Coccidian oocyst parasitism in the blue-black grassquit: influence on secondary sex ornaments and body condition

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Female choice for conspicuous secondary sexual traits, which often decrease male survival, is greatly debated in the sexual selection literature. The parasite-mediated sexual selection hypotheses suggest that parasite-resistant males should show greater expression of secondary sexual traits. Males in better condition should also have more extravagant ornamentation. According to these hypotheses, females that favour more conspicuous secondary sexual traits are choosing less parasitized males and/or those in better condition and should have offspring with better chances of surviving. We assessed interactions between the expression of secondary sexual traits, parasite load and body condition in the blue-black grassquit, *Volatinia jacarina*. We trapped and banded males and sampled intestinal parasites (coccidian oocysts). We considered plumage (percentage of blue-black coverage and coloration of feathers on rump and breast), display characteristics (frequency of events and height/frequency of leaps) and body condition (body mass and size, haematocrit and total plasma protein levels). There was a negative correlation between the number of oocysts and two secondary sexual traits, blue-black coverage and leap frequency. A negative correlation was also found between oocysts and two measures of body condition, body mass and size index. We found no correlation between body condition and secondary sexual traits. Results support parasite-mediated but not condition-dependent sexual selection. There were no correlations between body condition and secondary sexual traits, so parasites may affect secondary sexual traits directly.

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During the last few decades, studies have shown that sexual selection is common in natural populations and that females favour conspicuous characters in their mating partners (Andersson 1994). Zahavi (1975, 1977) suggested that females choose males with exaggerated ornaments because these traits are costly to produce (i.e. hinder survival by increasing the risk of predation), and thereby serve as honest signals of male viability. Chosen males would thus be of better quality if they could survive despite the handicap of the exaggerated ornaments, and this viability would be a heritable characteristic passed on to the female's offspring.

One variation of the handicap principle suggests a coadaptive cycle between parasites and hosts (Hamilton & Zuk 1982). Parasites in this context would act as a selecting agent for more resistant host genotypes, and females that chose more resistant males would produce offspring with greater chances of survival. According to this hypothesis, females would choose more resistant males by using cues indicating better health and vigour, such as plumage brightness or elaborate displays. A female using these cues to select a mate could benefit by: (1) producing offspring that are resistant to parasites (Hamilton & Zuk 1982); (2) minimizing contagion by parasites (Borgia & Collis 1989); (3) associating with a healthier male that could provide better resources (Hamilton 1990). All these possibilities have been encompassed within the term parasite-mediated sexual selection (PMSS).

The PMSS hypotheses yield some logical predictions: (1) a male's ornamentation should decrease as the infestation by parasites increases; (2) female choice should favour males that are more ornamented (also true in other models for female choice); and (3) females should choose males with lower rates of parasitism.

Zahavi (1975, 1977) proposed that only males in good physical condition would be capable of having exaggerated sexual ornaments. It is conceivable that those that are more physically fit may have more developed sexual characters, independent of their level of parasite

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infestation (Folstad & Karter 1992). The relation between characteristics signalling male condition and secondary sexual characters has been the subject of several studies (reviewed by Andersson 1994). For instance, the synthesis of carotenoids depends upon carotenoid pigments in the diet, which tend to be limited in the habitat. Thus, sexual coloration produced by carotenoids may depend upon males having good physiological condition that allows them to acquire the pigments (Kodric-Brown 1985; Hill 1990, 1991; Hill & Montgomerie 1994). The supply of carotenoid pigments, experimentally manipulated in zebra finches, *Taeniopygia guttata*, provoked changes in cell-mediated immune function and sexual attractiveness (Blount et al. 2003).

The idea that structural coloration may signal the male's condition is more controversial (Smiseth et al. 2001). In contrast to plumage colours produced by chemical pigments, structural colours depend upon nanometre-scale biological structures on the surface of the feather that differentially reflect wavelengths of light (Prum et al. 2003). Structural colour can be either iridescent or noniridescent, and these may involve entirely different mechanisms of production (Prum et al. 2003). Gray (1996) suggested that structural or melanic colours were poor candidates as honest signals for male viability, because few direct metabolic costs are associated with their production. However, structural colours may reveal developmental stability during the growth of feathers (Fitzpatrick 1998), and evidence supports this suggestion (Keyser & Hill 1999; Doucet 2002; Shawkey et al. 2003). In certain birds, such as blue tits, *Parus caeruleus*, females are sensitive to plumage ultraviolet (UV) reflectance in males, resulting in preference for males with more brilliant crown patches (Andersson et al. 1998; Hunt et al. 1998). In a study of the blue-UV structural coloration of male eastern bluebirds, *Sialis sialis*, for instance, birds with more ultraviolet hues fledged more offspring, suggesting that melanin-based and structural-based plumages can function as honest indicators of male quality and parental care (Siefferman & Hill 2003). In the blue-black grassquit, *Volatinia jacarina*, Doucet (2002) found a positive correlation between structural coloration and body condition of males. In another study of body condition, male brown-headed cowbirds, *Molothrus ater*, that were subjected to stressful nutritional conditions grew less colourful, structurally based iridescent plumage (McGraw et al. 2002). Finally, in an experimental study of wild turkeys, *Melagris gallopavo*, males infected with coccidial oocysts showed proportionately less UV reflectance in their plumage, indicating that iridescent coloration may be a reliable indicator of male health (Hill et al. 2005).

