixed procedure in SAS, SAS Institute 1996) in which sex, age and period were entered as fixed factors and year was entered as a random effect. The associations between body size and condition with colour variables were tested by running general linear models in which sex, period and year were entered as fixed effects. Analyses on nests were performed with general linear models (laying date and clutch size) and logistic regressions (nest failure probability, Genmod procedure in SAS) including period and year as fixed effects. Past research demonstrated that the number of helpers at the nest influence reproductive success (Valencia et al. 2003). Therefore, we control for effects of helpers on nesting failure probability by including the number of helpers as a covariable. Only some individuals were recorded in the colony during the reproduction in the same year in which the feathers were collected which reduced our sample size for testing relationships between magpie coloration and nest traits (i.e. laying time, clutch size and nest failure probability). Model selection was carried out by removing, one by one, the effects that were the furthest to statistical significance. Due to low sample size we disregarded testing interactive effects between independent variables.

## **Results**

### **Plumage coloration**

The wing coverts of azure-winged magpies reflect most strongly in the violet and the blue regions of the bird-visible spectrum (Fig. 1). The average hue for the sampled feathers was  $427 \pm 29.0$  nm, well within the blue range (400–475 nm; Fig. 1). Mean values of secondary wing covert colour predictors and of morphological traits are reported in Table 1. Variability of colour predictors was similar for female and male azure-winged magpies (Table 1).

### **Plumage coloration in relation to age, sex and period**

We did not find any sexual or age related variation in brightness and UV-blue hue (Table 2). UV/blue chroma, however, was significantly related to sex (Table 2): males have more saturated UV/blue coloration than females (Table 1).

Blueness varied seasonally (Table 2). Feathers collected from birds captured from October to February had more brilliant and saturated blue coloration and showed a more violet hue than those collected afterwards (brightness (mean  $\pm$  SD), freshly moulted  $2571.1 \pm 47.0$  vs late moulted:  $2289.5 \pm 26.3$ ; UV-blue chroma (mean  $\pm$  SD), freshly moulted  $0.55 \pm 0.0$  vs late moulted:  $0.54 \pm 0.0$ ;

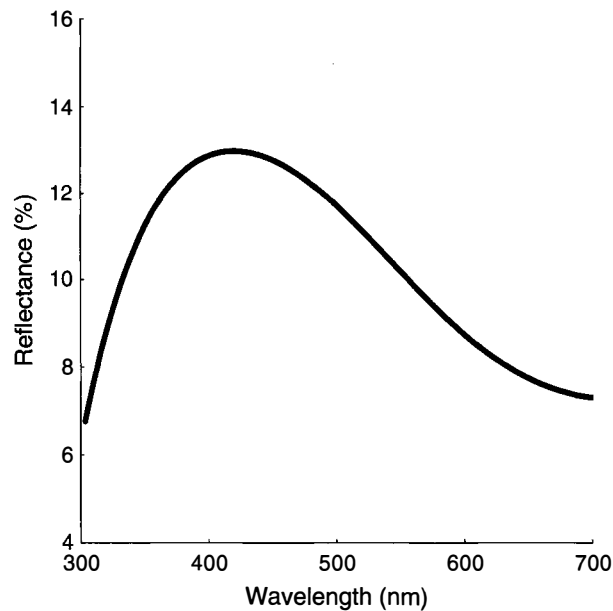


Fig. 1. Average reflectance spectra for the azure-winged magpie secondary wing cover feathers ( $n = 45$  individuals). Values are means of the means for each individual (i.e. three measures for each individual).

UV-blue hue (mean  $\pm$  SD), freshly moulted  $416.8 \pm 6.61$  vs late moulted:  $430.7 \pm 2.36$ .

### Plumage coloration and pairing

We did not find evidence for assortative mating with respect to blue coloration (brightness:  $F_{1,7} = 0.00$ ,  $P = 0.95$ ; UV-blue chroma:  $F_{1,7} = 2.55$ ,  $P = 0.15$ ; UV-blue hue:  $F_{1,7} = 1.87$ ,  $P = 0.21$ ). However, there was assortative mating with respect to tarsus length ( $r = 0.72$ ,  $F_{1,6} = 6.40$ ,  $P = 0.04$ ), and body condition ( $r = 0.87$ ,  $F_{1,6} = 18.93$ ,  $P = 0.004$ ). These results could not be driven by age-assortative pairing since yearlings were similar in size ( $F_{1,139} = 3.07$ ,  $P = 0.08$ ), and had similar condition than adults ( $F_{1,134} = 0.006$ ,  $P = 0.93$ ).

