

ANIMAL BEHAVIOUR, 2006, **72**, 423–430 doi:10.1016/j.anbehav.2006.01.016

Available online at www.sciencedirect.com



# SCIENCE d DIRECT\*

# A complex plumage pattern as an honest social signal

GARY R. BORTOLOTTI\*, JULIO BLAS\*, JUAN J. NEGRO† & JOSÉ L. TELLA†

\*Department of Biology, University of Saskatchewan †Estación Biológica de Doñana, C.S.I.C. Sevilla, Spain

(Received 31 August 2005; initial acceptance 7 November 2005; final acceptance 30 January 2006; published online 23 June 2006; MS. number: A10236R)

Although birds are highly patterned animals, the efficacy and information content of complex colour signalling by combining multiple feathers has rarely been investigated. We examine a melanin-dependent plumage pattern in the red-legged partridge, *Alectoris rufa*, in a captive breeding facility in Spain. Both males and females are striking in that the majority of the entire side of the body is covered with a bold brown, black and white vertical pattern. During agonistic displays the feathers are aligned in nearly perfect, continuous dorsoventral 'flank bands'. We report on the unusual morphology and developmental complexity of the pattern, including the number and anatomical location of the display feathers. We investigate whether the area of any of the three colours of individual feathers was a signal of a bird's quality. Black alone varied significantly with age, sex and mass scaled to body size in both sexes, as well as the heterophil/lymphocyte ratio of females. Birds in poor condition appear not to be able to pigment the feather completely to its edge. The negative correlation between the black areas of the display feathers of mated pairs and the negative correlation between male ornament size and reproductive success suggest various behavioural associations with colour. Plumage patterns provide many more potential opportunities for signalling an individual's quality than overall colour alone, and they have the advantage that they can be turned off and on at the appropriate time through behavioural modification.

© 2006 The Association for the Study of Animal Behaviour. Published by Elsevier Ltd. All rights reserved.

Although there are numerous studies of the social significance of avian coloration (Hill & McGraw 2006a, b), the vast majority of these deal with variation either in hue or intensity, or in the amount of body area that is pigmented. Although the developmental complexity of patterns within individual feathers has been subject to investigation (Prum & Williamson 2002), the information content of complex colour signalling by combining multiple feathers has rarely been investigated. Possible exceptions may be the train of the peacock, Pavo cristatus, where the visual effect of multiple feathers may be a signal (Petrie et al. 1991; but see Manning & Hartley 1991), and recent studies of immaculate plumage (e.g. Ferns & Hinsley 2004). Studies of the plumage spotting of barn owls, Tyto alba, have shown the importance of linking specific attributes of individual feathers that contribute to the overall appearance of a plumage to fitness (Roulin 2004). However, birds are highly patterned organisms,

Correspondence: G. Bortolotti, Department of Biology, University of Saskatchewan, 112 Science Pl., Saskatoon, SK S7N 5E2, Canada (email: gary.bortolotti@usask.ca). Juan Negro and José Tella are at the Estación Biológica de Doñana, C.S.I.C. Avda. M<sup>a</sup> Luisa s/n, Pabellón del Perú, Apdo 1056, E-41013 Sevilla, Spain.

combining colours among feathers in distinct, speciestypical fashions that often vary with age and sex, as do most sexually selected traits. The coordination of multiple feathers to form a colour pattern presents an additional level of complexity and developmental difficulty to a signal, which in itself may advertise attributes of quality of individual birds.

Analyses of colour patterns will differ from those of hue or intensity because they are often dependent on specific behaviours to create or enhance the pattern. This coordination of display behaviour and coloration has the added advantage of directing researchers to possible functions of the pattern. General coloration could be used by any number of observers for any number of reasons, but the linking of a specific display to a plumage pattern that results from that display limits who the receiver of the signal may be and allows predictions of the function of colour (e.g. Petrie et al. 1991). Here we examine a bold melanin-dependent plumage pattern in the red-legged partridge, Alectoris rufa, and by using an increasingly reductionist approach, we question how the pattern is formed and what attributes of the colours of individual feathers may be informative as a social signal. We show that colour varies in a fashion typical of sexually selected traits, in that it is dependent on age, sex and

423

condition of the individual. Fitness consequences associated with coloration suggest a link between behaviour and plumage ornamentation.

#### **METHODS**

# Study Species and Description of the Display Plumage

Alectoris comprises seven species that are similar in appearance and range over Europe, the Middle East and Asia. These partridges are atypical galliforms in that both sexes have brightly coloured ornaments but with only a modest degree of dichromatism generally not visible to the eye (Villafuerte & Negro 1998). Most monomorphic species of the order are cryptically coloured (del Hoyo et al. 1994). The red-legged partridge has a variable mating system with most pairs being monogamous and having long-term pair bonds, but multiple mates have been recorded for both sexes (Cramp & Simmons 1980). In the nonbreeding season they form flocks of 10–40 birds (Cramp & Simmons 1980).