We examined the relation between secondary sexual characters, body condition and parasites in the blue-black grassquit, a small, granivorous bird found in Mexico, Central America and most of South America (Sick 1997). During the reproductive season, males moult to produce black, glossy plumage (a result of iridescent structural colour produced in the barbules of the feathers) with white underwing patches; females and young males are inconspicuously brown. Males display by leaping from elevated perches, exposing white underwing patches and calling (Weathers 1986; Sick 1997). Males display in aggregations, similar in appearance to leks (Weathers 1986; Almeida & Macedo 2001). Paternal investment and female use of the male territory for nesting and feeding indicate that the blue-black grassquit has an essentially socially monogamous mating system rather than the polygynous system associated with lekking (Almeida & Macedo 2001).

We tested some of the predictions generated by the parasite-mediated sexual selection hypotheses and examined male condition, potentially mediated by parasites, for the blue-black grassquit under field conditions. We tested the following predictions: (1) parasite infestation and extravagance of secondary sexual characters of males are negatively correlated; (2) body condition and extravagance of secondary sexual characters of males are positively correlated; (3) parasite loads and body condition in males are negatively correlated.

METHODS

We collected the field data from October 2001 to February 2002, within the campus of Universidade de Brasília, Brasilia ($15°46'$S, $47°52'$W), Brazil, in an area of altered native vegetation (cerrado sensu stricto). The study period coincided with the rainy season, which is the reproductive period of the blue-black grassquit. We captured males with mist nets and marked them with unique colour combinations of plastic leg bands. Standard morphological measurements (weight: $N = 94$; wing length: $N = 94$; tarsus: $N = 93$; tail length: $N = 88$) were taken to determine body condition. We collected faecal, blood and feather samples and measured the size of the underwing white patch (procedures detailed below). We also collected ectoparasites (unpublished data) and endoparasites, as described below. The whole procedure took approximately 30 min per bird, after which the birds were freed at the same place where captured. All individuals flew off without apparent ill effects. Capture of birds and their manipulation was authorized by IBAMA/Brazil (DIFAS permits).

Collection of Endoparasites

To determine the presence of intestinal parasites, we confined each bird in a paper bag for 15 min between 1500 and 1800 hours, when many birds, including grassquits, excrete oocysts maximally (Brawner & Hill 1999; unpublished data), and collected faeces from the bag. Faecal samples were preserved in a 2.2% solution of potassium dichromate (McDougald & Reid 1997). In the laboratory the faecal samples were subjected to a standard flotation test (Greve 1996). With the objective of determining the number of oocysts per gram of faeces (Brawner & Hill 1999), we weighed some jars containing potassium dichromate before and after introduction of faecal samples (nearest 0.001 g). However, given the small faecal volume and their weight loss after collection, it was impossible to determine the exact weight of the collected material. Thus, we used a regression analysis, with the weight of the jars after samples were introduced and the number of oocysts, to determine whether oocyst number was

correlated with faecal volume, and the result was not statistically significant (Pearson correlation: $r_{36} = 0.02$, $P = 0.95$). Hence, we used the absolute count of oocysts per faecal sample in this study. Coccidian oocysts were counted under $400\times$ magnification. Given the difficulty of identifying the different coccidian species through their oocysts, they were considered in a single category (Buchholz 1995). Numbers were estimated by counting in groups of tens (for approximately 50 in a field), hundreds (for approximately 200 in a field) or two-hundreds (for approximately 500 in a field), depending on their concentration.

To quantify haemoparasites, we collected 50 µl of blood from the medial metatarsal vein with an EDTA-treated capillary tube. One drop of blood was smeared on a glass slide, which was air-dried and later fixed with methanol and stained with May-Grunwald Giemsa in the laboratory. We subsequently examined 200 visual fields ($1000\times$ magnified) to check for the occurrence of haemoparasites.

Secondary Sexual Characters

### White underwing patch and structural coloration

To determine the areas of the left and right white underwing patches of each captured individual, we gently flattened each wing against an acrylic plate and traced the outline of the area on an overlaid acetate sheet (Weatherhead et al. 1993). Each drawing was digitized (150 dots per inch) and the areas determined with Scion Image software (Beta 3b version: NIH image program with free distribution at http://www.scioncorp.com). The average area of the patches of both wings was used in the analyses.