### Plumage coloration and body size and condition

Neither colour variable was retained in final models predicting body size and condition (colour variables excluded with  $0.31 < P < 0.86$ ).

### Plumage coloration and breeding outcome

Female and male blue coloration was unrelated to laying date and/or clutch size (laying date females: colour variables excluded with  $0.16 < P < 0.77$ ; laying date males: colour variables excluded with  $0.63 < P < 0.73$ ; clutch size females: colour variables excluded with  $0.07 < P < 0.90$ ; clutch size males: colour variables excluded with  $0.26 < P < 0.71$ ). However, after deleting non significant terms, nests of females with brighter plumages were less likely predated during the season ( $\chi^2_{1,26} = 6.15$ ,  $P = 0.01$ , Fig. 2). In addition, nests attended by brighter males were less likely predated ( $\chi^2_{1,36} = 4.81$ ,  $P = 0.03$ , Fig. 2).

### Discussion

Our results provide support for the existence of sex-related expression of blue coloration in the azure-winged magpie since males tended to have more saturated UV-blue coloration than females. Among individuals, the expression of blue plumage colour was not correlated with body condition or size, and based on a reduced number of nests we found no support for assortative mating in relation to blueness. Female and male blueness did not predict laying date or clutch size. However, nests of brighter females were those more likely escaping to predation. Furthermore, nests attended by brighter males were less predated than those attended by duller males.

We have found that the feather colour of the azure-winged magpie changed seasonally. Wing coverts became less UV-blue reflectant with time, while overall brightness decreased and feathers tended to become greener. The pattern of variation in plumage coloration reported in this study was qualitatively similar to that reported by Örnborg et al. (2002) for the crown feathers in blue tits *Parus caeruleus* (see also Delhey et al. 2006). A further practical consideration of this result concerns the need of statistical control of seasonal changes in coloration (Delhey et al. 2006), such as we have done, to avoid biasing other statistical comparisons in studies of plumage signalling.

Based on only 9 pairs we did not find evidence that pairing patterns in azure-winged magpies were based on blue plumage coloration. However, azure-winged magpies did not mate randomly since individuals in better condition tended to mate with individuals of similar quality. Although low sample size makes unlikely any firm conclusion, perhaps, azure-winged magpies rely in other elaborate traits and or behaviours not measured in this study. Alternatively, it could be possible that there was a positive correlation

Table 1. Structural plumage coloration descriptors and morphological traits (mean  $\pm$  SD) of adult male and female azure-winged magpies.

Trait	Males			Females		
	Mean	SD	n	Mean	SD	N
Brightness	2368.4	320.9	84	2339.4	265.8	59
UV-blue chroma	0.6	0.02	84	0.5	0.03	59
UV-blue hue (nm)	426.6	29.5	84	428.7	28.6	59
Tarsus length (mm)	34.9	0.9	82	34.3	1.1	59
Bill length (mm)	54.0	1.0	27	52.3	1.4	29
Wing length (mm)	132.6	3.5	27	127.5	3.1	29

Table 2. The influence of age, sex and period on structural blue coloration of the azure-winged magpie.

Effect	Brightness	UV-blue chroma	UV-blue hue
Sex	$F_{1,137} = 1.53, P = 0.22$	<b><math>F_{1,137} = 4.72, P = 0.03</math></b>	$F_{1,136} = 0.20, P = 0.66$
Age	$F_{1,136} = 0.17, P = 0.67$	$F_{1,136} = 0.03, P = 0.87$	$F_{1,137} = 0.89, P = 0.35$
Age x sex	$F_{1,135} = 0.12, P = 0.73$	$F_{1,135} = 0.03, P = 0.87$	$F_{1,135} = 0.02, P = 0.88$
Period	<b><math>F_{1,138} = 19.93, P &lt; 0.001</math></b>	<b><math>F_{1,137} = 3.42, P = 0.06</math></b>	<b><math>F_{1,138} = 6.22, P = 0.013</math></b>
Year	$Z = 0.84, P = 0.19$	$Z = 0.17, P = 0.43$	$Z = 0.27, P = 0.84$

Model selection was carried out by removing, one by one, the effects that were the furthest to statistical significance, starting with the highest-order interactions down to the main effects. Fixed terms included in the final model are in bold.

between female and male blueness only at the time of pair formation. Indeed, in another study we have found that differences in blueness between breeders and helpers are maximal in winter, when pairing presumably occurs (Solis et al. 2008).