The red-legged partridge and other members of the genus are striking in appearance in that the majority of an entire side of the body is covered with a bold flank banding pattern. The feathers of these flank bands overlap the wing even at rest, but are especially prominent during a number of displays. This disposition of feathers high over the wing is unusual when compared to other species (G. R. Bortolotti & J. Blas, personal observation). The agonistic and sexual behaviour of Alectoris spp. have been analysed in detail (Goodwin 1953, 1954, 1958; Stokes 1961, 1963). Like many other birds with melanin ornaments (Jawor & Breitwisch 2003), partridges use their coloration in agonistic interactions (both sexes) and courtship (males). Upon meeting an opponent or potential mate, a bird immediately turns laterally. The feathers of the patch are held slightly erect in a 'vertical plane so as to exhibit them as fully as possible' (Goodwin 1953, page 589). The bands are evenly spaced and approximately equal in dorsal-ventral length, giving a squarish appearance to the patch (Fig. 1). The banding pattern is disrupted during submissive displays such as crouch and wing flap (Goodwin 1958; Stokes 1963).

Of 49 male and 36 female red-legged partridges observed in a relaxed position, 47 (96%) and 34 (94%), respectively, had eight distinct vertical black bands, while the remainder had nine, as appears to be typical for the species. The visible portion of the feathers that create the flank bands (i.e. feather tips not hidden by adjacent feathers) is made up of three distinct colours. From distal to proximal, there is a 5-mm deep patch of brown, a welldefined 2-mm-wide black bar running across the feather, and a border of white and beige about 3-5 mm wide. The remainder of each display feather, hidden as a result of overlap, is light grey and brown. Any or all of the exposed colours of the display feathers may be informative as a signal, but the black bar is particularly striking because it aligns with other bars near perfectly to form the uninterrupted dorsoventral flank bands. The entire tricoloured



**Figure 1.** Display of the red-legged partridge showing conspicuous flank bands. The body is oriented such that the side of the body is at a right angle to an opponent.

tip grows in about 14 days, and the black bar is deposited over 3-4 days starting on about day 8.

#### **Study Population and Feather Collection**

We studied partridges at the Lugar Nuevo breeding facility in southern Spain, near Andújar (province of Jaen). We refer to partridges 1 year old as yearlings, and all older birds as adults. The adults were paired and housed in separate outdoor breeding cages  $(120 \times 80 \times 50 \text{ cm})$  with an attached nestbox. These partridges lay eggs between April and June, and moult in late summer and autumn, as they do in the wild in Spain. Each year, eggs were collected every day or two until the birds naturally terminated laying. It is not known whether such methodology invalidates the interpretation of results for reproductive performance as compared to birds in the wild. Records were kept of the date of initiation of laying and total number of eggs laid.

One feather from the display patch was collected per adult from 33 breeding pairs in January 1998. These birds had been mated for at least 2 years. For consistency, we plucked the most posterior tricoloured feather. However, we subsequently determined from close inspection of morphology that four birds must have lost that specific feather, because we collected the wrong one; therefore, we excluded these four birds from the analysis. As there is little or no anterior—posterior variation in the area of black (see Results), the posteriormost feather should be representative of the display patch. Mass and tarsus length were taken to evaluate body condition using the residuals from a reduced major axis (Green 2001). We returned in April 1999 and collected feathers from 35 yearlings; however, we did not collect mass data because these birds had just been paired in breeding cages, and mass data collected during this period of stress would be unreliable.

A blood smear was taken at the same time that feathers and mass data were collected from the adults in January 1998. Smears were stained with Wright-Giemsa and examined by light microscopy using a  $40 \times$  objective. Total white blood cell count (WBC  $\times 10^9$ /litre) was estimated based upon the average number of leucocytes in five fields. The WBC count was standardized to the number of red blood cells (RBCs), and to control for variability in smear thickness, only the fields with an evenly distributed monolayer of RBCs were counted. The proportions of heterophils, lymphocytes, monocytes, eosinophils and basophils were determined from a sample of 100 leucocytes counted at the feathered edge of the smear.

In 2003 we evaluated the anatomy of the display feather patch as a whole. Feathers were counted and the display area dissected on one adult female and one adult male that had died, as well as three live yearling males. Precise counts of feathers are somewhat difficult in this species because individuals show a pronounced tendency to shed body feathers when handled. Although the sample is small, it suffices for the intended purpose of illustration. Each tricoloured feather was numbered and the exact point of insertion into the skin was marked with reference to its external anteroposterior position (i.e. band number) as well as its dorsoventral position.

In May 2003 we plucked and collected one tricoloured feather from the left display tract in a sample of 20 males. The previous skin dissections gave us a reference point to select a specific feather in the central portion of the display tract (mid-position within the fourth flank band), which was the same for all individuals. Regenerated feathers were collected in July 2003 to compare with originals. To avoid accidental feather shedding, the birds were isolated from March 2003 onwards in individual cages.