We estimated the proportion of black or blue-black feathers ($\pm 5\%$) in the crown, rump, mantle and breast regions of each bird (as proposed by Keyser & Hill 1999). For statistical analyses we used the average proportion of the four areas.

To determine the structural plumage reflectance of each male, we removed approximately 15 small feathers from the central areas of the rump and breast with forceps and arranged them on adhesive tape replicating the conformation of the feathers on the bird's body. We measured the feathers' reflectance with a spectrophotometer (Ocean Optics S2000) under a pulsed xenon light source (Ocean Optics PX-2; 220–800 nm range). All spectral measures are expressed as a percentage of reflectance relative to a barium sulphate white standard, which reflects 99% of incident light. We measured the reflectance of feathers from the two body regions (rump and breast) with a fibre-optic probe. The feathers were placed under a small opening cut out from black velvet paper to standardize the reading surface. The probe was positioned over the feathers and kept at a $90°$ angle at 1 cm from the surface with a metal support that excluded all external light from the area being measured. We calculated the reflectance curves with a reading at every 0.3 nm from 320 to 700 nm. The restriction of measurements to this range is based on evidence that, below 320 nm, there is an abrupt drop in light transmission in the eyes of birds (Goldsmith 1990; Maier 1994), and that 700 nm is the upper spectral limit for vertebrates (Lythgoe 1979; Jacobs 1981).

Reflectance data were calculated by using the values for brightness, intensity, hue and contrast for the two body regions (Keyser & Hill 1999; Doucet 2002). Brightness was calculated as the sum of percentage reflectance from 320 nm to 700 nm, and is an indicator of the total light reflected from the feather surface. Colour intensity was defined as the maximum reflectance attained. Hue was estimated as the wavelength of the maximum reflectance. Contrast was calculated as the difference between maximum and minimum reflectance, and describes colour saturation.

The frequency distribution of peak wavelengths for 82 sampled birds (Fig. 1) showed a disruptive pattern, with a concentration of individuals in the blue range (320–500 nm) of the spectrum, and other individuals distributed above this range, both for rump (Fig. 1a) and breast (Fig. 1b) feathers. Thus, birds with maximum reflectance in the brown colour (above 550 nm) were excluded from
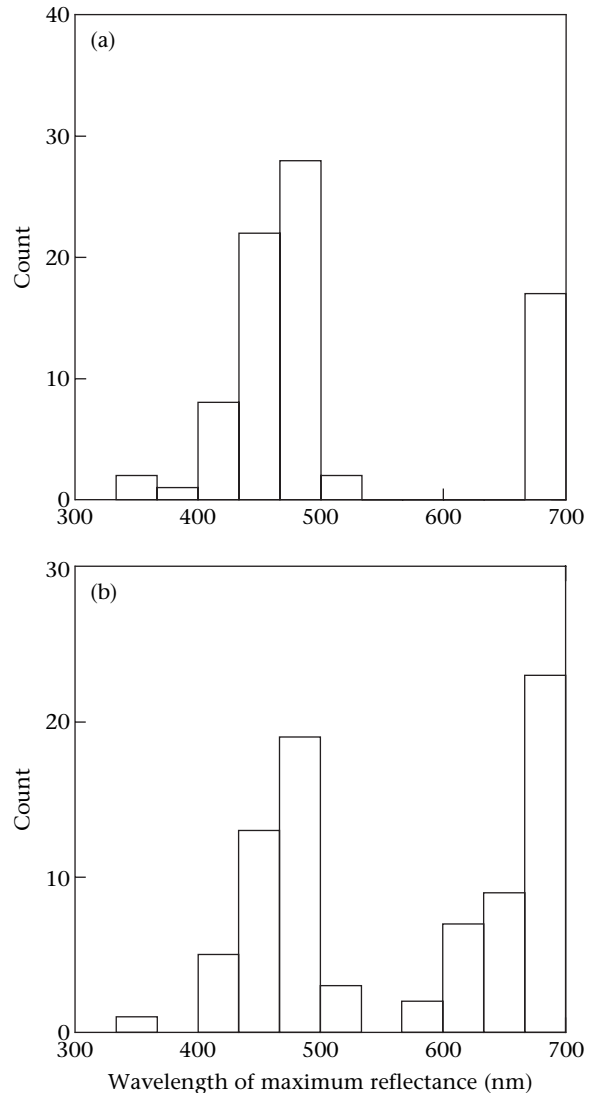
Figure 1. Distribution of peak wavelength reflected by the plumage of male blue-black grassquits on (a) the rump and (b) the breast.

analyses (41 birds were excluded for breast colour and 19 for rump colour analysis).