Plumage brightness in female and male azure-winged magpies serves as a predictor of nestling failure probability.

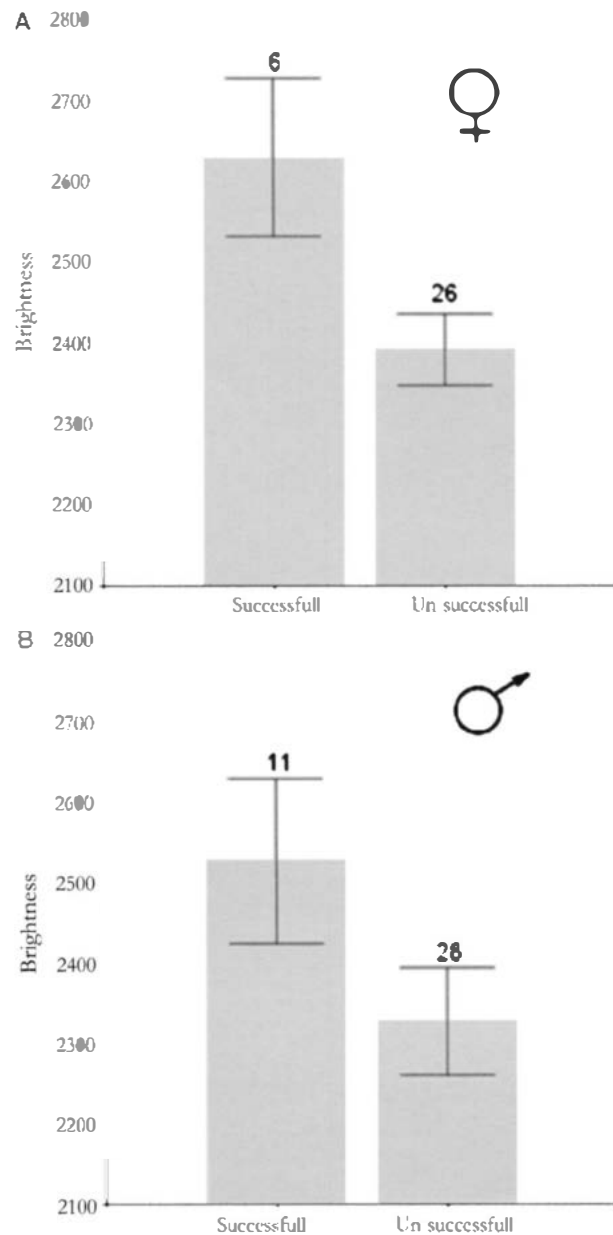


Fig. 2. Female (a) and, male (b) plumage brightness (mean  $\pm$  SE) in relation to nesting failure.

The positive relationship between structural plumage coloration of females and males and nestling survival indicates that both sexes could use the expression of this trait to identify partners that will effectively avoid depredation during the nestling development. A number of studies have previously tested for a relationship between female ornamentation and reproductive success in birds (reviewed in Roulin 2004, Amundsen and Pärn 2006). Indeed, studies with the tawny owl *Strix aluco* reported a link between female melanin-based colour plumage and reproductive success (Roulin et al. 2003), while studies in the barn swallow *Hirundo rustica* (Cuervo et al. 1996), and the bluethroat *Luscinia svecica* (Smiseth and Amundsen 2000) have failed to find such a link. Only Siefferman and Hill (2005) have previously found evidence for a positive relationship between breeding success and structural coloration in females of the eastern blue bird. Our results could be due to a higher female investment in predator avoidance when mated with a highly ornamented male (Burley 1986). However, there was no evidence of assortative mating with respect to blueness in our population (but only nine pairs). Alternatively, plumage brightness may signal capacity of nest defence against predators irrespective of sex. Finally, this result was unaffected for helper presence which has previously been demonstrated to influence breeding success in azure-winged magpies (Valencia et al. 2003).

In conclusion, we have found support for the view that structural blue coloration could be favoured by sexual selection and may have a signalling function in both sexes of the azure-winged magpie. In addition, our results suggest a link between the ability of avoiding predators and plumage coloration in this species. Experimental studies are required to understand how this blue plumage coloration is selected and maintained in the azure-winged magpies.

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