# Colour Area Measurements and Statistical Analyses

Feather lengths were measured with a ruler, and widths were measured with digital calipers. The areas of black, white and brown in the feather tips, and pennaceous area of the entire feather, were quantified digitally. Feathers were photographed with a Nikon coolpix 990 digital camera under constant conditions. Digital images were edited with Adobe Photoshop, and the portions of black, white and brown and total pennaceous area were exported as TIFF files. Area measurements were performed on processed TIFF files using the Analyze Particles command of the public domain NIH Image program (developed at the US National Institutes of Health and available on the Internet as http://rsb.info.nih.gov/nih-image/).

Statistical tests were performed using general linear models (ANOVA/ANCOVA) following a backwards, stepwise procedure. From initial saturated models that included all the independent variables and interactions, we sequentially removed nonsignificant terms until we obtained a minimum adequate model where all the retained effects were significant.

# RESULTS

## **Creation of the Visual Effect of Flank Bands**

Colour patterns across the feather coat are largely dependent on the arrangement of feathers in definite areas of growth (pterylosis). For both of the dead birds, there was a total of 49 tricoloured feathers (per side) in the display feather patch. Three other live birds had 46–49 per side. Surprizingly, the display patch was not made up of likesized feathers evenly arranged along the body's length. Instead, the entire patch was a cluster of feathers in a small area of skin on the upper breast. Even when relaxed the display patch normally covers most of the side of the body, an area of approximately  $84 \text{ cm}^2$ ; however, the feather follicles emerge from a small piece of skin (relaxed) about  $1 \times 4$  cm. The follicles were arranged in an evenly spaced, grid-like pattern across the skin surface, solely within the pectoral tract (see also Lucas & Stettenheim 1972).

Display feathers were clustered anteriorly, so we examined how the visual effect of eight vertical and evenly spaced bands along the length of the body, approximately equal in length dorsoventrally, is created. The alignment of the bands, and how these feathers are held somewhat apart from the body during display is probably accomplished by the typical fashion of muscular contraction, but perhaps coupled with a lowered wing pressing against these feathers (Fig. 1). The labelling and plucking of each feather in a display patch revealed that the number of feathers per band decreased from eight to three, and feather length progressively increased anteroposteriorly (Fig. 2a). The feathers of the posterior flank bands were able to cover the rear of the bird, yet insert anteriorly, by being over twice as long as feathers in the anterior bands. There was a significant increase from the anterior end to the posterior end in the area of white (Spearman rank correlation:  $r_s = 0.78$ , N = 45, P < 0.001), brown ( $r_s = 0.31$ , N = 45, P = 0.04) and total pennaceous area ( $r_s = 0.86$ , N = 45, P < 0.0001; Fig. 2b). However, there was no significant change in the area of the black bar ( $r_s = 0.25$ , N = 45, P = 0.10).

# Information Content of Flank Bands

It is unknown what visual component of a vertical band is informative; however, the fact that there was no increase in area of black (but there was for the other colours) across the display patch (Fig. 2b), and the perfect dorsoventral alignment of the black area to create the flank bands themselves, implies regulation of the black area. We hypothesized that, given the need for coordination of feathers of the same length (Fig. 2a), a vertical flank band may signal an individual's ability to develop feathers consistently during moult or when feathers are shed or pulled out by predators or conspecifics. To test this, we compared the lengths, widths and areas of black, brown, white and total pennaceous area in original feathers to those that were regrown



**Figure 2.** (a) Box plots showing median and quartiles for feather lengths in the display patch from the anterior end (flank band 1) to the posterior end (flank band 8). (b) Mean areas of brown, black and white portions of display feathers (same as in part a above) that are visible during display, as well as the nondisplay portion. The tip of a display feather is shown with colours that match the bars in the figure.

after experimental plucking. If replacement feathers grew in shorter or longer, a zigzag pattern would result. Changes in feather width or area of colours would also alter the visual signal. Using paired t tests we found that feather length remained unchanged ( $t_{19} = 0.82$ , P = 0.42), but feather width showed a highly significant decrease ( $t_{19} = 19.45$ , P < 0.0001). Replacement feathers were on average over 5 mm (22%) narrower, and this had important consequences because there was a significant decrease in white  $(21\%: t_{19} = 8.41, P < 0.0001)$ , brown  $(22\%: t_{19} = 5.82, P < 0.0001)$ 0.0001) and black (21%:  $t_{19} = 6.73$ , P < 0.0001). Therefore, the tips of the replacement feathers had black bars that remained vertically aligned within the flank band, but they contained less colour. Furthermore, all variables except black showed a very strong correlation before and after plucking (Pearson's correlation: length:  $r_{19} = 0.80$ , P < 0.0001; width:  $r_{19} = 0.64$ , P = 0.002; brown:  $r_{19} =$ 0.59, P = 0.006; white:  $r_{19} = 0.76$ , P < 0.0001; black:  $r_{19} =$ 0.39, P = 0.09). This result suggests that black is influenced by different factors than those of the other colours. The information content of the signal of the display may thus be associated with the coloration of individual feathers.