To evaluate variability of blue-black coloration among males, we compared the four spectral feather variables measured (brightness, intensity, hue and contrast) with nonparametric correlations. These analyses showed a significant correlation between the variables of intensity, contrast and brightness for both regions (Table 1), but no correlation between any of these variables and hue (peak wavelength). Thus, we chose to use spectral intensity as our measure of blue-black coloration for subsequent analyses. The average intensity values for the rump region were significantly higher than those for the breast region (paired t test: $t_{40} = -2.55$, $P = 0.02$).

Behaviour

The mean $\pm$ SD interval between capture and the first day of observing a bird in the field was 35.22 ± 24.56 days (range 4–113 days, $N = 27$). Observations were made between 0600 and 1100 hours Brasilia Time (BRT), which is the peak activity period for this species (Almeida & Macedo 2001), and at distances from 15 to 30 m. It was not possible to observe all birds on different days, so some analyses are based on only 1 day of observation. We observed 27 birds on 1–3 days (15 birds for 1 day; 11 birds for 2 days; one bird for 3 days). In a previous study, intensity of displays (displays per minute) did not vary with progression of the season (Almeida & Macedo 2001). Nevertheless, we verified whether our results varied seasonally by comparing the averages of display measures (frequency of displays, frequency of leaps and height of leaps) between the first and last days of observations for those males that we were able to monitor for more than 1 day ($\overline{X} \pm SD = 12.75 \pm 13.36$ days, range 1–39 days, $N = 12$ males). We found no significant differences in these measures between first and last days of observation (paired t test; frequency of leaps: $t_{11} = -0.67$, $P = 0.52$; frequency of displays: $t_{11} = -1.13$, $P = 0.28$; leap height: $t_9 = 1.12$, $P = 0.29$). Duration of observation bouts per bird averaged 2.08 min (range 0.23–9.95 min). Of those birds observed only on 1 day ($N = 15$), there were two that were observed for only a single bout, but these were unusually long bouts (4.6 min and 5.7 min). For remaining birds sampled on only 1 day but during repeated bouts, total observation times ranged from 1.67 min to 12.70 min. For each individual, the observation period ended when the bird stopped display activity or changed to a new perch, at which point a new observation period was initiated.

We recorded whether the male vocalized while executing the leap ('complete display'), or vocalized without executing the leap ('incomplete display'). We used the sum of complete and incomplete displays divided by observation time (min) to calculate the frequency of displays per minute. We used the number of complete displays divided by observation time (min) to calculate the frequency of leaps per minute. To measure the height of the leaps during complete displays, we used a horizontal and a vertical ruler fixed upon a tripod. The horizontal ruler furnished a fixed distance for the observer to rest his/ her chin to verify, using the fixed vertical ruler, the height (mm) of the leap of the displaying bird. We then measured the distance between the tripod and the bird's display perch. For calculation purposes, we considered the leap and the vertical ruler as parallel and the distance between the tripod and the perch as a fixed straight line. With these values we calculated the height of the leap by triangulation. For each observation period of a bird, we registered at least 10 height measures and used the average for statistical analyses.

Other variables may also affect courting activity, so we estimated the height of the perch (for males that were too high) or directly measured the height of each perch used by males during their display, and we also recorded the number of vocalizations that we could hear from other males in the vicinity of the focal male for 30 s following each observation period. This last measure was considered an estimate of abundance of other territorial males at the moment that the focal male was being observed.

Body Condition

Except for tarsus and wing length, which were positively correlated (Pearson correlation: $r_{85} = 0.22$, $P = 0.04$), none of the other body measurements showed significant correlations (weight versus wing length: $r_{85} = 0.14$, $P = 0.20$; weight versus tarsus: $r_{85} = 0.13$, $P = 0.25$; weight versus tail length: $r_{85} = 0.03$, $P = 0.76$; wing length versus tail length: $r_{85} = -0.09$, $P = 0.42$; tarsus versus tail length: $r_{85} = 0.004$, $P = 0.97$). Thus, to estimate body condition, we used three criteria: (1) body mass; (2) a size index, which was calculated as mass divided by tarsal length; (3) wing length, because poor body condition may lead to deterioration of size characters

Table 1. Descriptive data and Spearman rank correlation of spectral plumage characteristics for male blue-black grassquits ($N = 41$ for breast region, $N = 63$ for rump region)

	Breast region					Rump region				
	Intensity	Contrast	Peak	Brightness	Mean$\pm$SD	Intensity	Contrast	Peak	Brightness	Mean$\pm$SD
Intensity	—	0.89*	−0.19	0.97*	7.85$\pm$2.76	—	0.92*	0.16	0.88*	8.94$\pm$2.91
Contrast		—	−0.31	0.80*	3.43$\pm$2.13		—	0.02	0.88*	4.42$\pm$2.05
Peak			—	−0.16	462.85$\pm$30.96			—	0.17	458.67$\pm$28.23
Brightness				—	7285$\pm$2320				—	8034$\pm$2409

Asterisk indicates a significant correlation (*$P < 0.01$; $\alpha = 2$).

(Andersson 1994). Birds in poor body condition, for example, may develop smaller feathers (Grubb 1989).

We also used haematocrit (HT) and plasma protein levels as indicators of nutritional status. Haematocrit is the relative quantity of red blood cells in total blood volume. Nutritional deficiencies can result in anaemia due to a shortage in essential amino acids. Other deficits in vitamins and minerals, such as riboflavin, cobalamine and copper may result in reduced or ineffective haematopoiesis (Watson & Canfield 2000). A decreased level of plasma proteins may signify malnutrition, resulting usually from a reduction in the level of plasma albumin (Ots & Hõrak 1998). We centrifuged capillary tubes containing blood samples for 5 min at 1500 revolutions/min; haematocrit was expressed as a percentage of the capillary tube occupied by blood cells relative to the total blood volume (Saino et al. 1997). The plasma was deposited upon a manual refractrometer to determine the level of plasma proteins.

Statistical Analyses

We used the Kolmogorov–Smirnov bicaudal test to verify the normality of all variables. The parasite counts did not have a normal distribution, so we used non-parametric statistics (Spearman correlation) for those variables. For all tests, the level of significance was 0.05 and tests of hypotheses were one tailed.

We used ANOVA to verify differences among males in the frequency of displays per minute, frequency of leaps per minute and height of the leaps during displays. We used each individual's mean values in correlation analyses between parasites and variation in body condition.

The Spearman correlation analyses using counts of oocysts and plumage or behavioural characters were based upon the raw data. However, to improve graphic visualization, we grouped counts of oocysts into categories, as detailed in the figure legends.

RESULTS

General Results

We captured 94 male grassquits and collected behavioural observations on 27 of these. Analyses use the maximum number of individuals for each pair of variables being analysed (Table 2). We identified four types of parasites (coccidian oocysts, lice, mites and diptera) in addition to an unidentified type designated as morphotype 5 (MT5). All of the birds examined were infested with coccidian oocysts. Infestation by lice (suborder Ischnocera), mites, diptera and MT5 was infrequent and not severe either in prevalence or intensity. Thus, in the present study, we considered only the associations between oocysts and secondary sexual and body condition variables.

The frequency of displays per minute, as well as the frequency of leaps per minute, differed significantly among individuals (ANOVA: $F_{26,82} = 3.28$, $P < 0.001$ and $F_{26,82} = 7.08$, $P < 0.001$, respectively). The height of leaps also differed among individuals ($F_{24,57} = 8.55$,

Table 2. Body condition and secondary sexual characters of blue-black grassquits

Characters	N	Mean ± SD
Body condition		
Weight (g)	94	10.1 ± 0.7
Tarsus (mm)	93	15.23 ± 0.58
Tail (mm)	88	46.90 ± 2.30
Bill length (mm)	92	9.79 ± 0.49
Left wing length (mm)	94	5.88 ± 1.7
Haematocrit level (%)	70	48 ± 4
Proteins (g/dl)	52	4.4 ± 0.8
Secondary sexual characters		
White underwing patch area (cm^2)	94	5.8 ± 1.7
Blue-black plumage coverage (%)	84	74 ± 25
Frequency of displays/min	27	14.0 ± 2.6
Frequency of leaps/min	27	9.3 ± 5.1
Height of leap (cm)	25	25 ± 9

$P < 0.001$). The abundance of other males during focal male displays was negatively correlated with leap height (Spearman correlation: $r_S = -0.25$, $N = 82$, $P < 0.05$) and positively correlated with leap frequency ($r_S = 0.45$, $N = 109$, $P < 0.001$). Thus, focal males in areas of high male density leaped more often but less high.

Relation between Parasites and Secondary Sexual Characters

White underwing patch and structural coloration

For this part of the study we conducted four correlational analyses, where we considered the associations between oocyst count and the following measures: area of white underwing patch, percentage of blue-black feathers, intensity of breast structural colour and intensity of rump structural colour. There was no correlation between the area of the white underwing patch and oocysts (Spearman correlation: $r_S = -0.15$, $N = 85$, $P = 0.08$). Oocyst count was negatively correlated with the percentage of blue-black feathers ($r_S = -0.26$, $N = 83$, $P < 0.01$; Fig. 2). There was a borderline significant negative association between intensity of reflectance and oocysts for the breast region ($r_S = -0.26$, $N = 38$, $P = 0.05$). This tendency toward a negative association, however, was not found for the rump region ($r_S = 0.17$, $N = 60$, NS).