#### Information Content of Individual Feathers

If feather colour is a sexually selected trait, then it should vary with both age and sex (Andersson 1994; but see Jawor & Breitwisch 2003). We used an ANCOVA with sex and age as factors, and feather width at the black bar (the widest part of the feather) and total pennaceous area as covariates. The latter two variables controlled for potential confounding effects of overall size, such as those resulting from sexual size dimorphism (males are slightly larger, Cramp & Simmons 1980), and allowed us to distinguish between size (total area) and shape (width) as determinants of colour area. The brown tip was explained by both total pennaceous area and width of feather, but not sex or age (Table 1). For white, only total pennaceous area was significant (Table 1). However, males had larger black areas than females, adults had larger black areas than yearlings, and feather width was a significant covariate, but total area was not (Table 1).

The above results highlight the importance of black, as well as the width of feathers. The importance of feather width was also suggested by the plucking/regrowth experiment (above), and by the fact that feather widths were more variable than lengths (Table 2). However, as a visual signal, the entire width of the signal portion of the feather may not be important. Overlap occurs in such a fashion

**Table 1.** Results from the general linear model (ANCOVA) analysing the effect of age, sex, pennaceous feather area and feather area and feather width on the three colour portions of the display feathers (N = 93 birds)

Parameter	Estimate	SE	Test	Р				
Total brown area								
Intercept	-67.1259	22.2647	$X_1^2 = 9.09$	0.0026				
Age								
Yearling	2.6521	3.5543	F <sub>1,88</sub> =0.56	0.4556				
Adult	0.0000	0.0000						
Sex	5 0077	2 0100	г <u>1</u> 70	0 10 22				
Fomalo	-3.0977	0.0000	$F_{1,88} = 1.70$	0.1925				
	0.0000	0.0000	E	0 0005				
Feather width	3 1853	1 0195	$F_{1,88} = 12.20$	0.0005				
	5.1055	1.0175	11,88-2.70	0.0010				
Total white area	2 1 2 5 2	10 (700	v <sup>2</sup> 0.05	0.0107				
Intercept	3.1353	13.6/33	$X_1^2 = 0.05$	0.8186				
Age	1 9550	2 1 9 2 9	E _0.72	0 2052				
Adult	0.0000	0.0000	$r_{1,88} = 0.72$	0.3752				
Sex	0.0000	0.0000						
Male	-3.5198	2.4012	$F_{1,00} = 2.15$	0.1427				
Female	0.0000	0.0000	1,88 2110					
Pennaceous area	0.0459	0.0092	$F_{1,88} = 25.07$	<0.0001				
Feather width	1.0312	0.6261	$F_{1.88} = 2.71$	0.0996				
Total black area			,					
Intercent	9 8849	13 5505	$X^2 - 0.53$	0 4657				
Age	2.0042	13.3303	$x_1 = 0.55$	0.4057				
Yearling	-6.2190	2.1632	F <sub>1 00</sub> =8.27	0.0040				
Adult	0.0000	0.0000	1,88					
Sex								
Male	5.8063	2.3796	F <sub>1,88</sub> =5.95	0.0147				
Female	0.0000	0.0000						
Pennaceous area	0.0057	0.0091	$F_{1,88} = 0.40$	0.5294				
Width	1.5086	0.6205	$F_{1,88} = 5.91$	0.0150				

		Μ	Males		Females		
	Feather portion	Adult <i>N</i> =30	Yearling <i>N</i> =18	Adult <i>N</i> =28	Yearling <i>N</i> =17		
Colour portion of display							
Black	Ventral	36.9	31.5	44.9	32.0		
	Dorsal	15.1	20.4	18.1	13.8		
Brown	Ventral	42.3	27.5	48.6	34.6		
	Dorsal	36.2	22.4	30.8	27.5		
White	Ventral	22.7	16.0	11.2	17.3		
	Dorsal	19.1	19.4	7.9	14.9		
Total area*	Ventral	17.9	10.2	19.2	10.6		
	Dorsal	14.5	12.7	14.0	12.3		
Feather length		4.9	4.4	4.0	3.5		
Feather width		7.6	7.7	7.0	7.8		
Wing chord		1.9	nd	2.5	nd		
Tarsus		4.1	3.3	3.0	2.7		
Mass		7.7	9.6	6.9	9.4		

 
 Table 2. Coefficients of variation for the colour of display feathers and morphometrics of red-legged partridges

nd = no data.