Behaviour

Three correlational analyses between oocyst count and behavioural characteristics were conducted. We found no correlation between oocyst count and the frequency of displays ($r_S = -0.16$, $N = 24$, $P = 0.23$) or between oocyst count and leap height ($r_S = -0.06$, $N = 22$, $P = 0.39$). However, oocyst count was negatively correlated with the frequency of leaps ($r_S = -0.45$, $N = 24$, $P = 0.02$; Fig. 3).
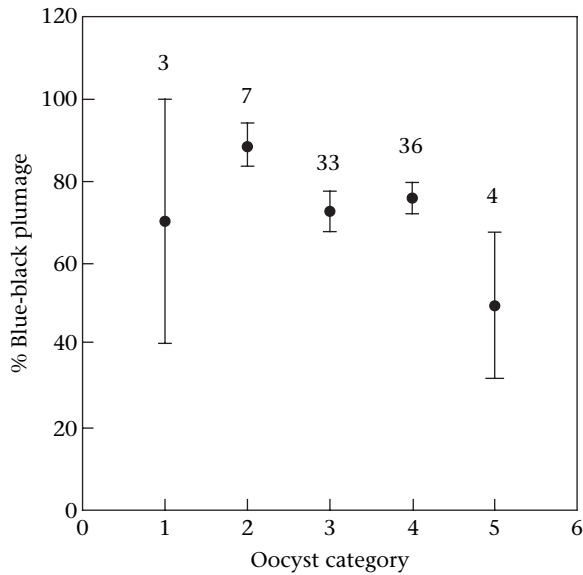
Figure 2. Relation between mean ± SE oocyst count and percentage of blue-black feathers for male blue-black grassquits. Oocyst category: 1 = 0–10 oocysts; 2 = 11–100 oocysts; 3 = 101–1000 oocysts; 4 = 1001–10 000 oocysts; 5 = >10 000 oocysts. Sample sizes are shown above data points.

Relation Between Body Condition and Secondary Sexual Characters

Of 35 Spearman correlational analyses used to examine possible positive associations between body condition and secondary sexual characteristics, none were significant. The area of white underwing patch, colour intensity and the percentage of blue-black feathers were not correlated with any of the body condition measures used (mass, size index, wing length, haematocrit, plasma protein levels;
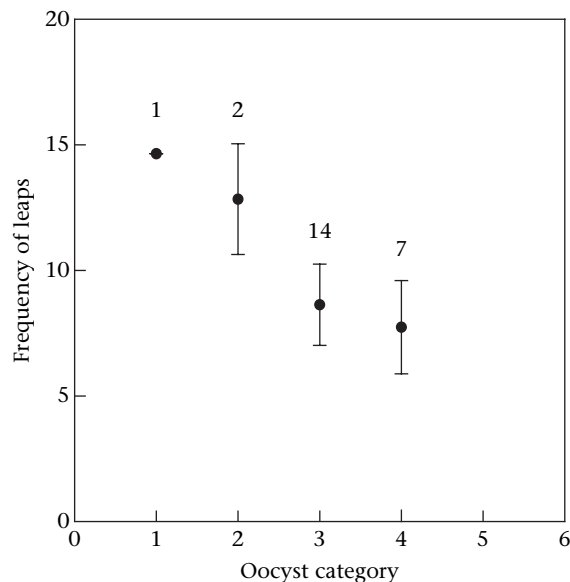
Figure 3. Relation between mean ± SE oocyst count and leap frequency for male blue-black grassquits. Oocyst categories as given in Fig. 2 legend. Sample sizes are shown above data points.

Appendix, Table A1). Furthermore, neither the frequency of displays and leaps nor the height of leaps was correlated with these measures of body condition (Appendix, Table A2).

Relation Between Parasites and Body Condition

We found negative correlations between oocyst count and body mass ($r_S = -0.19$, $N = 85$, $P = 0.04$) as well as size index ($r_S = -0.22$, $N = 85$, $P = 0.02$; Fig. 4). The other body condition variables (wing length: $r_S = 0.08$, $N = 84$, $P > 0.50$; haematocrit: $r_S = -0.18$, $N = 69$, $P > 0.50$; plasma protein levels: $r_S = 0.05$, $N = 51$, $P > 0.50$) were not associated with oocyst count.

DISCUSSION

In this field study, we investigated the three-way relation that may occur between parasite infestation, the expression of secondary sexual characters and body condition in the blue-black grassquit. Considering first the possible relation between parasites and secondary sexual
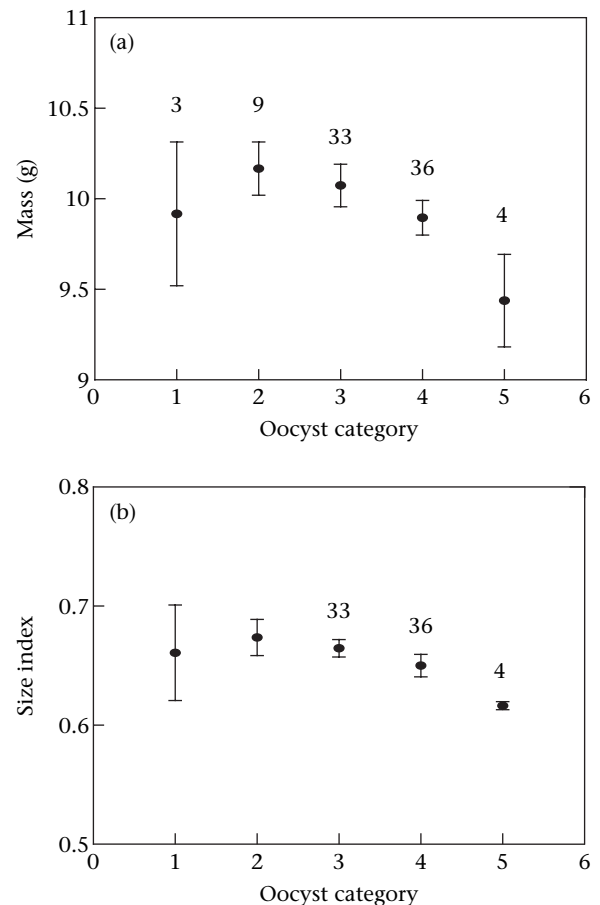
Figure 4. Relation between mean ± SE oocyst count and (a) body mass, and (b) size index for male blue-black grassquits. Oocyst categories as given in Fig. 2 legend. Sample sizes are shown above data points.

characters, the results of our field study corroborate the hypothesis that secondary sexual characters signal male quality (Zahavi 1975, 1977; Kodric-Brown & Brown 1984), insofar as we consider a heavy parasite load to be an indicator of inferior quality. One well designed laboratory experiment that involved greenfinches, *Carduelis chloris*, infected with coccidian parasites, for instance, clearly demonstrated the negative effects upon both the physiology as well as expression of carotenoid-based plumage coloration (Hõrak et al. 2004). In blue-black grassquits, we found an inverse relation between coccidian oocyst count and some of the secondary sexual characters, such as percentage of blue-black plumage coverage, frequency of leaps and reflectance of feathers in the breast region. We found no relation between the size of the white underwing patch, shown during displays, and parasite counts.

Although males more heavily infested with parasites leapt less frequently, the height of the leaps was unrelated to parasite count and may result from other determining factors. For instance, the fact that the vocalization is uttered at the height of the leap may result from its broadcasting properties: the higher the leap, the further away the vocalization can be heard (Wilczynski et al. 1989). Thus, perch height may be of great importance, and a higher perch should make it less necessary for the male to invest energy into increasing the height of the leap. Another study of blue-black grassquits that also measured leap and perch heights showed a negative correlation between these variables (Carvalho 2002). The abundance of males in an area may also influence the display characteristics. We found an inverse relation between leap height and the abundance of other males in the vicinity of focal males, and a positive relation between male abundance and leap frequency. Thus, it appears that males can regulate the height of the leaps relative to leap frequency. A male in an area with a higher density of males leaps more often but saves energy by decreasing the height of the leaps. The frequency of the leaps may be important not only in interactions between the sexes but also between males.

The variation in structural coloration found among males was extensive, and endorses the idea that the dimorphic structural coloration of birds, similarly to the carotenoid-produced colours, is a sexually selected characteristic that indicates male quality (Keyser & Hill 1999; Doucet 2002). Structural colour may be an important attribute used by females and other males, but they may use it in combination with other traits, including behavioural ones (Keyser & Hill 1999). This hypothesis is consistent with our finding that, in addition to the negative relation between breast structural colour and oocyst count, birds with fewer parasites leapt more frequently.

Secondly, in our study, we considered the possible relation between body condition and expression of secondary sexual characters. As an honest advertisement of male quality, secondary sexual characters should involve production and maintenance costs (Kodric-Brown & Brown 1984). Only individuals in peak condition should be able to invest resources in the production of these characters. Such a positive association has been both supported as well as refuted in different studies, depending upon the characters and species investigated. For instance, body condition and secondary sexual characters are significantly positively associated in passerines (Møller 1991; Harper 1999). Similarly, Doucet (2002) found a significantly positive correlation between male body condition and plumage colour in the blue-black grassquit. Other studies, however, have shown a significant, although weak, association between structural coloration and body condition (e.g. blue grosbeak, *Guiraca caerulea*: Keyser & Hill 1999).