\*Total area of pennaceous part of feather.

that dorsal feathers fall on top of more ventral feathers as is typical in birds. The ventral portion of each feather (i.e. using the rachis as the line of division) should be entirely visible. The feathers of the display patch are naturally asymmetrical with the ventral area being, on average, 85% of the dorsal area for males ( $\overline{X} \pm SE$  area: dorsal:  $605 \pm 12.8 \text{ mm}^2$ ; ventral:  $516 \pm 11.6 \text{ mm}^2$ ; paired t test:  $t_{47} = 9.99$ , P < 0.0001) and 89% for females (dorsal:  $524 \pm 7.6 \text{ mm}^2$ ; ventral:  $459 \pm 7.4 \text{ mm}^2$ ;  $t_{44} = 8.41$ ,  $P < 1000 \text{ mm}^2$ ;  $t_{44} = 1000 \text{ mm}^$ 0.0001). If visibility of the colour is important, then there may be a differential amount of information in dorsal and ventral portions of the feathers. To examine this we looked at the variation between sides of individual feathers. Firstly, there were highly significant correlations between the dorsal and ventral areas that were brown for both sexes (Pearson's correlation: males:  $r_{28} = 0.49$ , P = 0.01; females:  $r_{26} = 0.57$ , P = 0.002), and white for  $(r_{28} = 0.50, P = 0.005;$  females:  $r_{26} = 0.24,$ males P = 0.22), implying that, for these colours, area may be determined primarily by feather size, as was also suggested by the pattern of anterior-posterior variation (Fig. 2) and the ANCOVA of age, sex and feather size. However, there was no significant association between the two sides of the black bar for either sex (males:  $r_{28} = 0.18$ , P = 0.33; females:  $r_{26} = 0.16$ , P = 0.42). Secondly, when comparing among individuals, coefficients of variation (CV) for colour areas, relative to feather size and some physical morphometrics, showed a high degree of variation (Table 2). More interesting, the visible, ventral portion was consistently more variable than its dorsal counterpart. The disparity between the two sides of the feather was on average 2.2 times more variable for black, but only 1.3 times more variable for brown and 1.1 times more variable for white.

Close inspection of feathers suggested an explanation for why there was a difference in variability in black between the two sides of a feather, and why only the black area did not correlate between dorsal and ventral portions. Many feathers had incomplete black bars (i.e. the pigment stopped abruptly short of the edge of the feather). The incomplete portion was always on the ventral (i.e. visible) side. Males and females showed the same proportion of incomplete barring in both age classes, but there was a tendency for yearlings to have a lower percentage of complete bars (28% of 36 birds) compared to adults (47% of 58 birds) (chi-square test:  $\chi_1^2 = 3.3$ , P = 0.07). These proportions are roughly comparable to a sample of feathers from the same position on the bodies of study skins (but of birds of unknown age) in the collection of the Estación Biológica de Doñana: 29% (9 of 31) of males and 30% (8 of 27) of females had complete black bars.

### Colour and Components of Individual Quality

An ANCOVA starting with all colour variables revealed that body condition increased with black ventral area (Fig. 3a), but there were no other significant relationships for other portions of colour for males ( $F_{1,27} = 8.58$ ,



Figure 3. (a) Association between the ventral black area of the display feather and mass scaled to body size condition index. (b) Mean  $\pm$  SE of condition indexes for males and females with complete and incomplete black bars on individual feathers.

P = 0.01) or females ( $F_{1,26} = 4.87$ , P = 0.04). Birds with complete black bars were also in significantly better condition than those without complete black bars for both sexes (males:  $F_{1,27} = 4.98$ , P = 0.03; females:  $F_{1,26} = 8.61$ , P = 0.01; Fig. 3b).

The black ventral area was correlated with the heterophil/lymphocyte ratio (H:L) for females (Pearson's correlation:  $r_{23} = -0.49$ , P = 0.01) but not for males ( $r_{24} = 0.22$ , P = 0.29). The significant relationship for females remained when partial correlations controlled for potential confounding effects of body condition ( $r_{22} = -0.53$ , P = 0.01) and the ventral black bar of their mate ( $r_{21} = -0.49$ , P = 0.02; for rationale see below), but again there was no correlation for males. The main influence on H:L was a decrease in heterophils, suggesting that females with large areas of black were under less stress (Ots et al. 1998).

### **Consequences of Colour**

In the confines of captivity, the behaviour of one member of the pair may influence the well-being of its mate. Using a partial correlation controlling for feather width, we found a significant negative correlation between the ventral areas of the black bars of mated pairs that had bred together for a few years (Pearson's correlation:  $r_{21} = -0.42$ , P = 0.04). No correlation existed for yearlings that had only just been paired ( $r_{10} = -0.26$ , P = 0.41).

Using data on reproduction in 1997, we tested whether reproductive effort compromised coloration. However, neither laying date nor total number of eggs laid explained black ventral area (ANOVA: males:  $F_{2,26} = 0.91$ , P < 0.42; females:  $F_{2,24} = 0.65$ , P < 0.94). For the 1998 breeding season (i.e. after we collected feathers), the dependent variables of laying date and eggs laid were examined in relation to the black ventral areas and condition indexes (in January) of both members of the pair. Timing of laying was not significantly explained by any variable ( $F_{4,21} = 1.94$ , P = 0.14). However, the number of eggs laid varied considerably (range 11–52) and decreased with an increase in both laying date ( $F_{1,21} = 30.52$ , P < 0.0001) and black ventral area of the male ( $F_{1,21} = 6.52$ , P = 0.02).

# DISCUSSION

# **Colour of Individual Feathers**

The black area of the display feather was quite consistent over the entire patch, despite the variation in feather length (Fig. 2). Although this might imply a high degree of regulation of pigmentation, as might be expected of a trait under strong genetic control, evidence suggests otherwise. How individual feathers responded to plucking, how black area varied with such factors as age, mass scaled to size, and for females, H:L and the colour of their mate, all suggest a developmentally flexible, condition-dependent trait.

Recent studies show melanins to be associated with a variety of physiological processes that can satisfy the requirement of honest signalling (Jawor & Breitwisch 2003; McGraw 2005; Hill 2006; but see Badyaev & Hill 2000). The plumage spottiness of barn owls is a reliable signal because of a variable amount of pigment produced per spot, rather than the production of number of spots (Roulin 2004). Our results suggest that partridges in poor condition may be incapable of producing pigmented barbs completely across the feather, even though the ventral portion is smaller than the dorsal portion. This counterintuitive finding needs to be explored from a developmental perspective if one is to reveal the true cost of this signal.

More colourful birds in good condition are generally expected to have higher reproductive success; however, our results and those of recent studies on a variety of species (Lemon et al. 1992; Qvarnstrom & Forsgren 1998; Griffith et al. 1999) show that males with smaller melanin ornaments have reproductive advantages. Based on these latter findings, and those showing that melanin coloration is testosterone dependent (but see below), Jawor & Breitwisch (2003) concluded that females may prefer less dominant or less aggressive males, or those that are high-quality parents. However, in our study, pairs were not self-selected. Nevertheless, differential allocation may explain why female partridges mated to males with a large black area laid fewer eggs (Sheldon 2000). Red-legged partridges often lay two separate but concurrent clutches, so that each member of the pair can incubate and raise a brood (Cramp & Simmons 1980; Green 1984). Hens may curtail laying if they perceive their mate as being too sexual, either because there is a risk of losing him to another female, or because he may be a poor parent (i.e. because high levels of testosterone are incompatible with parental behaviour; Hegner & Wingfield 1987).

Behavioural incompatibility within a pair may be a more parsimonious explanation for our results. Only male plumage predicted reproductive performance. If males with more black are dominant, more aggressive, or more sexual, their mates may be harassed. The fact that females mated to colourful males had smaller bands and elevated H:L, suggests that mating with these males negatively impacted females. Studies of red-legged partridges (Bottoni et al. 1993) as well as other galliforms (e.g. Lupo et al. 1990) have shown that pairs allowed to select their own mates have better reproduction than those paired by humans, although why this the case is unknown. In addition, pen size for red-legged partridges influences the number of eggs laid (Gaudioso et al. 2002), perhaps, given our results, because females may or may not be able to avoid a mate that is too aggressive or too sexual. Although our results on mated pairs could thus, in part, be an artefact of housing conditions, it has a biologically meaningful parallel. Moult occurs postreproduction at a time when partridges begin to flock and form important dominance hierarchies. The subsequent agonistic interactions, with associated consequences for plumage colour, could have important consequences for winter survival and mating success in the following spring. It is plausible that this signal evolved through either intrasexual or intersexual mechanisms of sexual selection, or to function in dominance relationships outside of the breeding season, or both.

The apparent link between male melanin coloration and aggressive behaviour would appear to be a classic example of a testosterone-mediated, sexually selected trait (Jawor & Breitwisch 2003; Pérez-Rodríguez et al. 2006). However, unlike in other species, this is not the case in red-legged partridge. We experimentally manipulated testosterone levels in red-legged partridge, then plucked a display feather, and yet found no effects in the area of black (J. Blas, G. Bortolotti, L. Pérez-Rodríguez, J. Viñuela, T. Marchant, unpublished data). Furthermore, it is known that male plumage in galliforms develops in the absence of oestrogen, rather than the presence of testosterone (Owens & Short 1995). Our study of testosterone and condition in partridges provides an alternative explanation. Changes in body condition modulate testosterone levels (Pérez-Rodríguez et al. 2006), so birds in good condition that do develop more black on the feathers would also have sufficient and rogen levels to support the aggressive nature of the displays that show the signal.

# Patterns and Behaviour

Plumage patterns provide many more potential opportunities for signalling an individual's quality than colour or size of colour patches alone. In this study we have just scratched the surface of possibilities of how individual feathers and their collective visual impact could be assessed by observers. Although it is very difficult to test the visual impact of feathers, the careful alignment of individual feathers dorsoventrally to create bands of approximate equal length and width, and their even spacing along the body suggest that the ability to develop such a complex trait could be a measure of a bird's quality. Peahens may analyse an attribute of the peacock's train (Petrie et al. 1991); however, it is also plausible that females respond to train symmetry, a measure of developmental stability of this costly plumage trait (Manning & Hartley 1991). Stripes or bands are common on remiges or retrices of birds, but when lines are evident, they are usually created simply by a change in colour at the tips of feathers of the same length along a well-defined and limited feather tract (G. R. Bortolotti, personal observation). We know of no zebra-like birds (i.e. those with flank stripes) outside of a few related galliforms, and none with patterns as well developed as in Alectoris.