Contrary to our expectations, we found no correlation between secondary sexual characters and body condition. Doucet (2002), studying this same species, and Keyser & Hill (1999), studying the blue grosbeak, found a direct relation between structural plumage coloration, a secondary sexual character, and feather growth rate, which may function as an indicator of nutritional condition at time of moult. In our study, it is possible that such an association (body condition versus plumage characteristics) was not found because feathers grown in the past do not correlate with measures of current body condition. Despite the variation in leap height shown by different males, this variable also was unrelated to body condition.

Another possible explanation for the lack of correlation between secondary sexual characters and body condition is that 'condition', as measured by some fat:metric ratio or simply mass, is not a good indicator of a bird's health or well-being, because these measures may change more rapidly than sexually selected characteristics or parasite loads. This explanation may be especially true for species where the optimum amount of fat is on the low side, because excessive weight could interfere with the performance of acrobatic displays (McDonald 1989). Male blue-black grassquits may be under similar selection, because in our study (unpublished data), males had larger wings and tails than females, but weighed less. Indeed, flight acrobatics may partly explain the smaller size of males in several waders and raptors (reviewed in Andersson 1994).

We found no relation between body condition and the area of white underwing patch that males expose during displays. This result may mean that this characteristic, even if it evolved in response to sexual selection, may not correspond to male quality and may be explained in other ways, such as Fisher's (1930) runaway selection process. If so, the characteristic would be purely ornamental and not necessarily associated with male condition. Alternatively, the white patch may serve as a badge of individual or species-specific recognition (Andersson 1994). We suggest that a more plausible explanation is that the white wing patch functions as an amplifier (sensu Hasson 1989) of the structural colour.

The third aim of our study was to evaluate the relation between body condition and parasites. Parasites may directly affect the physical condition of hosts, decreasing their capacity to obtain or compete for resources. Coccidian parasites, for example, may be pathogenic, causing weight loss, emaciation and diarrhoea in canaries (Dorrenstein 1997) and domestic birds (Ruff & Reid 1977). For the grassquits in our study, a higher infestation by coccidian parasites was associated with a lower body mass as well as a lower body size index. Both characteristics may indicate a chronic deficiency in nutrients caused by

coccidian infection. However, the other measures of body condition used, such as haematocrit and plasma protein levels, showed no association with oocyst count.

In conclusion, our data are consistent with the PMSS prediction that a male's ornamentation should decrease as parasite infestation increases. We considered secondary sexual traits of the blue-black grassquit, encompassing both courtship displays as well as characteristics of the plumage, and found negative correlations between these variables and coccidian oocyst count. These parasites may also have a detrimental effect on the bird's health, because we found a negative correlation between intensity of infection and measures of body condition. Our results, however, do not show that birds with superior measures of body condition also have more extravagant secondary sexual characteristics. Future study of blue-black grassquits should address the question of whether females indeed choose males with more extravagant ornaments, and whether this choice is associated with direct or indirect benefits to the female resulting from lower levels of parasites in preferred males.

Acknowledgments

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Appendix

Table A1. Spearman rank correlations between body condition variables and plumage characteristics of blue-black grassquits

	White patch			Rump colour			Breast colour			% Blue-black		
	r	N	P	r	N	P	r	N	P	r	N	P
Mass	0.09	94	0.19	−0.23	63	>0.50	0.12	41	0.22	−0.14	84	>0.50
Size index	0.06	93	0.28	−0.32	63	>0.50	0.17	41	0.15	−0.17	84	>0.50
Wing length	0.03	94	0.37	0.04	63	0.36	0.20	41	0.45	0.08	84	0.24
Haematocrit	0.09	70	0.22	−0.02	50	>0.50	−0.14	31	>0.50	0.04	69	0.36
Plasma protein	−0.03	52	>0.50	−0.14	39	>0.50	0.03	27	0.43	−0.18	51	>0.50

Table A2. Spearman rank correlations between body condition variables and behavioural characteristics of blue-black grassquits

	Frequency displays			Frequency leaps			Height leaps		
	r	N	P	r	N	P	r	N	P
Mass	−0.07	27	>0.50	−0.12	27	>0.50	0.31	25	0.07
Size index	0.07	27	0.37	−0.13	27	>0.50	0.30	25	0.08
Wing length	−0.05	27	>0.50	−0.03	27	>0.50	−0.08	25	>0.50
Haematocrit	−0.12	20	>0.50	0.10	20	0.34	0.27	18	0.14
Plasma protein	0.09	15	0.38	−0.26	15	>0.50	0.02	14	0.47