When cost is addressed in most studies of avian coloration, the focus has largely been on the synthesis and expression of pigments (e.g. McGraw et al. 2002, 2005; Jawor & Breitwisch 2003). There is rarely any analysis or description of the number and size of feathers, or their pterylosis. Even the fact that feathers overlap to either hide or expose colour has not been discussed. Such a shortcoming may prevent any truly comprehensive analysis of the physiological cost of colour and how such costs could be spread over the course of a moult. Although we did not quantify it thoroughly, the number of tricoloured feathers and flank bands varied little. The display patch may have been under intense directional selection to cover the entire side of the body, perhaps for maximal visual impact. We did not feel confident in counting the total number of feathers from a large sample of birds because this species readily sheds feathers when handled. Such feather loss may be an adaptation to

confuse predators (Lindstrom & Nilsson 1988). Undoubtedly, many feathers are lost during frequent agonistic encounters. Gaps created by the loss of feathers, or the smaller areas of colour in replacements as demonstrated by our plucking experiment, may provide information to conspecifics on yet another potential measure of individual quality, the rate of stressful physical encounters.

Regardless of the information contained, plumage patterns as a signal have some distinct advantages in communication. While many colourful fish, reptiles and invertebrates can change colour almost instantly, birds, with minor exceptions for facial skin, are much more limited. Feather colour is fixed in the short term, but colour patterns are not. A patterned signal, unlike overall plumage coloration, can be turned off and on through behavioural modification of feather arrangements. A conspicuous colour pattern can be hidden or revealed at the appropriate times, thus avoiding potential costs associated with a signal that is always in view (conspicuousness to predators, constant challenge to conspecifics). Many birds erect feathers during displays; however, little attention has been given to the nature of the information that may be contained in such an act. Elevated feathers could simply function to make the bird more conspicuous. Alternatively, as suggested by our results, erect feathers may make colour available for assessment by an observer. A major advantage in studying a behaviourally induced colour pattern is that the link between signal and social context is clearer, and allows for a more direct approach when formulating predictions of function.

#### Acknowledgments

We thank the staff of the partridge farm at Lugar Nuevo (Consejeria de Medio Ambiente, Junta de Andalucia) especially Paloma Prieto, for their cooperation and assistance, R. Villafuerte for help in data collection, H. Kalyn and L. Ma for analysing photographs, K. Skelton for analysing blood smears, J. Cabot of the Estación Biológica de Doñana for access to study skins, and L. Bortolotti for comments on the manuscript. We are especially grateful to F. Hiraldo for substantial logistical support. Funding was provided by the Natural Sciences and Engineering Research Council of Canada (to G.R.B.), Ministerio de Educación, Cultura y Deporte (to J.B.) and the Isabel Maria Lopez Martinez Memorial Scholarship (to J.B.).

#### References

- Andersson, M. 1994. Sexual Selection. Princeton, New Jersey: Princeton University Press.
- Badyaev, A. V. & Hill, G. E. 2000. Evolution of sexual dichromatism: contribution of carotenoid- versus melanin-based coloration. *Biological Journal of the Linnean Society*, **69**, 153–172.
- Bottoni, L., Massa, R., Lea, R. W. & Sharp, P. J. 1993. Mate choice and reproductive success in the red-legged partridge (*Alectoris rufa*). Hormones and Behavior, **27**, 308–317.
- Cramp, S. & Simmons, K. E. L. 1980. Handbook of the Birds of Europe the Middle East and North Africa. Vol. 2. Oxford: Oxford University Press.
- Ferns, P. N. & Hinsley, S. A. 2004. Immaculate tits: head pattern as an indicator of quality in birds. *Animal Behaviour*, 67, 261–272.

- Gaudioso, V. R., Alonso, M. E., Robles, R., Garrido, J. A. & Olmedo, J. A. 2002. Effects of housing type and breeding system on the reproductive capacity of the red-legged partridge (*Alectoris rufa*). *Poultry Science*, **81**, 169–172.
- Goodwin, D. 1953. Observations on voice and behaviour of the red-legged partridge *Alectoris rufa*. *Ibis*, **95**, 581–614.
- Goodwin, D. 1954. Notes on captive red-legged partridges. Avicultural Magazine, 60, 49–61.
- Goodwin, D. 1958. Further notes on pairing and submissive behaviour of the red-legged partridge *Alectoris rufa*. *Ibis*, **100**, 59–66.
- Green, A. J. 2001. Mass/length residuals: measures of body condition or generators of spurious results? *Ecology*, 82, 1473–1483.
- Green, R. E. 1984. Double nesting of the red-legged partridge Alectoris rufa. Ibis, 126, 332-346.
- Griffith, S. C., Owens, I. P. F. & Burke, T. 1999. Female choice and annual reproductive success favour less-ornamented male house sparrows. *Proceedings of the Royal Society of London, Series B*, 266, 765–770.
- Hegner, R. E. & Wingfield, J. C. 1987. Effects of experimental manipulation of testosterone levels on parental investment and breeding success in male house sparrows. Auk, 104, 462–469.
- Hill, G. E. 2006. Environmental regulation of ornamental colors. In: Bird Coloration. Vol. I. Measurements and Mechanisms (Ed. by G. E. Hill & K. J. McGraw), pp. 507–560. Cambridge, Massachusetts: Harvard University Press.
- Hill, G. E. & McGraw, K. J. (Eds) 2006a. Bird Coloration. Vol. I. Measurements and Mechanisms. Cambridge, Massachusetts: Harvard University Press.
- Hill, G. E. & McGraw, K. J. (Eds) 2006b. Bird Coloration. Vol. II. Function and Evolution. Cambridge, Massachusetts: Harvard University Press.
- del Hoyo, J., Elliott, A. & Sargatal, J. 1994. Handbook of the Birds of the World. Vol. 2. Barcelona: Lynx Edicions.
- Jawor, J. M. & Breitwisch, R. 2003. Melanin ornaments, honesty and sexual selection. Auk, 120, 249–265.
- Lemon, R. E., Weary, D. M. & Norris, K. J. 1992. Male morphology and behavior correlate with reproductive success in the American redstart (*Setophaga ruticilla*). *Behavioral Ecology and Sociobiology*, 29, 399–403.
- Lindstrom, A. & Nilsson, J. A. 1988. Birds doing it the octopus way: fright moulting and distraction of predators. *Ornis Scandinavica*, 19, 165–166.
- Lucas, A. M. & Stettenheim, P. R. 1972. Avian Anatomy. Integument Part I. Agriculture Handbook 362. Washington, D.C.: U.S. Government Printing Office.

- Lupo, C., Beani, L., Cervo, R., Lodi, L. & Dessi-Fulgheri, F. 1990. Steroid hormones and reproductive success in the grey partridge (*Perdix perdix*). *Bollettino di Zoologia*, **57**, 247–252.
- McGraw, K. J. 2005. The antioxidant function of many animal pigments: are there consistent health benefits of sexually selected colourants? *Animal Behaviour*, **69**, 757–764.
- McGraw, K. J., Hill, G. E. & Parker, R. S. 2005. The physiological costs of being colourful: nutritional control of carotenoid utilization in the American goldfinch, *Carduelis tristis. Animal Behaviour*, 69, 653–660.
- McGraw, K. J., Mackillop, E. A., Dale, J. & Hauber, M. E. 2002. Different colors reveal different information: how nutritional stress affects the expression of melanin- and structurally based ornamental plumage. *Journal of Experimental Biology*, **205**, 3747–3755.
- Manning, J. T. & Hartley, M. A. 1991. Symmetry and ornamentation are correlated in the peacock's train. *Animal Behavior*, **42**, 1020–1021.
- Ots, I., Murumagi, A. & Horak, P. 1998. Haematological health state indices of reproducing great tits: methodology and sources of natural variation. *Functional Ecology*, **12**, 700–707.
- Owens, I. P. F. & Short, R. V. 1995. Hormonal basis of sexual dimorphism in birds: implications for new theories of sexual selection. *Trends in Ecology and Evolution*, **10**, 44–47.
- Pérez-Rodríguez, L., Blas, J., Viñuela, J., Marchant, T. & Bortolotti, G. R. 2006. Condition and androgen levels: are conditiondependent and testosterone-mediated traits two sides of the same coin? *Animal Behaviour*, **72**, 97–103.
- Petrie, M., Halliday, T. & Sanders, C. 1991. Peahens prefer peacocks with elaborate trains. Animal Behaviour, 41, 323–331.
- Prum, R. O. & Williamson, S. 2002. Reaction-diffusion models of within-feather pigmentation patterning. *Proceedings of the Royal Society of London, Series B*, 269, 781–792.
- Qvarnstrom, A. & Forsgren, E. 1998. Should females prefer dominant males? *Trends in Ecology and Evolution*, **13**, 498–501.
- Roulin, A. 2004. Proximate basis of the covariation between a melanin-based female ornament and offspring quality. *Oecologia*, 140, 668–675.
- Sheldon, B. C. 2000. Differential allocation: tests, mechanisms and implications. *Trends in Ecology and Evolution*, **15**, 397–402.
- Stokes, A. W. 1961. Voice and social behavior of the chukar partridge. Condor, 63, 111-127.
- Stokes, A. W. 1963. Agonistic and sexual behaviour in the chukar partridge (Alectoris graeca). Animal Behaviour, 11, 121–134.
- Villafuerte, R. & Negro, J. J. 1998. Digital imaging for colour measurement in ecological research. *Ecology Letters*, 1, 151–154.